

**Integrating enhanced efficiency fertilizers and nitrogen rates to improve Canada Western
Red Spring wheat production in the Canadian prairies**

by

Adam Corbett Fast

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Department of Agricultural, Food and Nutritional Science
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Abstract

Canada Western Red Spring (CWRS) wheat (*Triticum aestivum* L.) is the most widely grown wheat class in western Canada. This is mainly due to its excellent milling and baking quality, concomitant with a high protein concentration. Adequate nitrogen (N) supply is important to achieve optimal CWRS grain yield and quality. In CWRS production, N is routinely applied as granular urea fertilizer during planting. Consequential N loss can arise when using unprotected urea fertilizer. Enhanced efficiency fertilizers (EEFs) aim to maintain the integrity of applied N by increasing plant nutrient bioavailability while reducing environmental N loss. To determine if the use of EEFs and different N rates can improve upon conventional methods, a CWRS wheat experiment was established in 2019 across four locations in Alberta and two in Saskatchewan, Canada. This experiment consists of two factors: (i) urea type [(urea; urea + urease inhibitor (Agrotain®); urea + nitrification inhibitor (eNtrench®); urea + dual inhibitor (SuperU®); urea + dual inhibitor (NBPT/DMPSA); and slow-release fertilizer (Environmentally Smart Nitrogen® (ESN®))], and (ii) N rate [60; 120; 180; and 240kg N ha⁻¹]. Results indicate urea type affected grain yield in Dark Brown Chernozem soils but not in Black Chernozem & Dark Grey Luvisol soils. In Dark Brown Chernozem soils, a dual inhibitor (SuperU®) increased grain yield by 3.3% relative to urea, while all other EEFs attained similar results. Furthermore, slight increases in net return were observed with the use of a dual (SuperU®) and urease inhibitor (Agrotain®). Grain protein content was not influenced by urea type; however, increasing N rate in both soil groups resulted in quadratic and linear increases in grain yield and protein content, respectively. Application of N fertilizer at a rate of 120 kg N ha⁻¹ was agronomically optimal and provided greatest net return. These results suggest growers who incorporate dual inhibitors in CWRS wheat production can achieve modest increases in grain

yield; moreover, the use of other EEFs will not reduce grain yield or protein content relative to conventional urea.

Preface

This thesis is an original work by me, Adam Fast. No part of this thesis has been previously published, except as conference abstracts corresponding to presentations made during my M.Sc. program. I also referenced this work in field day, industry, and guest lecture presentations. I co-conducted the experiment described in chapter 2 with the assistance of Agriculture and Agri-Food Canada (AAFC) technical staff at the Lethbridge Research and Development Centre in Lethbridge, AB. Experimental site data collection and maintenance were led by the following individuals: Dr. Sheri Strydhorst (Barrhead & Edmonton, AB, 2019); Dr. Guillermo Hernandez Ramirez, Sarah Anderson (Edmonton, AB, 2020-2021); Greg Semach (Beaverlodge, AB); Adam Fast, Ryan Dyck, Steve Simmill, Warren Taylor (Lethbridge, AB); Laurel Thompson, J.P. Pettyjohn, Daniel Pucko (Vermilion, AB); Chris Holzapfel (Indian Head, SK); and, Jessica Enns (Scott, SK). All locations depended on assistance from student and other research staff.

I co-collected and compiled data. I conducted the literature review, statistical analysis, and wrote the first draft of chapters 1, 2, and 3. Statistical guidance was provided by Dr. Brian Beres and Dr. Dean Spaner. Significant editorial revisions were made to my original drafts by Dr. Dean Spaner, Dr. Guillermo Hernandez Ramirez, Dr. Brian Beres, and Dr. Edward Bork. I incorporated these suggestions into the thesis, prior to final submission.

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The experiment described in chapter 2 is one component of a larger study entitled “Integrating N fertilizer technologies with superior genetics to optimize protein in CWRS wheat without compromising yield, 4R principles, and environmental health” led by Dr. Brian Beres of AAFC Lethbridge (2019-2023). Once all data is collected, scientific publications will be prepared and submitted.

Dedication

I, Adam Fast, dedicate this thesis to my grandparents, Otto and Irene Fast, and to my late (to be) father-in law, Gerard Bergeron. You have taught me the value of hard work and achieving goals you set in life.

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List of Abbreviations

°C	Degrees Celsius
4R	Right Fertilizer Source, Right Rate, Right Time, Right Place
AAC	Agriculture and Agri-Food Canada
AB	Alberta
AC (used in cultivar name)	Agriculture Canada
AC	Ammonium Carbonate
AMO	Ammonia Monooxygenase
AN	Ammonium Nitrate
AS	Ammonium Sulphate
b ₀	Base of 0°C
BBCH	Biologische Bundesanstalt, Bundessortenamt und Chemical Industry
BP	Before Present
C/N ratio	Carbon/Nitrogen Ratio
CDC	Crop Development Centre
CERS	Canada Eastern Red Spring
CH ₄	Methane
cm	Centimeter
CO ₂	Carbon Dioxide
CRF	Controlled-Release Fertilizer
Cu	Copper
CV	Coefficient of Variation
CWRS	Canada Western Red Spring
DCD	Dicyandiamide
DI	Dual Inhibitor
DMP	3,4,-dimethylpyrazole
DMPP	3,4,-dimethylpyrazole phosphate
DMPSA	2-(N-3,4-dimethyl-1H-pyrazol-1-yl) succinic acid isomeric mixture
EEF	Enhanced Efficiency Fertilizer
ESN®	Environmentally Smart Nitrogen®
Fe	Iron
g	Gram
g 1000 kernels ⁻¹	Grams per 1000 kernels (of wheat)
g cm ⁻³	Grams per Cubic Centimeter
GHG	Greenhouse Gas
GWP	Global Warming Potential
ha	Hectare

HRS	Hard Red Spring
kg	Kilogram
kg CO ₂ kg ⁻¹ N	Kilogram of CO ₂ per Kilogram of N
kg ha ⁻¹	Kilograms per Hectare
kg hL ⁻¹	Kilograms per Hectolitre
kg m ⁻³	Kilograms per Cubic Meter
kg N ha ⁻¹	Kilograms of Nitrogen per Hectare
kg N ha ⁻¹ yr ⁻¹	Kilograms of Nitrogen per Hectare per Year
kg N ₂ O-N ha ⁻¹	Kilograms of Nitrous Oxide Derived Nitrogen per Hectare
kPa	Kilopascal
LSD	Least Significant Difference
m ²	Meters squared
meq 100g ⁻¹	Milliequivalents per 100 grams of soil
mg H ₂ O kg ⁻¹	Milligrams of Water per Kilogram of Soil
mm	Millimeter
Mg	Megagram
Mg ha ⁻¹	Megagram per Hectare
N	Nitrogen
N ₂	Dinitrogen
N ₂ O	Nitrous Oxide
NBPT	N-(n-butyl) thiophosphoric triamide
NBPTO	N-(n-butyl) phosphoric triamide
NH ₃	Ammonia
NH ₄ ⁺	Ammonium
NI	Nitrification Inhibitor
NO ₃ ⁻	Nitrate
NS	Not Significant
NUE	Nitrogen Use Efficiency
OMAFRA	Ontario Ministry of Agriculture, Food and Rural Affairs
P	Probability
ppb	Parts Per Billion
PCU	Polymer Coated Urea
pH	Potential of Hydrogen
PRS®	Plant Root Simulator®
r	Correlation Coefficient
SK	Saskatchewan

SNF	Stabilized Nitrogen Fertilizer
SRF	Slow-Release Fertilizer
Tg N yr-	Teragram of N per Year
UAN	Urea Ammonium Nitrate
UI	Urease Inhibitor
USA	The United States of America
USDA	United States Department of Agriculture
yr	Year
ZGS	Zadoks Growth Scale

1.0 Enhanced efficiency fertilizers: A review of nitrogen dynamics and grain quality in Canada Western Red Spring wheat production

1.1 Introduction

Reliance on sustainable crop intensification will be critical to address a rising global population and corresponding food demand. Wheat (*Triticum aestivum* L.) is one of the three most important global grain crops. Conventionally produced wheat in western Canada is reliant on adequate nitrogen (N) supply in the form of fertilizer. The most commonly used N fertilizer is urea, which is typically applied in subsurface bands or on the soil surface. Consequential N loss can result from the use of urea. Several avenues of ureal N loss can transpire if conducive conditions are present (Buresh and Baanante 1993). These can be affected and exacerbated by the choice of application method and timing, as well as environmental conditions. With increased N loss, plant N uptake is reduced resulting in lower above-ground biomass, grain protein concentration and grain yield (Brown et al. 2005). An increased risk of pollution and ecosystem degradation is also possible with increased N loss (Snyder et al. 2009).

Enhanced efficiency fertilizers (EEFs) have been developed to improve plant nutrient uptake and combat environmental loss associated with conventional fertilizers. This is achieved through the augmentation of traditional fertilizers with chemicals or coatings, to reduce the likelihood of certain types of N loss. By reducing N loss and providing greater N availability to the plant, EEFs aim to increase yield components and preserve environmental health (Olson-Rutz et al. 2011). The following literature review outlines the importance of wheat in global and Canadian agriculture, the dynamic nature of N and associated losses from urea, the different EEFs that have been developed for use in modern crop production, and their potential for increasing wheat productivity in western Canada while also preserving the environment.

1.2 Wheat (*Triticum aestivum* L.)

1.2.1 Global and historic perspective

Maize (*Zea mays* L.), wheat, and rice (*Oryza sativa* L.) are the three most important staple grain crops. In 2021, approximately 1100, 776, and 507 million Mg of grain, respectively, was harvested in each of these crops globally (United States Department of Agriculture [USDA]

2022a). Of these crops, wheat is grown on the greatest land base of roughly 221 million ha in 2021 (United States Department of Agriculture [USDA] 2022a). Wheat (*Triticum*), rye (*Secale*) and barley (*Hordeum*), are members of the tribe Triticea, a subdivision of the grass family Poaceae (Breiman and Graur 1995). Like other field crops, wheat is thought to have originated in the mountainous regions of the Fertile Crescent (Charmet 2011). Modern bread wheat is also thought to have undergone two polyploidization events. In between 500,000-150,000 years before present (BP), the first event occurred between *Triticum urartu* and a closely related wild species, *Aegilops speltoides*, creating a novel amphitetraploid species with 14 chromosome pairs (*Triticum turgidum* L. *ssp diccoides* (*T. diccoides*)) (Charmet 2011). This novel species was domesticated and evolved as *T. turgidum ssp dicocum* Schübl. (*T. dicocum*), which became the eventual progenitor of *Triticum durum* L. (durum wheat) (Charmet 2011). A second polyploidization event occurred around 10,000 BP, between the domesticated tetraploid *T. dicocum* and the wild diploid species *T. tauschii* to create a hexaploid species, *Triticum aestivum* L. (bread or common wheat) (Charmet 2011). Globally, bread and durum wheat constitute 92 and 7% of total production, respectively (Aquino et al. 1999). Additionally, although all wheat plant species are C₃ in nature, they can further be distinguished by growth habits of spring or winter.

1.2.2 Canadian wheat production and basic growth needs

Nearly all Canadian wheat is grown in the prairie provinces of Alberta, Saskatchewan, and Manitoba, with a relatively small portion in British Columbia and eastern Canada (McCallum and DePauw 2008). In 2021, approximately 22 million Mg of wheat (Statistics Canada 2022) were grown on 9.5 million ha across Canada (Statistics Canada 2021). Alberta, Saskatchewan, and Manitoba accounted for 29, 51, and 12% of this production, respectively (Statistics Canada 2021). As described by McCallum and DePauw (2008), Canadian wheat is divided into nine market classes based on growth habit (winter or spring) and quality factors (protein concentration, gluten strength, kernel hardness, colour); of which, Canada Western Red Spring (CWRS) is the largest. Canada Western Red Spring wheat is typically used for making high volume pan bread due to its relatively high protein content but is also suitable for blending (Iqbal et al. 2016). In the United States of America (USA), a Hard Red Spring (HRS) wheat class complements the Canadian CWRS class, albeit with slightly lower bread making quality. Of the

approximate 15 million hectares of wheat harvested annually in the USA, HRS typically constitutes 25% and is mainly found in the northern Plains (Minnesota, Montana, North Dakota, South Dakota) (United States Department of Agriculture [USDA] 2022b).

Suitable environmental conditions are needed to facilitate wheat production. Optimal wheat growth is achieved between 17-23°C (Porter and Gawith 1999). Adequate plant growth can be attained from a wide range of precipitation input (250 to 1750 mm yr⁻¹) (Enghiad et al. 2017); however, the distribution of rainfall within a growing season is typically a greater yield determinant than total annual precipitation (He et al. 2013).

1.3 Nitrogen and urea in agricultural systems

Nitrogen is one of three major macronutrients and 17 essential nutrients needed for plant growth. In the environment, N is cycled in several processes which influence its availability in the soil-plant system (Whalen et al. 2021). Nitrogen is often regarded as the single most important nutrient limiting plant growth (Shah 2008); however, it is often limiting in most ecosystems due to variations in soil type and distribution (Courty et al. 2015). These limitations can lead to N deficiency illustrated as slow stunted growth and leaf chlorosis, which reduces overall plant health and yield potential (Western Plant Health Association [wph] 2023). The application of N-containing fertilizer can correct and prevent plant N deficiencies (Tucker 1999), thereby underscoring the importance of proper N management in cropping systems.

Approximately 90% of soil N is contained within organic matter, while the remaining fraction is inorganic and available for plant use (Trenkel 2010). In cultivated soils, N in the form of ammonium (NH₄⁺) and nitrate (NO₃⁻) ions are mainly taken up by plants, fungi, and bacteria (Jackson et al. 2008; Guo et al. 2019). Nitrate plant uptake is more energy-demanding since it must be taken up actively (Haynes and Goh 1978) and then be biochemically reduced to NH₄⁺ before assimilation inside the plant (Li et al. 2013; Courty et al. 2015). Conversely, NH₄⁺ uptake is passive and can be directly assimilated (Haynes and Goh 1978). In soil solution, NH₄⁺ is in equilibrium with ammonia (NH₃) (Li et al. 2013) and because most soils have considerably low pH, NH₃ and associated NH₄⁺ concentrations are also low. Additionally, despite NH₄⁺ being energetically preferred, most plants will develop toxic effects if presented solely with NH₄⁺

nutrition (Guo et al. 2019). Ammonium toxicity is a poorly understood phenomenon (Esteban et al. 2016; Sun et al. 2021); however, it often inhibits crop biomass (Guo et al. 2019), yield, and overall photosynthetic capacity (Du et al. 2021). Therefore, the major inorganic N source for arable crops is NO_3^- due to its higher mobility and greater soil concentration (Courty et al. 2015). Acknowledging the disadvantages of each, the co-provision of both NO_3^- and NH_4^+ in cropping systems is needed and often fosters greater plant growth than the sole provision of either (Britto and Kronzucker 2002).

Nitrogen-based fertilizers have long been employed to provide arable crops with useable inorganic N. Urea, anhydrous ammonia, and urea-ammonium nitrate (UAN) are common contemporary synthetic N fertilizers. Nearly 20% of western Canadian growers use anhydrous ammonia, while the remaining majority use urea (Lyseng 2018). Despite being used relatively the least, UAN has recently increased in popularity in Saskatchewan (Government of Saskatchewan). Urea ($(\text{NH}_2)_2\text{CO}$), also known as carbamide, is the most commonly applied inorganic N fertilizer in global crop production due to its relatively high N content (46%) and lower transportation cost per unit (Engel et al. 2011; Nikolajsen et al. 2020). The application of urea to field crops can be achieved in subsurface bands, nests, or on the soil surface through broadcasting (Engel et al. 2010). Regardless of application method, ureal N can be lost through NH_3 volatilization, denitrification and NO_3^- leaching prior to plant uptake (Cameron et al. 2013) (Figure 1.1). After application and soil contact, urea will hydrolyze rapidly with water and the urease enzyme to produce ammonium carbonate (AC) (Mikkelsen 2009). Ammonium carbonate is an unstable compound which easily decomposes into NH_4^+ and further releases NH_3 gas (Adams et al. 2018). In soil (du Plessis and Kroontje 1964) and aqueous solutions (Korner et al. 2001), NH_4^+ and NH_3 are in equilibrium with concentrations being primarily influenced by increases in pH and temperature (Emerson et al. 1975). When AC is produced, nearby soil pH rises thereby shifting the equilibrium to favour NH_3 production (Mikkelsen 2009; Lasisi et al. 2020a). This shift also increases the risk NH_3 volatilization and N loss (Nikolajsen et al. 2020) (Figure 1.1). Instead of *gassing off* via volatilization, NH_4^+ cations can also be oxidized to NO_3^- anions via nitrification (Figure 1.1). This process is considered a gateway for the two other avenues of N loss (Moreau et al. 2019), as NO_3^- is prone to denitrification or leaching depending on environment. Firstly, denitrification is the stepwise conversion of NO_3^- up to and including:

nitrite (NO_2^-), nitric oxide (NO), nitrous oxide (N_2O), and dinitrogen (N_2) gas (Whalen et al. 2021) (Figure 1.1). This conversion often occurs in saturated and anoxic soils, as NO_3^- becomes a terminal electron acceptor during bacterial respiration (Cameron et al. 2013). Due to the emission of the potent greenhouse gas (GHG) N_2O , denitrification is commonly regarded as the most important avenue of agricultural N loss. Secondly, NO_3^- leaching typically occurs if water supply becomes greater than the soil's water-holding capacity (Angus and Grace 2017) (Figure 1.1). The anionic and highly soluble nature of NO_3^- reduces its sorption to soil exchangeable sites (Padilla et al. 2018; Mahmud et al. 2021), thereby increasing its mobility and susceptibility to environmental loss. The primary consequence of leached N from agricultural soils, is nutrient overloading and eutrophication of nearby aquatic ecosystems (Padilla et al. 2018). In soils experiencing oversaturation, NO_3^- leaching is more frequent in those with poor and coarse structure while finer-textured soils can expect greater denitrification (Cameron et al. 2013).

1.3.1 Conventional nitrogen rates in spring wheat

Adequate N supply is needed to achieve high grain quality and yield in CWRS production. As established by Karamanos (2013), a standard 2.69 Mg ha^{-1} spring wheat crop in western Canada will partition 95 kg ha^{-1} of N between its seed (67 kg ha^{-1}) and straw (28 kg ha^{-1}). Since cereal N recovery varies between 25-50% (Kubota et al. 2017), higher fertilizer rates must be used. According to the Alberta Fertilizer Guide (2004), irrigated CWRS benefits from N rates between $40\text{-}125 \text{ kg N ha}^{-1}$. This can be compared to spring wheat recommendations of $70\text{-}100 \text{ kg N ha}^{-1}$ for growers in Ontario (Ontario Ministry of Agriculture Food and Rural Affairs [OMAFRA] 2017). Standard N rates have also been suggested to allow growers to independently determine their own fertilizer applications based on anticipatory yields. According to Dinkins and Jones (2019), every kg of wheat grown in Montana should be fertilized with 55 g of N. Gelderman and Lee (2019) suggest a comparable 40 g for growers in South Dakota. The wide range of recommended N rates stem from factors affecting soil N availability such as: preceding crops, mineralization, soil texture and water supply (Alberta Agriculture Food and Rural Development [AAFRD] 2004). Since variability of soil N exists, residual N levels must be accounted for to facilitate optimal crop fertilization; however, only 10% of western Canadian farmers use soil nutrient testing nutrients (Karamanos and Cannon 2002). The main advantage of soil fertility testing is to determine current soil nutrient levels and avoid over- or under-

fertilization (Gill 2019). An example of soil fertility testing in western Canada, is the use of Plant Root Simulator (PRS®) probes (Western Ag Innovations, Saskatoon, SK) (Hangs 2002; Qian and Schoenau 2005; Owens et al. 2020) or determination by the Kjeldahl method (Bremner 1960; Matejovic 1995). Economic and environmental N loss can be reduced by compensating for residual soil N based on soil testing. Soils with high initial NO_3^- concentrations needed no additional N or a maximum application of 50 kg N ha^{-1} to obtain good wheat quality (Abad et al. 2005). Conversely, soils low in initial NO_3^- concentration required 100 kg N ha^{-1} to obtain similar results (Abad et al. 2005). This trend is supported in other studies (Beres et al. 2008b; Rial-Lovera et al. 2016; Gill 2019). Generally, sufficient yield components in small grain crops are achieved with soil test-recommended N rates (Gill 2019). Furthermore, increasing application rates beyond those prescribed does not typically benefit yield (Beres et al. 2010). Adequate soil fertility testing and compensation are needed to avoid fertilization errors and associated N loss.

High yielding CWRS and HRS production in the Northern Great Plains is also often reported with similar but consistent N rates. In western Canada, recent research by Hucl et al. (2022) indicated that four CWRS cultivars and five CWRS low-protein analogues, achieved highest grain yield when side-banded with urea at a rate of at least 140 kg N ha^{-1} , while the lowest was observed at 0 kg N ha^{-1} . Grain protein content also increased from 13.1 to 14.9% with an increase in N rate from 0 to 280 kg N ha^{-1} (Hucl et al. 2022). Walsh et al. (2018) observed that rain-fed HRS in Idaho achieved similar grain yield and protein content when fertilized with urea at rates of 90 or 135 kg N ha^{-1} . This suggests that 90 kg N ha^{-1} was agronomically optimal and is in accordance with the results of Karamanos et al. (2014), who found rates of at least 80 kg N ha^{-1} were needed to maximize yields in central Saskatchewan. Similar industry standard rates of 100 kg N ha^{-1} are seen for bread wheat production in Quebec (Yergeau et al. 2020). Another study by Walsh and Walsh (2020b) in northcentral Montana, indicated that increasing N rates up to 140 kg N ha^{-1} resulted in increasing grain yield, protein content and nitrogen use efficiency (NUE) of HRS. In southern Alberta, a fertigation study reported that CWRS grain yield did not improve with additional fertigated N beyond the previously banded urea at 120 kg N ha^{-1} (Smith et al. 2019). Furthermore, grain protein content increased with N rate via banding or fertigating up to 210 kg N ha^{-1} (Smith et al. 2019). A similar response was observed in durum and bread wheat, in

which, split-applications were indifferent if they exceeded available soil levels of 100 kg N ha⁻¹ (Beres et al. 2008a). In Ontario, similar conclusions were drawn in Canada Eastern Soft Red Winter (CESRW) wheat where the most profitable N rate was 105 kg N ha⁻¹ in one location, and varied with topography (91, 104, 120 kg N ha⁻¹ for upper, mid, and lower, respectively) in the other (Denys et al. 2006). In North Dakota, no differences in grain yield were observed between four HRS cultivars fertilized with 140 and 224 kg N ha⁻¹ (rain-fed site) or 168 and 280 kg N ha⁻¹ (irrigated site) (Otteson et al. 2007). Likewise in Minnesota, grain yield of four HRS varieties significantly improved when urea supply increased from 67 to 134 kg N ha⁻¹, but no differences were seen between the 134 and 201 kg N ha⁻¹ treatments (Farmaha et al. 2015). Thapa et al. (2015) also reported no significant HRS yield difference between urea applied at 146 and 168 kg N ha⁻¹ in south central Minnesota. In southern Alberta rain-fed CWRS, grain yield improved with increasing N rates up to 90 kg N ha⁻¹ while protein content improved up to the highest rate of 120 kg N ha⁻¹ (Beres et al. 2012a). Lastly, a prairie-wide Canadian study reported in 24 out of 26 site years, CWRS grain yield remained unaltered with urea applied at 60, 75, 120 and 135 kg N ha⁻¹ in bands at planting; moreover, protein content was significantly greater at the two highest N rates relative to the two lowest (McKenzie et al. 2006).

Collectively and over time, research in the Northern Great Plains show similar optimal N rates for high-yielding wheat production. Nitrogen rates of approximately 100-130 kg N ha⁻¹ return optimum grain yield return on a reoccurring basis. Intensive N input (~200 kg N ha⁻¹) often reports little improvement. These generalities are reported by others (Nyiraneza et al. 2012; Kostić et al. 2021). It should be noted that water can often be a greater yield-limiting factor than N (Mon et al. 2016; Long et al. 2017; Walsh et al. 2018). A 30-year analysis on Brown Chernozemic soils in southern Saskatchewan determined that precipitation from seeding to anthesis, was most crucial to achieve high wheat yields (He et al. 2013). They also elucidated that due to greater water holding capacity, clayey soils often produced higher and more stable yields relative to nearby silt loam soils (He et al. 2013). Adequate water supply will also often synergize with proper nutrient management to optimize wheat yield components (Long et al. 2017). Lastly, a Quebecois bread wheat study found no influence of seven rates of split-applied ammonium nitrate (AN) fertilizer on yield or bread quality (Yergeau et al. 2020). Instead, they reported a significant positive correlation between yield components and soil microbial richness

and diversity (Yergeau et al. 2020); therefore, a soil health index based on microbial community information remains an important component of soil fertility, leading to improved crop production.

1.3.2 Timing and placement of nitrogen fertilizer

In wheat production, N fertilizer can be applied in different ways and at different times. In Canada, conventional N fertilization occurs entirely at planting to limit in-field disturbance and reduce operational costs (Ma et al. 2006). Nitrogen applied around planting is however subject to increased risk of N loss prior to plant uptake (Subedi et al. 2007). Additionally, since wheat N uptake follows a sigmoidal path throughout the growing season, applying all N fertilizer around planting may not sufficiently match the crops overall seasonal demand. Split N fertilizer applications are employed to mitigate this problem (Subedi et al. 2007). In wheat, the majority of N uptake occurs before flowering. This N is used primarily for increasing yield potential by increasing the number of seed-bearing tillers and seeds per tiller (Brown et al. 2005). Nitrogen allocated for grain protein development is also taken up during this time; however, it is only translocated to developing kernels during the grain filling stage (Jones and Olson-Rutz 2012) as driven by sink-source relationships (Bancal 2009). Additional N can be accrued during and after heading, though it generally benefits grain protein content since yield potential has already been determined (Brown et al. 2005). Post-emergence application of N can be achieved with granular (urea), foliar, dribble or irrigated (UAN, liquid urea) fertilizers (Beres et al. 2008a).

As established by Howard et al. (2002), the application of N fertilizer to wheat must occur before the accelerated uptake stage in order to obtain highest yields and NUE. Generally, N applied before the booting stage increases grain yield, while applications as late as the milk stage can raise grain protein content (Brown et al. 2005). Using the Zadoks Growth Scale (ZGS) for cereals (Zadoks et al. 1974), common post-emergent N applications in wheat are performed around the start of stem elongation (ZGS30) and near the booting phase (ZGS45) to achieve increases in grain yield (Howard et al. 2002) and protein content (Karamanos et al. 2005), respectively. The simultaneous improvement of both typically occurs with increasing N rate prior to reaching a cultivars max yield potential (Fowler 2003). Conversely, further N input promotes their negative correlation where one variable will diminish (Iqbal et al. 2007; Bogard et

al. 2010). Individual genotype and environment (Iqbal et al. 2007) are considered the primary influencers of this relationship. Two hypotheses have been outlined as the basis of this phenomena. First, a competition between C and N for energy exists (Munier-Jolain and Salon 2005), where N uptake, assimilation, and translocation demands energy that can hinder seed production. Second, a N dilution effect occurs as yield increases (Acreche and Slafer 2009). Fowler (2003) established that increasing N in high-yielding wheat is appropriate for increasing protein content; however, simultaneous yield improvement is unlikely. Additionally, the critical point where wheat shifts from yield to protein improvement is heavily genotype specific (Fowler 2003; Long et al. 2017). Fowler (2003) also stresses the environmental specificity of each genotype's yield-protein correlation. Because of this negative correlation, as N rates increase beyond those needed to actualize yield potential, grain yield often stagnates while protein continually increases (Campbell et al. 1977; Long et al. 2017; Ghimire et al. 2021). Lastly, some studies report improvement in both grain yield and protein by increasing the amount of N applied (Beres et al. 2008a; Mohammed et al. 2013), thereby underscoring the dominant influence of genotype and environment on wheat yield components.

Mixed reviews on split-applications of N fertilizers are often reported. In southeast Saskatchewan, Lafond et al. (2008) determined that if at least a third of total prescribed N was applied at seeding, post emergent N applications up to the 5.5-leaf stage (ZGS15) maintained CWRS grain yield. Across four CWRS cultivars grown in southern Alberta, AN split applied at the 4-leaf stage (ZGS14) improved grain yield compared to the fertilized control; furthermore, a split-application near anthesis increased grain protein concentration (Beres et al. 2008a). A similar protein response was reported when soil-applied AN at seeding (90 kg N ha^{-1}) was supplemented with a foliar urea application (10 kg N ha^{-1}) at the booting phase in Canada Eastern Red Spring (CERS) wheat (Ma et al. 2006). In Brazil, HRS grain yield reduced while protein concentration increased when total N (70 kg N ha^{-1}) was split applied between tillering and heading, relative to the conventional practice of a sole application at tillering (Corassa et al. 2018). Similarly, split N applications in several Canadian wheat classes have provided slight increases in protein content while reducing grain yield, relative to a single application at planting across Manitoba (Heard 2017) and Saskatchewan (Hall et al. 2020). In north central Montana, dryland HRS top-dressed with two N rates (45 or 90 kg N ha^{-1}) during the flowering stage did

not improve grain yield or protein content compared to a single application at planting (Walsh et al. 2018). Another study reported highest HRS grain yield and protein content at the highest N rate (140 kg N ha^{-1}) applied entirely at tillering, while delaying N fertilization until the flag leaf stage (ZGS37) resulted in significantly lower yield components (Walsh and Walsh 2020a). A two-year field experiment in eastern Ontario indicated that split-applications of AN (60 kg N ha^{-1} preplant + 40 kg N ha^{-1} top-dress or foliar spray at ZGS45) did not benefit CERS grain yield or protein relative to a single preplant application (100 kg N ha^{-1}) (Subedi et al. 2007). Supporting CERS results were found by Zebarth et al. (2007) in New Brunswick. Split N applications can also pose greater lodging risks under high yielding and irrigated systems (Wu et al. 2019). Lastly, in dryland regions common in Palliser's triangle, the late application of granular urea can be ineffective due to the absence of precipitation required to move the granules into the root zone (McKenzie et al. 2006). In these situations, foliar N applications had the ability to improve fertilizer efficacy (Karamanos et al. 2005; McKenzie et al. 2006), while irrigated environments still benefitted from soil-applied N (Beres et al. 2008a). In general, mixed reviews regarding split N applications in spring wheat uphold the conventional practice of a single application during planting.

1.3.3 Nitrogen fertilizer and the environment

A consequence to synthetic N fertilizer use is climate change. As defined by the United Nations, climate change “refers to long-term shifts in temperatures and weather patterns” (United Nations). Natural solar cycle variations mainly caused climate change prior to the 19th century; however, recent anthropogenic activities have been reported to principally drive climate change (United Nations). Climate change is linked to global warming, severe weather variability, declining biodiversity (United Nations) and increased pollution (Suddick et al. 2013). A large contributor to this, is the production, use, and N cycling, of conventional synthetic N fertilizers in agriculture (Stuart et al. 2015). Synthetic fertilizer production is dependent on NH_3 . Industrial NH_3 production needed for fertilizer is achieved via the Haber-Bosch process. Globally, the Haber-Bosch process accounts for nearly 80% of total NH_3 production (Wang et al. 2018). Consequently, its energy dependent nature uses nearly 2% of global energy supply (Wang et al. 2018) and maintains a massive carbon footprint (Ghavam et al. 2021). Approximately 2.6 Mg of life cycle GHGs are emitted from the production of 1 Mg of NH_3 (Liu et al. 2020). In Canada,

fertilizer manufacturing emits roughly $1.3 \text{ kg CO}_2 \text{ kg}^{-1}$ of N produced (Natural Resources Canada 2008).

Following the production of synthetic N fertilizer, their use in-field contributes further to climate change. Carbon dioxide (CO_2), methane (CH_4), and nitrous oxide (N_2O) are three GHGs linked to agricultural production with respective 100-year global warming potentials (GWP) of 1, 27, and 273 (Forster et al. 2021). Due to N_2O 's high GWP, it is considered the most potent biogenic GHG and precursor of ozone layer depletion (Ravishankara et al. 2009; Cameron et al. 2013). Contemporarily, N_2O emissions account for approximately 6% of anthropogenic radiative forcing (Myhre 2013). Coupled with an atmospheric lifetime of >100 years (Suddick et al. 2013; Yahya 2018), the climate warming impact of N_2O is great. Nitrous oxide is produced via nitrification and denitrification; both being avenues of N loss from fertilizer application in field crops (Cameron et al. 2013) (Figure 1.1). Over the past 150 years, atmospheric N_2O concentrations have increased roughly from 270 ppb to over 330 ppb in 2018 (Tian et al. 2020). Globally, 7.3 Tg N yr⁻¹ of N_2O are emitted from anthropogenic sources (Tian et al. 2020) and of this, 60% is reported from agricultural soils (Del Grosso et al. 2022). In Canada, agriculture accounts for 70% of anthropogenic N_2O emissions, mostly caused by the application of synthetic nitrogen fertilizers such as urea (Environment and Climate Change Canada 2020). Agriculture has also been outlined as the major source of N pollution worldwide (Suddick et al. 2013).

Apart from N_2O emissions, other avenues of N loss can cause environmental consequences. Nitrate leaching and runoff can facilitate nutrient overloading and eutrophication of nearby aquatic ecosystems (Padilla et al. 2018), thereby reducing local biodiversity and creating zones of hypoxia. The release of NH_3 gas through volatilization, and nitric oxide from denitrification into the atmosphere (Figure 1.1), eventually deposits on the ground. This surface deposition of reactive N can lead to water and soil acidification, eutrophication, and pollution (Suddick et al. 2013; Stuart et al. 2015; Skorupka and Nosalewicz 2021). Additionally, reactive N released from inefficient fertilizer use can exacerbate climate change and vice versa (Suddick et al. 2013). The amount of N applied is directly related to the probability of high N load in surface runoff (Hou et al. 2019) and gaseous N loss (Shcherbak et al. 2014).

Reducing N pollution is needed to reduce climate change and limit the aforementioned consequential outcomes (Suddick et al. 2013). Models demonstrate that increased GHG emissions, climate change degradation, and pollution are likely if anthropogenic agricultural activities continue as they do (Scherger et al. 2022; Vitousek et al. 2022); therefore, a change must occur to reduce environmental impact and promote sustainability (Stuart et al. 2015). The greatest opportunities for reductions are apparent in agricultural systems, due to its often-inefficient use of N (Suddick et al. 2013). In most annual grain crops, fertilizer uptake is less than 40% of what is applied (Cassman et al. 2002). The majority of remaining N is lost through air, and above- or below-ground pathways (Follett and Delgado 2002), while a small portion may be bound organically (Stuart et al. 2015). Low N-use efficiency at both the plant and cropping system level contribute to N loss, reactive N cycling, and further impacting climate change (Stuart et al. 2015). Improving the efficacy of N fertilizer can lead to increased plant nutrient uptake and lower environmental loss (Chen et al. 2008; Grant and Wu 2008); thereby, reducing associated N pollution and climate change (Cameron et al. 2013; Suddick et al. 2013; Angus and Grace 2017). Insufficient production of N fertilizer in relation to expected consumption has also been predicted up to 2050 (Yahya 2018); therefore, enhancing the efficiency of fertilizer is also needed to aid in production demand. Crop producers can reduce GHG emissions and improve NUE by incorporating best management practices and 4R principles for fertility management (Snyder et al. 2009; Tenuta et al. 2019). A method of achieving both, is to incorporate EEFs in on-farm nutrient management (Akiyama et al. 2010; Trenkel 2010).

1.4 Enhanced efficiency fertilizers

As established by Olson-Rutz et al. (2011), EEFs “are fertilizers that reduce [nutrient] loss to the environment and/or increase nutrient availability compared to conventional fertilizers” (p.2). Similarly, Akiyama et al. (2010) succinctly put that EEFs are used to “increase the efficiency of fertilizer use by crops” (p.1837). While certain EEFs have been available since the 1960s (Reddy 1964), their understanding and adoption have increased largely over the past few decades. In 2019, approximately 24% of total N fertilizer tonnage sold in the USA was classified as an EEF (The Fertilizer Institute 2019). Focusing on N-based EEFs (hereafter referred to as EEF(s)), there are two main categories: (i) slow- or controlled-release nitrogen fertilizers, and (ii) nitrogen stabilizers (Trenkel 2010). Grant and Wu (2008) provide a detailed review of common

EEFs and their appropriate use on the Canadian prairies, while Trenkel (2010) provides an exhaustive addendum of many EEFs along with their trade name and manufacturer in each major crop producing region around the world. It should be noted, that EEFs can be associated with many fertilizers, nutrients, and nutrient blends; however, this review will solely focus on N-based EEFs developed to augment urea (hereafter referred to as EEF(s)).

1.4.1 Slow- or controlled-release nitrogen fertilizers

The terms slow- and controlled-release fertilizers are often interchanged. Trenkel (2010) provided a division between the two, where slow-release fertilizers have nutrient release patterns which are fully dependant on soil and weather conditions and cannot be predicted; conversely, controlled-release fertilizers have some degree of predictableness in their nutrient release. Despite this, to maintain in line with most of the supporting literature, we will consider both slow- (SRF) and controlled-release fertilizers (CRF) as one common group (SRF) for this review. Slow-release fertilizers are likely the most popular type of EEF used in modern crop production. According to Tolescu and Iovu (2010), a SRF must contain at least one nutrient that either (i) delays its assimilation and use by plants after application, or (ii) is available for the plant to use over a significantly longer period of time than its conventional alternative. From this, we can establish that SRFs release nutrients at a slower rate than conventional urea.

Slow-release fertilizers are conventional soluble fertilizers (i.e., urea) encapsulated with a protective coating (Trenkel 2010). These coatings can be made of water-insoluble, semi-permeable or impermeable-with-pores type material (Trenkel 2010) and aim to restrict the dissolution rate of the soluble materials within them (Naz and Sulaiman 2016). In western Canada, SRFs aim to release N at a prolonged rate matching the crops seasonal demand (Grant et al. 2012). By accomplishing this, NUE can be improved (Haderlein et al. 2001) and environmental N loss be reduced (Grant et al. 2012). The most common SRF used in crop production in western Canada and the northern Plains, is Environmentally Smart Nitrogen® (ESN®) produced by Nutrien Ltd.. Environmentally Smart Nitrogen® is a polymer coated urea (PCU) which allows water to infiltrate and dissolve urea, then slowly release the solubilized solution. Environmentally Smart Nitrogen® has a light green colour, bulk density of 769 kg m⁻³, pH of 7.0, and is very lightly soluble in hot or cold water due to its polymer coating (Golden et

al. 2011). The manufacturer of ESN® characterizes that 80% N release is achievable between 30 and 60 days at 23°C (Golden et al. 2011). Dowbenko (2006) also states that if western Canadian and northern Plain growers apply ESN® at seeding, they can expect 35-50% N release within the first 25-30 days after application and the remainder be evenly released over the following 30-40 days. These statements are supported by the findings of Golden et al. (2011). They observed slight differences in release rate between seven Arkansas soils, resulting in 75-83% by day 40. Soil moisture had the greatest initial influence on release rate, but no significant differences were observed between all treatments (125-380 mg H₂O kg⁻¹) by day 40. Lastly, they concluded temperature to be the greatest influence on release rate where percentage of N remaining in ESN® granules was 71, 13, 14 and 14%, at 15, 20, 25 and 30°C, respectively (Golden et al. 2011).

The dependency of ESN® on temperature for N release is well documented (Huett and Gogel 2000; Dimkpa et al. 2020), with Gandeza et al. (1991) providing two explanations: (i) an increase in temperature equates to an increase in the difference between the surroundings of the granule and its internal surface, and (ii) an increase in temperature increases the moisture permeability of the polymer coating. It should also be noted that soil moisture can greatly affect PCU N release rate. Soil matric potential drier than -60kPa in two Australian soils (silty clay, sandy loam) was reported to significantly reduce N release rate relative to wetter conditions (Verburg et al. 2021). Soil temperature and moisture remain the two largest influencers on the release rate of SRFs.

1.4.2 Stabilized nitrogen fertilizers

Stabilized nitrogen fertilizers (SNFs) are the other major category of EEFs. Nitrogen stabilizers are substances which prolong the duration of soil applied N in the ureal or ammoniacal form (Trenkel 2010). Stabilized nitrogen fertilizers are conventional fertilizers (i.e., urea) that have been augmented with a nitrogen stabilizer. In western Canada, SNFs are divided into urease (UI) and nitrification inhibitors (NI). Trenkel (2010) provides a detailed overview of the characteristics that UIs and NIs must possess; however, they all pertain to maintaining environmental health while improving plant nutrient availability.

1.4.2.1 Urease inhibitors

Following the application of N fertilizer, NH_3 volatilization is the first encountered avenue of N loss. After hydrolysis transforms a urea granule into NH_4^+ cations, if near the soil surface, these cations can be converted to NH_3 and be volatilized to the atmosphere. Urease inhibitors are compounds which delay urea hydrolysis. As established by Byrne et al. (2020), this delay is achieved by impeding the urease enzyme's active site through several mechanisms. The practical goal for a UI is to allot more time for the urea granule to be incorporated into the soil and become adsorbed to exchangeable sites in the form of NH_4^+ , before possible volatilization. Urease inhibitors can be divided into two categories based on components: (i) metal complexes and (ii) organic compounds. Before the latter half of the 20th century, metal complexes were widely used as UIs (Shaw 1954; Tabatabai 1977). These complexes functioned by reacting with the sulfhydryl groups of the urease enzyme, creating insoluble sulfites which inhibited the enzymes performance (Cantarella et al. 2018; Byrne et al. 2020). As ranked by Byrne et al. (2020), the following metallic ions are listed in decreasing order of efficacy at inhibiting urease activity: $\text{Ag}^+ \sim \text{Hg}^{2+} > \text{Cu}^{2+} > \text{Cd}^{2+} > \text{Ni}^{2+} > \text{Co}^{2+} > \text{Fe}^{2+} > \text{Mn}^{2+}$. Today, most commercially available UIs are based on organic compounds which can be divided into two classes of structural urea analogs: (i) hydroxamic acid and (ii) compounds that impact the urea hydrolysis reaction mechanism (Byrne et al. 2020). Within the latter, organophosphorus compounds are mostly used in agriculture and within them, phosphoramides are included (Byrne et al. 2020). Phosphoramides act as strong UIs due to their binding to the active metalcentre site of the urease enzyme (Kafarski and Talma 2018). N-(n-butyl) thiophosphoric triamide (NBPT) is the most widely used phosphoramide in agriculture (Modolo et al. 2018). N-(n-butyl) thiophosphoric triamide has been most notably traded under the name Agrotain® since the mid 1990's (Cantarella et al. 2018) and is available in over 13 products to over 52 countries, Europe and South America (Koch Fertilizer). The standard Agrotain® 20% nitrogen stabilizer product is a green liquid with a pH of 8-9.5 and NBPT concentration between 10-30% (Koch Fertilizer 2021b). Lastly, the non-aqueous, liquid formulation of Agrotain® allows it to be injected into molten urea before granulation, applied to prills, or added to an UAN solution (Trenkel 2010).

The inhibitory effects of NBPT are complex and relatively short-lived. By blocking three active sites of the urease enzyme and forming a tridentate bond with two nickel and one oxygen center (Byrne et al. 2020), NBPT forms a carbamate bridge thereby increasing the difficulty for urea to reach the active nickel center of the urease enzyme (Manunza et al. 1999). Despite this, NBPT is not the direct inhibitor of urease; however, its oxygen analog N-(n-butyl) phosphoric triamide (NBPTO) is (Cantarella et al. 2018). NBPTO is not directly applied however, due to its faster degradation (Hendrickson and Douglass 1993) and greater instability relative to NBPT (Byrne et al. 2020). Furthermore, the conversion from NBPT to NBPTO is poorly understood but does occur faster under aerobic conditions (Cantarella et al. 2018). The effects of NBPT usually last between three and seven days, at which time new enzyme production overwhelms the old inhibitor (Byrne et al. 2020). Research on three dimensional dynamics of soil N showed that banded NBPT treated urea greatly inhibited ureolytic activity for seven days within 75 mm of the fertilizer band (Janke et al. 2020). During this time, concentrations of urea-N were 16-fold higher in the fertilizer band of the NBPT treatment (Janke et al. 2020). The authors concluded that NBPT's inhibitory effects lasted 34 days (Janke et al. 2020). NBPT efficacy will also reduce as soil pH decreases (Engel et al. 2013; Engel et al. 2015). The onset of degradation is also affected by soil temperature, as NBPT begins degrading faster in warmer soils (2-4 days (Soares et al. 2012)) compared to cooler soils (up to 10-15 days (Watson et al. 2008)). Storage effects on NBPT treated fertilizers also indicate lower efficacy after being stored in warmer compared to cooler temperatures (Watson et al. 2008; Cantarella et al. 2016); however, storage up to six months at ambient temperature had no alterations on its performance (Lasisi et al. 2020c).

1.4.2.2 Nitrification inhibitors

Once in the soil, nitrification can enable the oxidation of NH_4^+ cations into NO_3^- anions. Nitrification inhibitors are compounds which delay the oxidation of NH_4^+ by suppressing the activity of soil nitrifiers (Subbarao et al. 2006). This results in greater inorganic N remaining as NH_4^+ cations (Lasisi et al. 2021), which are more likely to be adsorbed to soil exchangeable sites and be protected from leaching than NO_3^- anions (Degenhardt et al. 2016). By hindering nitrification, the risk of denitrification is also indirectly lowered (Nikolajsen et al. 2020). Dicyandiamide (DCD ($\text{C}_2\text{H}_4\text{N}_4$)) and nitrapyrin ($\text{C}_6\text{H}_3\text{Cl}_4\text{N}$) are the two most common NIs used in global crop production. Dicyandiamide is more common in Europe and Asia, while nitrapyrin

is in North America and Australia (Trenkel 2010). Both stabilizers function by inhibiting the ammonia monooxygenase (AMO) enzyme needed for the rate limiting first step of nitrification (Degenhardt et al. 2016; Yang et al. 2016). This is accomplished by chelating copper (Cu) ions, which removes a major cofactor of the AMO enzyme (Duncan et al. 2017; Torralbo et al. 2017). Lastly, both NIs are classified as bacteriostatic (Di and Cameron 2017; Woodward et al. 2021).

Like UIs, NIs have been used in crop production for many decades. Dicyandiamide has been investigated since the 1960s in Canada (Reddy 1964) and 1970s in the USA (Bock). Dicyandiamide is a crystalline, water-soluble, and non-volatile (Di and Cameron 2016) powder, suitable for use in granular fertilizer coatings or incorporation with solid N fertilizers (Subbarao et al. 2006). As established by Amberger (1989), the inhibitory effects of DCD persists for 4-8 weeks depending on soil temperature, water content, texture, organic matter and pH. It is also well documented that DCD's efficacy is negatively correlated with increasing soil temperature (Kelliher et al. 2008; McGeough et al. 2016). Irigoyena et al. (2003) concluded that DCD in soil temperatures of 30, 20 and 10°C, inhibited nitrification for a week, month, and more than 3 months, respectively. Dicyandiamide also degrades faster at higher levels of soil organic matter (Elrys et al. 2020) due to microorganismal assimilation of DCD-derived N (Subbarao et al. 2006). The efficacy of DCD also reduces as pH rises (Puttanna et al. 1999) since ammonia-oxidizing bacterial growth increases in alkaline conditions (Robinson et al. 2014; Wakelin et al. 2014). In summation, DCD is affected by various soil characteristics and inhibition efficacy is often environment specific (Elrys et al. 2020). This statement is supported by others (Di and Cameron 2016; McGeough et al. 2016; Di and Cameron 2017).

Nitrapyrin is the other predominant NI in commercial crop production. It is the active ingredient in two common stabilizer products sold in North America. Produced by CortevaTM Agriscience, their traded names are Instinct® and eNtrench®, which contain nitrapyrin concentrations of 16.95% (Corteva Agriscience 2015) and 25.97% (Corteva Agriscience 2021), respectively. Instinct® has a pH of 8.51 and a liquid density of 1.12 g cm⁻³ at 20°C (Corteva Agriscience 2015). eNtrench® has a pH of 8.54 and a liquid density of 1.196 g cm⁻³ at 20°C (Corteva Agriscience 2021). Nitrapyrin was first introduced in the Midwest region of the USA in the early 1960s (Goring 1962) and was registered for use in 1974 (Huber et al. 1977). Due to its

highly volatile nature, nitrapyrin was only used successfully in soil-injected fertilizers such as anhydrous ammonia, aqua ammonia, and urea-ammonium nitrate (UAN) (Goos and Guertal 2019). Within the past decade, microencapsulated nitrapyrin products (i.e., Instinct®, eNtrench®) have been developed. These products are involatile and can be impregnated into urea and used accordingly (Goos and Guertal 2019). Similar to DCD, the degradation of nitrapyrin enhances as soil temperature rises (Wolt 2004). Typically, nitrapyrin will degrade within 30 days in warm soils but can persist well in cold soils and be used successfully in fall applications (Subbarao et al. 2006; Byrne et al. 2020). Initial laboratory incubation studies found nitrapyrin to inhibit nitrification for 6 weeks in 74 out of 87 soils across the USA and one province in Canada (Goring 1962). More recent research by Chen et al. (2010), reported the inhibition of nitrification in a brown vertisol soil for 42 days at 40 and 60% water filled pore space, and at 5, 15 and 25°C. Another study observed significant inhibitory effects over 14 days at 20 and 40°C, with a 45% water filled pore space similar to regional rainfall conditions in Australia (Fisk et al. 2015).

In Europe, another NI called 3,4,-dimethylpyrazole phosphate (DMPP) based on the 3,4,-dimethylpyrazole (DMP) compound is commonly used. Developed in 1995 by Badische Anilin und Soda Fabrik (BASF) in Germany, DMPP is mostly traded under the name Entec® (Trenkel 2010). The efficacy of DMPP has been well documented in European research (Zerulla et al. 2001; Pfab et al. 2012; Huérfano et al. 2015; Huérfano et al. 2018). Recently, a novel NI called the 2-(N-3,4-dimethyl-1H-pyrazol-1-yl) succinic acid isomeric mixture (DMPSA) has emerged. Improving on DMPP, DMPSA is more stable in alkaline conditions and can be used with calcium ammonium nitrate or diammonium phosphate fertilizers (Huérfano et al. 2016; Rodrigues et al. 2018). Developed in 2015, DMPSA is relatively unknown and has not been extensively researched (Huérfano et al. 2018). Though the inner mechanisms are not fully understood, recent research by Corrochano-Monsalve et al. (2021) demonstrated that DMPSA and DMPP act as Cu chelating compounds like the more mainstream NIs, DCD and nitrapyrin.

1.4.2.3 Dual inhibitors

Typically, the efficacy of nitrogen stabilizers is based on environment (Woodward et al. 2021). Variable and unique conditions can warrant the use of multiple inhibitors. Dual inhibitors

(DIs) combine NIs and UIs into a single product. In theory, the combination of both inhibitors will reduce NH_3 volatilization and NO_3^- leaching, and indirectly denitrification and N_2O emissions (Afshar et al. 2018). In North America, a popular DI is traded under the name SuperU® by Koch™ Fertilizer. SuperU® is a stabilized fertilizer that conglomerates NBPT, DCD and urea. It is sold as pre-coated urea granules that are blue in colour, have a pH of 7.2 and a density of 753 kg m^{-3} (Koch Fertilizer 2021a). Like urea, SuperU® can be applied on the soil surface or placed in seed-row or fertilizer bands (Afshar et al. 2021). Agrotain® Plus is another popular DI traded mainly in the USA by Koch™ Agronomic Services. Agrotain® Plus has the same active ingredients as SuperU®; however, it is sold in a concentrated powder or granular form and must be incorporated with N fertilizers before use (Koch Fertilizer 2015).

Despite the broad advantage of DIs, the duration of their inhibitory effects are not well documented; nonetheless, inferences can be made based on their active ingredients. Afshar et al. (2018) did note that SuperU® broadcasted in rosette staged camelina (*Camelina sativa* L.), delayed urea hydrolysis for approximately five days compared to urea. In a laboratory incubation study by Dell et al. (2014), SuperU® and Agrotain® Plus were both found to inhibit nitrification for 21 days compared to urea and UAN. Additionally, NO_3^- accumulation was higher in soil with 30% than 18% water content, thereby suggesting DIs degrade faster in wetter soils (Dell et al. 2014). Lastly, similar trends were found with the same active ingredients applied to winter wheat in the United Kingdom (Fu et al. 2020).

1.5 Impact of enhanced efficiency fertilizers on carbon footprint

Recently, a worldwide meta-analysis comprised of 182 peer-reviewed publications concluded UIs to reduce NH_3 volatilization by 51%, and NIs to reduce N_2O emissions by 49% (Fan et al. 2022). A meta-analysis by Akiyama et al. (2010) reported similar reductions (>34%) in N_2O and NO emissions from the use of NIs and polymer-coated fertilizers; however, indifferent results from UIs relative to urea were noted. This work was supported by Thapa et al. (2016), who analyzed 43 peer-reviewed studies and also revealed DIs to be significant N_2O reducers, performing better than SRFs but less than NIs. Snyder (2017) also provided a detailed review from over 35 peer-reviewed publications of the effects of EEFs on crop yields and

reductions in nitrate leaching, NH₃ volatilization, and direct N₂O emissions. On a global GHG emission scale, EEFs are shown to reduce harmful flux and aid in environmental preservation.

In the northern Great Plains, EEFs have been employed to reduce GHG emissions. Maize grown in central Colorado had reduced N₂O emissions (>40%) with the use of ESN® and SuperU® over urea (Halvorson et al. 2014). Similar trends were observed in CWRS in Manitoba (Gao et al. 2015). Conversely, winter wheat grown in southern Alberta had indifferent N₂O emissions during the non-winter portion of its life cycle when fertilized with three EEFs (ESN®, Instinct®, SuperU®) or urea (Owens et al. 2020). The authors also noted that a split-application in the late fall increased N₂O emissions relative to split-applying in the spring or only banding at planting (Owens et al. 2020). These results were supported in two more studies (An et al. 2020; An et al. 2021). In Montanan winter wheat, surface-applied NBPT coated urea reduced total NH₃ loss by 66% compared to uncoated urea (Engel et al. 2011). Furthermore, volatilization protection lasted 2-3 weeks on acidic soils and more than 7 weeks on alkaline soils during the winter months (Engel et al. 2011). Correspondingly, surface applied NBPT treated urea (Agrotain® Advanced & ARM UTM) in Manitoba reduced NH₃ volatilization by 65 and 40% for fall and spring applications, respectively, when averaged over two years (first season – spring wheat, second season – canola (*Brassica napus* L.)) (Lasisi et al. 2020b). In this same study, urea treated with a DI (ARM UTM Advanced) attained statistically similar results for the fall, but 27% less reduction in the spring application (Lasisi et al. 2020b). This contests the results of Thapa and Chatterjee (2017) who in 2015, observed greater spring NH₃ volatilization with the use of a NI (Instinct®) than DI (SuperU®). They also observed reduced N₂O emissions of 43 and 53% in DI and NI treated urea, respectively (Thapa and Chatterjee 2017). Similar results from a novel DI (NBPT + DMPSA) fertilizer in a Spanish wheat crop indicated reduced gaseous NH₃ and N₂O loss by 50.5 and 91.6%, respectively (Guardia et al. 2021). Lastly, Goos and Guertal (2019) carried out a laboratory and two greenhouse experiments to assess urea hydrolysis, NH₃ volatilization and nitrification reduction potential of several EEFs (Nutrisphere-N®, NZone®, Agrotain® Ultra, Instinct®, SuperU®) using three North Dakota soils. Their first experiment exhibited significant delay in urea hydrolysis (up to 10 days) from urea treated with NBPT (Agrotain® Ultra). These results translated similarly to their NH₃ volatilization experiment, where NBPT significantly reduced volatilization loss the entire 14-day period following

application. Lastly, their nitrification experiment reported the NI (Instinct®) and DI (SuperU®) treatments to significantly reduce nitrification for two weeks in each soil (Goos and Guertal 2019).

1.5.1 Pollution swapping

Despite the beneficial goals of EEFs, secondary repercussions can transpire. Pollution swapping is the most environmentally consequential indirect effect of EEFs. This occurs from the use of NIs which indirectly increase NH_3 volatilization. Since NIs retain inorganic soil N in the form of NH_4^+ , the $\text{NH}_4^+ - \text{NH}_3$ equilibrium can shift to producing NH_3 and increase volatilization risk (Zaman and Nguyen 2012). A global meta-analysis of 62 peer-reviewed publications reported that although applying NIs reduced NO_3^- leaching, N_2O , and NO emissions by approximately 48, 44, and 24%, respectively, NH_3 emissions rose by roughly 20% (Qiao et al. 2015). They also concluded that total gaseous N flux increased by 14.3% when using NIs (Qiao et al. 2015). Similar trends have been observed in other meta-analyses (Lam et al. 2017; Fan et al. 2022). Pollution swapping can also be found when DIs increase NH_3 volatilization relative to UIs, thereby diminishing their purpose. In six laboratory experiments, three representative soils of the Mideastern Corn Belt were tested over 2 weeks for NH_3 volatilization with surface applied urea treated with UIs (NBPT), NIs (DCD, nitrapyrin), and DIs (NBPT + DCD, NBPT + nitrapyrin) (Frame 2017). In five out of six trials, the sole application of either NI resulted in higher gaseous NH_3 loss relative to granular urea. When applying either DI, its NBPT component minimized NH_3 loss but was less effective than unaccompanied NBPT (Frame 2017). These findings have been supported elsewhere (Awale and Chatterjee 2017; Lasisi et al. 2020a). Due to possible secondary repercussions stemming from the use of EEFs, careful nutrient management based on anticipatory environmental conditions should be emphasized.

1.6 Impact of enhanced efficiency fertilizers on yield components

Aside from reducing environmental N loss, EEFs are implemented to increase and/or prolong plant N availability. In theory, this would lead to greater plant growth; however, in-field results are not always supportive. Globally, Thapa et al. (2016) concluded a 7% overall cereal crop (maize, wheat, rice) yield increase with the use of NIs. This supported the previous meta-evaluation by Wolt (2004). Additionally, DIs increased mean yields in alkaline (2.0%), coarse-

textured (5.7%), and irrigated soils (2.0%), while SRFs had no overall effect (Thapa et al. 2016). Conversely, another meta-analysis by Fan et al. (2022) reported UIs achieving greater crop yields relative to NIs or DIs. In southern Alberta winter wheat, the substitution of urea with a SRF by 50 or 100% increased grain yield by an average of 4.3% across all site years, despite a drop in protein content of 1.3% (Beres et al. 2010). Similar results were observed in Montanan HRS by Walsh and Girma (2016), where just a SRF or a 50:50 mix resulted in increased grain yield in one quarter of site-years. In Southern Great Plain winter wheat, grain yield and protein content were highest with a DI (SuperU®) broadcast at 70 kg N ha⁻¹ relative to a banded SRF (ESN®) or broadcasted urea: moreover, when N rates were 30 kg N ha⁻¹, urea yielded slightly higher than the other N sources while the SRF caused slightly higher grain protein (Adams et al. 2018). In two western Canada winter wheat studies, a DI consistently improved and stabilized grain yield and protein relative to other common EEFs and urea (Beres et al. 2018; Wang et al. 2022). In Denmark, UAN treated with a DI (NBPT + DMPSA) or UI (NPBT) increased wheat grain yield by 7 and 14%, respectively (Nikolajsen et al. 2020). Similarly, under drip irrigation in China, wheat fertilized with a DI (NBPT + nitrapyrin) achieved the same grain yield as wheat fertilized with urea while using 20% less fertilizer (Tao et al. 2021). Lastly, grain yield of two waterlogged summer maize cultivars increased with the use of a NI (nitrapyrin) in China (Ren et al. 2017). Depending on agronomic management and environment, crop production can be optimized with EEFs.

Despite the benefits that EEFs can provide agricultural systems, reports of inconsistent benefits are frequent in publication and contribute to the hinderance of their widespread adoption (Li et al. 2018; Verburg et al. 2022). Thapa and Chatterjee (2017) did not observe any grain yield or protein content response of Minnesotan rain-fed spring wheat to the addition of broadcasted inhibitors (SuperU®, Instinct®), which supported their previous findings (Thapa et al. 2015). A Manitoban investigation of several EEFs (ESN®, SuperU®, Agrotain® Plus) in CWRS indicated that slope position and seeding date were greater influencers on grain yield and protein content than N form (Grant et al. 2016). Another CWRS study in Manitoba reported no difference in grain yield despite reductions in N₂O emissions from the use of EEFs (SuperU®, ESN®) (Gao et al. 2015). In rain-fed Pennsylvania maize production, grain yield did not differ between EEFs (ESN®, SuperU®, Agrotain® plus) over 4 years compared to urea (Dell et al.

2014). Similarly, banding and broadcasting a DI (SuperU®) or urea at 90 kg N ha⁻¹ did not alter rain-fed HRS grain yield or protein concentration in Montana (Afshar et al. 2021). Lastly, ammonium sulphate (AS) treated with a novel NI (DMPSA) applied to a Mediterranean wheat crop did not differ in grain yield or protein content compared to untreated AS at 120 or 180 kg N ha⁻¹ regardless of application method (Huérffano et al. 2016). Furthermore, DMPP obtained similarly indifferent results in wheat (Huérffano et al. 2015), maize and ryegrass (*Lolium perenne* L.) (Huérffano et al. 2018).

Another niche for certain EEFs is the ability to be used as seed-row placed fertilizers. The seed safety nature of ESN® has been well documented (McKenzie et al. 2007; Beres et al. 2012b; Malhi and Lemke 2013; Qin et al. 2014) ; however, improvement in yield components are not consistent in spring (Walsh and Girma 2016) or winter wheat (McKenzie et al. 2010; Rajkovich et al. 2017). Across southern Alberta, a seed-placed UI (NBPT) was also found to inhibit cereal seedling damage despite only showing increased grain yield in eight of 39 site-years (Karamanos et al. 2004). In a Canada-wide study, Grant et al. (2012) reported no consistent improvement in grain yield or N use efficiencies with the use of ESN® or split-applications in relation to standard regional timing and placement of conventional granular urea at planting. Environmentally Smart Nitrogen® has also been found to reduce grain yield in certain environments, due to the nutrient release rate being too prolonged for plant uptake (Beres et al. 2010). Conversely, side-banded ESN® improved CWRS grain yield during periods of drought in southern Alberta relative to urea (Beres et al. 2010). This suggests that the slower N release rate of ESN® can prevent early vigorous plant growth that would deplete early-season soil moisture, thereby hindering future water uptake over the growing season, carbohydrate grain partitioning (Angus and van Herwaarden 2001) and grain yield (Passioura 2006). It should be noted however, that EEF performance is most often environment specific. Several publications emphasize the importance that environmental and soil conditions play to the overall efficacy of any EEF (Grant and Wu 2008; McGeough et al. 2016; Elrys et al. 2020; Woodward et al. 2021; Fan et al. 2022) and that insignificant results likely stem from a lack of conducive conditions for N loss (McKenzie et al. 2010; Grant et al. 2016; Verburg et al. 2022). This contributes to management problems in relation to EEF use, since environmental conditions cannot be accurately predicted prior to applying fertilizer.

1.7 Challenges with enhanced efficiency fertilizers

1.7.1 Cost

The increased cost of EEFs also hinders wide-spread consumer adoption (Thapa et al. 2016). To our knowledge, no peer-reviewed publication describes a detailed economic analysis of NIs, UIs, or DIs in CWRS cropping systems of western Canada. Adams et al. (2018) did provide a net profit analysis of ESN®, SuperU® and urea in Texan winter wheat, whereby they reinforced the notion that treatments with higher input costs fared worse in each price level. They also showed that in most cases, urea provided the highest net profit followed by SuperU® then ESN®. In western Canada, ESN® was shown to provide no economic benefit over the use of urea across six locations (Khakbazan et al. 2013); however, mixing urea and ESN® has proven economically viable for wheat production in Minnesota (Farmaha and Sims 2013). Depending on global location and type, SRFs can cost 2.5-10 times greater than conventional fertilizers (Shaviv 2001; Davidson and Gu 2012). Trenkel (2010) also notes, that PCU is typically threefold higher in price. In southern Alberta, approximate costs (\$CAD Mg⁻¹) for urea and common EEFs are: 1100 – urea; 1223.5 – Agrotain®; 1102.6 – eNtrench®; 1200 – SuperU®; 1240 – ESN® (Fast 2023). Lastly, and despite higher initial costs, SRFs can be cost reducing overall since they can facilitate a single N application and thereby reduce fuel and labour costs associated with split-applications (Akiyama et al. 2010). This same logic has been outlined by Trenkel (2010) for NIs and could also be applied to UIs and DIs. As concluded by Buresh and Baanante (1993), EEFs have the ability to be cost effective in environments with high N fertilizer response, high price of conventional fertilizers, and a large degree of preventable N loss from the use of EEFs.

1.7.2 Coating damage

Another challenge with the use of pre-coated EEFs, is the possibility of coating damage. Before an EEF reaches the soil, it must pass through various machinery and be handled several times. Since ESN® is the most widely used EEF with a factory fertilizer coating, it has been the main subject of investigation. Beres et al. (2012b) evaluated handling damage to the polymer coating of ESN®; in which, they concluded that substantial abrasion occurred when the product was transferred through machinery with scaly deposits, used in seeders configured with manifold-header systems operating at high air fan speeds, and broadcasted via an air boom applicator. The increased coating damage relayed into greater N release and lesser performance

as a SRF (Beres et al. 2012b). Similar results relating to air boom applicator damage of PCU has been observed by others (Parish 2001a; b; Bierman et al. 2015). Since coating damaged-SRFs allow for greater N release, consequences can transpire if used improperly. Coating damaged-ESN® can also reduce seed safety if applied in the seed row (Beres et al. 2012b), or possibly increase the risk of volatilization if surface-applied. In southern Alberta, the application of seed-row placed ESN® with increased N release (comparable to increased coating damage from excessive handling (Beres et al. 2012b)), reduced plant stand establishment in CWRS, triticale (*X Triticosecale* Wittmack) and canola (Qin et al. 2014). These effects were also exacerbated by increasing N rates and cereal grain yields were greatly hindered (Qin et al. 2014).

1.8 Summary

Wheat production is an important component of global food security and is often limited by soil-available N. In western Canada, N is often supplied to cereal crops through fertilizer applications during seeding. Conventional N fertilizers can experience environmental N loss which also suppresses plant development and yield components. Enhanced efficiency fertilizers have the potential to mitigate N loss, thereby improving environmental health and optimizing crop production. Slow-release fertilizers can gradually release soluble N fertilizer at a rate which mimics the demand of the crop, synchronizing supply with seasonal plant uptake. Their release rates can also be too prolonged for optimal plant uptake and their coatings can be damaged from excessive handling. Urease inhibitors can preserve ureal integrity to avoid NH₃ volatilization loss and are most effective when used in alkaline soils or surface applications. Nitrification inhibitors can prevent the conversion of NH₄⁺ to NO₃⁻ and allow for greater NH₄⁺ adsorption to soil exchangeable sites. These inhibitors are most optimal for use in saturated soils prone to NO₃⁻ leaching or denitrification. A concern when using NIs is the possibility of pollution swapping, as they can increase the risk of NH₃ volatilization. This can also negate the reduction efforts of a UI component in a DI product. The functionality of EEFs is highly dependent on environmental conditions and varies between crop and cultivars. Higher costs of EEFs along with inconsistent reported benefits also suppress their widespread adoption. Furthermore, genotype and environment specific research is needed due to the wide variability of both factors influencing EEF efficacy; namely, modern CWRS cultivars in the major growing regions of western Canada.

1.9 Figures

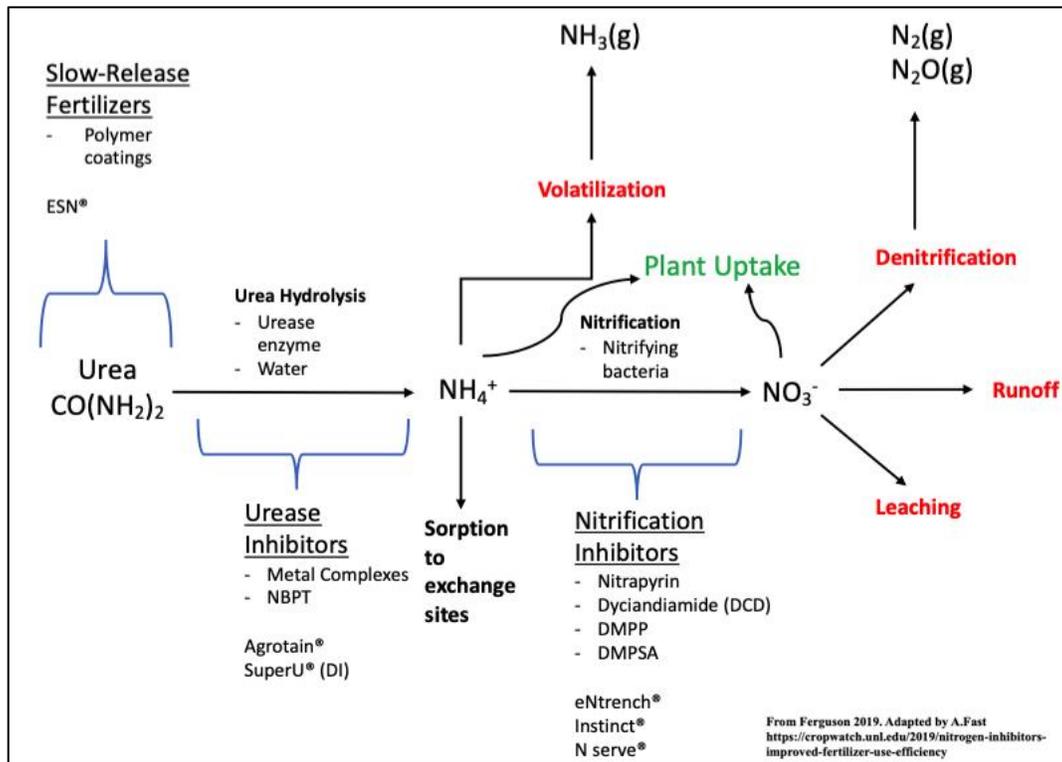


Figure 1. 1. Visual representation of the nitrogen cycle based on urea fertilizer application in cropping systems. **Red lettered** words equate to avenues of nitrogen loss. **Green lettered** words equate to nitrogen uptake by the plant. (Adapted by A. Fast from: Ferguson, R. 2019. <https://cropwatch.unl.edu/2019/nitrogen-inhibitors-improved-fertilizer-use-efficiency>)

1.10 Present study

1.10.1 Research objectives

Given the need to elucidate potential yield component benefits that enhanced efficiency fertilizers may afford Canada Western Red Spring wheat production in western Canada, the following two objectives of this research are outlined:

- 1) To determine if Canada Western Red Spring wheat yield components can benefit from the use of several common enhanced efficiency fertilizers over conventional urea.
- 2) To investigate the nitrogen requirements of a modern high-yielding Canada Western Red Spring wheat cultivar to optimize grain yield and protein content.

1.10.2 Null hypotheses

Based on the previously outlined objectives, their null hypotheses are summarized as:

- 1) Null: Enhanced efficiency fertilizers provide no benefit to Canada Western Red Spring wheat yield components relative to urea.
- 2) Null: Grain yield and protein content of a modern high-yielding Canada Western Red Spring wheat cultivar cannot be optimized with nitrogen management.

2.0 Integrating enhanced efficiency fertilizers and nitrogen rates to improve Canada Western Red Spring wheat production in the Canadian prairies.

2.1 Introduction

Bread or common wheat (*Triticum aestivum* L.) is one of three staple grain crops forming the basis of modern global diet and agriculture. Globally, 776 million Mg of wheat were produced in 2021 (United States Department of Agriculture [USDA] 2022), with nearly 22 million Mg (Statistics Canada 2022) grown in Canada. Of this, roughly 92% of Canadian wheat production occurred in the three prairie provinces of Alberta, Saskatchewan, and Manitoba (Statistics Canada 2021). In Canada, nine market classes of wheat are grown with the Canada Western Red Spring (CWRS) class the largest (McCallum and DePauw 2008). Grain protein content is typically regarded as the single most important quality characteristic of the CWRS class (Hucl et al. 2022). Furthermore, CWRS is typically grown for high volume pan bread production or used in blending (Iqbal et al. 2016). A minimum protein content of 10.0% is required for CWRS no. 1 grading in Canada (Canadian Grain Commission 2022); however, markets are often based on a protein content of 13.5%. Premium pricing can be applied to wheat that overachieve this mark. Achieving greater protein content without compromising grain yield is an important target of CWRS production.

Adequate nitrogen (N) fertilization is required to realize high protein content in CWRS wheat. A conventional wheat crop in western Canada will partition approximately 95 kg ha⁻¹ of aboveground N between its seed (67 kg ha⁻¹) and straw (28 kg ha⁻¹) (Karamanos 2013); however, higher overall N availability must be facilitated due to variable (25-50%) cereal N use efficiency (Kubota et al. 2017). The two primary sources of synthetic N fertilizer in Canada are urea and anhydrous ammonia. In western Canada, nearly 20% of growers use anhydrous ammonia (Lyseng 2018), while the remaining majority mostly use urea. The conventional method of applying N fertilizer in Canada achieved with granular urea at or near the time of planting (Ma et al. 2006). Optimal CWRS and Hard Red Spring (HRS) wheat application rates for urea have been previously published. These reports focus primarily on optimizing both grain yield and protein content. Karamanos et al. (2014) reported optimal CWRS (cv. CDC Imagine) grain yield return with a N rate of 80 kg N ha⁻¹, while continually increasing N rate linearly increased protein content in central Saskatchewan. A southern Alberta study reported similar grain protein

content trends; however, a 90 kg N ha⁻¹ rate achieved greatest grain yield in CWRS (cv. AC Lillian) (Beres et al. 2012). A HRS (cv. Choteau) study led by Walsh et al. (2018) reported a decrease in grain yield and protein content when N rate increased from 90 to 135 kg N ha⁻¹ in northcentral Montana. Conversely, another similar study reported greatest grain yield and protein content at a rate of 140 kg N ha⁻¹ (Walsh and Walsh 2020).

Nitrogen loss is a reoccurring challenge associated with the use of synthetic N fertilizer. Ammonia (NH₃) volatilization, denitrification, and nitrate (NO₃⁻) leaching are the primary pathways of this N loss (Cameron et al. 2013). These losses reduce N availability and associated plant uptake, thereby reducing plant growth and yield components (Western Plant Health Association [wph] 2023). While both urea and anhydrous ammonia can experience each type of N loss, greater overall loss is typically observed when using anhydrous ammonia (Fernandez et al. 2015; Eagle et al. 2017). The use of enhanced efficiency fertilizers (EEFs) in on-farm nutrient management, is one method of combating ureal N loss. The two main purposes of EEFs are to: (i) reduce environmental nutrient loss and (ii) increase plant nutrient availability (Olson-Rutz et al. 2011). The labelling of an EEF can be applied to many nutrients; however, our focus is on urea-based EEFs (hereafter referred to as EEF(s)). Three principal EEF categories exist in western Canadian crop production. First, slow- or controlled-release fertilizers simply provide a rate of nutrient release that is slower than normal release fertilizers (i.e. urea) (Trenkel 2010). Next, urease inhibitors are compounds which delay the process of urea hydrolysis (Byrne et al. 2020). With this delay, greater time is afforded for the fertilizer to be incorporated into the soil and become adsorbed as ammonium (NH₄⁺) ions to exchangeable sites. Lastly, nitrification inhibitors suppress soil nitrifiers and delay the oxidation of NH₄⁺ (Subbarao et al. 2006). This facilitates a greater likelihood for the NH₄⁺ ions to be adsorbed to soil exchangeable sites, rather than be lost to the environment through denitrification or NO₃⁻ leaching (Degenhardt et al. 2016). Furthermore, there are also dual inhibitor EEFs containing both urease and nitrification inhibitor components.

Previous studies have examined the agronomic performance of several EEFs. A global meta-analysis by Thapa et al. (2016), reported a 7% overall cereal (maize, wheat, rice) crop yield improvement with the use of a nitrification inhibitor. Additionally, mean yields increased with

the use of a dual inhibitor in alkaline, (2.0%) coarse-textured (5.7%) and irrigated (2.0%) soils only, while slow-release fertilizers did not improve yield (Thapa et al. 2016). In western Canadian winter wheat (cvs. AC Radiant, CDC Ptarmigan, AAC Wildfire), stabilized and improved grain yield and protein content resulted from the use of a dual inhibitor (SuperU®) over other EEFs and urea (Beres et al. 2018; Wang et al. 2022). Positive grain yield response has also been observed with the substitution (50 or 100%) of urea with a slow-release fertilizer (ESN®) in Montana (Walsh and Girma 2016) and southern Alberta (Beres et al. 2010). Lastly, canola (*Brassica napus* L.) yield improvements of up to 10% were reported in one third of encountered site-years in Alberta and Saskatchewan, with the use of a slow-release fertilizer (ESN®) instead of urea (Blackshaw et al. 2011).

An experiment was initiated to determine the yield component benefits that EEFs may afford CWRS production in western Canada. The objectives of this experiment were twofold: (i) to determine if CWRS yield components can benefit from the use of several common EEFs over conventional urea; and (ii) investigate the N requirements of a modern high-yielding CWRS cultivar to optimize grain yield and protein content. Correspondingly, our hypotheses were: (i) that EEFs may facilitate greater CWRS production over urea; and (ii) that an increased N rate may be required to realize grain yield and protein content in a modern high-yielding CWRS cultivar.

2.2 Materials and Methods

2.2.1 Experimental site description, design, and agronomic management

A field experiment was conducted at six sites across western Canada from 2019-2021, generating a total of 18 site-years. Site location varied slightly by year but were situated near the communities of Beaverlodge, Barrhead, Edmonton, Lethbridge and Vermilion, AB, and Indian Head and Scott, SK (Figure 2.1). Soil and environment data for each site are listed in Table 2.1. Agronomic, soil and environment data for each site-year are listed in Table 2.2. The treatment structure consisted of a factorial randomized complete block design with 25 total treatments per block, with four blocks per site. Treatment combinations were comprised of six urea types and four N rates, with an additional null control treatment plot that did not receive any N fertilizer (hereafter referred to as 0-N applied). A modern high-yielding CWRS cultivar, ‘AAC Viewfield’ (Cuthbert et al. 2019) was managed with six types of urea: (i) urea; (ii) urea + urease inhibitor

(Agrotain®); (iii) urea + nitrification inhibitor (eNtrench®); (iv) urea + dual inhibitor (SuperU®); (v) urea + dual inhibitor (NBPT/DMPSA); and (vi) slow-release fertilizer (Environmentally Smart Nitrogen®(ESN®)) in a 75:25 blend with urea (Table 2.3). The N rate treatment consisted of four fertilizer rates applied entirely at planting in mid- or side-row fertilizer bands: (i) 60; (ii) 120; (iii) 180; and (iv) 240kg N ha⁻¹. Other macronutrient fertilizer amendments were made based on pre-plant soil fertility testing (PRS® Soil Test System - Western Ag Labs, Saskatoon, SK, Canada). If needed, the applied fertilizer forms were triple superphosphate (0-45-0) (The Mosaic Company, Tampa, FL, USA), potassium chloride (0-0-60) (The Mosaic Company, Tampa, FL, USA), and magnesium sulfate (0-0-0-12) (Agriculture Solutions Inc., Sebringville, ON, Canada).

Configurations of seeding equipment differed by site, but resembled the drill designed and built by Agriculture and Agri-Food Canada at the Lethbridge Research and Development Center. This drill utilized ConservaPak™ knife openers (8) (Model CP129, Vale Industries, Indian Head, SK, Canada) spaced at 24cm, a Valmar™ air delivery system (Valmar Air Inc., Ellie, MB, Canada), a Raven™ hydraulic seed calibration and product control system (Raven Industries Inc, Sioux Falls, SD, USA) and Morris™ seed cups (Morris Industries Ltd., Saskatoon, SK, Canada). Prior to planting, all seed was treated with a fungicide to protect against seed- and soil-borne diseases at a rate of 325 mL 100kg of seed⁻¹ (Raxil PRO—tebuconazole [(RS)-1-*p*-chlorophenyl-4,4-dimethyl-3-(1H-1,2,4-triazol-1-ylmethyl)pentan-3-ol] 3.0 g L⁻¹ + prothioconazole [(RS)-2-[2-(1-chlorocyclopropyl)-3-(2-chlorophenyl)-2-hydroxypropyl]-2,4-dihydro-1,2,4-triazole-3-thione] 15.4 g L⁻¹ + metalaxyl [metyl *N*- (methoxyacetyl)-*N*-2,6-xylyl-DL-alanite] 6.2 g L⁻¹ Bayer Crop Science Canada Inc., Calgary, AB, Canada). Plots were seeded directly into previous crop stubble in a uniform fashion to achieve a desired seeding rate of 400 seeds m⁻². Plots were seeded to a desired length with an additional 50cm on either end and were then trimmed or rototilled to create the final desired plot length prior to harvest. Total plot area varied by site-year but ranged between 4.59-23.16m². Preceding crop stubble consisted of either canola, barley (*Hordeum vulgare* L.) silage, or chemical fallow, with the aim of creating a low background soil N environment; no experimental plots were seeded into wheat stubble.

Weed control varied by site, weed prevalence, and encountered conditions. The Biologische Bundesanstalt, Bundessortenamt und Chemical Industry (BBCH) crop growth staging scale was used to determine key wheat growth stages throughout the growing season (Federal Biological Research Centre for Agriculture and Forestry 2001). Herbicide application typically consisted of one pre-plant and two in-crop applications (BBCH 12-22) (if needed) based on full label rates. At BBCH 30-32, a plant growth regulator was applied to all plots at a rate of 1.8 L ha⁻¹ (Manipulator--chlormequat chloride (CCC) [2-chloroethyl-trimethyl-ammonium chloride] Eastman Chemical Company, Kingsport, TN, USA). A fungicide application near heading timing (BBCH 61-63) was made based on site conditions and disease presence. A pre-harvest desiccant was applied when grain moisture content was <30% to assist with harvest management if needed (RoundUp--glyphosate [N-(phosphonomethyl) glycine] Bayer Crop Science Canada Inc., Calgary, AB, Canada). All post-emergent herbicide and fungicide applications were made using motorized sprayers calibrated to deliver a carrier volume of 45 L ha⁻¹ at 275 kPa pressure.

2.2.2 Data collection

Post emergent plant counts were performed between BBCH 20-29 to determine the number of viable plants by staking and counting 1 m sections of the second, third, second last and third last rows of the plot. Plant density (plants m⁻²) were calculated by dividing the sum of viable plants counted, by the row spacing multiplied by the number of rows counted. These same row locations were counted again to determine spike density (heads m⁻²), and heads (n plant⁻¹) were calculated by dividing the number of heads by the number of plants in each staked row section. Above ground plant biomass was harvested by hand using sickle type instruments. Quadrats were placed near the previously staked portions of the plots, to determine the area to be harvested. Biomass harvest area varied by plot size, between 0.25-1.22 m² among sites. Harvested biomass bundles were weighed (g) prior to and after a dry down period. Dried bundles were threshed with a stationary thresher and the harvested grain was weighed (g). Harvest index was then obtained by dividing grain weight (g) by the dried biomass weight (g) of each sample. Days to maturity was determined when the majority of the plot reached physiological maturity, where kernel moisture in the lower third of a spike was <40% and kernels could not be easily severed when pinched between thumb and fingernails.

The entirety of each plot was harvested using a Wintersteiger Nurserymaster Elite (Wintersteiger AG Salt Lake City, UT, USA) or similar plot combine equipped with a straight cut header, pickup reel, and crop lifters. Grain yield was determined for each plot by weighing dried samples and were corrected to 13.5% grain moisture content. These values were used to calculate total grain yield (Mg ha^{-1}). A two kg subsample of grain was retained to determine seed weight ($\text{g 1000 kernels}^{-1}$) and test weight (kg hL^{-1}) as per industry standards (Canadian Grain Commission 2022). Whole grain protein concentration was determined using near infrared reflectance spectroscopy technology from the same subsample (Foss Decater GrainSpec, Foss Food Technology Inc., Eden Prairie, MN, USA).

2.2.3 Data analysis

There was a total of 18 site-years in this experiment. The Lethbridge rain-fed/dry and Vermilion sites in 2021 suffered severe growth impairment due to drought and heat, and their data were removed from analysis. Due to supply issues, one of the original urea type treatments (urea + urease & nitrification inhibitor (NBPT/DMPSA)) changed inhibitor composition across years and was also removed from analysis. The following two analyses are therefore based on 16 site-years, five urea types and four N rates. The first analysis was performed with all data, including the 0-N applied plots. This analysis compared plots with 0-N applied to the average response of all treatments with N applied, to determine initial differences between the application of N fertilizer or lack thereof (Table 2.4). The second analysis included only the full factorial portion of the experiment and excluded the 0-N applied plots. This analysis was performed to elucidate differences in yield components between different N management treatments in CWRS production (Tables 5, 6 and 7). Data were split into and presented as two groups based on soil zone (Table 2.1), due to similar biological conditions and treatment response trends. The Lethbridge (Irrigated and dry), AB and Scott, SK site-years (7) were combined to form the Dark Brown Chernozem Soil group. The Barrhead, Beaverlodge, Edmonton and Vermilion, AB, and Indian Head, SK site-years (9) were combined to form the Black Chernozem & Dark Grey Luvisol Soil group.

Data were analyzed using the MIXED procedure of SAS (version 9.4, Cary, NC, USA). Homogeneity of variance was tested, and any outlier observations detected using tests for normality were removed prior to analysis using the UNIVARIATE procedure (SAS Institute Inc 2019). Environment (site-year), replication and associated interactions were considered random effects, while treatment effects (Urea type, N rate, Urea type×N rate) were considered fixed and significant if $P \leq 0.05$ (Steel et al. 1997). A Kenward-Roger approximation for degrees of freedom was used. If fixed factors were declared significant, mean separation tests were performed using Fisher's protected LSD. The LSMEANS statement was used to generate least squares means of the fixed effects (SAS Institute Inc 2019). The CONTRAST statement was used to perform a regression analysis to determine linear, quadratic, and cubic relationships between N rate and the response variables (SAS Institute Inc 2019). The CONTRAST statement was also used to conduct single degree of freedom contrasts between plots with N applied and 0-N applied (SAS Institute Inc 2019).

A grouping methodology was used to explore system responses and variability of 'AAC Viewfield' grain yield (Francis and Kannenberg 1978). The means and coefficients of variation (CV) were estimated for each combination of treatments. Means were plotted against CV for each combination of treatments. The overall treatment mean, and CV were used to categorize the biplot data into four quadrants: high mean and low variability (Group I), high mean and high variability (Group II), low mean and high variability (Group III) and low mean and low variability (Group IV). Mean-CV biplots provide a general overview of system stability; thereby, supplementing the most important response variable results (i.e., grain yield) (Wang et al. 2022).

A simple economic analysis was conducted to determine net return associated with increasing N rate, based on the N rates and grain yield results of this experiment. A CWRS wheat price of \$435.68 Mg⁻¹ was obtained from Alberta market values for CWRS no.1 13.5% protein content (Alberta Wheat Commission 2022). Fertilizer costs were based on December 2022 quotes obtained from agricultural input suppliers and manufacturer representatives located nearest to Lethbridge, AB (Table 2.1). Net return was calculated as:

$$N = YP - CR$$

where N is the net return (\$CAD ha⁻¹), Y is crop yield (Mg ha⁻¹), P is CWRS price (Mg⁻¹), C is cost of urea (Mg⁻¹), and R is N rate (Mg N ha⁻¹).

A second analysis was conducted based on urea type and grain yield results of this experiment in the Dark Brown Chernozem Soils at a N rate of 120 kg N ha⁻¹. Net return was calculated as:

$$N = YP - CR$$

where N is the net return (\$CAD ha⁻¹), Y is crop yield (Mg ha⁻¹), P is CWRS price (Mg⁻¹), C is cost of each urea type (Mg⁻¹), and R is N rate (0.12 Mg N ha⁻¹). These equations were adapted from other agronomic studies (O'Donovan et al. 2001; Mason et al. 2007; Beres et al. 2018).

2.3 Results

2.3.1 Environmental conditions

A wide array of weather conditions occurred during the different growing seasons of this experiment (Table 2.2). At all sites in 2019, normal precipitation was exceeded. Conversely, in 2021, all sites received below normal water input, except for the Lethbridge irrigated site. The Edmonton, Vermilion, and Scott locations received above-normal precipitation in 2020, while the Lethbridge dry site received near normal rainfall. Indian Head in 2020 benefited from soil moisture reserves gained in the previous growing season, despite receiving slightly under half of the normal precipitation that year. Growing degree days at Beaverlodge and Indian Head in 2021 were greater than 110% of their normal values, while all other sites-years remained within 10% of normal. Furthermore, all sites in 2019 and 2021 under- and over-achieved their growing degree days, respectively. Mean temperatures for all sites in 2019 and Indian Head in 2020 were below normal, while all other site-years experienced above average temperatures. All planting and harvest dates in 2019 and 2021 reflected regionally-adapted practices; however, in 2020, several locations (Lethbridge Irrigated & Dry, Indian Head, and Scott) were delayed by up to four weeks due to workplace logistical constraints stemming from the onset of the COVID-19 pandemic.

2.3.2 Plant stand

In both soil zones, spike density ($P < 0.05$) and heads plant⁻¹ ($P < 0.05$) increased when N was applied; however, plant density did not increase with the addition of applied N (Table 2.4). Plant density was affected by urea type in the Dark Brown Chernozem soils ($P < 0.05$) (Table 2.5). The urease inhibitor treatment increased plant density more so than the nitrification inhibitor or slow-release fertilizer, while the dual inhibitor attained similar results to both groups (Table 2.6). Urea achieved the lowest plant density; however, remained comparable to the nitrification inhibitor and slow-release fertilizer treatments. In the Black Chernozem & Dark Grey Luvisol soils, plant density was unaltered by urea type; however, spike density was greatest ($P < 0.0001$) when fertilized with a dual or urease inhibitor, and lowest when supplied with a nitrification inhibitor, slow-release fertilizer, or urea (Table 2.7). Spike density remained unaltered by urea type in the Dark Brown Chernozem soil group (Table 2.5). The greatest number of heads plant⁻¹ in the Dark Brown Chernozem soils was seen in the urea, nitrification, and dual inhibitor treatments (Table 2.6). Conversely, in the Black Chernozem & Dark Grey Luvisol soils, the nitrification inhibitor treatment solely produced the greatest heads plant⁻¹ (Table 2.7). The lowest number of heads plant⁻¹ was observed in the urease inhibitor and slow-release fertilizer treatments in the Dark Brown ($P < 0.01$) and Black Chernozem & Dark Grey Luvisol soils ($P < 0.05$), respectively.

Plant density decreased linearly ($P < 0.01$) while heads plant⁻¹ increased linearly ($P < 0.01$) as N rate increased in the Dark Brown Chernozem soil zone (Table 2.6, Figure 2.2). Spike density remained unaltered by changes in N rate (Table 2.5). Plant density in the Black Chernozem & Dark Grey Luvisol soil zone was greatest at the 120 kg N ha⁻¹ rate and lowest at the 60 kg N ha⁻¹ rate, while the other rates were similar to both groups illustrating an overall cubic relationship ($P < 0.05$) (Table 2.7, Figure 2.2). Spike density increased curvilinearly ($P < 0.05$) and heads plant⁻¹ ($P < 0.01$) increased linearly with increasing N rate, respectively (Table 2.7, Figure 2.2).

2.3.3 Grain yield and quality

Test weight, thousand kernel weight and harvest index did not improve with the application of N in either soil zone; however, grain yield ($P < 0.01$) and protein content ($P < 0.01$) were improved (Table 2.4). In the Dark Brown Chernozem soil zone, increasing N rate linearly

decreased test weight ($P < 0.05$), while thousand kernel weight and harvest index were unaltered (Table 2.6, Figure 2.3). Conversely, grain yield ($P < 0.01$) and protein content ($P < 0.0001$) increased linearly with increasing N rate (Table 2.6, Figure 2.4). Furthermore, grain yield increased curvilinearly due to the significance of its quadratic relationship ($P < 0.05$) with N rate, where yields reduced as N rates increased from 180 to 240 kg N ha⁻¹. In the Black Chernozem & Dark Grey Luvisol soil zone, similar linear ($P < 0.01$) and quadratic ($P < 0.05$) relationships were seen between N rate and grain yield (Table 2.7, Figure 2.4). In each soil zone, the three highest N rates improved grain yield relative to the 60 kg N ha⁻¹ rate, while maintaining similar results between them. The linear relationships in grain protein content ($P < 0.0001$) and test weight ($P < 0.001$) were also similar between soil groups (Tables 6 and 7, Figures 2.3 and 2.4). Furthermore, thousand kernel weight and harvest index were not affected by N rate in either soil zone.

Grain yield differed based on urea type in the Dark Brown Chernozem soils only ($P < 0.05$) (Table 2.5). Here, the greatest and lowest grain yields were achieved with the dual inhibitor and slow-release fertilizer, respectively (Table 2.6, Figure 2.5). All urea types shared similarity to at least one other; the following list ranks them in descending grain yield order: dual inhibitor, urease inhibitor, urea, nitrification inhibitor, and slow-release fertilizer (Table 2.6, Figure 2.5). Urea type was also not impactful on grain protein content, test weight, thousand kernel weight or harvest index in the Dark Brown Chernozems (Table 2.6). In the Black Chernozem & Dark Grey Luvisol soil zone, urea type did not alter any grain yield or quality characteristics. Despite a lack of statistical difference to the other urea types, the slow-release fertilizer treatment in the Black Chernozem & Dark Grey Luvisol soils returned the lowest grain yield similar to the Dark Brown Chernozem soil group (Table 2.7).

Biplot analysis in both soil zones indicated greater grain yield and lower variability with a N rate of 120 kg N ha⁻¹ using a dual or urease inhibitor, or with a N rate of 180 kg N ha⁻¹ using a NI inhibitor (Figure 2.6, Group I). In the Dark Brown soils, other Group I treatment combinations included urea applied at 120 and 180 kg N ha⁻¹, and the slow-release fertilizer applied at 240 kg N ha⁻¹ (Figure 2.6). In the Black Chernozem & Dark Grey Luvisol soils, a nitrification inhibitor applied at 180 kg N ha⁻¹ also was categorized in Group I (Figure 2.6). The

treatment combinations with the lowest grain yield and highest variability in both soil zones included the nitrification inhibitor applied at 60 kg N ha⁻¹ and the slow-release fertilizer applied at 120 kg N ha⁻¹ (Figure 2.6, Group III). Additionally, the nitrification inhibitor applied at 240 kg N ha⁻¹ and the slow-release fertilizer applied at 60 and 180 kg N ha⁻¹ were categorized in Group III in the Dark Brown Chernozems.

Treatment interactions (Urea type×N rate (T×R)) were not deemed significant for any variable in either soil group (Table 2.5). Mean grain yield, protein content, plant and spike density, and heads plant⁻¹, were greater with N applied than 0-N applied in both soil groups (Table 2.4). Conversely, test weights were greater with 0-N applied in both groups, thousand kernel weight was greater with N applied in the Black Chernozem & Dark Grey Luvisol soils only, and harvest indices were similar in the Dark Brown Chernozem but greater with 0-N applied in the Black Chernozem & Dark Grey Luvisol soils (Table 2.4).

In the Dark Brown Chernozem soil zone, conventional urea achieved a net return of \$2394.9 ha⁻¹ (based on a CWRS price of \$435.68 Mg⁻¹ (Alberta Wheat Commission 2022) and fertilizer costs listed in Table 2.3) (Figure 2.7). Relative to this, the urease and dual inhibitor treatments increased net return by \$33.1 and \$70.8 ha⁻¹, respectively. Net return was also reduced by \$9.0 and \$51.8 ha⁻¹ with the use of nitrification inhibitor or a slow-release fertilizer, respectively. Focusing on N rate, net return increased up to 120 kg N ha⁻¹, beyond which diminishing returns were observed independent of soil zone (Figure 2.8).

2.4 Discussion

2.4.1 Enhanced efficiency fertilizers

2.4.1.1 Plant stand

The primary uses of EEFs in crop production are twofold: (i) to reduce N loss and (ii) increase N availability for plants (Olson-Rutz et al. 2011). With N being regarded as the most important plant nutrient responsible for growth (Shah 2008), increasing N supply can therefore translate into increasing plant growth. Plant density was lowest in the slow-release fertilizer, nitrification inhibitor and urea treatments in the Dark Brown Chernozem soils. The urease and dual inhibitors improved upon urea, thereby suggesting a urease inhibitor component can facilitate greater early season N availability and plant uptake. Since slow-release fertilizers are

designed to prolong N release as the growing season progresses (Golden et al. 2011), early-season N availability can be impaired. It is likely that lower initial N availability decreased plant density with the use of the slow-release fertilizer relative to the urease or dual inhibitors. Despite the nitrification inhibitor producing relatively lower density, it remained on par with urea. Karamanos et al. (2004) observed greater plant density with a seed-placed urease inhibitor in several crops relative to urea, while others have found no difference when urea or slow-release fertilizers were side-banded in CWRS (Mangin et al. 2022b) or winter wheat (McKenzie et al. 2007; Beres et al. 2010; Beres et al. 2018). The lack of a difference in plant density due to urea type in the Black Chernozem & Dark Grey Luvisol soils, can suggest that greater mineralization rates stemming from higher organic matter concentrations and residual soil-N (Grzyb et al. 2020) provided sufficient N at the start of the growing season for adequate and indistinguishable stand establishment.

Spike density was unaffected by urea type in the Dark Brown Chernozems; however, in both soil groups, N applied produced a higher density than 0-N applied, similar to the findings of Mangin et al. (2022b). In the Black Chernozem & Dark Grey Luvisol soils, the slow-release fertilizer produced the lowest spike density and heads plant⁻¹; credible to insufficient early-season N availability as previously mentioned. With limited early season N, reduced heads plant⁻¹ have been reported from increased tiller mortality (Longnecker et al. 1993; Otteson et al. 2008). These results support the findings of several western Canadian winter wheat experiments, where other common EEFs and urea produced greater heads plant⁻¹ than a slow-release fertilizer (Beres et al. 2018; Wang et al. 2022).

2.4.1.2 Grain yield and quality

Grain protein content, test weight, thousand kernel weight and harvest index were unaffected by urea type, regardless of soil zone. Since these variables remained unchanged, all urea types were likely sufficient in facilitating N availability and plant uptake throughout the growing season to substantiate similar grain quality (Brown et al. 2005). Additionally, conducive environmental conditions to promote N loss were likely not encountered; thereby, reducing efficacy of these EEFs (Verburg et al. 2022). Grain yield and protein content are the primary focus of publications centered on EEF agronomic performance and other yield components such

as test weight, thousand kernel weight, and harvest index, are typically excluded from their analyses. Despite this, Mangin et al. (2022b) reported no difference in thousand kernel weight between three urea type treatments. Furthermore, inconsistent differences have been reported between winter wheat test weight and thousand kernel weight among several liquid and granular EEFs (Owens et al. 2022; Wang et al. 2022).

In western Canada, unremarkable improvement in grain protein content has been reported with the use of several EEFs in winter wheat studies (McKenzie et al. 2007; McKenzie et al. 2010; Wang et al. 2022). Furthermore, Beres et al. (2010) observed a protein penalty with the use of a slow-release fertilizer:urea blend. Spring wheat (CWRS and HRS) protein content has also been reported to lack any benefits from the applications of EEFs (Thapa et al. 2015; Afshar et al. 2021; Mangin et al. 2022a), which is supported by these results. Mixed reviews on grain yield in spring and winter wheat supplied with EEFs are also frequent in publication, where some studies report improvement over conventional urea (Beres et al. 2018; Wang et al. 2022) and others don't (McKenzie et al. 2007; Afshar et al. 2021; Mangin et al. 2022a). In our experiment, urea type affected grain yield in the Dark Brown Chernozem soils only. Here, the dual inhibitor treatment increased grain yield relative to urea. Additionally, in both soils, the dual inhibitor applied at a rate of 120 kg N ha⁻¹ resulted in greatest stability (Figure 2.6, Group I). Beres et al. (2018) and Wang et al. (2022) reported similar findings in winter wheat, where a fall applied dual inhibitor produced greater yield and increased stability. A greater overall reduction in N loss is expected with the use of a dual inhibitor than either of its components by themselves. This likely translates to greater overall N uptake by the plant and is credible to the yield and stability increase observed here. The other EEFs attained similar results to urea. The lowest overall grain yield was provided by the slow-release fertilizer, while also reducing grain yield relative to the dual and urease inhibitor treatments. This suggests that relatively inferior plant N uptake and grain partitioning occurred, likely attributable to inadequate N availability from the overly prolonged release of the slow-release fertilizer in Dark Brown Chernozem soils (McKenzie et al. 2007), when applied in a separate fertilizer row. The N release of a slow-release fertilizer is dependent on increasing temperature (Golden et al. 2011) and available moisture to imbibe the granular coating (Verburg et al. 2021). An insufficiency of either requirement can lead to an overly-prolonged N release rate (Golden et al. 2011); thereby, hindering bioavailability during

the earlier stages of the growing season. Wheat grain yield and N use efficiency become impaired without adequate N supply prior to the accelerated N uptake phase (Howard et al. 2002), which may be compromised with a slow-release fertilizer applied in separate fertilizer rows. Additionally, in the Dark Brown Chernozems, the slow-release fertilizer applied at 60-180 kg N ha⁻¹ resulted greater instability (Figure 2.6, Group III). Lastly, application timing of EEFs in CWRS has been investigated in a companion study of our experiment. There, preliminary results show no difference in grain yield or protein content with the use of different EEFs (Beres 2022). Additionally, greatest return and stability regarding yield components have been observed when applying N entirely at seeding instead of two or three split applications (Beres 2022). These results agree with the findings of Walsh et al. (2018) and the fundamental N application method of this experiment.

2.4.1.3 Fertilizer efficacy or lack thereof

In the field of agronomy, agricultural products are typically judged on their ability to improve a growers return on investment. This is achieved primarily with improvements in grain yield or protein content in wheat production. Mixed reviews of EEF agronomic performance continue to limit their widespread adoption in modern crop production. These more or less negative reports typically stem from a reoccurring lack of conducive environmental conditions for N loss (Buresh and Baanante 1993; Li et al. 2018; Verburg et al. 2022). Evidently, an EEF designed to limit a certain avenue of N loss will not perform well, without having encountered environmental conditions that promote that certain avenue of N loss. Modern farming practices such as no-till seeding, diverse crop rotations and the incorporation of 4R nutrient stewardship principles already help growers reduce the likelihood of significant N loss; moreover, side-banding N fertilizer is an effective method for reducing gaseous N loss relative to surface applications (Snyder et al. 2009). Additionally, reduced yield components from N loss can be masked by the continual use of (overly) high N rates (Li et al. 2018). Greater crop N availability can be facilitated from increasing residual soil-N and mineralization rates (Grzyb et al. 2020). Consequently, this can balance out encountered N loss to the extent where crop performance is not impaired (Li et al. 2018). Gauging the use of EEFs based solely on variables such as yield, is difficult to accomplish when their ability to reduce unnoticeable N loss (i.e., nitrous oxide (N₂O) emissions) may also be present (Snyder et al. 2009; Fan et al. 2022). Certain reports are

advocating for the social cost of EFFs (i.e., ability to reduce N₂O emissions) to be included in future agricultural policy; namely, in the form of subsidies or tax exemptions which would promote their adoption (Gu et al. 2021; Lam et al. 2022). Additional information regarding why EEF benefits are inconsistent in field experiments can be found in the reports of Li et al. (2018) and Verburg et al. (2022).

2.4.2 Increasing nitrogen rates

2.4.2.1 Plant stand

In the Dark Brown Chernozem soil zone, plant density decreased linearly with increasing N rate; moreover, a cubic relationship between N rate and plant density was observed in the Black Chernozem & Dark Grey Luvisols (Figure 2.2). In a laboratory study, wheat plant growth impairment has been reported with high rates of N supply stemming from reduced carbon fixation and distribution in photosynthesis, and lower plant nutrient uptake overall (Guo et al. 2019). Furthermore, these consequences were exacerbated with NH₄⁺ supply causing toxic effects (Guo et al. 2019). In field experiments, increased seedling damage in spring wheat has been associated with NH₃⁻ forming fertilizers placed closer to the seed (Deibert 1993; Mooleki et al. 2010). Despite this, modern air-delivered seed drills are typically proficient in providing adequate seed to fertilizer separation in mid- or side-row bands (Mooleki et al. 2010). Additional reports of unaltered plant density in response to the amount of N applied in separate fertilizers rows are commonplace (McKenzie et al. 2007; Otteson et al. 2008; Beres et al. 2012; Mangin et al. 2022b).

As expected and observed by others (Abad et al. 2005; Otteson et al. 2008; Beres et al. 2012), a linear increase in spike density was apparent with increasing N rate in the Black Chernozem & Dark Grey Luvisol soil group (Table 2.7). Despite no difference in spike density in the Dark Brown Chernozems, both soil groups displayed linear increases in heads plant⁻¹ with increasing N rate. This suggests that lower spikes per area are associated with decreasing N availability. Decreased tillering and associated heads plant⁻¹ have been reported with reduced early season N availability, stemming from lower N rates (Longnecker et al. 1993; Otteson et al. 2008). Conversely, greater tillers, heads, and kernels, have been observed with more intensive bread wheat fertilization (Abad et al. 2005).

2.4.2.2 Grain yield and quality

In both soil zones, thousand kernel weight and harvest index did not differ with respect to N rate. This contests previous results (Otteson et al. 2007; Beres et al. 2012), where thousand kernel weight decreased with increasing N rate; however, our findings support another similar study (Otteson et al. 2008). Wheat yield components such as thousand kernel weight and harvest index, have been described to be dependent on and affected by environmental conditions at relevant developmental stages overtop of N application method (Koppensteiner et al. 2022). The indifference of these variables to increasing N rate can possibly be attributable to environmental idiosyncrasies encountered during pertinent growth stages. Nyiraneza et al. (2012) also noted thousand kernel weight response to differ based on soil texture; whereby, loamy soils benefited, and clayey or sandy soils were hindered with increasing N rates. Harvest index in wheat has also been reported to remain unchanged with additional N, due to the simultaneous increase in grain weight and dry plant matter production (Davidson and Campbell 1984; Koppensteiner et al. 2022). Both soil zones followed similar linear decreases in test weight as N rate increased, thereby supporting previous reports in HRS (Otteson et al. 2007; Walsh and Walsh 2020) and oat (*Avena sativa* L.) production (Lafond et al. 2013; May et al. 2020). Despite this downward trend, all treatments remained above the CWRS no. 1 test weight threshold grade of 75.0 kg hL⁻¹ (Canadian Grain Commission 2022). Furthermore, other Canadian prairie studies have observed no alterations in test weight based on increasing N rate (Beres et al. 2012; Hucl et al. 2022), while Beres et al. (2008) found modest improvements in Soft White Spring Wheat.

As expected, grain protein content increased linearly with N rate. This relationship is well defined and supported in previous studies (Beres et al. 2012; Walsh and Walsh 2020; Hucl et al. 2022). All treatments, including the 0-N applied plots, exceeded the minimum CWRS no. 1 protein grade in Canada of 10.0% (Canadian Grain Commission 2022). Furthermore, all applied N rates achieved greater than 12.0% protein, a common HRS quality level (U.S. Wheat Associates 2022), while rates ≥ 120 kg N ha⁻¹ surpassed the *standard* protein premium level of 13.5% for CWRS in Canada. Grain yield followed a curvilinear response to increasing N rate in both soil zones, supporting the findings of Nyiraneza et al. (2012) and Walsh et al. (2018). A large increase in grain yield from the 60 to 120 kg N ha⁻¹ rate, followed by reducing

improvements as N rate increased mirrors other findings (Walsh and Walsh 2020; Ghimire et al. 2021; Hucl et al. 2022). These observations exemplify the law of diminishing returns regarding wheat grain yield (Tilman et al. 2002), which can generally be credited to two related sources. First, an inverse relationship between grain yield and protein exists. Here, as N supply increases, N uptake and grain partitioning can shift towards protein development at the expense of augmenting yield (Iqbal et al. 2007). Observations of this effect are continually reported (Long et al. 2017; Corassa et al. 2018; Ghimire et al. 2021), despite individual variability based on wheat class and cultivar (Fowler 2003). Second as described by Martre et al. (2006), grain N is first supplied from excess stem N and N released through natural leaf senescence. Generally, this initial supply of N is insufficient and additional N is then remobilized throughout the plant and obtained through accelerated leaf senescence (Masclaux-Daubresse et al. 2010). This creates a *yield dilemma* since lower grain yields are often caused by greater leaf senescence and associated N remobilization during the same time as grain filling (Masclaux-Daubresse et al. 2010; Have et al. 2017). Furthermore, since whole-plant senescence overlaps with grain filling, quality factors such as test weight and thousand kernel weight can be negatively affected (Distelfeld et al. 2014), likely attributing to the inverse relationship between N rate and test weight observed here (Figure 2.3). Lastly, other reported sources of diminishing grain yield in cereals in response to increasing N supply include: lodging (Berry et al. 2004), soil acidification (Guo et al. 2010), and late maturity (Yang et al. 2017).

2.5 Economic implications

Broader adoption of EEFs has been limited due to mixed reviews on efficacy and greater input costs. Based on the results presented here, the use of a dual or urease inhibitor resulted in slightly higher net returns than urea in the Dark Brown Chernozem soil zone (Figure 2.7). Conversely, the slow-release fertilizer lowered net return relative to urea, supporting the results of Khakbazan et al. (2013). Net return calculations based on urea type are difficult to establish for broader interpretations. This analysis is limited since the values used are based on approximate quotes from three industry sources at a certain time (December 2022) and location (Lethbridge, AB). Nitrogen fertilizer costs change weekly and are different geographically. Additionally, certain EEFs (eNtrench®) require additional labour to impregnate urea granules with chemical, which is difficult to budget. Despite this, the range of net return relative to urea

was within approximately 70 \$ ha⁻¹, which could attract growers to use certain EEFs (urease or dual inhibitors) in the Dark Brown Chernozems. Lastly, net returns could increase further if the social cost of EEFs were factored into future agricultural policy (Gu et al. 2021).

Although a number of factors were not considered in either economic analysis (e.g., transportation costs, yield and market fluctuations, storage, chemical mixing), a N rate of 120 kg N ha⁻¹ appears to be most advantageous (Figure 2.8). Protein premiums were also not factored into this analysis as these values change yearly based on markets, geography and purchasing company; however, in recent years, premiums have been historically low on the order of three cents to zero cents (CAD ¢) per 1/10 percentage point of protein (Neil Blue, Alberta Agriculture and Irrigation, personal communication, 2023) and would likely not influence the overall economic trends observed here. The N rate results support the findings of Nyiraneza et al. (2012).

2.6 Recommendations

Several recommendations can be generated from the results of this investigation. Enhanced efficiency fertilizers provide valuable protection against N loss if the anticipated conditions of loss are encountered (Verburg et al. 2022); however, in terms of grain yield and quality in wheat, they are ineffectual when used in incorrect conditions. Despite not encountering favourable N loss scenarios, the use of EEFs will not result in any limitation to modern high-yielding CWRS production or grain quality when the fertilizer is applied in side- or mid-row bands. Modest agronomic and economic improvements are also possible across the different soil zones studied here. Furthermore, a dual inhibitor fertilizer likely provides optimal protection against overall N loss while delivering improved yield and net return benefits relative to urea in Dark Brown Chernozem soils (Beres et al. 2018; Wang et al. 2022). While the use of a slow-release fertilizer in the Dark Brown Chernozem soil zone did not reduce grain yield relative to urea, it did not achieve the same returns as some other EEFs. Additionally, the slow-release fertilizer and nitrification inhibitor achieved a lower net return compared urea. Others have noted that in colder and drier climates, the use of a slow-release fertilizer may be limiting (Golden et al. 2011; Wang et al. 2022). Grain yield was also improved with the use of higher N rates return to a point. More intensive N supply (>180 kg N ha⁻¹) proved diminishing to grain yield, while

initial increases provided greatest return (Nyiraneza et al. 2012). Grain yield stability was generally greater at rate N rate of 120 or 180 kg N ha⁻¹, however varied with urea type (Figure 2.6). Very high grain protein content (>15.0%) was also achievable with the most intensive N rate (240 kg N ha⁻¹), thereby supporting the notion that growers may pursue higher protein premiums with increased N fertilizer input if that is their focus. Increasing N rates up to 240 kg N ha⁻¹ is acceptable with regards to test weight, since CWRS no. 1 grading was maintained despite higher inputs resulting in slightly lower quality. In this study, the use of a 120 kg N ha⁻¹ rate is considered optimal for N application in side- or mid-row bands for CWRS production in Western Canada. This N rate achieved the greatest net return in both soil zones, while maintaining premium level protein content (Nyiraneza et al. 2012). Lastly, the application of N fertilizer is vital to increasing grain yield, protein content (Karamanos et al. 2014) and net return; however, the absence of applied N did not negatively affect other grain quality characteristics or plant density in this study.

2.7 Conclusions

Improved CWRS grain yield and protein content suitable for market, is largely dependent on appropriate N fertilization. Enhanced efficiency fertilizers were developed to mitigate environmental N loss, with hopes of affording greater plant N uptake relaying into increased yield components. Our results showed mixed yield component response in relation to EEF use in western Canadian CWRS wheat. The use of a dual inhibitor product in the Dark Brown Chernozem soil zone can provide increased grain yield returns over conventional urea, whereas other EEFs attained similar results. Net return improvements are possible with the use of a dual or urease inhibitor in the Dark Brown Chernozems. Grain yield was unaltered by using different EEFs in the Black Chernozem & Dark Grey Luvisol soil zone, while grain protein content remained unaffected independent of soil location. An optimal seeding N rate of 120 kg N ha⁻¹ in side- or mid-row bands was elucidated for balancing grain yield and stability, protein content, and net return. Additional N input also continued to provide greater grain protein content and would facilitate growers to pursue greater pricing premiums. The use of an EEF in conjunction with proper N rates can help optimize contemporary CWRS production in western Canada; however, conventional urea continues to maintain validity.

2.8 Acknowledgements

The authors would like to thank Dr. Sheri Strydhorst, Sarah Anderson, Greg Semach, Ryan Dyck, Steve Simmill, Warren Taylor, Laurel Thompson, J.P. Pettyjohn, Daniel Pucko, Chris Holzapfel, and Jessica Enns for their technical assistance. We also acknowledge Jens Klemptner, Brad Haubrich and Brent Nilsson for facilitating fertilizer cost information, and Neil Blue for protein premium pricing information. This research was funded by Alberta Agriculture and Forestry, the Alberta Wheat & Barley Commissions, the Saskatchewan Wheat Commission, NutrienTM, and CortevaTM Agriscience. In-kind funding was provided by Agriculture and Agri-Food Canada, Alberta Agriculture and Forestry, Lakeland College, and the University of Alberta.

2.9 Tables

Table 2. 1. Soil and environment data (May 1 to October 1) for experimental sites in Alberta and Saskatchewan, grouped by soil zone.

Location	Soil Zone	Soil Order	Soil Texture	Soil Organic Matter Content (%)	Soil pH	Normal Growing Degree Days ₅₀ ^{a,b}	Normal Precipitation (mm) ^a	Normal Temperature (°C) ^a
Dark Brown Chernozem Soils								
Lethbridge, AB	Dark Brown	Typic Boroll	Clay loam	2.7-2.9	6.3-8.0	2300	256	14.9
Scott, SK	Dark Brown	Typic Boroll	Sandy loam	3.7-4.1	5.3-6.3	2148	247	14.0
Black Chernozem & Dark Grey Luvisol Soils								
Barrhead, AB	Dark Grey	Boralfic Boroll	Loam	5.7	5.2	2083	302	13.4
Beaverlodge, AB	Dark Grey	Boralfic Boroll	Clay loam	7.7	6.4	1953	259	12.6
Edmonton, AB	Black	Udic Boroll	Silty clay	9.4-14.0	5.4-6.1	2155	282	13.8
Vermilion, AB	Black	Udic Boroll	Silt loam	4.6	5.8	2138	274	13.8
Indian Head, SK	Black	Udic Boroll	Clay	4.4-5.7	7.7-8.1	2103	275	14.7

^aNormal values were calculated using 30-yr historical climate norms from the nearest geographical provincial weather station (Alberta data obtained from <http://agriculture.alberta.ca/acis/weather-data-viewer.jsp>; Saskatchewan data obtained from https://climate.weather.gc.ca/climate_normals/index_e.html). ^bGrowing degree days were calculated using a base temperature of 0°C as described by Cao and Moss (1989).

Table 2. 2. Soil and environment data (May 1 to October 1) for experimental site-years (2019-2021) in Alberta and Saskatchewan, grouped by soil zone.

Location Year	Latitude, Longitude	Soil Cation Exchange Capacity (meq 100g ⁻¹)	Soil Nitrate Level 0-15cm (kg ha ⁻¹)	Seeding Date	Harvest Date	Growing Degree Days _{b0} ^{a,b}	Percent of Normal GDD _{b0} ^a	Total Precipitation (mm) ^a	Percent of Normal Precipitation (%) ^a	Mean Temperature (°C) ^a
Dark Brown Chernozem Soils										
Lethbridge, AB (Dry)										
2020	49°69'N, 112°76'W	23 ^c	46.0	26-May	12-Sep	2352	102	249	97	15.4
Lethbridge, AB (Irrigated)										
2019	49°42'N, 112°41'W	23 ^c	65.0	02-May	18-Sep	2255	98	370 (178) ^d	144	14.7
2020	49°42'N, 112°41'W	23 ^c	77.3	29-May	25-Sep	2352	102	350 (102) ^d	137	15.4
2021	49°42'N, 112°41'W	23 ^c	-	20-Apr	16-Aug	2496	108	334 (229) ^d	130	16.4
Scott, SK										
2019	52°22'N, 108°52'W	14	39.2	21-May	24-Sep	1997	93	278	112	13.0
2020	52°22'N, 108°52'W	11.9	15.7	02-Jun	06-Oct	2086	97	289	117	13.7
2021	52°22'N, 108°52'W	15.2	14.6	14-May	29-Aug	2301	107	155	63	15.1
Black Chernozem & Dark Grey Luvisol Soils										
Barrhead, AB										
2019	54°4'N, 114°21'W	12.4	78.7	13-May	10-Oct	2004	96	318	105	12.9
Beaverlodge, AB										
2021	55°11'N, 119°23'W	25 ^b	48.2	06-May	28-Aug	2165	111	177	68	14.3
Edmonton, AB										
2019	53°43'N, 113°33'W	27.7	51.2	07-May	07-Oct	1967	91	366	130	13.0

	53°44'N, 113°35'W	25 ^b	26.1	19-May	30-Sep	2125	99	367	130	13.9
2020										
	53°45'N, 113°26'W	25 ^b	104.0	13-May	17-Sep	2252	104	208	74	15.3
2021										
Vermilion, AB										
	53°09'N, 110°57'W	16.15	56.4	19-May	09-Oct	2142	100	363	132	14.0
2020										
Indian Head, SK										
	50°31'N, 103°36'W	45.9	12.3	21-May	07-Sep	2100	99	334	121	13.9
2019										
	50°33'N, 103°35'W	50.7	11.2	01-Jun	06-Sep	2242	107	128	47	14.8
2020										
	50°33'N, 103°34'W	46.7	9.0	11-May	30-Aug	2386	113	164	60	15.7
2021										

^aYearly values are reported from the nearest geographical provincial weather station (Alberta data obtained from <http://agriculture.alberta.ca/acis/weather-data-viewer.jsp>; Saskatchewan data obtained from https://climate.weather.gc.ca/historical_data/search_historic_data_e.html).

^bGrowing degree days were calculated using a base temperature of 0°C as described by Cao and Moss (1989). ^cValues obtained from <https://mangomap.com/gis4ag/maps/26449/alberta-soils#>. ^dTotal irrigation + precipitation amount (irrigation amount). ‘-‘ = missing data.

Table 2. 3. List of urea type treatment information.

Urea Type	Acronym	Brand Name	Manufacturer	Active Ingredient	Cost ^b
Urea	-	-	various	-	1100.0 ^c
Urease Inhibitor	UI	Agrotain®	Koch TM Fertilizer	NBPT (N-(n-Butyl)thiophosphoric triamide)	1223.5 ^{d,f}
Nitrification Inhibitor	NI	eNtrench®	Corteva TM Agriscience	Nitrapyrin	1102.6 ^{e,f}
Dual Inhibitor	DI	SuperU®	Koch TM Fertilizer	NBPT (N-(n-Butyl)thiophosphoric triamide) + DCD (dicyandiamide)	1200.0 ^d
Slow-Release Fertilizer ^a	SRF	ESN®	Nutrien®	Polymer Coating	1205.0 ^c

^aThe Slow-Release Fertilizer treatment is a 75:25 blend of SRF:urea, and cost is split 75:25. ^bCosts (\$CAD Mg⁻¹) are based on December 2022 quotes from agriculture inputs suppliers and manufacturer representatives located nearest to Lethbridge, AB (quoted values: ^cJens Klempnauer, AgroPlus Inc., personal communication, 2022; ^dBrad Haubrich, KochTM Fertilizer Canada, personal communication, 2022; ^eBrent Nilsson, CortevaTM Agriscience Canada, personal communication, 2022).

^fDoes not factor in cost of impregnating or mixing chemical onto urea granules. ESN® = Environmentally Smart Nitrogen®. ‘-’ = none.

Table 2. 4. Analysis of variance results and least square means for each data set for main effects related to the application of nitrogen or lack thereof, in agronomic yield components in AAC Viewfield.

Effect	Yield (Mg ha ⁻¹)	Protein (%)	Test Weight (kg hL ⁻¹)	Thousand Kernel Weight (g) ^a	Harvest Index ^b	Plant Density (m ⁻²)	Spike Density (m ⁻²)	Heads (n plant ⁻¹)
Dark Brown Chernozem Soils								
0-N Applied	3.95	11.4	80.9	34.3	0.43	205	306	1.50
N Applied	5.76	13.8	80.5	34.1	0.43	215	376	1.73
P value	0.008	0.0011	NS	NS	NS	NS	0.0163	0.0245
LSD _{0.05}	1.14	1.0	-	-	-	-	52	0.19
Black Chernozem and Dark Grey Luvisol Soils								
0-N Applied	3.37	11.3	79.6	35.1	0.44	293	414	1.49
N Applied	5.25	13.9	79.0	35.6	0.42	298	606	2.11
P value	0.0018	0.0003	NS	NS	NS	NS	0.0048	0.011
LSD _{0.05}	0.95	1.0	-	-	-	-	114	0.44

$\alpha = 0.05$. ^aDoes not include data from Barrhead, AB, 2019. ^bDoes not include data from Indian Head, SK, 2019, Lethbridge (Dry or Irrigated), AB, 2020, Beaverlodge or Edmonton, AB, 2021. (LSD_{0.05}) Least significant difference at P < 0.05. ‘-’ = P>0.05. NS = Not Significant.

Table 2. 5. Probability values from the analysis of variance results for each data set for main effects related to urea type and nitrogen rate responses of agronomic yield components in AAC Viewfield. Environments, replications within environments, and associated interactions between random and fixed effects are considered to be random.

Effect	Yield (Mg ha⁻¹)	Protein (%)	Test Weight (kg hL⁻¹)	Thousand Kernel Weight (g)^a	Harvest Index^b	Plant Density (m⁻²)	Spike Density (m⁻²)	Heads (n plant⁻¹)
Dark Brown Chernozem Soils								
Urea Type (T)	0.0174	0.16	0.26	0.88	0.73	0.0196	0.35	0.0077
Nitrogen Rate (R)	0.0132	<0.0001	0.0164	0.48	0.87	0.0294	0.17	0.0137
R _{Linear}	0.0097	<0.0001	0.0019	0.15	0.50	0.0063	0.19	0.0021
R _{Quadratic}	0.0283	0.07	0.87	0.59	0.85	0.27	0.16	0.49
R _{Cubic}	0.70	0.80	0.77	0.89	0.68	0.53	0.21	0.42
T × R	0.41	0.53	0.37	0.34	0.40	0.61	0.95	0.32
Black Chernozem and Dark Grey Luvisol Soils								
Urea Type (T)	0.73	0.54	0.98	0.11	0.61	0.13	<0.0001	0.0318
Nitrogen Rate (R)	0.0104	<0.0001	0.0009	0.64	0.40	0.0423	0.0004	0.0106
R _{Linear}	0.0054	<0.0001	<0.0001	0.25	0.28	0.36	<0.0001	0.0019
R _{Quadratic}	0.0427	0.06	0.99	0.83	0.59	0.08	0.0469	0.21
R _{Cubic}	0.75	0.96	0.88	0.64	0.22	0.031	0.47	0.78
T × R	0.99	0.30	0.79	0.61	0.65	0.75	0.94	0.60

$\alpha = 0.05$. ^aDoes not include data from Barrhead, AB, 2019. ^bDoes not include data from Indian Head, SK, 2019, Lethbridge (Dry or Irrigated), AB, 2020, or, Beaverlodge or Edmonton, AB, 2021.

Table 2. 6. Analysis of variance results and least square means for main effects related to urea type and nitrogen rate responses of agronomic yield components in AAC Viewfield grown near Lethbridge Dry (2020) and Irrigated (2019-2021)), AB and Scott 2019-2021, SK.

Dark Brown Chernozem Soils								
Urea Type (T)	Yield (Mg ha ⁻¹)	Protein (%)	Test Weight (kg hL ⁻¹)	Thousand		Plant Density (m ⁻²)	Spike Density (m ⁻²)	Heads (n plant ⁻¹)
				Kernel Weight (g)	Harvest Index ^a			
Urea	5.80	14.0	80.6	34.1	0.44	208	374	1.78
Urease Inhibitor	5.91	13.8	80.6	34.2	0.44	227	375	1.65
Nitrification Inhibitor	5.78	13.9	80.4	34.0	0.43	214	382	1.78
Dual Inhibitor	5.99	13.9	80.4	34.2	0.44	221	385	1.73
Slow-Release Fertilizer	5.71	13.7	80.4	34.1	0.43	214	371	1.71
P value	*	NS	NS	NS	NS	*	NS	**
LSD _{0.05}	0.17	-	-	-	-	11	-	0.08
Nitrogen Rate (R)								
60 kg N ha ⁻¹	5.44	12.1	81.1	34.6	0.44	220	369	1.66
120 kg N ha ⁻¹	5.94	13.5	80.7	34.2	0.44	221	383	1.73
180 kg N ha ⁻¹	6.03	14.6	80.2	33.9	0.43	216	378	1.74
240 kg N ha ⁻¹	5.94	15.1	79.9	33.9	0.43	211	379	1.78
P value	*	***	*	NS	NS	*	NS	*
LSD _{0.05}	0.37	0.6	0.7	-	-	7	-	0.07
R _{Linear}	**	***	**	NS	NS	**	NS	**
R _{Quadratic}	*	NS	NS	NS	NS	NS	NS	NS
R _{Cubic}	NS	NS	NS	NS	NS	NS	NS	NS
T × R	NS	NS	NS	NS	NS	NS	NS	NS

^aDoes not include data from Lethbridge (Rain-Fed or Irrigated), AB, 2020.

α = 0.05. (***) Significant at P<0.001. (**) Significant at P<0.01. (*) Significant at P<0.05. (NS) Not significant. (LSD_{0.05}) Least significant difference at P<0.05. ‘-’ = P>0.05.

Table 2. 7. Analysis of variance results and least square means for main effects related to urea type and nitrogen rate responses of agronomic yield components in AAC Viewfield grown near Barrhead 2019, Beaverlodge 2021, Edmonton 2019-2021, and Vermilion 2020, AB, and Indian Head 2019-2021, SK.

Black Chernozem & Dark Grey Luvisol Soils								
Urea Type (T)	Yield (Mg ha ⁻¹)	Protein (%)	Test Weight (kg hL ⁻¹)	Thousand		Plant Density (m ⁻²)	Spike Density (m ⁻²)	Heads (n plant ⁻¹)
				Kernel Weight (g) ^a	Harvest Index ^b			
Urea	5.21	13.9	79.0	35.3	0.41	300	600	2.08
Urease Inhibitor	5.24	13.9	79.0	35.8	0.42	302	614	2.12
Nitrification Inhibitor	5.27	14.0	79.1	35.5	0.42	288	603	2.26
Dual Inhibitor	5.25	14.0	79.0	35.9	0.41	301	623	2.14
Slow-Release Fertilizer	5.20	14.0	79.0	35.8	0.41	301	593	2.02
P value	NS	NS	NS	NS	NS	NS	***	*
LSD _{0.05}	-	-	-	-	-	-	12	0.09
Nitrogen Rate (R)								
60 kg N ha ⁻¹	4.81	12.3	79.8	35.8	0.42	293	548	1.95
120 kg N ha ⁻¹	5.31	13.7	79.3	35.7	0.42	304	613	2.09
180 kg N ha ⁻¹	5.44	14.7	78.7	35.7	0.40	297	628	2.19
240 kg N ha ⁻¹	5.37	15.3	78.2	35.4	0.41	299	637	2.19
P value	*	***	***	NS	NS	*	***	*
LSD _{0.05}	0.39	0.6	0.8	-	-	7	39	0.15
R _{Linear}	**	***	***	NS	NS	NS	***	**
R _{Quadratic}	*	NS	NS	NS	NS	NS	*	NS
R _{Cubic}	NS	NS	NS	NS	NS	*	NS	NS
T × R	NS	NS	NS	NS	NS	NS	NS	NS

^aDoes not include data from Barrhead, AB, 2019.

^bDoes not include data from Indian Head, SK, 2019, or Beaverlodge or Edmonton, AB, 2021.

α = 0.05. (***) Significant at P<0.001. (**) Significant at P<0.01. (*) Significant at P<0.05. (NS) Not significant. (LSD_{0.05}) Least significant difference at P<0.05. ‘-’ = P>0.05.

2.10 Figures

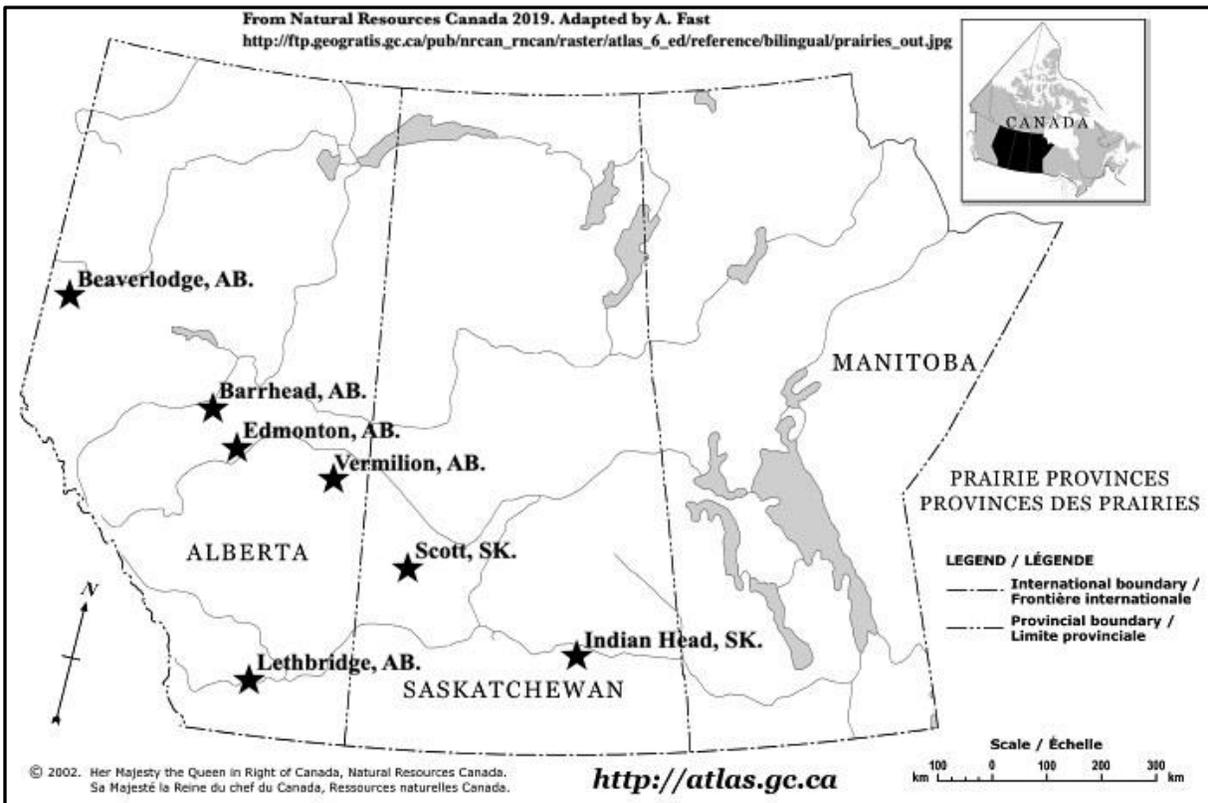


Figure 2. 1. Geographical distribution of experimental site locations to investigate urea type and nitrogen rate on Canada Western Red Spring wheat in western Canada, 2019-2021 (Retrieved from: https://ftp.maps.canada.ca/pub/nrcan_rncan/raster/atlas_6_ed/reference/bilingual/prairies_out.jpg).

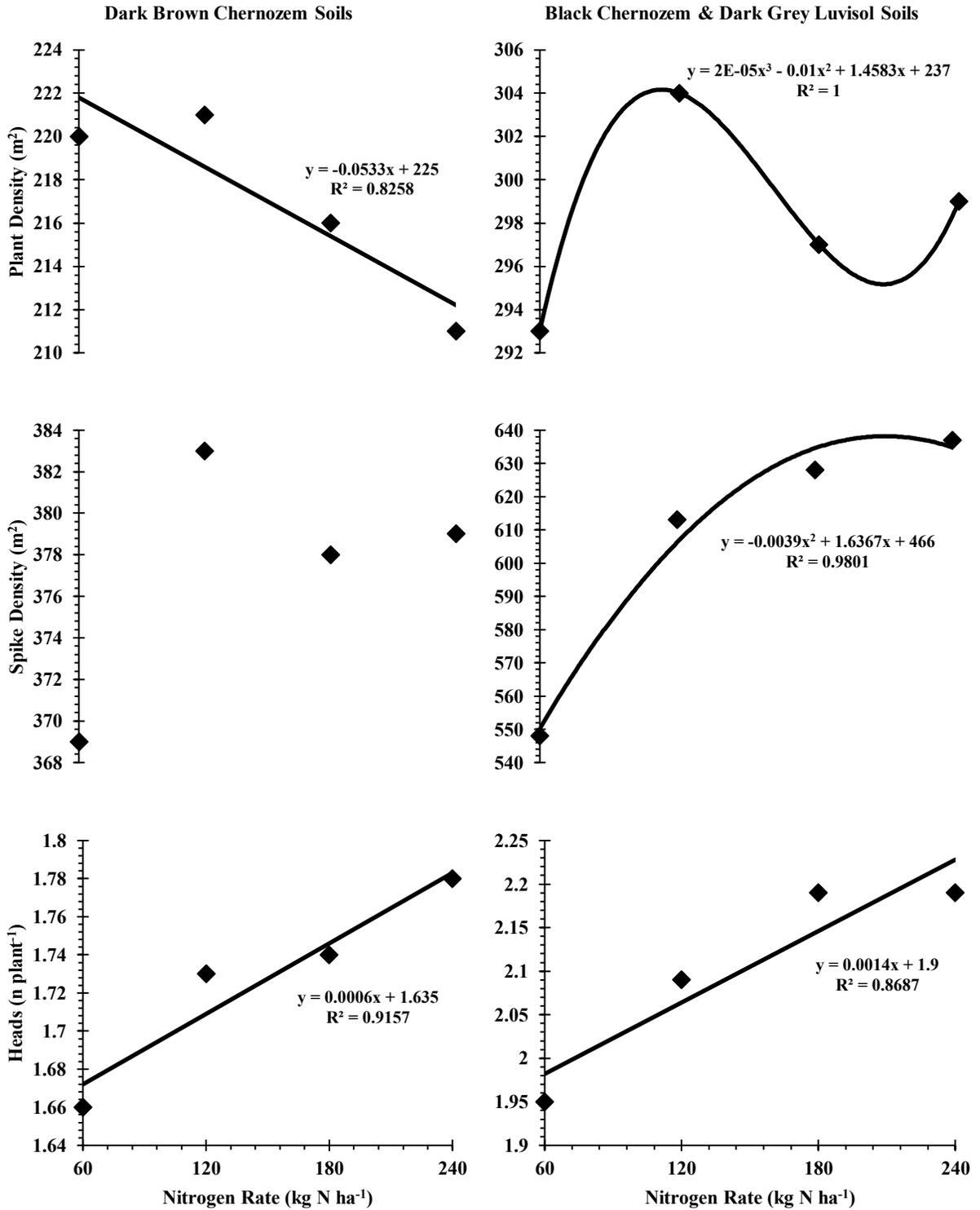


Figure 2. 2. Summary of main effect means for plant and spike establishment. Regression lines are presented when $P < 0.05$ (2019-2021).

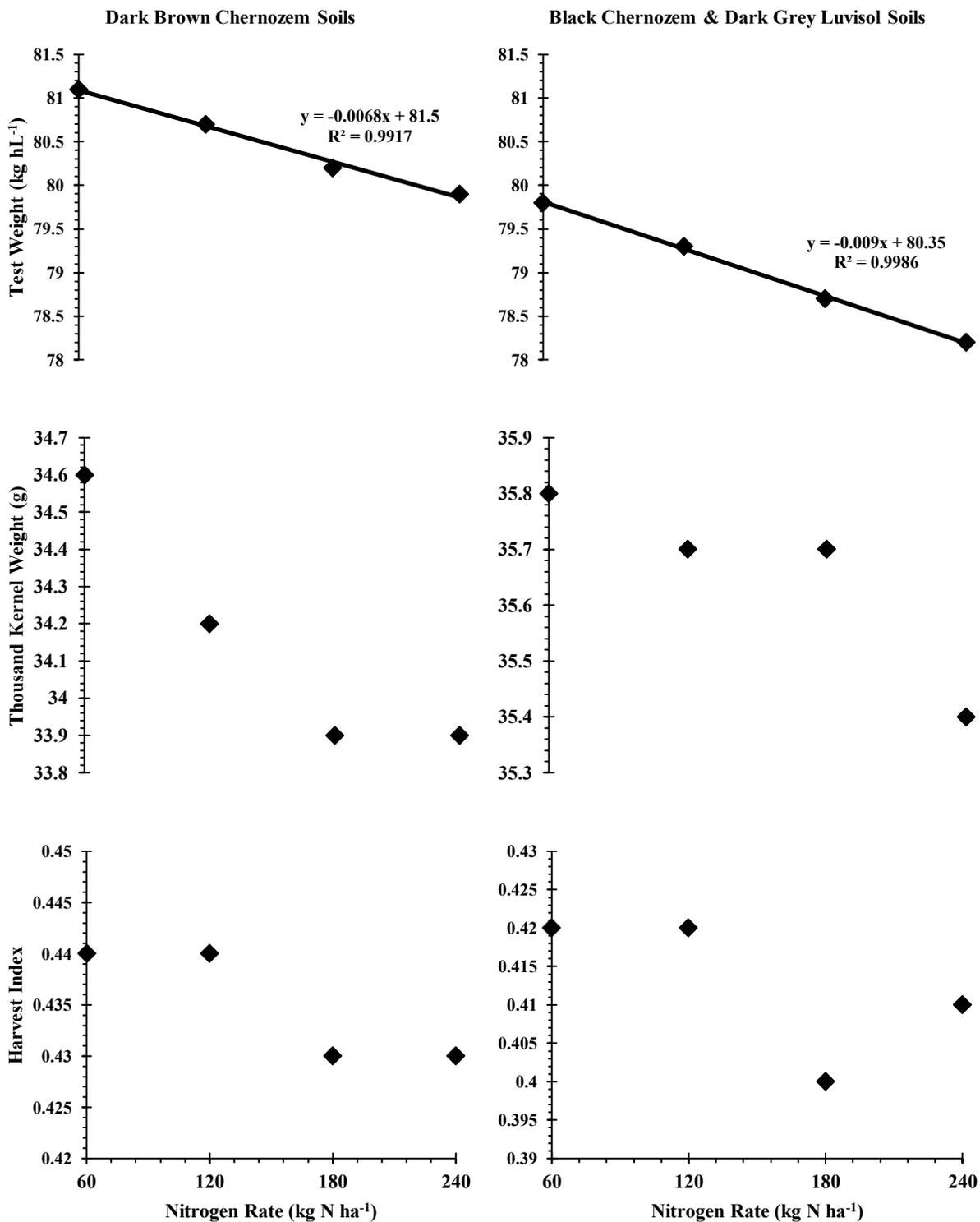


Figure 2. 3. Summary of main effect means for grain quality characteristics. Regression lines are presented when $P < 0.05$ (2019-2021).

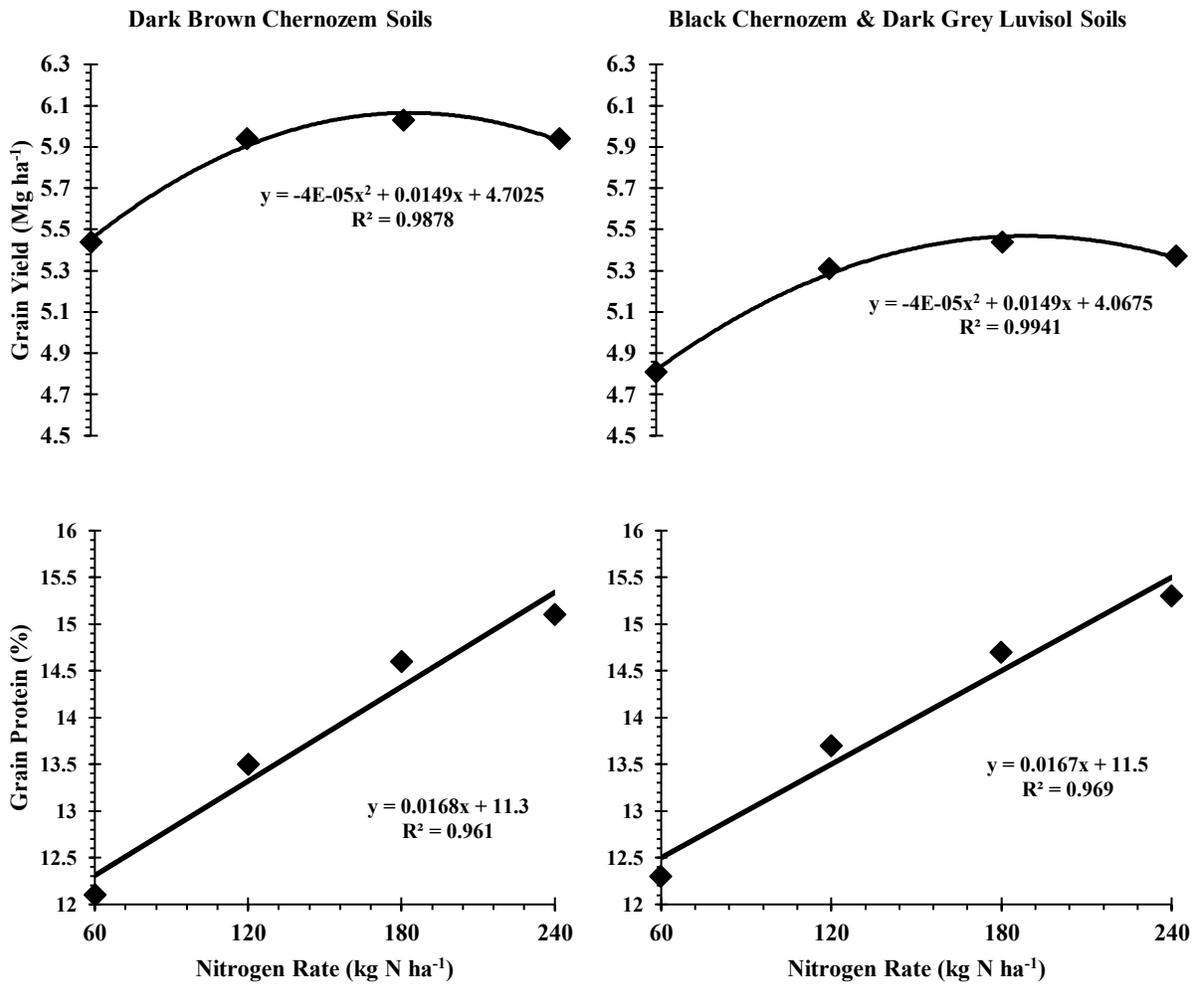


Figure 2. 4. Summary of grain yield and protein content means in response to nitrogen rate. Regression lines are presented when P<0.05 (2019-2021).

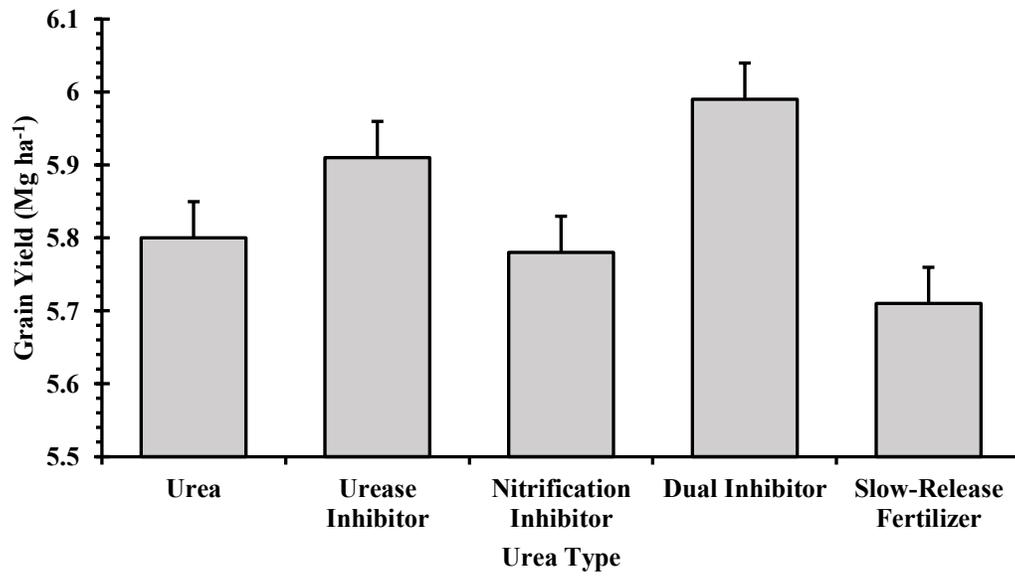


Figure 2. 5. Graphical representation of grain yield response to urea type in the Dark Brown Chernozem Soil group. Error bars represent standard error of the urea type effect. (*) Significant at $P < 0.05$ (2019-2021).

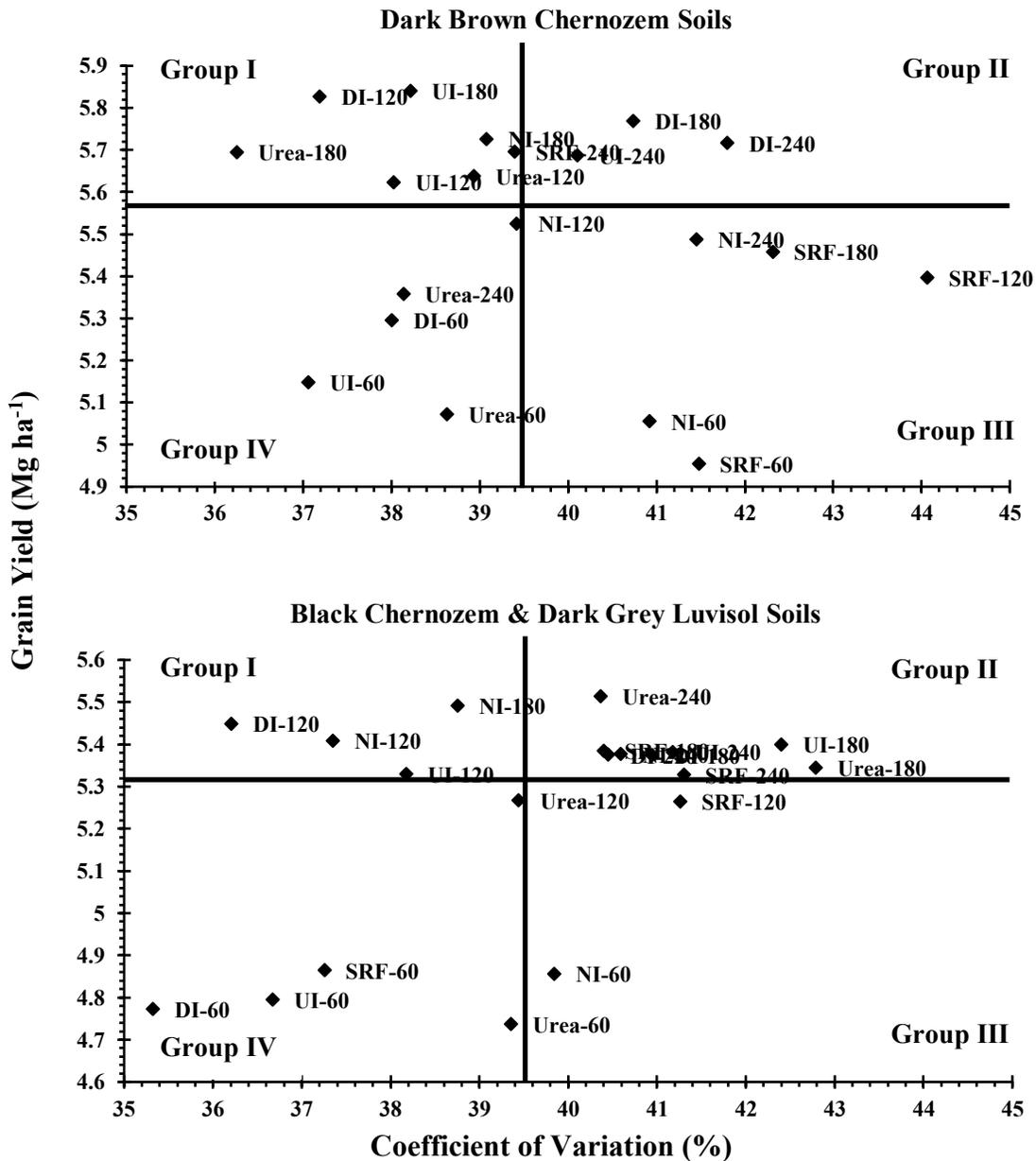


Figure 2. 6. Biplot summarizing the effects of urea type and nitrogen rate on mean grain yield of ‘AAC Viewfield’ compared with its respective coefficient of variation (CV). Group I: High mean, low variability (optimal); Group II: High mean, high variability; Group III: Low mean, high variability (poor); Group IV: Low mean, low variability. Labels are as follows: DI is dual inhibitor, NI is nitrification inhibitor, UI is urease inhibitor, SRF is slow-release fertilizer, Urea is urea, 60 is 60 kg N ha⁻¹, 120 is 120 kg N ha⁻¹, 180 is 180 kg N ha⁻¹, 240 is 240 kg N ha⁻¹ (2019-2021).

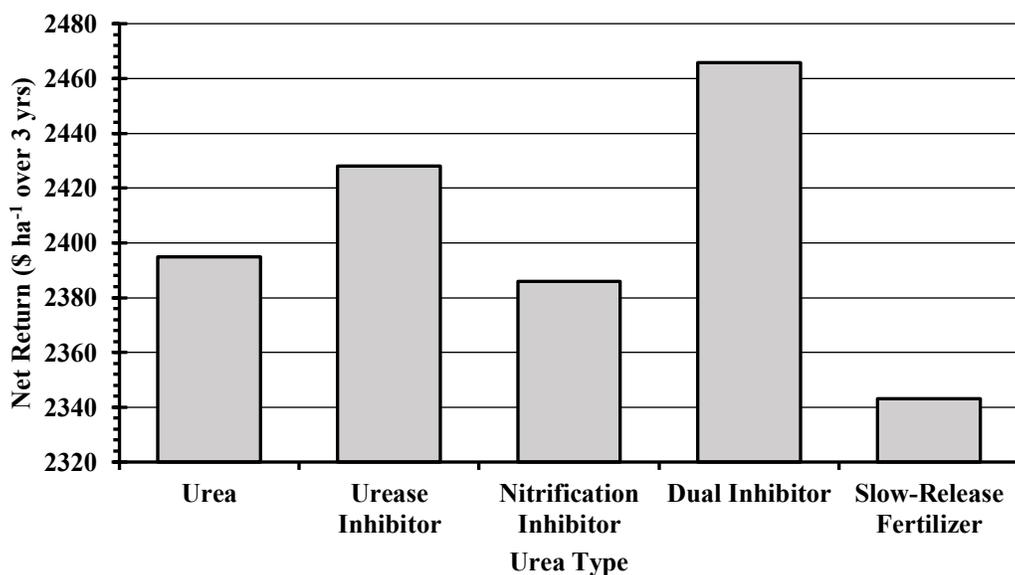


Figure 2. 7. Graphical representation of a simplified net return response to urea type in the Dark Brown Chernozem Soil group. Values are based on a nitrogen rate of 120 kg N ha⁻¹ (0.12 Mg N ha⁻¹), average yields observed in this experiment (2019-2021), a CWRS price of \$435.68 Mg⁻¹, and fertilizer prices listed in Table 2.3.

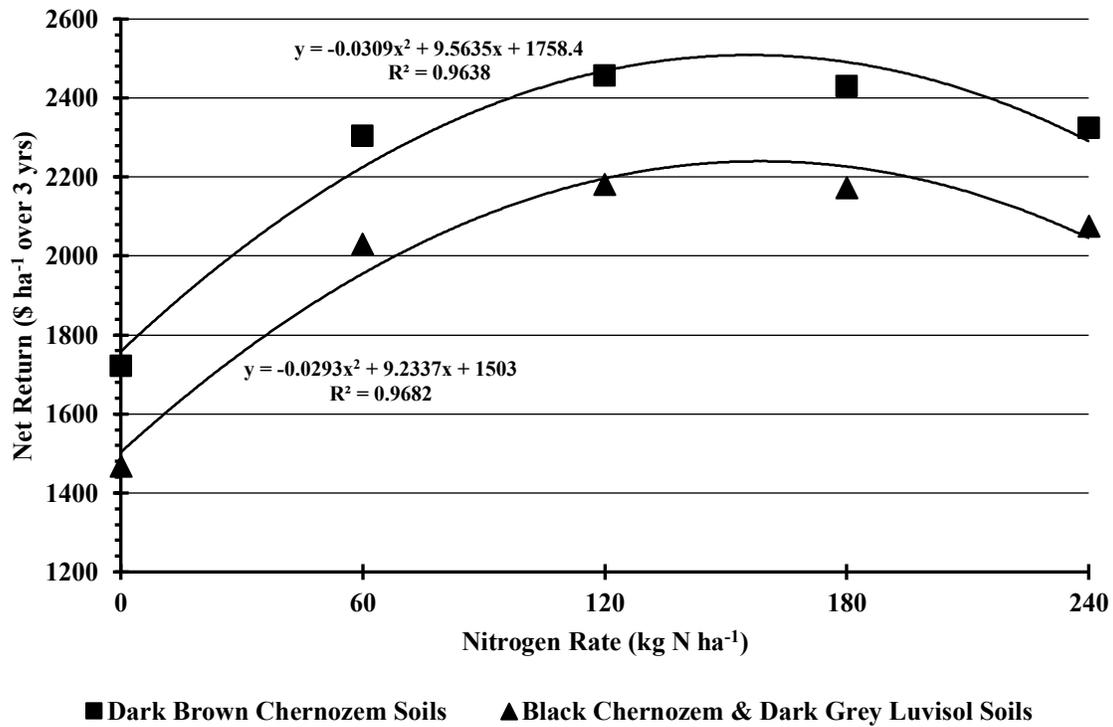


Figure 2. 8. Graphical representation of a simplified net return response to increasing nitrogen rate. Quadratic equations and R^2 values are presented for each data set. Values are based on combined mean urea type yields observed in this experiment, a CWRS price of \$435.68 Mg⁻¹, and a urea price of \$1100 Mg⁻¹ (2019-2021).

3.0 General discussion and conclusions

The incorporation of proper N rate and urea type are needed in order to optimize grain yield and protein content in CWRS production in western Canada. Nitrogen is vital to plant growth and increased N supply can lead to increased yield components. Nitrogen supply in western Canadian cropping systems, is typically achieved with granular urea fertilizer; however, N loss can arise under certain environmental conditions. To mitigate this, EEFs have been developed to increase plant-available N and mitigate environmental N loss. Different EEFs target certain microbes or processes that occur in the N cycle and only work well against those specific conditions; therefore, it is crucial to understand the nature of N loss based on environment and choose the appropriate EEF for anticipated conditions. Combining an appropriate EEF at a N rate of 120 kg N ha⁻¹, can contribute to improved CWRS production in western Canada.

The objectives of this study were:

- 1) To determine if Canada Western Red Spring wheat yield components can benefit from the use of several common enhanced efficiency fertilizers over conventional urea.
- 2) To investigate the nitrogen requirements of a modern high-yielding Canada Western Red Spring wheat cultivar to optimize grain yield and protein content.

The objectives of this study are explained by the following summaries:

- 1) Canada Western Red Spring yield components were inconsistently affected by the use of different enhanced efficiency fertilizers relative to urea. Furthermore, except for the use of a dual inhibitor in the brown soils (grain yield), all enhanced efficiency fertilizers attained similar grain yield and protein content to conventional urea.
- 2) Nitrogen rates of 120 kg N ha⁻¹ were observed to optimize grain yield while also providing at least 13.5% protein content in AAC Viewfield. Additionally, increasing N

rate from 60-240 kg N ha⁻¹ linearly and quadratically increased grain protein content and yield, respectively.

3.1 Recommendations for future work

- 1) The use of a SRF resulted in the lowest grain yield return independent of soil group, despite not being significantly different than conventional urea. It could be beneficial to investigate other ratios of SRF:urea that are less than the 75:25 mixtures used here.
- 2) Research into seed-placed SRF fertilizer has proven beneficial to increase seed safety and N rates. Similar limited research with urease inhibitors has also shown possible benefits; therefore, further investigation into seed-placed urease (Agrotain®) and dual inhibitors (SuperU®) should be considered.
- 3) Continual research into newer cultivars (e.g., AAC Russell VB or CDC Silas) is needed to continually update nutrient management packages for growers in western Canada. As mentioned, the critical point of shifting grain yield response to gain in protein depends on cultivar and environment; therefore, future research on new and improved varieties will be needed.

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