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Article

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Abstract

Urease inhibitor (specifically, *N*-(*n*-butyl) thiophosphoric triamide, NBPT) and nitrification inhibitors (NIs) have been used to minimize nitrogen (N) loss from urea. However, their effects on improving crop N use efficiency (NUE) are usually inconsistent. A 2-year study was conducted to determine the best combination of NBPT and different NIs on urea that will maximize NUE while reducing nitrate leaching. Treatments consisted of untreated urea, NBPT-treated urea, and six types of (NBPT + NI)-treated urea that were surface applied at 80 kg N ha^{-1} on plots seeded to canola (2019) and wheat (2020) at Carman and Portage in Manitoba, Canada. Plots at Carman had lysimeters installed to measure leached water and nitrate. The sites had at least 35% lesser rainfall than climate normal during each growing season. At each site, average grain yields, N removal, and residual nitrate were not significantly different between untreated urea and inhibitor-treated urea. Over the 2 years, there was no significant benefit of NBPT or NBPT + NI on crop NUE at each site. Cumulative leached nitrate (19–40 kg N ha^{-1}) did not differ significantly among urea treated with and without inhibitors. This is because >50% of the precipitation occurred when the effectiveness of NI had elapsed. Although NBPT and NI are known to reduce N losses to the atmosphere, this study suggests that the agronomic benefit and nitrate leaching prevention by NI applied in the spring may be limited in regions where large precipitation occurs later in the growing season or during non-growing season.

Key words: NBPT, nitrification inhibitor, nitrate leaching, nitrogen use efficiency

Résumé

L'inhibiteur de l'uréase (plus précisément le *N*-triamide de l'acide thiophosphorique triamide, ou NBPT) et les inhibiteurs de la nitrification (IN) servent à réduire les pertes de N venant de l'urée. Malheureusement, ces composés n'accroissent souvent l'efficacité de l'utilisation de l'azote (EUA) que de manière variable. Les auteurs ont entrepris une étude de deux ans pour déterminer la combinaison NBPT+IN qui optimisera le mieux l'EUA tout en réduisant la lixiviation des nitrates. Les traitements étaient les suivants : application en surface d'urée non traitée, d'urée conditionnée avec du NBPT et d'urée traitée avec six mélanges de NBPT+IN à raison de 80 kg de N par hectare sur des parcelles ensemencées avec du canola (2019) ou du blé (2020), à Carman et à Portage, au Manitoba (Canada). Un lysimètre avait été installé sur les parcelles de Carman pour mesurer les pertes d'eau et de nitrates. Les sites ont enregistré des précipitations inférieures d'au moins 35 % à la normale pendant chaque période végétative. Le rendement grainier moyen, la quantité de N utilisée et les résidus de nitrates ne différaient pas significativement entre l'urée non traitée et l'urée conditionnée avec un inhibiteur, aux deux sites. Au cours des deux années de l'étude, les auteurs n'ont relevé aucun avantage marquant pour le NBPT ou le mélange NBPT+IN sur le plan de l'UEA, aux deux endroits. Le volume cumulatif de nitrate perdu par lixiviation (de 19 à 40 kg par hectare) n'a pas varié de façon sensible entre l'urée avec ou sans inhibiteur aux deux sites, car plus de la moitié des précipitations sont survenues quand l'IN avait perdu son efficacité. Bien que le NBPT et les IN atténuent les dégagements de N dans l'atmosphère, l'étude laisse croire que l'application d'un IN au printemps ne présente qu'un faible avantage en ce qui concerne l'agronomie et la lutte contre la lixiviation des nitrates, dans les régions où les pluies abondantes se manifestent plus tard pendant la période végétative ou la saison morte. [Traduit par la Rédaction]

Introduction

Nitrogen (N) fertilizer, particularly urea, is used to supplement soil N to enhance crop growth. However, applied N is susceptible to losses through different pathways such as ammonia volatilization, nitrate leaching, nitrous oxide emission, and dinitrogen gas emission, thereby causing low crop N

use efficiency (NUE) of N fertilizers (Fageria and Baligar 2005). With urea fertilizer, N loss due to ammonia volatilization occurs during the hydrolysis of urea to ammonium by urease enzymes (Chien et al. 2009). The ammonium produced following hydrolysis of urea may be taken up by plants, becomes immobilized by soil microorganisms, becomes fixed on the non-exchangeable clay surface, and/or gets converted to nitrate by nitrification process. Nitrification is a microbial sequential transformation of ammonium to nitrite and then to nitrate (Subbarao et al. 2006). While nitrate may be taken up by plants, continuous accumulation of nitrate in the soil may increase its risk of leaching when its presence coincides with a high amount of rainfall or irrigation (Di and Cameron 2002). Leaching of nitrate is mainly due to its greater mobility in soil than ammonium as a result of its negative charge, which does not allow it to be held by the negatively charged soil particle (Meisinger and Delgado 2002). Other factors such as soil texture, rate of nitrification, soil porosity, cropping system, and type of N source also influence the magnitude of nitrate leaching (Meisinger and Delgado 2002; Cameron et al. 2013).

Nitrate leaching from agroecosystems is the primary cause of N contamination of water bodies (Randall and Mulla 2001). Apart from water contamination, nitrate may become denitrified to dinitrogen gas, a process where nitrous oxide may be emitted (Nikièma et al. 2016). Moreover, the nitrification process itself emits nitrous oxide to the atmosphere (Wrage et al. 2001). This is of environmental concern since nitrous oxide is a greenhouse gas whose potential global warming effect is 265 times greater than carbon dioxide (IPCC 2014; Adelekun et al. 2019).

In a bid to reduce N losses from urea fertilizers, particularly when urea is broadcasted without incorporation, coating of urea with urease inhibitor has been encouraged in line with the 4R nutrient stewardship (Johnston and Bruulsema 2014). The most widely used urease inhibitor is N-(n-butyl) thiophosphoric triamide (NBPT), which can reduce ammonia volatilization from urea by over 50% across different soil and environmental conditions (Silva et al. 2017; Lasisi et al. 2019). Also, nitrification inhibitors (NIs) have been used to inhibit the activities of ammonia-oxidizing organisms that convert ammonium to nitrite, thereby reducing N loss due to nitrate leaching and nitrous oxide emission (Wissemeier et al. 2001; Zerulla et al. 2001; Subbarao et al. 2006). NIs include dicyandiamide (DCD), 2-chloro-6 (trichloromethyl) pyridine (commonly known as nitrapyrin, NPN), 3,4-dimethyl pyrazole phosphate (DMPP), and 2-amino-4-chloro-6-methyl pyrimidine (AM).

The use of NBPT plus NI (double inhibitor) on urea has the potential of simultaneously reducing ammonia volatilization, nitrous oxide emission, dinitrogen gas emission, and nitrate leaching. While the reduction of nitrous oxide emission from urea was greater with double inhibitors than with NBPT only (Harty et al. 2016), the reduction of ammonia volatilization from urea was greater with NBPT only than with double inhibitors (Soares et al. 2012; Lasisi et al. 2020a). Despite the reduction of N losses from urea by NBPT and NIs, the conserved N has not consistently increased crop yield and/or NUE particularly in small grains with relatively low N requirement (McKenzie et al. 2010; Grant 2014; Mohammed et al. 2016; Lasisi et al. 2020b; Tao et al. 2020; Thilakarathna et al. 2021). Most of the yield responses to NBPT and NIs have been in crops such as corn that require large amounts of N (Drury et al. 2017; Martins et al. 2017; Liu et al. 2019). On the Canadian prairie, particularly in Manitoba, yield response to inhibitor usage is sometimes masked by high soil fertility status (Lasisi et al. 2020b). In addition, evapotranspiration exceeds precipitation during the growing season (Campbell et al. 1993; Agriculture and Agri-Food Canada 2016), which makes precipitation a limiting factor for nutrient uptake to maximize yield (Duan et al. 2004). We are not aware of any study that simultaneously compared the efficiency of different double inhibitors in improving agronomic parameters while measuring nitrate leaching with a field core lysimeter under a typical Manitoba condition. Although DCD and NPN are the most studied and commonly used NIs in crop productions, DMPP, which is less often used, has a greater stability, persistence, and inhibitory effect over a longer period than DCD and NPN (Wissemeier et al. 2001). In the case of AM, which is rarely used as a NI, the bactericidal effect on Nitrosomonas is similar to NPN but with less volatility than NPN (Subbarao et al. 2006), which may make it more effective. This 2-year (2019–2020) study was conducted to determine which of the four common NIs (DCD, NPN, DMPP, and AM) when combined with NBPT maximizes yield, N removal, and N uptake as well as reduces nitrate leaching from surface-applied urea fertilizer.

Materials and methods

Site characteristics

A 2-year study (2019 and 2020 growing seasons) was conducted at two locations (Carman and Portage la prairie) on two contrasting soils in Manitoba, Canada. The Carman location (49°29.619′N, 98°2.204′W) has sandy loam soil, while Portage la Prairie herein referred to as Portage (49°57.907′N, 98°15.922′W) has clay loam soil. The two locations were mapped as Black Chernozem on Canadian Soil Classification System (Michalyna and Smith 1972; Mills and Haluschak 1993). Details of the soil (<2 mm) characteristics at the two sites are shown in Table 1.

Experimental setup

The Carman site was a long-term plot of annual (cereal–canola rotation) and perennial (a mixture of orchard and timothy forage grasses) cropping systems (2009–2018). Each plot was 10 m × 4.5 m with a buffer of 5 m between the replicates and 2 m between the plots. Each plot had a field core lysimeter (54 cm i.d. and 106 cm deep) installed at the Southeast corner of the plot along the 10 m strip to directly measure the amount of water and nitrate that moved beyond the root zone. Attached to the bottom of the lysimeters were two drainpipes (0.64 cm i.d.); one of the pipes (water pipe) was meant to collect leached water from the lysimeter by suction and the other pipe (air pipe) was meant to equalize pressure during suction. The lysimeter in each plot had the same cropping history as the plot in which it was installed. Detailed descriptions of the plots and the lysimeter had been previously

Table 1. Physical and chemical properties of soil (0–15 cm) at Carman and Portage.

Property	Carman	Portage
Soil series	Hibsin	Neurhorst
Soil pH$_{water}$	6.5	7.96
Electrical conductivity (μS cm^{-1})	412	596
Organic matter (g kg^{-1})	17	71
Field capacity (m^3 m^{-3})	0.33	0.44
Cation exchange capacity (cmol kg^{-1})	9.8	36
Urease activity (mg NH$_4$-N kg^{-1} soil h^{-1})	24	88
Bulk density (Mg m^{-3})	1.2	1.1
Soil texture	Sandy loam	Clay loam
Sand (g kg^{-1})	799	269
Silt (g kg^{-1})	47	343
Clay (g kg^{-1})	154	388

Table 2. Types and rates of concentration of urease (NBPT) and nitrification inhibitors applied on urea.

Treatments	Label	NBPT (mg kg^{-1} urea)	Nitrification inhibitors (mg kg^{-1} urea)
Urea	UR	—	—
Urea + NBPT	UR$_{NBPT}$	600	—
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	600	8500
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	600	2100
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	600	90
Urea + NBPT + AM	UR$_{NBPT+AM}$	600	3000
Super U	Super U	600	8500[*]
Urea + ARM U advanced	UR$_{AD}$	360	90
Control (no urea)	Control	—	—

Note: DCD, NPN, DMPP, and AM are nitrification inhibitors; DCD is dicyandiamide; NPN is 2-chloro-6 (trichloromethyl) pyridine; DMPP is 3,4-dimethyl pyrazole phosphate and AM is 2-amino-4-chloro-6-methyl pyrimidine; NBPT is *N*-(*n*-butyl) thiophosphoric triamide.
[*]The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively.

reported (Nikièma et al. 2013; Karimi et al. 2017; Lasisi et al. 2017). In the fall of 2018, we collected soil samples from the 0–60 cm depth at an increment of 15 cm from the plots to determine the residual or initial nitrate, which was an average of 29 kg N ha^{-1}. Following soil sampling, the perennial forage grasses on the perennial cropping system plots were tilled and incorporated into the soil in preparation for the spring of 2019. In the spring of the first year of this study (2019), both the annual and perennial plots were tilled and seeded to canola (*Brassica napus* L.; var: L255PC) on 13 May 2019. In the second year, the plots were seeded to wheat (*Triticum aestivum* L. 'AC Brandon') on 25 May 2020. Similar to the field plots, the lysimeters were also tilled and seeded to the same crop as the field plot.

The Portage site had canola–wheat–soybean rotation and was seeded to soybean in 2018. In the spring of 2019, we collected soil samples from the 0–60 cm depth at an increment of 15 cm at Portage to determine the soil residual or initial nitrate, which was on average 94 kg N ha^{-1}. Similar to Carman, Portage was seeded to canola in the first year (14 May 2019) and wheat in the second year (22 May 2020).

Both sites were set up as a randomized complete block design with four replications. At Carman, the setup was such that two of the replicates were on the plots that were previously seeded to perennial forage grasses, while the other two replicates were on the plots that were previously seeded to annual crops.

Treatment description

There were nine treatments in all and consisted of untreated urea, urea treated with the urease inhibitor (NBPT), urea treated with six types of double inhibitors (NBPT plus NI), and a control with no urea. The double inhibitors used were: (1) NBPT + DCD, (2) NBPT + NPN, (3) NBPT + DMPP, (4) NBPT + AM, (5) Super U, and (6) ARM U Advanced. Details of the types and concentrations of the inhibitors are given in Table 2. While analytical grades of NIs were used, the source of NBPT was Agrotain Advanced formulation (30% NBPT; Koch Agronomic Services LLC, KS). Super U was a commercially prepared urea treated with NBPT and DCD. ARM U Advanced

(24% NBPT and 6% DMPP; Active AgriScience Inc., BC) was a commercial double inhibitor formulation.

Analytical grade of each NI was used to coat urea before coating with Agrotain Advanced. An appropriate amount of NI was dissolved in dimethylformamide and then sprayed on the urea. The NI coated urea was air-dried in a fume hood. This was followed by coating the NI-treated urea with Agrotain at a rate of 2 mL per kg urea. ARM U Advanced was used to coat urea at a recommended rate of 1.5 mL per kg urea (Table 2).

Treatment application and agronomic practices

Following seeding of both sites and lysimeter to canola (2019) or wheat (2020), N treatments were applied at a rate of 80 kg N ha^{-1} by a broadcast method. In addition, sulfur (20 kg S ha^{-1}) and potassium (59 kg K$_2$O ha^{-1}) were applied as sulfate of potash fertilizer. Also, 20 kg P$_2$O$_5$ ha^{-1} (13.5 kg P$_2$O$_5$ ha^{-1} with the seed and 6.5 kg P$_2$O$_5$ ha^{-1} broadcast after seeding) was applied to each plot and lysimeter. During the growing season, appropriate herbicides and insecticides were applied to the plots based on the recommendation from the Manitoba crop protection guideline.

Above-ground biomass sampling

At Carman during harvest, the above-ground biomass of canola was sampled from three adjacent rows of a length of 5 m (for a total area of 2.6 m^2) with a hand sickle. In the second year (2020), the above-ground biomass of wheat was sampled with a sickle mower (blade width of 1.12 m and cut length of 10 m) from each plot for a total area of 11.2 m^2. In each year, the total weight of the harvested above-ground biomass was measured on the field. A subsample of the biomass was placed in a bag and weighed to allow its moisture content to be determined. Similarly, the above-ground biomass of plants within each lysimeter was harvested and bagged.

Fig. 1. Growing season monthly precipitation in the 2 years of study at Carman and Portage (https://web43.gov.mb.ca/climate/SeasonalReport.aspx). [Colour online.]
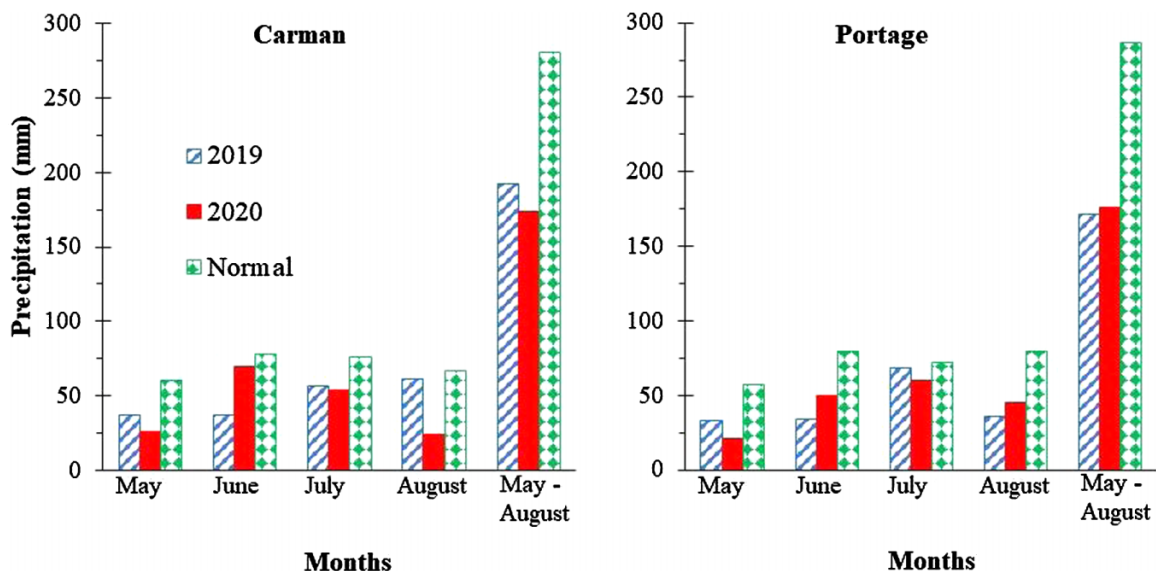
Table 3. Effect of urease (NBPT) and nitrification inhibitors on canola (2019) and wheat (2020) grain yields at Carman and Portage.

Treatments		Canola (Mg ha^{-1})		Wheat (Mg ha^{-1})	
		Carman	Portage	Carman	Portage
±N					
	Urea amended	2.13a	2.12a	3.05a	4.02a
	Control	1.07b	1.79b	1.77b	2.52b
+N					
Urea	Urea	2.31	2.22	2.94	4.15
Urea + NBPT	UR$_{NBPT}$	2.11	2.22	3.07	4.05
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	2.17	2.17	3.05	4.14
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	2.02	2.24	2.94	3.89
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	1.97	2.16	2.89	4.15
Urea + NBPT + AM	UR$_{NBPT+AM}$	2.16	1.82	3.44	3.77
Super U	Super U	2.27	2.24	2.87	4.29
Urea + ARM U advanced	UR$_{AD}$	1.98	1.91	3.19	3.70

Model	df	Probability values			
±N	1	**0.0001**	**0.0353**	**<0.0001**	**<0.0001**
+N	7	0.9343	0.2539	0.0781	0.4835

CONTRAST	Probability values			
Urea versus UR$_{IN}$	0.3868	0.4627	0.3876	0.4698
UR$_{NBPT}$ versus UR$_{DI}$	0.9450	0.3746	0.9454	0.7853

Note: ±N is a model effect between urea-amended plots versus control plot; +N is a model effect among urea-amended plots without the control plot. Probability values are significant at <0.05. Means with different letters within a column are significantly different at a probability level of <0.05. Probability values in bold are significant. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is N-(n-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. UR$_{IN}$, urea treated with inhibitors; UR$_{DI}$, urea treated with urease and nitrification inhibitors.

Table 4. Effects of urease (NBPT) and nitrification inhibitors on N removal from canola (2019) and wheat (2020) plots at Carman and Portage.

Treatments		Canola (kg N ha^{-1})		Wheat (kg N ha^{-1})	
		Carman	Portage	Carman	Portage
±N					
	Urea amended	70a	68a	75a	105a
	Control	30b	55b	35b	57b
+N					
Urea	Urea	74	68	76	106
Urea + NBPT	UR$_{NBPT}$	66	76	73	107
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	78	68	76	105
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	63	70	70	104
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	70	71	70	109
Urea + NBPT + AM	UR$_{NBPT+AM}$	71	59	83	99
Super U	Super U	76	74	71	114
Urea + ARM U advanced	UR$_{AD}$	66	61	78	95
Model	**df**	**Probability values**			
±N	1	**0.0001**	**0.0099**	**<0.0001**	**<0.0001**
+N	7	0.8877	0.1624	0.1368	0.6270
CONTRAST		**Probability values**			
Urea versus UR$_{IN}$		0.6936	0.9488	0.7617	0.8195
UR$_{NBPT}$ versus UR$_{DI}$		0.6583	0.0511	0.7607	0.6852

Note: ±N is a model effect between urea-amended plots versus control plot; +N is a model effect among urea-amended plots without the control plot. Probability values are significant at <0.05. Means with different letters within a column are significantly different at a probability level of <0.05. Probability values in bold are significant. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is N-(n-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. UR$_{IN}$, urea treated with inhibitors; UR$_{DI}$, urea treated with urease and nitrification inhibitors.

At the Portage site, canola (2019) or wheat (2020) above-ground biomass within an area of 6.72 m^2 was sampled with a sickle mower (blade length of 1.12 m) by cutting two adjacent 3 m strips. The total weight of the sampled above-ground biomass within this area was immediately determined. A subsample of this was bagged and weighed on the field.

The bagged subsamples from each site were dried in a drying room at 35 °C for at least 2 weeks. The dry weight of the biomass was determined and used to calculate the moisture content and total dry biomass on a dry weight basis. The dried biomass samples were threshed and separated into grains and straws. The grains and straws were ground, and their total N concentrations were determined using a wet oxidation method (Parkinson and Allen 1975).

The above-ground biomass, grains, and straws (all on dry weight basis) from each plot were scaled up to Mg ha^{-1}. Grain N removal (kg ha^{-1}) was calculated as the product of N concentrations in the grain and the grain yield. N uptake (kg ha^{-1}) was calculated as the sum of grain N removal and N in the straw (product of N concentrations in the straw and the straw biomass). The apparent NUE over the 2 years from each site was calculated as described by Fageria and Baligar (2005):

$$(1) \quad NUE = \frac{N_{up(N)} - N_{up(0)}}{N_{app}} \times 100$$

where $N_{up(N)}$ is the cumulative N uptake from urea-amended plot, $N_{up(0)}$ is the cumulative N uptake from control plot, and N_{app} is the cumulative N applied as urea with and without inhibitors.

Soil sampling

After harvest, we collected soil samples from both sites with a Dutch auger (4 cm in diameter) at 0–15, 15–30, 30–45, and 45–60 cm depth intervals. Six grams of fresh moist sample were extracted with 2 mol/L KCl to determine nitrate concentrations (Maynard et al. 2008). Gravimetric moisture content was also determined to correct for the moisture in the extracted soil. The nitrate concentrations were scaled up to kg ha^{-1}. The quantity of nitrate in the four depths on each plot was summed as residual nitrate.

Leachate sampling

In 2019, leachate was collected from the lysimeters at Carman on 28 May, 3 July, 31 July, and 11 September. In 2020,

Table 5. Effect of NBPT and nitrification inhibitors on canola (2019) and wheat (2020) N uptake and N use efficiency at Carman and Portage.

Treatments		Canola (kg N ha^{-1})		Wheat (kg N ha^{-1})		Nitrogen use efficiency (%)	
		Carman	Portage	Carman	Portage	Carman	Portage
±N							
	Urea amended	106a	105a	98a	129a	NA	NA
	Control	46b	84b	45b	70b	NA	NA
+N							
Urea	Urea	113	106	98	130	75	51
Urea + NBPT	UR$_{NBPT}$	100	117	94	134	64	61
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	116	105	98	129	77	50
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	98	108	91	127	61	50
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	109	108	94	132	55	54
Urea + NBPT + AM	UR$_{NBPT+AM}$	104	95	108	123	75	40
Super U	Super U	112	112	94	140	71	61
Urea + ARM U advanced	UR$_{AD}$	98	92	102	115	68	33
Model	**df**	**Probability values**					
±N	1	**0.0002**	**0.0199**	**<0.0001**	**<0.0001**	NA	NA
+N	7	0.9390	0.3831	0.1747	0.6649	0.2073	0.2871
CONTRAST		**Probability values**					
Urea versus UR$_{IN}$		0.6011	0.9104	0.8332	0.8521	0.3092	0.8496
UR$_{NBPT}$ versus UR$_{DI}$		0.6545	0.0691	0.4374	0.5189	0.5134	0.1631

Note: ±N is a model effect between urea-amended plots versus control plot; +N is a model effect among urea-amended plots without the control plot. Probability values are significant at <0.05. Means with different letters within a column are significantly different at a probability level of <0.05. Probability values in bold are significant. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is N-(n-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. UR$_{IN}$, urea treated with inhibitors; UR$_{DI}$, urea treated with urease and nitrification inhibitors; NA, not applicable.

we collected leachate on 27 May, 15 July, 9 September, and 2 November. Detailed descriptions of methods of leachate collection had been previously described in Nikièma et al. (2013). The volume of leachate during each collection time was determined and a subsample of the leachate was analyzed for nitrate concentration by a cadmium reduction method (Clesceri et al. 1998). The quantity of leached nitrate was determined by multiplying nitrate concentration (mg N L^{-1}) by the volume of leachate and expressed in kg N ha^{-1} using the area of the lysimeter. In 2019, about one half of the lysimeters did produce leachate at every sampling period and the lysimeters that had leachate had a very small volume. In 2020, all the lysimeters had an appreciable volume of leachate at each sampling period. As such, cumulative leachate was calculated by summing the leachate collected during the 2 years of study and expressed in millimeters using the area of the lysimeters. Also, cumulative leached nitrate was determined by summing the leached nitrate measured during the 2 years. Flow-weighted mean concentration of nitrate was calculated by dividing cumulative nitrate leached by cumulative leachate volume (Karimi et al. 2017).

Statistical analysis

Statistical analysis was conducted in SAS statistical package (SAS Institute 2014) by site and year in a factorial plus one control design. The factorial plus one control allowed comparison in two ways: (i) comparison between urea-amended plots and control plot and (ii) comparison among urea-amended plots without the control plot. A one-way analysis of variance (ANOVA) with PROC GLIMMIX was used to determine the effects of treatment on grain yield, N removal, N uptake, residual nitrate, cumulative leachate, and cumulative leached nitrate. Treatment was a fixed effect, while block (replicate) was a random effect. All variables were normally distributed except the residual nitrate at Portage that followed a lognormal distribution, and a lognormal distribution was specified in its model. Also, PROC GLIMMIX (beta distribution) was used to test the effect urea with and without inhibitors on NUE over the 2 years. At Carman, crop history was included as a random effect in all analyses to account for the differences in plot history. CONTRAST in PROC GLIMMIX was used to elucidate the effect of NBPT with and without NIs on variables. The ANOVA was deemed significant at a probability value of <0.05. When ANOVA was significant, treatment means comparison was performed with Fisher's least significant difference.

Results

Environmental conditions

According to Manitoba Agriculture weather data, Carman has a climate normal mean annual temperature of 2.8 °C

Table 6. Effects of urease (NBPT) and nitrification inhibitors on residual nitrate from canola (2019) and wheat (2020) plots at Carman and Portage.

Treatments		Canola (kg N ha^{-1})		Wheat (kg N ha^{-1})	
		Carman	Portage	Carman	Portage
±N					
	Urea amended	35	96	49a	55
	Control	28	94	36b	42
+N					
Urea	Urea	33	108	49b	52
Urea + NBPT	UR$_{NBPT}$	31	132	52ab	44
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	40	105	50b	50
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	28	115	41b	46
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	29	89	39b	64
Urea + NBPT + AM	UR$_{NBPT+AM}$	42	57	67a	57
Super U	Super U	37	83	41b	79
Urea + ARM U advanced	UR$_{AD}$	35	79	52ab	50
Model	**df**	**Probability values**			
±N	1	0.1115	0.6742	**0.0239**	0.1426
+N	7	0.0980	0.2546	**0.0422**	0.4261
CONTRAST		**Probability values**			
Urea versus UR$_{IN}$		0.6713	0.5421	0.9769	0.7164
UR$_{NBPT}$ versus UR$_{DI}$		0.2676	**0.0397**	0.6297	0.2944

Note: ±N is a model effect between urea-amended plots versus control plot; +N is a model effect among urea-amended plots without the control plot. Probability values are significant at <0.05. Means with different letters within a column are significantly different at a probability level of <0.05. Probability values in bold are significant. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is *N*-(*n*-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. UR$_{IN}$, urea treated with inhibitors; UR$_{DI}$, urea treated with urease and nitrification inhibitors.

and a climate normal annual precipitation of 521 mm. The climate normal mean annual temperature and annual precipitation at Portage were 2.6 °C and 513 mm, respectively. Also, Carman and Portage have a climate normal precipitation of 281 and 287 mm, respectively, during the growing season (May to August) of small grain crops (wheat and canola) (Fig. 1). Carman received 32% and 38% less precipitation than climate normal during May to August of 2019 and 2020, respectively, while Portage received 40% and 38% less precipitation than climate normal during May to August of 2019 and 2020, respectively. In 2019 at Carman, the amount of precipitation in September after our last leachate collection (on 11 September) when the crop had been harvested was 123 mm, which was 55% of the total precipitation from 1 May to 11 September.

Grain yield

In the first year and as expected, ANOVA showed the positive effect of urea amendment when compared to the control plot (with no urea added) on canola grain yield at both sites (Table 3). Average canola grain yield from urea-amended plots (with and without inhibitor) was greater than the canola grain yield from the control plot by 99% and 18% at Carman and Portage sites, respectively. In contrast, canola grain yield was not significantly different among the urea-amended plots at both sites (Table 3). At Carman, canola grain yield from urea-amended plots ranged from 1.97 to 2.31 Mg ha^{-1} (Table 3). At Portage, canola grain yield from urea-amended plots ranged from 1.82 to 2.24 Mg ha^{-1} (Table 3). Overall, CONTRAST analysis showed that there was no significant benefit of NBPT or double inhibitor on canola grain yield at each site (Table 3).

Similar to results obtained in the first year, the effect of urea application was significant on wheat grain yield at both sites in the second year (Table 3). Wheat grain yield from urea-amended plots was 72% and 60% greater than that from the control plot at Carman and Portage sites, respectively (Table 3). At each site, ANOVA did not show a significant effect of treatment on wheat grain yield among the urea-amended plots. At Carman, wheat grain yield from urea-amended plots ranged from 2.87 to 3.44 Mg ha^{-1} (Table 3). At Portage, wheat grain yield among the urea-amended plots ranged from 3.70 to 4.29 Mg ha^{-1} with no significant differences among the urea-amended plots (Table 3).

Grain nitrogen removal and uptake

There was a significant difference in canola grain N removal (N in grain) and N uptake (N in grain plus N in straw)

Table 7. Effect of urease (NBPT) and nitrification inhibitors on cumulative leached water and nitrate from 2019 to 2020 at Carman.

Treatments		Cumulative leached water (mm)	Cumulative leached nitrate (kg N ha^{-1})	Flow-weighted mean concentration (mg N L^{-1})
±N				
	Urea amended	174	30a	16a
	Control	176	11b	6b
+N				
Urea	Urea	166	20	13
Urea + NBPT	UR$_{NBPT}$	188	24	13
Urea + NBPT + DCD	UR$_{NBPT+DCD}$	168	39	21
Urea + NBPT + NPN	UR$_{NBPT+NPN}$	190	40	19
Urea + NBPT + DMPP	UR$_{NBPT+DMPP}$	166	28	16
Urea + NBPT + AM	UR$_{NBPT+AM}$	167	33	18
Super U	Super U	174	19	9
Urea + ARM U advanced	UR$_{AD}$	170	36	20

Model	df	Probability values		
±N	1	0.9355	**0.0428**	**0.0183**
+N	7	0.9676	0.5968	0.5615

CONTRAST	Probability values		
Urea versus UR$_{IN}$	0.7042	0.1123	0.1609
UR$_{NBPT}$ versus UR$_{DI}$	0.5335	0.2298	0.1524

Note: ±N is a model effect between urea-amended plots versus control plot; +N is a model effect among urea-amended plots without the control plot. Probability values are significant at <0.05. Means with different letters within a column are significantly different at a probability level of <0.05. Probability values in bold are significant. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is N-(n-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. UR$_{IN}$, urea treated with inhibitors; UR$_{DI}$, urea treated with urease and nitrification inhibitors.

between urea-amended plots versus control plots at both sites in the first year (Tables 4 and 5). Canola grain N removal was greater in urea-amended plots than control plots by 40 and 13 kg N ha^{-1} at Carman and Portage sites, respectively. Among the urea-amended plots, ANOVA did not show any significant effect of treatment on canola grain N removal at each site. Canola grain N removal from urea-amended plots ranged from 66 to 78 kg N ha^{-1} at Carman and 61–76 kg N ha^{-1} at Portage (Table 4). N uptake by canola at both sites followed a similar pattern to that of grain N removal, with N uptake among the urea-amended plots ranging from 98 to 116 kg N ha^{-1} at Carman and 92–117 kg N ha^{-1} at Portage (Table 5).

In the second year, there was also a significant difference in wheat grain N removal and N uptake between the urea-amended plots versus control plots at each site (Tables 4 and 5). Wheat grain N removal from urea-amended plots was significantly greater than wheat grain N removal from control plots by 114% and 84% at Carman and Portage sites, respectively. At Carman, wheat grain N removal among the urea-amended plots ranged from 70 to 83 kg N ha^{-1}, with ANOVA showing a lack of significant treatment effect. (Table 4). Similarly, wheat grain N removal among the urea treated with and without inhibitor plots ranged from 95 to 114 kg N ha^{-1} at Portage with no significant treatment effect (Table 4). At each site, the pattern of wheat N uptake among the urea-amended plots was similar to their wheat grain N removal (Table 5).

In each year, CONTRAST analysis did not show significant differences in N removal and N uptake for untreated urea versus urea treated with inhibitors at each site (Tables 4 and 5). Similarly, the average N removal or N uptake in urea treated with NBPT only versus urea treated with double inhibitors was not significantly different at each site.

Apparent nitrogen use efficiency

During the 2 years of study, NUE was not significantly different between the untreated urea and urea treated with inhibitors at Carman and Portage (Table 5). Similarly, the NUE from urea treated with NBPT versus urea treated with double inhibitors was not different at Carman and Portage (Table 5). At Carman, NUE ranged from 55% to 77% (Table 5). At Portage, NUE ranged from 33% to 61% (Table 5).

Residual nitrate

In the first year, ANOVA showed that there was no significant difference in residual nitrate between urea-amended plots versus control plots at each site (Table 6). Also, there was no significant effect of treatments among the urea treated with and without inhibitor plots. While residual nitrate ranged from 28 to 42 kg N ha^{-1} at Carman, it ranged from 57 to 132 kg N ha^{-1} at Portage (Table 6).

In the second year, there was a significant difference in residual nitrate between urea-amended plots and control plots at Carman but not at Portage (Table 6). At Carman, resid-

Fig. 2. Field core lysimeter leachate volume and nitrate leached during each sampling period from May 2019 to November 2020 at Carman. ∗ denotes the first sampling date in 2020. DCD, NPN, DMPP, and AM are nitrification inhibitors, while NBPT is a urease inhibitor. NBPT is *N*-(*n*-butyl) thiophosphoric triamide, DCD is dicyandiamide, NPN is 2-chloro-6 (trichloromethyl) pyridine, DMPP is 3,4-dimethyl pyrazole phosphate, and AM is 2-amino-4-chloro-6-methyl pyrimidine. The nitrification inhibitors in Super U and ARM U advanced were DCD and DMPP, respectively. [Colour online.]
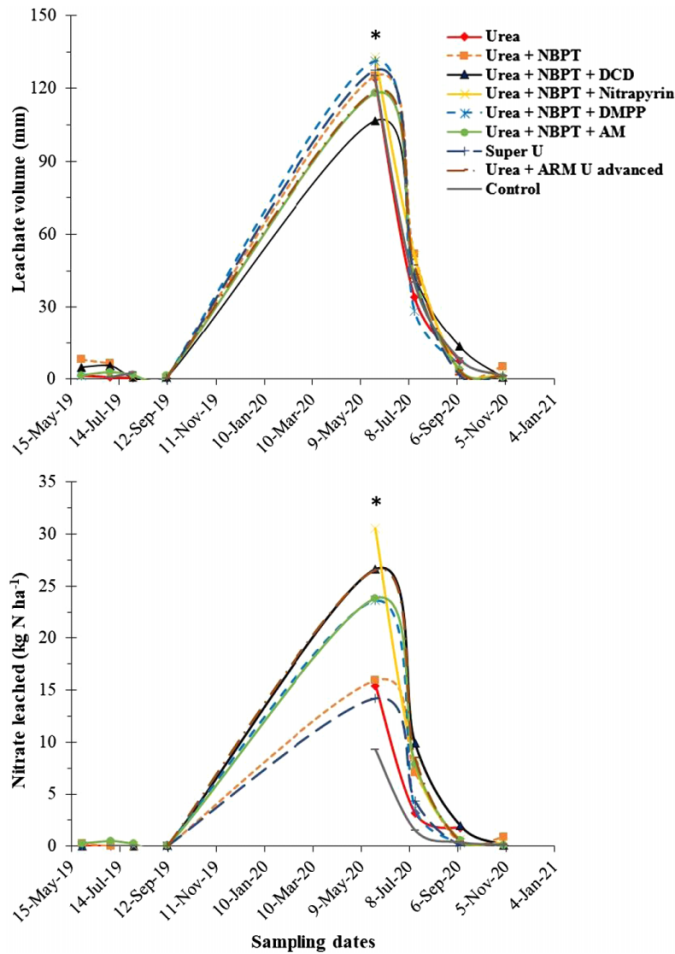
ual nitrate from urea-amended plots was 36% greater than the residual nitrate from the control plot. Also, ANOVA showed a significant effect of treatment on residual nitrate among the urea-amended plots at Carman but not at Portage. At Carman, residual nitrate from $UR_{NBPT+AM}$ plot was 34%–72% greater than the residual nitrate from all urea-amended plots except UR_{NBPT} and UR_{AD} (Table 6).

Contrast analysis showed that the average residual nitrate from urea treated with inhibitors versus untreated urea was not significantly different for each site in each year (Table 6). Also, average residual nitrate from urea treated with double inhibitor versus urea treated with NBPT only was not significantly different in each year and site, except in the first year at Portage where the average residual nitrate was lower in

urea treated with double inhibitor than urea treated with NBPT only (Table 6).

Cumulative leached water and nitrate

There was no significant effect of urea amendment on cumulative leached water at the end of the 2 years of study (Table 7). The cumulative leached water ranged from 166 to 190 mm with only 1% (on average) of the cumulative leached water collected during the 2019 growing season (Fig. 2). Also, 52%–79% of the cumulative leached water was collected during the first sampling in the spring of the second year (27 May 2020; Fig. 2). For the cumulative leached nitrate, there was a significant effect of urea application on the amount of leached nitrate. Cumulative leached nitrate from urea-amended plots was 173% greater than the cumulative leached nitrate from the control plot (Table 7). The cumulative leached nitrate from urea-amended plots ranged from 19 to 40 kg N ha^{-1}. Most of the cumulative leached nitrate occurred during fall precipitation in 2019 as shown by the amount of nitrate (38%–86% of the cumulative leached nitrate) measured during the first sampling in the spring of 2020 (Fig. 2). Although not statistically significant, cumulative leached nitrate from $UR_{NBPT+NPN}$ and $UR_{NBPT+DCD}$ was twice as much as the cumulative leached nitrate from untreated urea (Table 7). Differences in flow-weighted mean concentrations of nitrate, which ranged from 9 to 21 mg N L^{-1} among urea-amended plots were not significant (Table 7). Overall, there was no significant benefit of inhibitor on cumulative leached nitrate and flow-weighted mean concentration of nitrate.

Discussion

The lack of significant effect of inhibitors on grain yield and N removal may be because the residual N plus soil mineralization supplied sufficient N to optimize grain yield, thereby masking any benefit due to inhibitor usage. For example, despite N fertilization at Portage, the canola grain yield in some of the urea-amended plots was similar to the control plot. This was in part due to the high initial residual nitrate at Portage (94 kg N ha^{-1}). The difference in the sum of N uptake and residual nitrate (after harvest) versus the sum of initial nitrate (at the start of the study) and applied N fertilizer was an indication of high soil N mineralization, which may mask any agronomic benefit from the use of inhibitors. Earlier studies have suggested that the benefit of urea treated with inhibitors may only be observed in cases where crop N requirement is large, and the inherent soil fertility is relatively low (Abalos et al. 2014; Rose et al. 2018; Liu et al. 2019; Lasisi et al. 2020b). Also, the less than normal precipitation during the two growing seasons may explain the lack of significant effect of treatments on yield as the crops might not have attained their potential optimum yield. Studies have shown that crop yields and nutrient removal are lower than the optimum in the absence of sufficient precipitation (Duan et al. 2004; Wang et al. 2010).

High variability in the leachate data reduced our ability to detect differences in nitrate leaching. The quantity of leached nitrate (38%–86% of the cumulative leached nitrate) on the

first sampling day in 2020 was an indication that the absence of crop during the non-growing season increases the potential for leaching in the fall and spring. Since the simultaneous presence of high nitrate concentration and large precipitation events are the two dominant factors responsible for nitrate leaching (Cameron et al. 2013), the fall and spring precipitation during the non-growing season favored leaching of the accumulated nitrate. Ironically, the environmental benefit of NIs to reduce nitrate leaching may not be observed in regions where evapotranspiration is greater than precipitation during the growing season, particularly in dry years. This is because the inhibitory effect of the NIs, which lasts 4–10 weeks after application (Subbarao et al. 2006), would have elapsed by the time large precipitation events occur in the fall (non-growing season) or even later in the growing season when most inorganic N would have been in nitrate form. A similar study in Ontario that measured nitrate leaching from a corn field fertilized with untreated urea and double inhibitor-treated urea reported negligible amounts of leached nitrate during the growing season and a lack of inhibitor effect on the amount of leached nitrate during the non-growing season (Pawlick et al. 2019). The lack of significant effect of treatment on nitrate leaching implied that N was not a limiting factor for crop yield in this study even in untreated urea. This is because the applied N will still be within plant reach during the growing season in the absence of sufficient precipitation to leach nitrate, particularly in dry years. A previous work (Karimi et al. 2017) at this study site that measured the vertical distribution of nitrate in soil showed that most of the applied N was present as nitrate within 0–30 cm soil depth during the growing season. As such, the leached nitrate measured in the spring was due to nitrate movement by precipitation events during non-growing season. In any case, the concentration of nitrate leached from all the plots (except Super U and control) was greater than Health Canada maximum acceptable concentration of 10 mg N L^{-1} (Health Canada 2013). However, NIs are known to be beneficiary to fall-applied N (particularly early fall N application) as they reduce nitrate accumulation in the soil at the time of large precipitation events, and reduce the potential for nitrate leaching in the fall and early spring (Tiessen et al. 2006). Under typical Canadian prairie conditions, the reduced nitrate accumulation in the presence of NIs before the soil freezes up would make nitrate a limiting factor for denitrification process in the following spring (Lin et al. 2017). This implies that much of the fall-applied N that could be potentially lost through leaching and denitrification would be saved for crop uptake during the growing season. Results of the current study agree with findings of a previous study at the Carman location where the use of inhibitors provided yield benefit for fall-applied urea but not for spring-applied urea (Lasisi et al. 2020b). While inhibitors may not always provide yield benefit to farmers, they allow flexibility in farm operations. Also, NI has been shown to reduce nitrous oxide emission irrespective of the time and conditions during application (Drury et al. 2017; Wagner-Riddle et al. 2020), an added benefit of NI that was not investigated in this study.

Conclusion

There is an ongoing effort in using NBPT and NIs to optimize crop NUE of urea-based fertilizers. In this 2-year study, there was no significant benefit of NBPT, with and without NIs on canola and wheat grain yields, N removal, N uptake, and NUE at the two sites. Also, we did not observe any significant benefit of inhibitors on the amount of leached nitrate from the lysimeters. Neither the commonly used NIs (DCD and NPN) nor the less used NIs (DMPP and AM) were effective in reducing nitrate leaching under our field study conditions. Our ability to observe any effect of inhibitors might have been hindered by the high residual N plus high soil N mineralization coupled with the low amount of precipitation during the growing season at both sites. This study suggests that the agronomic and nitrate leaching reduction benefits of NIs applied in the spring may be limited in regions where growing season precipitation is below crop optimum need. This implies that there is a need for accessing the potential benefit of inhibitor usage on a regional level basis.

Article information

Author information

References

Abalos, D., Jeffery, S., Sanz-Cobena, A., Guardia, G., and Vallejo, A. 2014. Meta-analysis of the effect of urease and nitrification inhibitors on crop productivity and nitrogen use efficiency. Agric., Ecosyst. Environ. **189**: 136–144. doi:10.1016/j.agee.2014.03.036.

Adelekun, M., Akinremi, O., Tenuta, M., and Nikièma, P. 2019. Soil nitrous oxide emissions associated with conversion of forage grass to annual crop receiving annual application of pig manure. Can. J. Soil Sci. **14**: 1–14.

Agriculture and Agri-Food Canada. 2016. Environmental sustainability of Canadian agriculture. Agri-environmental indicator report series. Report #4. Agriculture and Agri-Food, Canada, Ottawa, ON.

Cameron, K.C., Di, H.J., and Moir, J.L. 2013. Nitrogen losses from the soil/plant system: a review. Ann. Appl. Biol. **162**: 145–173. doi:10.1111/aab.12014.

Campbell, C.A., Zentner, R.P., Selles, F., and Akinremi, O.O. 1993. Nitrate leaching as influenced by fertilization in the brown soil zone. Can. J. Soil Sci. **73**: 387–397. doi:10.4141/cjss93-041.

Chien, S.H., Prochnow, L.I., and Cantarella, H. 2009. Developments of fertilizer production and use to improve nutrient efficiency and minimize environmental impacts. Adv. Agron. **102**: 267–322. doi:10.1016/S0065-2113(09)01008-6.

Clesceri, L.S., Greenbert, A., Eaton, A.D., and Franson, M.A. 1998. Standard methods for the examination of water and wastewater. 20th ed. American Public Health Association, Washington, DC.

Di, H.J., and Cameron, K.C. 2002. The use of a nitrification inhibitor, dicyandiamide (DCD) to decrease nitrate leaching and nitrous oxide emissions in a simulated grazed and irrigated grassland.pdf. Soil Use Manage. **18**: 395–403. doi:10.1111/j.1475-2743.2002.tb00258.x.

Drury, C.F., Yang, X., Reynolds, W.D., Calder, W., Oloya, T.O., and Woodley, A.L. 2017. Combining urease and nitrification inhibitors with incorporation reduces ammonia and nitrous oxide emissions and increases corn yields. J. Environ. Qual. **46**: 939–949. doi:10.2134/jeq2017.03.0106. PMID:28991976.

Duan, Z., Xiao, H., Dong, Z., Li, X., and Wang, G. 2004. Combined effect of nitrogen-phosphorus-potassium fertilizers and water on spring wheat yield in an arid desert region. Commun. Soil Sci. Plant Anal. **35**: 161–175. doi:10.1081/CSS-120027641.

Fageria, N.K., and Baligar, V.C. 2005. Enhancing nitrogen use efficiency in crop plants. Adv. Agron. **88**: 97–185. doi:10.1016/S0065-2113(05)88004-6.

Grant, C.A. 2014. Use of NBPT and ammonium thiosulphate as urease inhibitors with varying surface placement of urea and urea ammonium nitrate in production of hard red spring wheat under reduced tillage management. Can. J. Plant Sci. **94**: 329–335. doi:10.4141/cjps2013-289.

Harty, M.A., Forrestal, P.J., Watson, C.J., McGeough, K.L., Carolan, R., Elliot, C., et al. 2016. Reducing nitrous oxide emissions by changing N fertilizer use from calcium ammonium nitrate (CAN) to urea based formulations. Sci. Total Environ. **563–564**: 576–586. doi:10.1016/j.scitotenv.2016.04.120. PMID:27155080.

Health Canada. 2013. Guidelines for Canadian drinking water quality: guideline technical document—nitrate and nitrite. Water and air quality bureau, healthy environments and consumer safety branch, Health Canada, Ottawa, Ontario (Catalogue No. H144-13/2-2013EPDF). [Online.] Available from https://www.canada.ca/en/health-canada/services/publications/healthy-living/guidelines-canadian-drinking-water-quality-guideline-technical-document-nitrate-nitrite.html. [Accessed 21 March 2021.]

IPPC. 2014. Summary for policymakers. In Climate change 2014: mitigation of climate change. Contribution of working group III to the fifth assessment report of the Intergovernmental Panel on Climate Change. Edited by O. Edenhofer, R. Pichs-Madruga, Y. Sokona, E. Farahani, S. Kadner, K. Seyboth , et al., Cambridge University Press, Cambridge and New York, NY.

Johnston, A.M., and Bruulsema, T.W. 2014. 4R nutrient stewardship for improved nutrient use efficiency. Procedia Eng. **83**: 365–370. doi:10.1016/j.proeng.2014.09.029.

Karimi, R., Akinremi, W., and Flaten, D. 2017. Cropping system and type of pig manure affect nitrate-nitrogen leaching in a sandy loam soil. J. Environ. Qual. **46**: 785–792. doi:10.2134/jeq2017.04.0158. PMID:28783787.

Lasisi, A.A., Akinremi, O.O., Tenuta, M., and Cattani, D. 2017. Short-term persistence of ammonium-N following pig manure application is greater with perennial forage grasses than annual crops. Can. J. Soil Sci. **97**: 769–782.

Lasisi, A.A., Akinremi, O.O., and Kumaragamage, D. 2019. Efficacy of a new N-(n-butyl) thiophosphoric triamide formulation in reducing ammonia volatilization from urea-based fertilizers. Can. J. Soil Sci. **99**: 395–405. doi:10.1139/cjss-2018-0072.

Lasisi, A.A., Akinremi, O.O., Zhang, Q., and Kumaragamage, D. 2020a. Efficiency of fall versus spring applied urea-based fertilizers treated with urease and nitrification inhibitors I. Ammonia volatilization and mitigation by NBPT. Soil Sci. Soc. Am. J. **84**: 949–962. doi:10.1002/saj2.20062.

Lasisi, A.A., Akinremi, O.O., and Kumaragamage, D. 2020b. Efficiency of fall versus spring applied urea-based fertilizers treated with urease and nitrification inhibitors II. Crop yield and nitrogen use efficiency. Soil Sci. Soc. Am. J. **85**: 299–313. doi:10.1002/saj2.20126.

Lin, S., Hernandez-Ramirez, G., Kryzanowski, L., Wallace, T., Grant, R., Degenhardt, R., et al. 2017. Timing of manure injection and nitrification inhibitors impacts on nitrous oxide emissions and nitrogen transformations in a barley crop. Soil Sci. Soc. Am. J. **81**: 1595–1605. doi:10.2136/sssaj2017.03.0093.

Liu, S., Wang, X., Yin, X., Savoy, H.J., McClure, A., and Essington, M.E. 2019. Ammonia volatilization loss and corn nitrogen nutrition and productivity with efficiency enhanced UAN and urea under no-tillage. Sci. Rep. **9**: 1–12. PMID:30626917.

Martins, M.R., Sant'Anna, S.A.C., Zaman, M., Santos, R.C., Monteiro, R.C., Alves, B.J.R., et al. 2017. Strategies for the use of urease and nitrification inhibitors with urea: impact on N_2O and NH_3 emissions, fertilizer ^{15}N recovery and maize yield in a tropical soil. Agric., Ecosyst. Environ. **247**: 54–62. doi:10.1016/j.agee.2017.06.021.

Maynard, D.G., Kalra, Y.P., and Crumbaugh, J.A. 2008. Nitrate and exchangeable ammonium nitrogen. In Soil sampling and methods of analysis. Edited by M.R. Carter and G. Gregorich. Taylor & Francis Group, Boca Raton, FL. pp. 71–80.

McKenzie, R.H., Middleton, A.B., Pfiffner, P.G., and Bremer, E. 2010. Evaluation of polymer-coated urea and urease inhibitor for winter wheat in southern Alberta. Agron. J. **102**: 1210–1216. doi:10.2134/agronj2009.0194.

Meisinger, J., and Delgado, J. 2002. Principles for managing nitrogen leaching. J. Soil Water Conserv. **57**: 485–498.

Michalyna, W., and Smith, R. 1972. Soils of the Portage la Prairie. Manitoba soil survey. Soils report No. 17. Manitoba Department of Agriculture, Winnipeg, MB.

Mills, G.F., and Haluschak, P. 1993. Soils of the Carman research station. Canada-Manitoba Soil Survey. Special report series 93-1. Manitoba soil survey unit, Manitoba land resource unit. Agriculture Canada and Manitoba Department of Agriculture, Winnipeg, MB.

Mohammed, Y.A., Chen, C., and Jensen, T. 2016. Urease and nitrification inhibitors impact on winter wheat fertilizer timing, yield, and protein content. Agron. J. **108**: 905–912. doi:10.2134/agronj2015.0391.

Nikièma, P., Akinremi, O.O., and Tenuta, M. 2016. Nitrous oxide emissions as affected by liquid and solid pig manures applied to annual and perennial forage crops on a sandy loam soil. Can. J. Soil Sci. **96**: 361–371. doi:10.1139/cjss-2015-0052.

Nikièma, P., Buckley, K.E., Enns, J.M., Qiang, H., and Akinremi, O.O. 2013. Effects of liquid hog manure on soil available nitrogen status, nitrogen leaching losses and wheat yield on a sandy loam soil of western Canada. Can. J. Soil Sci. **93**: 573–584. doi:10.4141/cjss2012-070.

Parkinson, J.A., and Allen, S.E. 1975. A wet oxidation procedure suitable for the determination of nitrogen and mineral nutrients in biological material. Commun. Soil Sci. Plant Anal. **6**: 1–11. doi:10.1080/00103627509366539.

Pawlick, A.A., Wagner-Riddle, C., Parkin, G.W., and Berg, A.A. 2019. Assessment of nitrification and urease inhibitors on nitrate leaching in corn (Zea mays L.). Can. J. Soil Sci. **99**: 80–91. doi:10.1139/cjss-2018-0110.

Randall, G.W., and Mulla, D.J. 2001. Nitrate nitrogen in surface waters as influenced by climatic conditions and agricultural practices. J. Environ. Qual. **30**: 337–344. doi:10.2134/jeq2001.302337x. PMID:11285893.

Rose, T.J., Wood, R.H., Rose, M.T., and Van Zwieten, L. 2018. A re-evaluation of the agronomic effectiveness of the nitrification inhibitors DCD and DMPP and the urease inhibitor NBPT. Agric., Ecosyst. Environ. **252**: 69–73. doi:10.1016/j.agee.2017.10.008.

SAS Institute. 2014. SAS/STAT 14.3 user's guide. SAS Institute, Inc., Cary, NC.

Silva, A.G.B., Sequeira, C.H., Sermarini, R.A., and Otto, R. 2017. Urease inhibitor NBPT on ammonia volatilization and crop productivity: a meta-analysis. Agron. J. **109**: 1–13. doi:10.2134/agronj2016.04.0200.

Soares, J.R., Cantarella, H., and Menegale, M.L.de C. 2012. Ammonia volatilization losses from surface-applied urea with urease and nitrifi-

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cation inhibitors. Soil Biol. Biochem. **52**: 82–89. doi:10.1016/j.soilbio.2012.04.019.

Subbarao, G.V., Ito, O., Sahrawat, K.L., Berry, W.L., Nakahara, K., Ishikawa, T., et al. 2006. Scope and strategies for regulation of nitrification in agricultural systems—challenges and opportunities. CRC Crit. Rev. Plant Sci. **25**: 303–335. doi:10.1080/07352680600794232.

Tao, R., Li, J., Hu, B., Shah, J.A., and Chu, G. 2020. A 2-year study of the impact of reduced nitrogen application combined with double inhibitors on soil nitrogen transformation and wheat productivity under drip irrigation. J. Sci. Food Agric. **101**: 1772–1781. doi:10.1002/jsfa.10791. PMID: 32892367.

Thilakarathna, S.K., Hernandez-Ramirez, G., Puurveen, D., Kryzanowski, L., Lohstraeter, G., Powers, L., et al. 2021. Nitrous oxide emissions and nitrogen use efficiency in wheat: N fertilization timing and formulation, soil N, and weather effects. Soil Sci. Soc. Am. J. **84**: 1910–1917. doi:10.1002/saj2.20145.

Tiessen, K.H.D., Flaten, D.N., Bullock, P.R., Burton, D.L., Grant, C.A., and Karamanos, R.E. 2006. Transformation of fall-banded urea: application date, landscape position, and fertilizer additive effects. Agron. J. **98**: 1460–1470. doi:10.2134/agronj2005.0304.

Wagner-Riddle, C., Baggs, E.M., Clough, T.J., Fuchs, K., and Petersen, S.O. 2020. Mitigation of nitrous oxide emissions in the context of nitrogen loss reduction from agroecosystems: managing hot spots and hot moments. Curr. Opin. Environ. Sustain. **47**: 46–53. doi:10.1016/j.cosust.2020.08.002.

Wang, Q., Li, F., Zhao, L., Zhang, E., Shi, S., Zhao, W., et al. 2010. Effects of irrigation and nitrogen application rates on nitrate-nitrogen distribution and fertilizer nitrogen loss, wheat yield and nitrogen uptake on a recently reclaimed sandy farmland. Plant Soil, **337**: 325–339. doi:10.1007/s11104-010-0530-z.

Wissemeier, A.H., Linzmeier, W., Gutser, R., Weigelt, W., and Schmidhalter, U. 2001. The new nitrification inhibitor DMPP (ENTEC®)—comparisons with DCD in model studies and field applications. *In* Development in plant soil science. Edited by W.J. Horst. Springer Publishing, Dordrecht, the Netherland. pp. 702–703.

Wrage, N., Velthof, G.L., Beusichem, M.L.V., and Oenema, O. 2001. Role of nitrifier denitrification in the production of nitrous oxide. Soil Biol. Biochem. **33**: 1723–1732. doi:10.1016/S0038-0717(01)00096-7.

Zerulla, W., Barth, T., Dressel, J., Erhardt, K., Horchler von Locquenghien, K., Pasda, G., et al. 2001. 3,4-dimethyl pyrazole phosphate (DMPP)—a new nitrification inhibitor for agriculture and horticulture. Biol. Fertil. Soils, **34**: 79–84. doi:10.1007/s003740100380.