Agronomic Practices to Reduce the Effects of Environmental Stresses on Spring Canola (*Brassica napus* L.) Establishment and Yield in Ontario

by

K. Meagan Griffiths

A Thesis
Presented to
The University of Guelph

In partial fulfillment of requirements
for the degree of
Master of Science
in
Plant Agriculture

Guelph, Ontario, Canada
© K. Meagan Griffiths, August, 2014

ABSTRACT

AGRONOMIC PRACTICES TO REDUCE THE EFFECTS OF ENVIRONMENTAL STRESSES ON SPRING CANOLA (*Brassica napus* L.) ESTABLISHMENT AND YIELD IN ONTARIO

K. Meagan Griffiths
University of Guelph, 2014

Advisor:
Professor H.J. Earl

Establishing an adequate plant stand is one of the primary challenges in the production of canola (*Brassica napus* L.), and plant population strongly affects crop morphology. We tested the hypotheses that i) pre-plant application of liquid dairy manure (LDM) improves crop establishment more than a fertilizer with similar nutrients, and ii) plant morphological changes correlated with low-density plant stands enhance crop tolerance of mid-season water stress. In greenhouse studies both LDM and fertilizer treatments enhanced seedling vigour over the untreated control, but in some cases percent seedling emergence was higher with LDM than the fertilizer treatments. Enhanced seedling emergence with LDM was not found in field trials, even at sub-optimal seeding rates. In a high-yielding greenhouse trial, low plant density shifted much of the pod load from main racemes to branch racemes, but this morphological change did not significantly reduce yield loss under water stress.

ACKNOWLEDGEMENTS

Firstly, I would like to extend a very large thank you to my supervisor, Dr. Hugh Earl, for his time, patience, support, optimism, humour, and guidance throughout the duration of my studies. I am continuously grateful for the opportunities that he has given me and will never forget the day that I walked into his office for advice, only to walk out of it with a new summer job and the opportunity to become his Masters student. That was a spectacular day! I am beyond grateful for his faith in me from that day on. I would also like to acknowledge the members of my advisory committee, Dr. John Lauzon and Dr. Eric Lyons. Their advice and extensive reviews of this work were very much appreciated.

This thesis could not have been completed without the diligent fieldwork of Godfrey Chu and Laxhman Ramsahoi. I can't imagine if I would have had to sample the cow poop and drive the manure tank across the fields by myself! I am so thankful for their kindness and willingness to help me out all the time, whatever the task. I would also like to thank Dietmar Scholz for his assistance in the greenhouse and Li Guo for her help and for being a cheerful face I could always count on. Thank you to the Ontario Canola Growers Association, the Ontario Ministry of Agriculture, Food and Rural Affairs and the Agriculture Adaptation Council for providing financial assistance.

In closing, I would like to thank my family, my friends, and everyone in the Plant Agriculture department. Throughout my time in the Crop Science Building at the University of Guelph, I've always felt part of the 'Plant Ag' family. Thank you all for your support and friendship!

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
B	Boron
BR	Branch Racemes
Ca	Calcium
CGR	Crop Growth Rate
Cl	Chlorine
Cu	Copper
D_{30}	Low-Density Treatment (30 plants m^{-2})
D_{90}	High-Density Treatment (90 plants m^{-2})
DAP	Days After Planting
Fe	Iron
HSD	Honestly Significant Difference
K	Potassium
KSW_{BR}	1000-Seed Weight on Branch Racemes
KSW_{MR}	1000-Seed Weight on Main Racemes
KSW_{TOT}	1000-Seed Weight for Total Plots
LA	Leaf Area
LDM	Liquid Dairy Manure
MAP	Monoammonium Phosphate
Mg	Magnesium
Mn	Manganese
Mo	Molybdenum

MR	Main Raceme
N	Nitrogen
NDRE	Normalized Difference Red-Edge
P	Phosphorus
POD_{BR}	Pods on Branch Racemes (m^{-2})
POD_{MR}	Pods on Main Racemes (m^{-2})
POD_{TOT}	Pods in Total Plots (m^{-2})
S	Sulfur
RCBD	Randomized Complete Block Design
SDW	Shoot Dry Weight
SPP_{BR}	Seeds Per Pod on Branch Racemes
SPP_{MR}	Seeds Per Pod on Main Racemes
SPP_{TOT}	Seeds Per Pod for Total Plots
$\%SW_{BR}$	Percentage of Total Seed Weight on Branch Racemes (g)
$\%SW_{MR}$	Percentage of Total Seed Weight on Main Racemes (g)
W_C	Water Control Treatment
W_S	Water Stress Treatment
Y_{BR}	Yield on Branch Raceme ($g\ m^{-2}$)
Y_{MR}	Yield on Main Raceme ($g\ m^{-2}$)
Y_{TOT}	Yield of Total Plot ($g\ m^{-2}$)
Zn	Zinc

CHAPTER 1

INTRODUCTION

1.1 General Introduction

Canola (*Brassica napus* L.), sometimes known as oilseed rape, is an oilseed crop in the mustard or cabbage family, also known as the Brassicaceae (CCC, 2012). Canola is a derivative of rapeseed and is distinguished by its desired oil qualities of low erucic acid and low glucosinolates. The first canola lines were bred in Canada, which is why the name "canola" was derived from "canadian oil, low-acid" (; Statistics Canada, 2009). Winter (i.e., fall-planted) and spring varieties of canola are available for production. Winter canola requires a period of vernalization and is not commonly grown in Canada due to winter survival issues (CCC, 2012).

Canola is a dicot plant with an epigeal emergence habit and produces large broad leaves, a deep taproot, a main raceme and lateral or branch racemes. Buds and yellow flowers, which are predominately self-pollinated, develop on all racemes into siliques (pods) that contain many small dark seeds (CFIA, 2012). An average canola seed contains 45% oil (Velasco et al., 1999; Statistics Canada, 2009). The protein and starch remaining in the meal following oil extraction are used in high-protein livestock feeds. Rapeseed cultivars that do not have the low-erucic acid quality are also produced, and are grown for non-food applications such as biodeisel and ink production.

Canada is the number one producer of canola oil in the world. A study released in 2013 by the Canola Council of Canada (2013a) reported that Canadian-grown canola contributes $19.3 billion to Canada's economy each year. Most of this economic impact is felt in Saskatchewan, Alberta, and Manitoba ($8.2 , $6.1, and $3.4 billion, respectively) though Ontario, British Columbia and Quebec are also significantly impacted ($7.7, $4.3, and $3.3 million,

respectively). The economic contribution to each province is produced from a combination of canola farming, seed development, handling and storage, crushing, refining, transportation, port activities, and livestock industry benefits.

Compared to 2012, canola production in Canada increased 29.5% in 2013 with a new record of 18.0 million tons (Statistics Canada, 2013). This is nearly a 60% increase from 2006 (Statistics Canada, 2012). This increase is from not only acreage, but also from increasing average yields. Average per-acre canola yields have risen 50% in 15 years (CCC, 2013b). Acreage to be seeded to canola in the 2014 growing season is forecasted at 18.2 million hectares (Statistics Canada, 2014).

The work described in this thesis focuses on two related aspects of canola agronomy: i) maximizing crop emergence and early vigour to ensure an adequate plant stand, and ii) investigating the effects of final plant populations on single-plant morphology and the associated consequences for crop tolerance of soil water deficits during the critical flowering period.

1.2 Early Establishment, Vigour and the Manure Effect

Early establishment and vigour of a crop can be important determinants of yield potential. Ensuring an adequate stand is a challenge in canola production due to the crop's small seed size (OMAFRA, 2009). The result of a small seed size is that the emerging seedling has fewer reserves compared to crops with larger seeds. For this reason, planting depth must be shallow (1 to 2 cm), leaving the seed more susceptible to environmental stresses such as soil erosion and low soil moisture. Canola seedlings also undergo epigeal emergence, where the cotyledons and

the shoot growing point emerge above the soil surface, increasing the plant's susceptibility to soil crusting (CCC, 2012).

Failure to establish an adequate and vigorous plant stand can lead to significant yield loss. For example, poor stand establishment reduces a canola crop's ability to compete with weeds at early stages of growth. Canola seedlings are sensitive to early weed competition, and yield reductions greater than 10% have been observed when weed-free conditions are not maintained between crop emergence and the four to six leaf stage of canola (Martin et al., 2001). Early seedling emergence and seedling vigour are two components identified as increasing the crop's ability to compete with weeds as well as to tolerate early predation by insects (Minotti and Sweet, 1981; Lamb, 1984).

During seedling development, from germination up until the rosette stage (5-6 leaves), biomass accumulation is particularly important for yield. Leaf area (LA) accumulation augments the main early source of assimilate production and is related to the final yield potential of the crop (Gallagher and Biscoe, 1978; OMAFRA, 2009; Government of Saskatchewan, 2012). Enhanced vigour of plant growth during early stages may therefore translate into enhanced crop yields.

The canola crop is more sensitive than other crops to environmental stresses at germination, emergence, and establishment and it is therefore normal for producers to manage the crop with greater intensity. To produce an adequate stand, canola requires a firm, moist and uniform seedbed for good seed to soil contact and therefore benefits from soil management

practices such as intensive tillage, cultipacking, and residue management (OMAFRA, 2009). Other factors such as variety, seeding date, seeding rate, fertility and weed control must also be considered at the time of crop establishment (CCC, 2005). To mitigate risks of inadequate plant stands, canola farmers may be increasing seeding rates to higher than necessary levels as insurance to achieve an adequate canola stand.

Researchers at the University of Guelph have observed short-term enhancement of canopy establishment and yield in winter canola with pre-plant application of manure (H. Earl, J Omeilan and D. Hume, unpublished results). In two separate studies, where canola stand establishment was otherwise inadequate, liquid dairy manure (LDM) application prior to seeding provided an improvement that was visible to the eye. These observations support the notion that canola benefits from manure application at establishment. In these previous trials, comparison with fertilizer treatments delivering the equivalent amounts of macronutrients suggested that the beneficial effects of the manure applications could not be attributed to the manure's macronutrient content alone. In other words, there may be a micronutrient or biological factor to explain the observed effect of manure in a canola crop. Effects of manure on stand establishment of canola have not been reported in the scientific literature and the physiological basis of the effect is unknown.

1.2.1 Hypotheses

Based on prior observations made by researchers at the University of Guelph, we hypothesize that LDM application prior to planting improves plant vigour and establishment of canola in early growth stages, to a greater extent than can be attributed to the manure's inorganic

macronutrient content alone. In addition, initial improvement of early season vigour is only important to final canola crop yields if the plant stand is otherwise marginal.

1.2.2 Research Objectives

1. To evaluate whether pre-plant application of manure has a positive effect on early growth and canopy establishment of spring canola.
2. To determine if the beneficial effects of manure on spring canola can be attributed to the manure's contribution of macronutrients, S, B or water.
3. To isolate biotic or abiotic factors that may have contributed to observed beneficial effects of manure.
4. To compare yield-limiting and non-yield-limiting canola stands in order to assess effects of early crop vigour on final crop yields.

1.3 Effects of Plant Density and Water Stress on Yield of Spring Canola

In is critical to optimize agronomic practices of canola production, as it is such an important crop to Canada. There is inconsistency in the literature as to how seeding rates impact canola yields (Clarke and Simpson 1978; Ozer, 2003; Lääniste, 2008; Mendham et al., 1981). It is necessary to investigate the crop further to understand its physiological responses to management and update recommendations.

Plant density has been the topic of research for many, with the end goals being i) to highlight the benefits and shortcomings of the use of various seeding rates and ii) to give local recommendations based on these findings. Canola can be seeded at a wide range of seeding rates

that will result in yields with no statistical difference (Donald, 1963; Morrison, 1990b; Diepenbrock, 2000). This occurs due to the plasticity that canola displays in response to its environment, which results in plants grown under low-density conditions tend to have larger leaves, more branch racemes and pods per plant compared to plants grown under high-density conditions (Angadi et al., 2003; Kuchtova and Vasak, 1998; Clarke and Simpson, l978; McGregor, 1987; Morrison, 1990a,b; Ozer, 2003; Ali et al., 1996; Degenhardt and Kondra, 1981)**.** Recommended seeding rates to producers can vary based on cultural factors such as planting date and equipment, along with local factors such as soil and climactic conditions including precipitation (OMAFRA, 2013a; Dupriez and De Leener, 1989; Hussain et al., 2003; Angadi et al., 2003).

Water stress is one of the most prominent yield-limiting abiotic factors for crops around the globe (Boyer, 1982). Canola yields generally increase with the amount of water the crop receives (Al-Jaloud et al., 1996; CCC, 2011). Canola crops receiving sufficient total moisture over the growing season may still experience yield reductions when the crop experiences a transient water stress (Tesfamariam et al., 2010).

The effect that a water stress event has on a canola crop depends on the genotype, intensity and duration of the stress, climactic conditions, and the stage of growth and development at which the stress occurs (Norouzi et al., 2008; Hashem et al., 1998; Robertson and Holland, 2004). Seasonal rainfall patterns in Ontario often expose a canola crop to water stress during the flowering and seed development stages, which typically begin in late June to early July (Kutcher et al., 2010). A transient water stress event at these stages results in

reductions of seed yield, ultimately as a result of the negative impact on plant processes that are vital for biomass accumulation (Issarakraisila, 2007; Ahmadi and Bahrani, 2009).

There are likely important differences in the stress responses of canola plants grown under low- versus high-density conditions, though this area is under-researched and not well discussed in the literature. Studies by Clarke and Simpson (1978) and McGregor (1987) report that in dry years, low-density plantings did not compensate up to yields produced by high-density plantings and attributed this to the lack of water resources available. Observations by Angadi et al. (2003) challenge these studies, since reducing plant population by 50%, from 80 to 40 plants m^{-2} did not significantly affect yields in a dry year.

There are several features of plant morphology in low-density plantings that suggest that such plantings may actually have advantages in the event of a transient stress. For example, increased branching and larger main racemes in low-density plantings tend to lead to an extended flowering time, since branch racemes and upper sections of main racemes flower later than the bases of main racemes (McGregor, 1987). Reproductive development is therefore less synchronized. An example of how this could be beneficial is in the occurrence of a stress during early flowering, which may compromise pod set on the main racemes and early-flowering branch racemes, but due to the non-synchronous flowering pattern, the plants may fully compensate by setting additional pods on later-developing branch racemes.

Primary organic molecules such as sugars and amino acids, also referred to as photosynthates or assimilates, are synthesized within the photosynthetic sources of the canola

plants: the leaves, stems or pods (Mengel and Kirkby, 2001). These assimilates are stored, transported and utilized by "sinks" such as fruits, seeds, stems, or roots. In addition to having an extended flowering period, canola plants under low-density conditions have increased efficiency of flower bud and pod retention (Angadi, 2003; McGregor, 1981), indicating a greater assimilate availability per plant in comparison to high-density plantings (Diepenbrock, 2000). McGregor (1987) suggests that this is a result of extended accumulation of assimilates in leaves, stems and pods. Similar conclusions were made by McWilliam et al. (1995), who suggested that better pod retention in low-density plantings can be attributed solely to the increased production and retention of leaves per plant, which are superior photosynthetic sources compared to pods and stems. The increased bud and pod retention observed for plants in low-density conditions contribute greatly to yield compensation up to levels achieved by high-density conditions and it is not clear whether these characteristics hold true under stress. If greater pod retention in low-density plantings does hold true at the time of an environmental stress, these plants may actually out-yield plants under high-density conditions.

With the morphological plasticity that canola plants express under different plant densities, it would be beneficial to understand how planting density affects the yield tolerance of the crop to water stress. This experiment will test how yield and individual yield components of canola plants grown under different plant densities respond to a water stress event during the critical period of flowering.

1.3.1 Hypotheses

The hypothesis is that increased assimilate sources and branching of individual plants under low-density populations result in an extended flowering time period and afford the plants more flexibility for re-allocation of resources to additional sinks and for pod retention, thus increasing the tolerance of the crop to a transient water stress event.

1.3.2. Research Objectives

1. To determine if the population density of a canola crop has an effect on the physiological response of plants to a transient water stress event.
2. To assess whether canola plants grown under low-density conditions have a greater tolerance to yield loss than those grown under high-density conditions under the circumstances of a transient water stress event.

CHAPTER 2

LITERATURE REVIEW

2.1 Nutrient Management in Canola Cropping Systems

Balanced and effective fertilizer management throughout the growing season is a vital part of any crop management strategy, having a large influence on sustaining soil productivity and optimizing crop yields and quality (Grant and Bailey, 1993; Mohammadi et al., 2011). Compared to wheat, another major crop in Canada, canola is thought of as a relatively high-maintenance crop, as it requires a greater amount of nutrients to produce optimal yields (Mohammadi et al., 2011). The amount of fertilizer required to meet the nutritional needs of a canola, as with any field crop, depends on the soil nutrients, yield potential, nutrient source or fertilizer type, application method, timing of application, and specific characteristics of the soil and climate of the location (Grant and Bailey, 1993; Berglund, 2007).

2.1.1 Sources, Rates and Application

Starter fertilizers can be applied using several different methods at or shortly prior to seeding. At seeding, fertilizers can be placed below ground as liquid through injection or as a granular by banding. Granular or liquid fertilizer is placed either in the row with the seed, in the row beneath the seed, or beside and below the row. Placing Nitrogen (N) fertilizer below ground in a concentrated band is advantageous in comparison to surface application, since it can reduce N immobilization, volatilization and denitrification (Tomar and Soper, 1987). If N is surface-applied, these effects can also be reduced by incorporation into the soil. Precise below-ground placement of Phosphorus (P) also has benefits, since it has low mobility in the soil and therefore close proximity to seedling roots can be crucial for early uptake (Miller et al., 1971; Qian and Schoenau, 2010). Starter Sulfur (S) fertilizer is also often recommended for canola, regardless of

a S soil test, since S is very important in the production of canola and the test can be unreliable due to variability in the soil profile (Jackson, 2000).

In many cropping systems, seed-placed fertilization, where fertilizer is placed in the same furrow as the seed, is cost-effective and an effective way to supply crops with early access to nutrients (Zentner et al., 2002); However, emerging canola plants are more susceptible to damage from seed-row placed fertilizers than many other crops (Bailey and Grant 1990; Qian and Schoenau, 2010; Mason, 1971). Although producers in Western Canada have adopted the use of starter in-row P fertilizer applications, significant reduction of germination and emergence due to toxicity have been reported from levels greater than 30 kg P_2O_5 ha^{-1} of monoammonium phosphate (MAP) (Qian and Schoenau, 2010)**.** Phosphorus fertilizers, as well as potassium (K) fertilizers, applied in close proximity to the seed can cause osmotic stress **(**Qian et al., 2012; Mason 1971; Qian and Schoenau, 2010; Nyborg, 1961)**.** This stress comes from the salt effect of these fertilizers, where soil osmotic potential is increased and water uptake by the seeds is subsequently reduced (Qian and Schoenau, 2010; Uhvits, 1946). Another way that seedling injury can occur is from N fertilizers, referred to as ammonia toxicity (Molberg 1961; Cooke 1962; Mason, 1971; Toews and Soper, 1978; Malhi and Gill, 2004; Wang et al. 1995; Grant et al., 2011; Nyborg 1961). Seedling damage by ammonia toxicity can have a major impact on stand density, crop yield and maturity (Karamanos et al., 2005).

Due to reported concerns of chemical injury, the general recommendation from the above research on canola fertility is that such starter fertilizers should not be placed in direct content with the canola seeds, unless using low rates. As an alternative, the best practice for early canola

nutrition is injection or banding beside and below the seed at planting, or broadcast and incorporating fertilizer prior to seeding, or a combination of both (Berglund, 2007; Hocking et al., 2003).

2.1.2 Macronutrients

Nitrogen is required in large amounts for crop growth and development, as it plays roles in plant tissue production, being present in proteins, amino acids, nucleotides, nucleic acids and chlorophyll (Grant and Bailey, 1993). Canola is very responsive to applications of N fertilizer, which has been shown to positively influence LA development, LA duration after flowering, branches per plant, flowers per plant and number and weight of pods and seeds per plant (Wright et al., 1988; Ramsey and Callinan, 1994; Hocking et al., 1997; Rathke et al., 2005; Allen and Morgan 1972; Berti and Mosca 1987; Marquard and Gendy 1989; Taylor et al., 1991).

A study at Michigan University by Gao et al. (2010) found that application rates greater than 84 kg N ha^{-1} did not result in any benefit to a canola crop, and that the high rates of 168kg N ha^{-1} negatively impacted oil content and quality. Copeland et al. (2001) had slightly different results, and recommend that 140 kg N ha^{-1} produces optimal yield for canola in the Michigan area. This is in agreement with the findings of Gan et al. (2007) in a study in the Great Plains area of Canada.

OMAFRA (2009) bases recommendations of N rates in Ontario on the "N:Canola Price Ratio" ($/kg N: $/kg canola). Based on the outcome of the ratio at the time, recommended rates range from 74 to 119 kg N ha^{-1}. This is an acceptable way to determine the amount of N to apply

to a crop, as the N soil test is criticized for being an inaccurate representation of plant available N in the soil over the growing season due to gains and losses such as mineralization and leaching **(**Ros, et al., 2011**)**. In addition to the N:Canola Price Ratio, N rates selected by producers should also consider the yield potential of the crop and factors specific to the soil such as the previous crop and if any other amendments were additionally applied (OMAFRA, 2009).

Under-application of N fertilizer to a canola crop can result in inadequate N, which leads to a reduction in vegetative growth and poor seed yields. Conversely, over-application of N, can lead to decreased seed oil content and quality (Gao et al., 2010; Brennan et al., 2000; Karamanos et al., 2005; Rathke et al., 2005) and potentially increasing the fraction of protein content in the seed, which is undesirable in a crop grown primarily for oil production (Gao et al., 2010). Excess N can also increase seed chlorophyll content and green seeds as a result of excess biomass production and subsequent delayed maturity from excess N application (Scott et al., 1973). Green seed decreases seed oil quality due high chlorophyll contents, as well as due to increased free fatty acids that reduce the oxidative stability of the oil (Abraham and DeMan, 1986; Tautorus and Low, 1994). Excess biomass production can also result in lodging, which reduces the movement of nutrients and moisture to the seed and creates logistical challenges and shattering at the time of harvest (Scott et al. 1973; Sheppard and Bates 1980; Wright et al. 1988; Bailey 1990).

Phosphorus is vital for energy metabolism and serves as a structural component in nucleic acids and phospholipids. Crop production requires P fertilization, since P has low mobility in the soil and it therefore may not always be available for plant uptake in sufficient

quantities (Grant and Bailey, 1993). With its abundance of long, thin root hairs, canola is effective in taking up P from the soil in locations where it may be deficient (Brewster et al., 1976; Foehse and Jungk, 1983). Although canola has a disadvantage in comparison to some other staple crops in Canada when it comes to P uptake since it is a non-mycorrhizal crop, it acidifies the rhizosphere through root exudates, which increases the solubilization of P, making it more available (Grinsted et al., 1982; Hoffland et al., l989a,b). Phosphorus fertilizer application according to a soil test remains a standard practice of canola producers, since canola requires available P from the soil for the entire growing season (Rose, 2008; OMAFRA, 2010). A limited P supply interrupts important physiological processes such as photosynthesis and oil and protein synthesis, resulting in decreased seed and oil yields (Tayo and Morgan, 1979).

Potassium plays a number of roles in plant nutrition and is required in large amounts for crop growth and development (Rengel and Damon, 2008; Fageria et al., 2011). Some of these roles include enzyme activation, cell metabolism, protein synthesis, solute transport, and physiological activities of the plant such as osmoregulation, cell extension, stomatal movement and stress signaling (Amtmann et al., 2006; White and Karley, 2010; Amtmann et al. 2008;Wang and Wu 2010; Mengel and Kirkby 2001; Marschner 1995). In field crop production specifically, K is most recognized for its crucial roles in photosynthesis and maintenance of cell turgor in plants and its application has been shown to improve crop yields and quality (Vyn et al., 2002; Sen Gupta et al. 1989; Govahi and Saffari, 2006). It is therefore standard practice to provide K in adequate quantities by fertilization according to a soil test (OMAFRA, 2010). Rose (2008) found that to reach optimal yields in a canola crop, this is especially important at growth stages up until early flowering.

2.1.3 Secondary Nutrients

Sulfur is considered to be a secondary nutrient, or even the fourth macronutrient in canola crop production (Govahi and Saffari, 2006; Grant and Baily, 1993). It is crucial for energy transfer, protein structure, and synthesis of protein and chlorophyll (Bidwell, 1979; Marschner, 1995; Grant and Bailey, 1993). Although S is important in all crops, it is especially important in the production of canola, as it is required for the biosynthesis of volatile oils. Crops belonging to the Brassicaceae family accumulate these oils in large quantities as S glucosinolates (Zhao et al., 1993; Marschner, 1995). To obtain optimal yields, S is therefore required in larger amount for application to canola compared to other crops.

There has been focus in the literature on S deficiency due to the interdependent relationship of S and N in canola production (Rendig et al., 1976; Jackson, 2000; Karamanos, 2007; Zhao et al., 1997). Sulfur is needed in canola production in an estimated 1:7 ratio with N, to play significant roles in reduction pathways and protein synthesis (Grant et al., 2003; Friedrich et al., 1977; Stewart and Porter, 1969; Bolton et al., 1976). In turn, insufficient plant-available S becomes a yield-limiting factor, and results in the reduction of N-use efficiency (Ceccoti, 1996; Grant and Bailey, 1993; Fismes et al., 2000).

Balancing S fertilization with other nutrients in agricultural production is important to obtaining optimum yields, especially in areas that may experience deficiency such as those with low pH, since S adsorption by soil is favourable in acidic conditions, where soils will adsorb plant-available S (SO_4^{2-}) by hydrous oxides of Al and Fe (Scherer, 2001; Asare and Scarisbrick, 1995; Jackson, 2000). Sulfur deficiency can also be of concern in water-logged soils since S can

be changed into unavailable sulfide-forms (e.g. pyrite), as well as in high drainage soils where it can be subject to leaching, and finally under cool spring temperature conditions when mineralization is slow (Fismes, 1999). Not surprisingly then, significant yield increases have been reported in response to S application (Janzen and Bettany, 1984; Zhao et al., 1993; Withers and O'Donnel, 1994; McGrath and Zhao, 1995; Nuttall et al., 1987; Jan et al., 2002).

In western Canada, S application has always been recommended for optimal yields in canola production due to low-S deposition in the area. In areas where plant-available S levels of agricultural soils are on the decline such as in southern Ontario, north eastern U.S. and Europe, canola producers have also been observing yield increases with the application of S (Grant et al., 2012). Reasons for the decline of soil S and the subsequent increased requirement for application of S in these areas include the use of low-S fertilizers, high-yielding crop varieties, intensive agriculture practices, declining use of S containing fungicides and declining atmospheric deposition (Scherer, 2001). Since the peak of S emissions in the mid-1980s, human-made sulfur dioxide (SO_2) emissions to the atmosphere have been reduced as a result of pollution control measures (Schöpp et al. 2003; Whelpdale, 1992). Sulfur deficiency in agricultural crops is occurring and will continue to accelerate if proper S-containing fertilizers are not applied (McGrath and Zhao, 1995). In the north east of Scotland, where soil S content as well as S atmospheric depositions are low, appropriate S application to rapeseed has been reported to result in quadrupling yields (Walker and Booth, 1992).

Although Calcium (Ca) and Magnesium (Mg) may not seem as crucial as S or the macronutrients discussed above, they are considered secondary nutrients, meaning that like S,

they are required in larger quantities than what is required of micronutrients. They are important in both structural and metabolic processes, where Ca is known to contribute to membrane stability and cell integrity as well as cell elongation and division (Marschner, 1995; Grant and Bailey, 1993). Magnesium is also known to have an important role in enzymatic reactions and as a component of chlorophyll (Bidwell, 1979).

2.1.4 Micronutrients

Micronutrients, which include Copper (Cu), Manganese (Mn), Iron (Fe), Boron (B), Zinc (Zn), Chlorine (Cl) and Molybdenum (Mo), are those that are required in extremely small quantities for plant nutrition (Romheld, 1991). Gupta (2008) referred to these elements as the "'spark plug' for the enzyme system in plants", as their primary roles are to function as components of prosthetic groups in metalloproteins and as activators of enzyme reactions. In the absence of these activation sources, micronutrient deficiencies can negatively impact crop quality and yields (Cakmak, 2002; Malakouti, 2007; Malakouti, 2008).

Specific micronutrients can become deficient in crops in soils with characteristics of calcareousness or high pH (Deckers and Steinnes, 2004). Deficiencies can also occur if soils are low in organic matter, which is a major source of micronutrients, or if soils are exposed to a climate with extreme wetness or dryness. In such conditions, action must be taken by producers to properly manage the land by monitoring soils through sampling and amending with inputs such as lime (Gupta, 1969; Gupta, 1972) and organic or synthetic fertilizers (Malakouti, 2008).

Intensive crop production can lead to micronutrient imbalances or deficiencies. To avoid this, soil chemical extractions are widely used to estimate the micronutrient status of soils, even though the correlation between extracted elements and what is plant-available for uptake is weak and varies widely, especially when compared over a range of soil types (Tandy et al., 2011). Soil tests involving chemical extraction have been reported to be useful in predicting B and Mo, whereas other methods such as Diffusive Gradients in Thin films (DGT), which measures the diffusive supply of elements, have proven to be more effective in estimating plant availability for Zn and Cu (Tandy et al., 2011). Another method commonly used to determine the nature of a micronutrient deficiency at a particular site is through tissue sampling of crops, which is useful in highlighting deficiencies as well as toxicities (Gupta et al., 2008).

Many studies in the literature display yield gains with the addition of micronutrients to crops including small cereals, corn, beans, forages and oil seeds (Malakouti and Tehrani, 2005; Malakouti, 2007; Gupta, 1979; Gupta, 1981; McFarlane et al., 1990; Takkar, 1993). In canola fertility studies on soils deficient in Zn, Fe and Mn, the correction of deficiencies through fertilizer application has increased yields and oil content (Hu et al., 1996; Singh Grewal et al., 1997; Singh Grewal and Grahma, 1999; Nabipour, 2009; Ahmadi and Javidfar, 1998; Nabipour, 2009; Bybordi, 2010).

Yang et al. (2009) report that deficiencies in Zn, Mo, and B have been shown to negatively affect the seed filling growth stage and subsequent yields of canola. Gupta (1979) and Adams (1997) also note that canola crops and other crops in the Brassicae family are particular responsive to B and Mo application.

Molybdenum deficiency is known to cause impaired pollen development (Kaiser et al., 2005; Schwarz and Mendel, 2006). This can limit yields, as reported by Sinha et al. (1990), who observed increases in seed yield, branches per plant, pods per plant, length of pods, seeds per pod and 1000-seed weight with its application. Decreases in canola 1000-seed weight from Mo deficiency has also been reported by Chen et al. (2005).

2.1.5 Boron: A Micronutrient "On Trial" in Canola Production

The practice of applying B to canola fields has been "on trial" around the globe, since being an oilseed crop, canola has a higher requirement for B compared to other crops such as cereals (Wooding, 1985; Ahmad et al., 2012). For example, the maximum recommendation for broadcasted or incorporated B in the Canadian Prairies is 1.7 kg B ha^{-1}, compared to 0.56 kg B ha^{-1} for cereal crops (Malhi and Karamanos, 2013).

Boron is a non-metal micronutrient, which is important during both vegetative and reproductive plant growth, as it is essential for cell structure (Ahmad et al., 2012; Warington, 1923; Gupta 1993). A B review article from Ahmad et al. (2012) highlights that it has other possible functions including roles in sugar transport, cell wall synthesis, lignification, cell wall structure integrity, carbohydrate metabolism, ribose nucleic acid metabolism, respiration, indole acetic acid metabolism and phenol metabolism (Parr & Loughman, 1983; Welch, 1995; Ahmad et al., 2009). In addition, research on B by the Ontario Canola Growers Association (OCGA), Ontario Ministry of Agriculture and Food (OMAF) and the University of Guelph has recently focused on the importance of B at the reproductive stage, where foliar application may provide a

yield advantage through reducing pod abortions caused by heat stress (Ramsahoi and Earl, 2011).

Investigation of B application to canola has also been under way in Western Canada, where it has been shown that B may have a role in increasing resistance to *Plasnodiophora brassicae* (clubroot) through slowing the development of the infection (Deora, 2011). Other fertility studies in Western Canada conducted by personnel at the Melfort Research Farm, University of Saskatchewan, University of Manitoba, and Alberta Agriculture have shown very limited evidence of a benefit of broadcast or incorporation of B to a canola crop. In these trials, there was no consistent yield response to B application, even on the sandy soils of the Gray and Dark Gray soil zones that have low organic matter and are suspected to be deficient in B (Malhi and Karamanos, 2013).

In contrast to what is being found in Western Canada, other areas that experience Boron deficiency in their soils have reported a benefit from B application to canola crops. For example, a study conducted in China by Hu et al (1994) showed that B application to a clay soil deficient in B improved pod-bearing branches and pod number per plant, seed number per pod, seed yield and oil content.

Though B deficiency in the initial vegetative stages of a canola crop may be difficult to diagnose, symptoms tend to show up in later reproductive phases as red margins and/or inter-venal mottling at flowering, failure of flower bud development, seed development limitations and delayed maturity (Nyborg and Hoyt, 1970; Grant and Bailey, 1993; Wooding, 1985). Boron

deficiency in these situations could be a product of specific features of the soil, climate, geology and fertilizer program of the site (Ahmad, 2012).

The soil concentration and availability of B ranges from soil to soil, depending on factors such as texture, pH, climate and parent material (Ahmad, 2012). Since organic matter is where soil B is mostly contained, crops grown on soils with low organic matter or low microbial activity due to texture or climate can experience B deficiency (Berger, 1962).

Plant-available soil B is normally in the form of boric acid (H_3BO_3 or $B(OH)_3$) and borate ($H_2BO_3^-$) (Camacho-Cristobal et al., 2008). Boric acid accounts for the majority of available B in soils, and since it is mobile, deficiencies can be caused during drought periods due to reduced mobility of B in the soil by mass flow to roots (Chiu and Chang, 1985; Chang et al., 1992; Barber, 1995; Scott et al., 1975). Another example of when B deficiency can occur is during periods of excess precipitation in areas with coarse textured soils, since the nutrient can leach to inaccessible depths (Berger, 1962; Shorrocks, 1997). In addition to these examples, although a soil may contain adequate B, pH level can alter plant availability of the micronutrient, making it less available in alkaline soils and toxic in acidic soils (Elrashidi and O'Connor, 1982; Takkar et al., 1989; Rashid et al., 1994; Niaz et al., 2002, Niaz et al., 2007).

Areas in South Australia, Egypt, Iran, Jordan, Libya California and Chile have experienced reduced crop yields due to high-B groundwater and subsequent toxic levels of B top soils (Yau et al., 1995). Boron toxicity in canola is uncommon however, with one study by Karamanos et al. (2003) showing that with only one exception, there was no impact of B rates as

high as 5.1 kg ha^{-1} on the yield of canola. This is 3.4 kg B ha^{-1} in excess of the maximum recommended application rates in western Canada.

2.1.6 Manure as an Amendment and Fertilizer Source

Manure contains organic components that are beneficial to plant and soil health. The use of manure in cropping systems has been shown to enhance soil biological activity and diversity (Altieri, 1999; Peacock et al., 2001) as well as soil organic matter (Sommerfeldt et al., 1988; Haynes and Naidu, 1998). Manure application also returns nutrients to the soil, reducing the need for synthetic fertilizers and improving the overall sustainability of agricultural practices (Mohammadi et al., 2011).

The majority of N in liquid manure is in the form of ammonium-N (NH_4^+) and organic-N (Beauchamp, 1986). The amount of N available to a crop after application depends on several factors such as animal source and diet, storage, handling, time of application, time to incorporation and factors affecting mineralization of organic matter (Paul and Beauchamp, 1993a). For example, liquid manure has a larger N to organic matter ratio in comparison to solid manure. Liquid manure also has a larger N to P_2O_5 ratio when compared to solid manure (Schroder, 2005). This is due to the fact that there is more N loss during production, storage and handling of solid manures (Dewes, 1995). Timing of incorporation of manure is another large factor, since greater than 50% of NH_4^+-N can be lost to ammonia volatilization if the manure is not incorporated below the surface quickly after application or injected (Chambers et al., 1999). Losses of available N supplied by manure due to leaching and immobilization must also be considered to successfully meet the nutritional needs of a crop.

Phosphorus and potassium availability in all manure types is generally high. When considering application rates, producers tend to use a P-based manure application, with 40 to 70% of total P applied considered to be plant-available in the first year of application (A & L Canada Laboratories Inc., 2014; OMAFRA, 2013b; Eghball et al., 2002). This is generally the trend, since N-based application results in excess P build-up in soils, which can be problematic especially in soils that are prone to erosion (Eghball and Power, 1999).

Potassium can also be supplied by manure as if it were a fertilizer. Potassium is very soluble, with greater than 70% being excreted in the urine component of the manure (Eghball et al., 2002). Potassium supplied by manure, depending on the source, can be almost 100% plant-available in the first year of application (A & L Canada Laboratories Inc., 2014).

Calcium and Magnesium, which are considered secondary nutrients, have been found to be approximately 55% plant-available from manure (Eghball et al., 2002). Though Ca and Mg are not regularly discussed in detail in terms of additional soil fertilization, S, which is also considered by many to be a secondary nutrient for a canola specifically, has received much more attention in the literature.

S is supplied to crops in both organic and inorganic forms upon application of manure. Inorganic S supplied by the manure is most commonly in the form of sulphate (SO_4^{2-}), which can be divided into SO_4^{2-} in soil solution (plant available S), and adsorbed SO_4^{2-} (Barber, 1995**)**. Inorganic sulfur is much less abundant in more agricultural soils than organic S, accounting for

less than 5% on average (Dick, 2008). Sulphate in solution is always in equilibrium with the adsorbed and mineral phase forms (adsorbed and S_2 forms) (Mengel and Kirkby, 2001; Dick, 2008**).** The full amount of S supplied by manure is not fully plant-available due to adsorption of S, based on the status of this equilibrium. Tabatabai and Chae (1991) found that the fraction of S that is released and plant-available from manure application ranges based on the soil pH, soil type, type of manure used, carbon to S ratio of the manure used, and S nutrient status of the soil. Estimates of plant available S fractions can range from 23% in swine manure to 50% in cattle manure, though it is generally less than 40% available in the first year (Eghball et al., 2002). Animal manures are not considered to be a good source of S, since the content can be low due to loss of mineral S by volatilization as hydrogen sulfide (H_2S) in storage and upon application (Oenema et al., 2007). In addition, the carbon to S (C/S) ratio of animal manures tends to be high, causing a large fraction of the S in the manure to be immobilized upon field application (Tabatabai and Chae, 1991). Generally, an organic material must have a C/S ratio of less than 200 to have a net release of plant available S (Dick, 2008). An organic material with a C/S ratio from 200 to 400 generally results in either no change or a small net increase or decrease in mineral soil-sulfur concentrations, and a C/S ratio greater than 400 will result in total S-immobilization.

Manure contains all plant-essential nutrients, and therefore can also be a source of micronutrients (Eghball and Power, 1994). Based on a chemical analysis of swine and cattle manure, Zn, Fe, Mn, Cu and B are less than 40% available from manure, and the short term mineralization of these elements in soils is limited (Eghball et al., 2002).

2.1.7 Manure For Nutrient Management in Canola Cropping Systems

Studies to date on manure management in canola are limited in the Canadian literature, and few studies analyze the effect of manure fertilization on establishment of canola. Recent studies in Australia and the United States report that canola yields from nutrient treatments of recommended NPK rates are improved further by addition of farmyard manure (Mohammadi et al., 2011; Gao et al., 2010). Mohammadi et al. (2011) concluded that manure application can reduce the need for chemical fertilizer usage as it was found to be a viable alternative to synthetic N fertilizer for increasing yields.

Studies investigating the effects of manure have also been conducted on a close relative of canola, Indian mustard (*Brassica juncea)*. Mustard yields are reported to be the highest with the combination of 100% recommended NPK plus farmyard manure application (Mandal et al., 2010; Mandal and Sinha, 2004). This is consistent with findings by Mohammahi et al. (2011). Mandal and Sinha (2004) found that several yield attributes such as plant height, number of branches per plant, number of siliques per plant, number of seeds per silique, 1000-seed weight, seed yield and oil yield of Indian mustard improved at 100% recommended rates of NPK plus 10 t ha^{-1} farmyard manure compared with only 100% recommended rates of NPK. This study also looked at combinations of NPK and farm yard manure with 10 kg ha^{-1} borax and 20 kg ha^{-1} $ZNSO_4$, and found that similarly improved yields resulted from application of 100% NPK plus borax and $ZnSO_4$, or 50% NPK plus farmyard manure, borax and $ZnSO_4$ (Mandal and Sinha, 2004).

2.1.8 Physical Attributes of Manure in Canola Production

Soil quality is especially important in canola planting. Preparation for planting must ensure that there is adequate moisture in the top 2.5 cm of the soil, and that it is level, firm and friable. These soil conditions will ensure uniform planting depth and emergence and give protection against soil crusting which can result in poor seedling establishment (OMAFRA, 2009). Many studies have investigated effects of manure on physical soil properties such as bulk density, aggregation and porosity, which affect crop growth and water relations such as hydraulic conductivity, water holding capacity and plant available water. The degree to which manure affects these properties depends on manure and soil type; however long term, as well as shorter term studies come to the consensus that organic fertilizer sources such as manure have positive effects on soil physical properties which will go on to benefit canola producers (Arriaga and Lowery, 2003; Celik et al., 2004).

2.2 Effects of Plant Density on Canola Growth, Morphology and Yield

2.2.1 Seeding Rate Recommendations Across Locations

A typical establishment estimate of canola is 50% of the seeding rate, which is lower than for many other crops with larger seed sizes (Earl, H.J. personal communication). Due to variations in climactic and field conditions within an area, as well as the plasticity that canola exhibits, the optimal plant population across locations is not always apparent. For example, recommended target populations for canola production in the Canadian prairies can range from 80 to 180 plants m^{-2} (Thomas, 1984). In Australia, a similar case is made since yields are reported to be unaffected by populations ranging from 50 to 130 plants m^{-2} (Potter et al., 1999). Mendham et al. (1981) states that in England, good canola yields can be obtained with as few as

8 plants m^{-2}. According to OMAFRA (2013), an optimal plant stand is 75 to 110 "healthy plants" m^{-2}.

OMAFRA (2009) seeding rate recommendations are based on seed size, in order to target a specific number of seeds ha^{-1}, rather than a weight of seeds ha^{-1}. In practice, however, most seeding of canola tends to be done by seed weight with no consideration of seed size. For Canadian producers, seeding rate ranges from 7 lbs $acre^{-1}$ (7.8 kg ha^{-1}) down to 4.5 lbs $acre^{-1}$ (5.0 kg ha^{-1}), or as low as 3.5 lbs $acre^{-1}$ (3.9 kg ha^{-1}) (OMAF, 2013). In Northern Iran, Shahin and Valiollah (2009) recommends a seeding rate of 4 kg ha^{-1}.

2.2.2 Plant Population Density and Intraspecific Competition

The microenvironment of individual canola plants is affected by the density at which the crop is planted, in that it alters plant competition for space, water and light (Donald, 1963). A high-density canola crop has greater intraspecific competition with potentially fewer resources of water and light per plant, as well as decreased area per plant. Even before seedlings experience such direct competition for resources, they may respond to early signals of potential future competition associated with high plant populations. Specifically, close proximity of neighboring plants alters the red:far-red ratio of horizontally propagated light in the crop canopy, due to the preferential absorption of red light by chlorophyll. The perceived reduction in the red:far red ratio induces light competition avoidance responses in developing seedlings, which may include altered plant height, leaf size and root:shoot dry matter ratios (Sadras and Calderini, 2009).

2.2.3 Effect of Population Density on Early Growth and Development of a Canola Crop

When comparing early vegetative growth in canola grown at various densities, high-density plantings have a greater crop growth rate (CGR), which is a measure of the rate of biomass produced per ground area (Angadi et al., 2003). This is caused by a more rapid canopy closure and consequently there is greater early-season light interception and utilization in high-density crops compared to low-density crops.

Morrison (1990a) points out that plants in low-density plantings have significantly higher net assimilation rates (NAP, a measure of carbon fixed per unit of LA) than plants in high-density plantings. This is in agreement with Clarke and Simpson (1978), and suggests that individual plants grown in low-density conditions are more photosynthetically efficient than plants grown in high-density conditions. This efficiency of individual plants in low-density conditions is due to greater LA establishment per plant, which contributes to branch and bud development and therefore to establishment of pod number (McGregor, 1987). Not only do individual plants in low-density plantings tend to have more leaf photosynthetic tissue, but since there is less shading of leaves, the tissues tend to also remain active longer than in high-density plantings.

The plasticity of a canola plant is highlighted when the effects of plant density on development and dry matter accumulation at early growth stages result in subsequent plant growth and morphology alterations at later stages. Since plants grown in low densities utilize more solar radiation per plant and therefore have an increased source, there is a greater amount of assimilate that can be allocated to sinks (Shahin and Valiollah, 2009). The primary

morphological response of plants in low densities is to increase branching per plant (Clarke and Simpson, l978a; McGregor, 1987; Morrison, 1990a,b; Diepenbrock, 2000; Ozer, 2003; Ali et al., 1996; Degenhardt and Kondra, 1981). For example, Angadi et al., (2003) reported that the mean number of primary branches increased from 5 to 9 per plant with a reduction in plant population from 80 to 5 plants m^{-2}.

2.2.4 Effect of Population Density on Canola Yield Components and Distribution

Seed yield of canola is a function of population density, or plants m^{-2}, as well as branch racemes per plant, pods per branch, seeds per pod and seed weight (McGregor, 1987; Kuchtova and Vasak; Leach et al., 1999; Angadi et al., 2003). Effects of plant density on early morphology of a canola crop go on to have subsequent effects on how final yields are attained, as well as how the yield is distributed on the plant (Angadi et al., 2003).

As branch racemes per plant increase in low-density populations, pods per plant also increase (James and Anderson, 1994; McGregor, 1987; Kuchtova and Vasak, 1998; Leach et al., 1999). This has been shown to be the primary yield component that is responsible for compensating yields, and is the most responsive to plant density (Diepenbrock, 2000; Clarke and Simpson 1978a,b; Kondra, 1975; Degenhardt and Kondra, 1981; McGregor, 1987; Shahin and Valiollah, 2009). A decrease in plant density has also been shown to result in increased seeds per pod and increased seed weight, although these effects are less pronounced than the increases in pods per plant and are not always present (Al-Barzinjy et al., 1999; Shahin and Valiollah, 2009).

Harvest Index, the fraction of crop biomass that makes up harvestable yield, has been shown to be stable across a range of populations, although it has been reported in some cases to be lower in high-density populations (Degenhardt and Kondra, 1981; Angadi et al., 2003). A larger fraction of this harvestable yield is carried on branch racemes of low-density populations when compared to high-density populations, which carry a greater fraction of total yield on the main raceme (Shahin and Valiollah, 2009; Morrison et al., 1990a; Leach et al., 1999). Pods produced on main racemes tend to have a greater number of seeds per pod, and increased 1000-seed weight when compared to that of branch racemes (Morrison, 1990b).

2.2.5 Final Seed Yield of Canola As Affected by Population Density

Final seed yield and seed oil content are primary factors that determine a canola crop's economic value. The literature reflects varying effects of planting density on these factors. In some circumstances, the literature shows that seeding rate does not significantly affect seed yields (Lääniste, 2008; Kondra 1975; Jurke and Fernando, 2008; Helps, 1971; Bengtsson, 1978; Singh and Yusuf, 1978), with final yields reportedly unaffected by densities of 50 to 130 plants m^{-2} (Potter et al., 1999), 40-80 plants m^{-2} (Angadi et al., 2003) and 20-80 plants m^{-2} (Mendham et al., 1981). In other studies, where plant population *per se* was not reported, the yield insensitivity to seeding rate also implies a lack of yield response to populations (e.g., Kondra, 1975 (rates of 3 – 12 kg / ha); Christensen and Drabble, 1984 (rates of 7 to 14 kg / ha)). In these cases, it is clear that individual plants in low-density populations were able to compensate through alteration of morphology and yield components.

Researchers such as Lythgoe et al. (2001) disagree with these findings, failing to see full yield compensation from low-density plantings and observing significantly lower yields with decreased plant density. This trend has also been reported in the literature by others (Ohlsson, 1972; Clarke and Simpson 1978; Bengtsson and Ohlsson, 1974; Kondra, 1975; Singh and Yusuf, 1978; May et al., 1994; Ozer, 2003; Degenhardt and Kondra, 1981; James and Anderson, 1994); However, there are also researchers who have shown evidence for the opposite, where low-density plantings were reported to out-yield high-density plantings (Morrison, 1990a)**.**

The ability of a low-density population of canola to have compensating yields is based primarily on increased number of pods per plant, which is determined by the development and survival of branch racemes and flower and pod retention, rather than by the potential number of flowers and pods (McGregor, 1981; McGregor, 1987; Angadi et al., 2003; Huhn and Schuster, 1975; Clarke and Simpson 1978; Shahin and Valiollah, 2009). The increased efficiency of flower bud and pod retention that has been observed in low-density plantings indicates a greater availability of assimilate per plant (Angadi et al., 2003; McGregor, 1981; Diepenbrock, 2000). McGregor (1987) suggests that this is a result of extended accumulation of assimilates in leaves, stems and pods, largely during and after flowering. Similar conclusions come from McWilliam et al. (1995), who suggested that better pod retention in low-density plantings can be attributed purely to the increased production and retention of leaves per plant, which are a superior photosynthetic source compared to pods and stems.

Yield can also be even further compensated in low-density plantings with increased seeds per pod and seed weight (Angadi et al., 2003). Ganeshaiah and Uma Shanker (1992) suggest that

seed number per pod in plants is a function of resource drawing ability of ovules; however, this form of compensation in canola can be weak in its effect on yield, and in some cases, it has been reported that this yield component was not affected by plant density (Shahin and Valiollah, 2009).

2.2.6 Seed Quality As Affected by Population Density

Although the timing of initial flowering tends to not be significantly affected by plant population, pod formation and maturation, which occurs from the bottom to the top of each raceme, tends to be earlier and more uniform in high-density plantings (McGregor, 1981; McGregor, 1987). Low-density plantings can result in delayed and uneven maturity at harvest, which leads to a higher chlorophyll concentration of seeds from immature pods (green seed) as well as possible yield loss due to shattering of over-mature pods (Clarke and Simpson,1978; Degenhardt and Kondra, 1981). Clarke (1977) reports similar results, in that high seeding rates result in a more synchronous maturity, contributing to lower seed chlorophyll content.

2.2.7 Population Density and Interacting Factors: Pests, Disease and Environmental Stress

As has been discussed, canola crops planted to different population densities are different in their growth and development, resulting in contrasting morphologies that affect yield components and final seed yield. In addition to this, there are other factors specific to the planting season and location, which interact with population density and affect final yield. How these factors interact with environmental conditions to influence seed production contributes to why there is a disconnect in the relationship between canola plant density and yields in the literature. Some of these factors include pests, disease, water and heat stress. When such factors

come together in a growing season, low- verses high-density plantings have advantages and disadvantages, and these will lead to either positive or negative effects on final yield and quality of the crop.

Weed and volunteer cereal competition have been shown in the canola literature to have negative effects on yields (Ohlsson, 1976; Marshall et al, 1989). In circumstances where weed pressure tend to be a problem, one reason to select a higher seeding rate is to be more competitive with weeds, by having a higher early CGR and earlier ground cover and canopy closure (Morrison, 1990b; O'Donovan, 1994). Duczek et al.(1996) supports these findings as reduced crop density was reported to encourage the growth of weeds.

Another issue that must be taken into consideration when selecting a target population density is fungus and disease. Sclerotinia stem rot infection has shown to be of concern in canola plantings, especially those of high density (Turkington et al.,1991). In these conditions, the crop canopy has a higher average relative humidity and an extended period of leaf wetness, resulting in an environment where sclerotinia can develop quickly (Turkington and Morrall, 1993). For example, Irvine and Duncan (2002) showed that sclerotina was a main factor in decreased yields at high-density plantings. Jurke and Fernando (2008) support the idea that manipulation of plant density is a form of cultural control for this disease and that decreasing seeding rates decreases the level of sclerotinia stem rot in canola. This is important, as sclerotinia has been reported to cause serious injury to the plants prior to full maturity, which results in increased chlorophyll content (Morrison, 1990b). Also, high-density plantings of canola are made up of more plants

with smaller stem diameters. These plants are potentially more susceptible to stem girdling diseases as well as lodging.

Lodging is another factor that a producer must consider, as a clear relationship has been established between plant density and stem diameter. High-density plantings have reduced stem diameters, making individual plants less able to structurally support themselves (Kondra, 1975; Morrison, 1990b). Morrison (1990b) stated that lodging in high-density plantings may in some circumstances be a result of the shallow root system in plants grown in high densities, as it was observed that many plants lodged from the base of the stem. In this study, these results were so extreme that lodging was considered a large factor in the significantly lower yields reported for high-density plant populations.

Lääniste et al. (2008) concludes that by "increasing plant densities, no yield increase can be predicted, but the yield will be more stabilized". From what is discussed above, one could argue with this statement, saying that in various environments, and under varying stresses there are adequate examples of how low- and high-density plant populations interact with other situational factors and result in positive or negative impacts on final seed yield. This view can be extrapolated to climactic factors as well, where there may be advantages and disadvantages of various population densities depending on fluctuating resources over a field season.

2.3 Physiological and Yield Responses of Canola to Water Stress

2.3.1 The Effect of a Water Stress Event on Physiological Processes

The primary process that is yield-limiting when a plant is affected by water deficit is photosynthesis (Ahmadi and Bahrani, 2009). This process is physically limited through the reduction of stomatal conductance, which leads to reduced carbon assimilation (Issarakraisila, 2007; Li et al., 1994). This creates a source limitation, which Ahmadi and Bahrani (2009) suggest is the main contributing factor to the resulting pod abortions. It has been shown that photosynthesis and stomatal conductance of a water-stressed canola plant can be 67 to 97 and 65 to 85 % lower, respectively, when compared to water replete plants (Hashem et al., 1998). Similar results have been reported by Dabas and Sheoran (1984). Non-stomatal limitations to photosynthesis can also occur under water stress, if the stress is severe enough to affect the photosynthetic machinery of the leaf, for example by inactivation of enzymes (Li et al., 1994).

In addition to the effects that water stress has on these gas exchange characteristics, there are other plant processes that can be affected, though they are less limiting to yield. An example of this is how water stress can negatively affect plant metabolism due to reduced transpiration and subsequent increased leaf temperatures (Mahan and Upchurch, 1988). Overall, the effect that water stress has on a canola crop is reduced dry mater production and seed yields (Wright et al., 1995).

2.3.2. Critical Periods of Water Stress in Canola

The timing of water stress is more influential than the intensity of the stress (Korte et al., 1983). A canola crop that is exposed to water stress in early vegetative stages can have reduced

LA due to decreased turgidity and LA expansion, as well as accelerated leaf senescence. As explained in Section 2.2.4, reduced LA results in a decreased source for individual plants, and therefore there is a reduced amount assimilate that can be allocated to sinks (Shahin and Valiollah, 2009). Decreased assimilate per plant can result in reduced branching and a reduction in subsequent yield components (Clarke and Simpson l978; McGregor 1987; Morrison, 1990a,b; Diepenbrock, 2000; Ozer, 2003; Ali et al., 1996; Degenhardt and Kondra, 1981); However, if the stress-induced loss of LA occurs during an early vegetative stage, the crop is capable of producing new leaves and recovering from the stress with relatively little yield loss (Tesfamariam et al., 2010; Gan et al., 2004; Hashem et al., 1998). When a water stress occurs during or after flowering, the crop has much less opportunity to recover (Morrison, 1993; CCC, 2011).

For a canola crop, flowering is known as the critical and most sensitive growth stage to water stress (Ahmadi and Bahrani, 2009; Rao and Mendham, 1991; Tesfamariam et al., 2010; Champolivier and Merrien, 1996; Hall, 1992; Sinaki et al., 2007; Kutcher et al., 2010). Due to the susceptibility of pollen development, anthesis and fertilization to water stress at this time, the canola crop may become sink limited as a result of increased flower and pod abortions (Ahmadi and Bahrani, 2009; Tesfarmariam et al., 2010; Mendham and Salisbury, 1995).

In addition to a reduced sink size, other results of water stress at flowering can include reduced duration of reproductive growth (Stoker and Carter, 1984; Hall, 1992), and since canola has an indeterminate growth habit, initiation of new branches. This can cause the plant to continue to produce vegetative growth into an extended growing season, resulting in delayed and

non-synchronous pod maturation. In these situations, problems can arise at harvest due to green seed in later-set pods and pod shattering of earlier pods (Tesfarmariam et al., 2010). Total yield loss from stress at flowering varies depending on the stress intensity and duration as well as other variables such as heat, disease and pests. As such, it is not surprising that water stress at flowering as been reported to decrease seed yield from 30 to 88% (Ghobadi et al., 2006; Hashem et al., 1998).

When a canola crop is exposed to a water stress following flowering (the seed-filling and development stage) the crop can become source-limited as a result of induced leaf shedding and hastened maturity (Tesfamariam et al., 2010; Gan et al., 2004). The amount of assimilates produced or available to be translocated for seed development is also decreased (Ghobadi et al., 2006; Ahmadi and Bahrani, 2009; Hashem et al., 1998). This can lead to substantial yield loss and is why the seed development or filling stage is next to flowering for sensitivity when it comes to water stress (Stoker and Carter, 1984; Nielsen, 1997; Sinaki et al., 2007; Ghobadi et al., 2006; Leon and Becker, 1995).

2.3.3 The Effect of Water Stress on Canola Yield Components

As mentioned, water stress can have an influence during early vegetative stages by decreasing the number of branches per plant (Diepenbrock, 2000; Halvorson et al., 2001); however, this is not often reported in the literature (Clarke and Simpson, 1978; Hashem et al., 1998). On the other hand, reduced pod number per plant is recognized to be the largest contributor to yield loss and is most sensitive to water stress at the time of flowering (Norouzi et al., 2008; Ghobadi et al., 2006; Champoliver and Merrien, 1996; Sinaki et al., 2007; Ahmadi and

Bahrani, 2009; Hashem et al., 1998). Water stress at flowering has been reported to reduce pod number per plant by 30 to 60 % compared to control treatments (Champoliver and Merrien, 1996; Norouzi et al., 2008; Hashem et al., 1998). As mentioned, this reduction is primarily due to flower and pod abortions and the result is a sink-limited crop. Seed number per pod and seed weight have also been reported to decline as a result of water stress, though to a lesser extent than pods per plant (Wright et al., 1995; Hashem et al., 1998; Ghobadi et al., 2006; Norouzi et al., 2008; Diepenbrock, 2000).

Reduction in seed weight occurs most often when the stress is experienced during seed development. At this time, sink capacity (seeds per plant) is set, and the stress can decrease the amount of assimilates produced or available to be translocated for seed fill (Ghobadi et al., 2006; Ahmadi and Bahrani, 2009; Hashem et al., 1998). The opposite has been reported from a stress during flowering, where an increase in seed weight has been reported due to the reduced seeds per plant (Kumar et al., 1994).

CHAPTER 3

EFFECTS OF MANURE AND BORON APPLICATION ON EARLY VIGOUR AND STAND ESTABLISHMENT OF CANOLA

3.1 Hypotheses and Research Objectives

3.1.1 Field Experiments

3.1.1.1 *Hypotheses*

The hypothesis is that LDM application prior to planting improves plant establishment and vigour of canola in early growth stages to a greater extent than can be attributed to the manure's macronutrient, S, B and water content alone. To test this, pre-plant nutrient applications of a LDM (T_{LDM}) and a synthetic nutrient solution with equivalent N, P, K, S and B (T_{FERT+B}) were compared for their effect on early crop growth and final crop yields. The effect of T_{LDM} application was also compared to the same volume of water (T_W) to confirm that effects observed are not due to the moisture content of the manure. If our hypothesis is correct, we expect that T_{LDM} will have an increased Normalized Difference Red-Edge (NDRE) canopy spectral reflectance index, a measure related to the ground cover and greenness of a crop, as well as increased crop yields, compared to T_{FERT+B} and T_W. In addition, since canola yields tend to be unaffected by a wide range of plant densities, we hypothesize that the benefits of manure on early crop vigour will be most pronounced under the circumstances of a yield-limiting plant stand.

3.1.1.2 *Research Objectives*

1. To evaluate the pre-plant manure application effect on canopy establishment and early growth of spring canola.
2. To determine if the beneficial effects of manure on spring canola can be attributed to the manure's contribution of macronutrients, S, B or water.

3. To compare yield-limiting and non-yield-limiting canola stands in order to assess effects of early crop vigour on final crop yields.

3.1.2 Greenhouse Experiments

3.1.2.1 *Hypotheses*

In the design of the 2012 field experiment it was overlooked that in preliminary experiments, where enhanced early crop growth of canola was observed in LDM treatments and not in fertilizer treatments, that B was not added to the fertilizer. Failure to show evidence of enhanced crop establishment by T_{LDM} over that of T_{FERT+B} in the 2012 field study therefore may have been due to the fact that T_{FERT+B} contained B. This led to a revised hypothesis that B content in the manure is responsible for observations in preliminary studies of increased canopy establishment and vigour of canola. We tested this hypothesis by applying nutrient treatments of T_{LDM}, T_{FERT+B}, and a fertilizer solution identical to T_{FERT+B} but without B (T_{FERT-B}) prior to seeding plants in a greenhouse setting. Early establishment and vigour of canola plants was assessed by harvesting plants during a late stage of vegetative growth and comparing plant emergence, LA, shoot dry weight (SDW) and leaf-level NDRE of plants. If our hypothesis is correct, we predict that plants under T_{LDM} and T_{FERT+B} will have increased LA and SDW in comparison to

T_{FERT-B}.

3.1.2.2 *Research Objectives*

1. To evaluate whether pre-plant application of manure has a positive effect on early growth and canopy establishment of spring canola, which exceeds the effect of a fertilizer treatment with the same macronutrient content.
2. To determine whether B is the component of the manure that was responsible for the additional enhancement of vigour and establishment observed in canola plants of preliminary studies during vegetative growth.

3.2 Materials and Methods

3.2.1 Field Experiments

3.2.1.1 *Experimental Design and Layout*

Field experiments were established in the spring of 2012 and 2013 at the Elora Research Farm (43°38'27.76"N, -80°24'20.43"W). The experiment was a modified split-plot design, with five main-plot soil nutrient treatments and two tiers in strips across the main plots (Figure 3.1). There were six experimental units within each main plot treatment and three experimental units within each tier. Three seeding rates were randomly assigned to the three experimental units within each tier. There were four repetitions used in this study. For a detailed description of tested effects and degrees of freedom in this specific experimental design, see the Appendix (A-1). Experimental units were 9 m^2 (1.5 m wide and trimmed to 6 m long) and were surrounded by border plots of canola on each side, planted at a rate of 120 seeds m^{-2}.

3.2.1.2 *Field Conditions and Treatments*

The field site used for the 2012 experiment is characterized as a London Loam soil type according to a soil survey of the Elora Research Farm (Grey-brown Luvisolic loam till) (Lauzon, J., personal communication). An analysis of a soil sample taken prior to planting indicated the organic matter content to be 4.5 % and the pH to be 7.2. Soil in this area is estimated to have a silt loam texture**.**

Prior to treatment applications, 60 kg ha^{-1} P_2O_5 and K_2O (0-20-20) was broadcast to the entire experiment area, as recommended based on a soil test (OMAFRA, 2002). For a detailed soil analysis, see the Appendix (A-2). On May 22nd, a Nuhn manure spreader was used to apply each of the soil nutrient treatments over the main plots. Treatments included:

1. Control (T_0)
2. 50,000 L ha^{-1} water (T_W)
3. 50,000 L ha^{-1} LDM (T_{LDM})
4. 50,000 L ha^{-1} of a fertilizer solution containing N, P, K, S and B (T_{FERT+B})
5. 25,000 L ha^{-1} LDM (T_{LDM25})

The LDM was obtained from the Elora Research Station's dairy facility. The rate of manure used is in the low to mid-range of manure application rates recommended to be applied to crops as recognized by OMAFRA (2002) and was based on what was used in preliminary trials where the beneficial effect of manure on early canola growth was originally observed. T_{FERT} contained nutrients in amounts that simulated the nutrient value of a sample analysis of the LDM source. Nutrient content of the solution was calculated from the total percentage of nutrients in the

manure on a weight basis (w/w %). For a detailed manure analysis, see the Appendix (A-4). The final solution was made with 17.4 kg urea (46-0-0), 7.3 kg ammonium sulfate (21-0-0-24), 12.3 kg MAP (11-52-0), 20.3 kg potash (0-0-60), and 87 ml of 10% B solution (Alpine Plant Foods, New Hamburg ON). Due to a miscalculation, this amount of B applied was 1.33 times the desired amount (the amount in the manure source)[*]. Nutrients were dissolved in warm water before being added to 7,250 L of water held within the manure spreader. This manure tank allowed for internal circulation used to mix the solution before application. On an area basis, T_{FERT} plots received 75 kg N ha^{-1}, 44 kg P_2O_5 ha^{-1}, 84 kg K_2O ha^{-1}, 12 kg S ha^{-1} and 800 g B ha^{-1}.

Each treatment was incorporated into the soil two days following application using a disk cultivator and the seedbed was prepared using a Brillian cultipacker. Canola was seeded to subplots on May 25th with a small-plot cone seeder at 1.5 cm depth. The variety used was a commercial glufosinate-tolerant spring canola hybrid, InVigour 5440, treated by the supplier with the insecticidal and fungicidal seed treatment Prosper (Bayer Crop Science, Guelph ON) (1000-sdwt = 5.25 g; germination = 95%). Seeding rates of individual plots was according to one of three assigned planting treatments of 15 (S_{15}), 40 (S_{40}) or 120 (S_{120}) seeds m^{-2}, which resulted in plots of various plant densities (Figure 3.2). The lowest seeding rate was to ensure a yield-limiting plant stand, in which differences in yield due to crop vigour could be detected. Emergence was seen four days after planting (DAP) and stand counts were performed one week after emergence.

[*]B in fertilizer solutions throughout field and greenhouse experiments in this study were calculated with the misinformation that the weight of the APFB solution was 1 kg L^{-1}, when the weight of the solution was in fact 1.31 kg L^{-1}. The amount of actual B in the APFB solution is 10%, and therefore it contained 131 g B L^{-1}, while calculations in this study were based on a solution with 100 g B L^{-1}.

At the 5-6 leaf stage (June 27th), the entire experiment was top-dressed with 150 kg N ha^{-1} as urea (46-0-0). Flea beetle and weed pressures were controlled on July 5th with application of 83 ml ha^{-1} Matador insecticide (Syngenta Canada, Guelph ON) and 2 L ha^{-1} Liberty herbicide (Bayer Crop Science, Guelph ON) in a volume of 150 L ha^{-1}.

In spring 2013, the 2012 field trial was repeated in a different field at the Elora Research Station, classified as a London Loam soil type, with an organic matter content of 4.3% and a pH of 7.5.

Prior to treatment application, a pre-plant fertilizer of 120 kg ha^{-1} P_2O_5 and K_2O (0-20-20) was applied to the entire field, since soil test results indicated very low P and K content. This is in excess of what is recommended by field production guides, to eliminate any possibility for deficiency, given that we did not have the opportunity for deep incorporation. For a detailed soil analysis, see the Appendix (A-2). Weeds were controlled with pre-plant incorporated Treflan herbicide (Dow AgroSciences Canada Inc., Calgary AB) at a rate of 2 L ha^{-1}.

Treatment application occurred on May 21st, using the same methods as reported for the 2012 field trial. Treatments were applied at a rate of 50,000 L ha^{-1} and included:

1. Control (T_0)
2. Water (T_W)
3. LDM (T_{LDM})
4. A fertilizer solution containing N, P, K, S and B (T_{FERT+B})
5. A second fertilizer solution that contained N, P, K and S, but did not contain B (T_{FERT-B}).

Fertilizer solution calculations were conducted as in the 2012 experiment, though based on a 2013 manure analysis. For a detailed manure analysis, see the Appendix (A-4). The solution was made up of 21 kg urea (46-0-0), 4.5 kg ammonium sulfate (21-0-0-24), 11.3 kg MAP (11-52-0) and 13.5 kg potash (0-0-60), which was added to 7,500 L of water held in the manure tank. On an area basis, the plots under the T_{FERT-B} and T_{FERT+B} treatments therefore received 79 kg N ha^{-1}, 39 kg P_2O_5 ha^{-1}, 54 kg K_2O ha^{-1} and 7.2 kg S ha^{-1}. Once T_{FERT-B} was applied to main plots, 54 ml of 10% B solution was added to the remaining 4130 L of solution for the application of T_{FERT+B}, and therefore plots under the T_{FERT+B} treatments received 872 g B ha^{-1} in addition to the N, P, K and S. One day after treatment application, treatments were soil-incorporated and the seedbed was prepared as in the 2012 field experiment.

InVigour 5440 was planted to subplots at three seeding rates (S_{15}, S_{40} and S_{120}) on June 9th. Emergence was seen three DAP and stand counts were performed two weeks after emergence. At the 5-6 leaf stage (July 15th), 150 kg N ha^{-1} as urea (46-0-0) was applied over the entire experiment. Additional pest control over the season included an application of 83 ml ha^{-1} Madador on June 25th for flea beetle control and 250 ml ha^{-1} Coragen (DuPont, Mississauga, ON) on July 12th for swede midge control.

3.2.1.3 *Field Measurements*

In 2012, canopy spectral reflectance data was collected for each experimental unit using a dual channel reflectance spectrometer (Unispec DC, PP Systems, Amesbury MA) on four dates throughout the seedling and rosette stages, up until budding (June 15, June 19, June 26 and July

6) (Figure 3.3). Five measurements from each subplot were used to calculate the mean NDRE for each plot. NDRE has been used as a measure of canopy cover and greenness, and relative to the Normalized Difference Vegetative Index, NDRE is more colour-sensitive and less affected by LA (Eitel et al., 2008). NDRE is calculated as:

$$NDRE = (R_{790} - R_{720}) / (R_{790} + R_{720}),$$

where R_λ is fractional canopy reflectance of incident solar radiation in the 3-nm waveband centered on λ.

Due to stress imposed on the crop by swede midge and drought during the critical reproductive period of the 2012 experiment, plots were not harvested to obtain yield data.

2013 reflectance data was collected for each subplot on five dates (June 24, July 2, July 6, July 13 and July 19) and plot means of NDRE were calculated. Subplots were harvested with a plot combine on October 11th, which was late for canola harvest. The late harvest was due to the late planting date and wet fall conditions. After harvest, seed was dried further for 3 days in a forced-air dryer at 60^{o}C. Moisture data for harvested seed was collected using a GAC II Grain analysis computer (Dicky-John Corporation, Auburn IL) and seed weight was corrected to 10% moisture content.

3.2.1.4 *Statistical Analysis*

NDRE values for 2012 and NDRE and yield values for 2013 were analyzed with SAS version 9.2 as a modified split-plot design, using a repeated measures approach for the NDRE data. A mixed model procedure at a type I error rate (α) of 0.05 was used. Missing data in the

2013 NDRE data set included one replication of reflectance measurements on the first sampling date. An Analysis of Variance (ANOVA) was conducted and assumptions of the ANOVA were met according to tests in an analysis of residuals. A Tukey's HSD (Honestly Significant Difference) test was used to obtain treatment means separations.

3.2.2 Greenhouse Experiments

3.2.2.1 *Experimental Design*

This experiment was designed as a randomized complete block (RCBD) with six replications and four soil nutrient treatments. Experimental units were made up of 51 x 17 x 15-cm box planters.

3.2.2.2 *Greenhouse Conditions and Treatments*

Plants were grown in a greenhouse at the University of Guelph in two experiments, one being in the winter of 2013 (February to March), and one in the summer of 2013 (July to August). Target day/night air temperature settings were 22/17^{o}C. A mixture of metal halide and high pressure sodium lamps provided a photosynthetically active photon flux density of approximately 600 μmol m^{-2} s^{-1} to the tops of plants, which supplemented incoming sunlight for 16 hours per day. The greenhouse settings allowed a shade cloth to deploy, and lamps to turn off if temperatures in the greenhouse exceeded 35^{o}C. Pest control was met throughout the experiment by integrated pest management (BioBee Biological Systems Ltd., Sde Eliyaho, Israel) where predators were applied to plants based on weekly scouting records.

Since the chemical and physical characteristics of potting mixes differ from those of field soil, field soil was used as the potting medium. Soil was taken from the top 15 cm of a field at the Elora Research Station. As in field studies, this soil was a London Loam soil type (Grey-brown Luvisolic loam till) (Lauzon, J., personal communication). An analysis of a soil sample indicated the organic matter content to be 3.2 % and the pH to be 7.6. Soil in this area is estimated to have a silt loam texture. For a detailed soil analysis, see the Appendix (A-3).

Soil was sieved through a 1 mm screen and stored in plastic bins that were lined and sealed with a thick plastic liner to ensure that the moisture content stayed consistent within each bin. Prior to treatment application, LDM was obtained from the Elora Research Station dairy facility.

On February 24th, box planters were filled with the sieved soil to a volume of 9 L and a weight of 7.5 kg dry soil. Experimental units within the same replication were filled from soil that was stored in the same bin, to ensure uniform soil moisture within each of the six replications. All box planters were supplemented with N as ammonium nitrate (34-0-0) at a rate equivalent to 100 kg ha^{-1}, which was mixed into the top 10 cm of the soil. It should be noted that this differs from the field protocol where additional N was applied as a split application prior to bolting. Four nutrient treatments were randomly applied to the boxes using a metal plate to simulate a spreading action and to ensure uniform application. A thin layer of soil was added to the top of each planter to simulate incorporation. Treatments were applied at a rate equivalent to 50,000 L ha^{-1} and included:

1. Control (water only) (T_W)

2. LDM (T_{LDM})
3. A fertilizer solution containing N, P, K, S and B (T_{FERT+B})
4. A second fertilizer solution that contained N, P, K and S, but did not contain B (T_{FERT-B}).

The nutrient content of the fertilizers were calculated as in the field studies, based on a manure sample analysis from February 19th, 2013. For a detailed manure analysis, see the Appendix (A-4). The fertilizer solutions were then prepared with 12.1 g urea (46-0-0), 2.8 g ammonium sulfate (21-0-0-24), 4.7 g MAP (11-52-0), and 9.3 g potash (0-0-60) added to 3,500 ml of warm water. Additionally, 0.05 ml of 10% B solution was included in the T_{FERT+B} solution only, which on an area basis works out to 950 g B ha^{-1}. Looking at the other nutrients on an area basis, each of the T_{FERT+B} and T_{FERT-B} treatments received 95 kg N ha^{-1}, 35 kg P_2O_5 ha^{-1}, 80 kg K_2O ha^{-1}, and 10 kg S ha^{-1}.

The experiment was seeded to spring canola three days after treatment application at a rate of 150 seeds m^{-2} (13 seeds per box) and 12 mm depth. The seed used was the same as for the field trials.

Using field soil as a medium for containers in a greenhouse setting can be challenging due to drainage and crusting issues. To address this, a target weight where the soil moisture content of individual boxes was equal to 80% of field capacity (FC) was calculated. This was done by first taking samples of the sieved soil to calculate the moisture content and dry weight of the soil of each replication. The FC of the silt loam soil used in this study is approximately 28% soil moisture by volume and therefore the daily target watering weight of boxes was established

at 22% moisture (Lauzon, J., personal communication). This target weight is in accordance with a controlled environment study by McGonigle et al. (1990), who used a similar soil type from a nearby location at the Elora research station.

This experiment was repeated for a second time in the same facility to clarify observed patterns in the first set of results. Six additional replications were planted on July 24th and harvested on August 21st, 2013. Nutrient content of the fertilizer solutions were re-calculated based on a new manure sample analysis from July 17th, 2013. For a detailed manure analysis, see the Appendix (A-4). The fertilizer solutions were prepared with 10.5 g urea (46-0-0), 2.7 g ammonium sulfate (21-0-0-24), 5.4 g MAP (11-52-0), and 7.3 g potash (0-0-60) added to 4,000 ml of warm water. 0.05 ml of 10% B solution was included in the T_{FERT+B} solution only, which on an area basis works out to 830 g B ha^{-1}. Looking at the remaining nutrients on an area basis, each of the T_{FERT} and T_{FERT-B} treatments received 75 kg N ha^{-1}, 35 kg P_2O_5 ha^{-1} 55 kg K_2O ha^{-1}, and 8 kg S ha^{-1}.

3.2.2.3 *Measurements*

Emergence was seen four DAP and the number of seedlings per box was recorded two weeks after emergence. One day prior to harvest, leaf spectral reflectance was measured using a Unispec SC reflectance spectrometer (PP Systems, Amesbury MA) (Figure 3.4). Leaf reflectance samples from newest fully expanded leaves of ten individual plants were randomly taken from each experimental unit. On March 25th (5-6 leaf stage), plants were harvested and LA was determined with a LA meter (LI-3100, LICOR, Lincoln NE). Fresh shoot biomass was collected from each experimental unit and SDW was taken after samples were dried to a constant weight at

80^{o}C in a forced-air drier.

3.2.2.4 *Statistical Analysis*

Mean plant number, LA, LA per plant, SDW, SDW per plant and NDRE of experimental units in winter and summer experiments were analyzed as a RCBD using SAS version 9.2. A mixed model procedure with a type I error rate (α) of 0.05 was used. Data from winter and summer experiments were not pooled due to significant crossover interactions between the treatments and experiments. An ANOVA was conducted and assumptions of the ANOVA were met according to tests in an analysis of residuals. A Tukey's HSD test was used to obtain treatment means separations.

3.3 Results and Discussion

3.3.1 Field Experiments

3.3.1.1 *Canopy Establishment*

When analyzed as a repeated measures study, the 2012 field trial experiment did not reveal an overall significant effect of soil nutrient treatments on canopy establishment as per NDRE data (P=0.06). Looking at individual sample dates, 42 DAP was the only date where there was a significant difference between nutrient treatments and the T_0 condition. Specifically, T_{FERT+B} had a significantly greater average NDRE than that of T_0 (Figure 3.5).

Soil treatments were altered slightly from 2012 to 2013 to test the hypothesis that the lack of difference observed between T_{FERT+B} and T_{LDM} in the 2012 field study may have been be due to the inclusion of B in the fertilizer treatment.

As in 2012, the 2013 field trial failed to reveal a significant effect of soil nutrient treatments on canopy establishment (P=0.06). In terms of individual sample dates, plots receiving T_{FERT+B} and T_{FERT-B} had significantly greater NDRE means than that of T_0 at 40 DAP (Figure 3.5). Plots receiving T_{FERT-B} also had a significantly greater average NDRE than T_W at both 34 and 40 DAP. This suggests that the effect of T_{FERT+B} on canopy establishment was not improved with the addition of B.

One trend over the experiments was for T_0 and T_W plots to have the lowest NDRE means on each measurement date. T_{LDM} did not significantly differ from these two treatments, nor from T_{FERT+B} and T_{FERT-B}. Although the aim of the study was to have the fertilizer solution in T_{FERT} mirror the nutrient analysis of T_{LDM}, the resulting NDRE means of T_{LDM} and T_{FERT} differed slightly. This may be explained by the fact that a substantial portion of N and S in the manure was not plant-available at the time of planting. These unavailable nutrient fractions, estimated to be up to 55% of total N and greater than 60% of total S (A & L Canada Laboratories, 2014), act as slow-release fertilizers, becoming available to plants through mineralization (Eghball et al., 2002). Mineralization is affected by temperature, moisture and microbial activity and therefore these nutrient fractions were subject to becoming available only later into the growing season. In addition to this, according a manure analysis conducted by A&L Laboratories, P (as P_2O_5) applied from this specific manure source was estimated to be only 40% plant-available in the first year of application. This is in accordance with estimations from OMAFRA (2013b). Fertilizer solutions in this experiment were calculated based on nutrient totals in the manure, and did not take into account unavailable fractions in order to test our hypothesis vigorously, since it

would be difficult to accurately estimate the amount of nutrients available during the canopy establishment period. From this, it is likely that greater fractions of plant available N, P and S nutrients in T_{FERT+B} and T_{FERT-B} compared to that available in T_{LDM} resulted in a greater means separation from T_C and T_W.

3.3.1.2 *Plant Populations and Yield*

Average percent emergence in the 2012 field experiment was not significantly affected by seeding rate or nutrient treatments (P = 0.79 and P = 0.17, respectively). S_{15}, S_{40} and S_{120} resulted in average plant stands of 12, 31 and 99 plants m^{-2}, respectively. Emergence was therefore 82, 79 and 82 %, respectively, which is high for canola at this location, where typical establishment rates are around 50% (H.J. Earl, personal communication).

As in 2012, average percent emergence in the 2013 study was not affected by seeding rate or nutrient treatments (P = 0.08 and P = 0.14, respectively). S_{15}, S_{40} and S_{120} resulted in average plant stands of 14, 36 and 99 plants m^{-2}, giving average percent emergence of 91, 90 and 82 %, respectively.

According to LRSA (2014) temperature data depicted in Figure 3.6, it is evident that the growing season of 2012 was slightly warmer than that of 2013, experiencing a greater number of days where the daily high was above 29.5^0C. This temperature has been identified by Morrison and Stewart (2002) as the critical temperature above which a canola crop to experiences heat stress. The crop of 2012 also experienced more stress due to drought, as can be observed from the cumulative precipitation of each season (Figure 3.6). From mid-June until the

end of July, a time period which encompasses the entire critical flowering period (Kutcher, 2010), the 2012 canola crop received only 8.5 mm of precipitation. For this reason, this crop failed and was not harvested for yield. During this same period in the 2013 field season, the crop received 150.5 mm of precipitation. The crop failure of 2012 due to drought stress reinforces what has been shown by many in the literature, that canola is highly sensitive to stress during the critical period of flowering (Ahmadi and Bahrani, 2009; Rao and Mendham, 1991; Tesfamariam et al., 2010; Champolivier and Merrien, 1996). The lack of control of swede midge in 2012 was also likely another factor that contributed to the crop failure.

Seeding rate had a significant effect on 2013 yields ($P<0.0001$). The lowest average yields were produced by S_{15}, as was expected, since this seeding rate treatment was included to serve as a yield-limiting factor (Table 3.1). Greatest yields were produced in S_{120}. This yield was average for Ontario, as the Ontario yield average in 2013 was 2,000 kg ha^{-1} (OMAFRA, 2013). A later than average planting date as well as damage from Swede midge (*Contarinia nasturtii* (Keiffer)) may have contributed to some yield loss in the 2013 experiment.

Yields produced in S_{40} were significantly lower than those produced in S_{120}. In agreement with these findings, researchers such as Lythgoe et al. (2001) and Ozer (2003) failed to see full yield compensation from low-density plantings, observing significantly lower yields with decreased plant density. Such findings are challenged in the literature by others who have shown that seeding rate does not significantly affect seed yields, with final yields reportedly unaffected by ranges of densities such as 50 to 130 plants m^{-2} (Potter et al., 1999), 40-80 plants m^{-2} (Angadi et al., 2003) and 20-80 plants m^{-2} (Mendham et al., 1981). Under these circumstances, individual

plants in low-density populations were able to compensate through alteration of morphology and yield components.

The 2013 yields were not significantly affected by the nutrient treatments (P=0.63). This indicates that although there were significant differences observed in canopy establishment between T_0 and the liquid starter fertilizer treatments (T_{FERT+B} and T_{FERT-B}), these were not large enough to affect yield. In other words, the superior early ground cover indicated by reflectance measurements of T_{FERT+B} and T_{FERT-B} plots did not translate to superior yields.

3.3.2 Greenhouse Experiments

Results of winter and summer 2013 greenhouse experiments were not combined for LA, SDW or leaf NDRE due to significant experiment x treatment interactions shown in the ANOVA summary (Table 3.2). The results for plant number per box, leaf area per plant (LAPP) and shoot dry weight per plant (SDWPP) did not show a significant experiment x treatment interaction and experiments of winter and summer were pooled together (Table 3.3).

3.3.2.1 *Plant Number*

Treatment means of plant number per box were pooled across experiments (Table 3.3), since results showed no treatment x experiment interaction (Table 3.2). There was a significant treatment effect on plant number per box, where T_0 and T_{LDM} resulted in a significantly greater plant number per box than T_{FERT+B} (Table 3.3). Plant number per box in T_{FERT-B} was not significantly different from that of T_{FERT+B}. Variations in plant number per box were likely a result of T_{FERT+B} and T_{FERT-B} having an injurious effect on germinating seeds. This can occur

when certain fertilizers in adequate concentrations are placed in close proximity to the seed, creating a toxicity or osmotic pressure change **(**Mason, 1971).

Recommendations for producers state that N (especially as urea) as well as S, K, and to a lesser extent, P are not to be placed in close contact with seeds, especially for crops with small seeds such as canola. As an alternative to seed-placed fertilizers, one can band two inches beside and below the seed, or broadcast the fertilizer. These recommendations are to reduce the potential for injury due to ammonia toxicity and increased osmotic pressures (salt content) of the soil solution in the vicinity of the seed (Malhi and Gill, 2004; Burglund et al., 2007; Molberg, 1961).

Although T_{LDM} solutions contained identical nutrients of N, P, K, S as T_{FERT+B} and T_{FERT-B} solutions, T_{LDM} did not show signs of fertilizer toxicity on seed germination. This can be explained by difference in the form of N in T_{LDM} compared to in the solutions of T_{FERT+B} and T_{FERT-B}. The source of N in T_{FERT+B} and T_{FERT-B} fertilizer solutions was urea. When applied to soil, urea [$CO(NH_2)_2$] is rapidly hydrolyzed to ammonium (NH_4^+) by the soil enzyme urease (Agehara and Warncke, 2005). Another product of this reaction is ammonia (NH_3^+), which is toxic, and the known compound responsible for the adverse effect that urea has on seed germination (Bremner and Krogmeier, 1989).

Though animal manures such as the one used for T_{LDM} contain NH_4^+, which undergoes hydrolysis by urease just as in the case of urea, it only makes up a fraction of total N. According to an analysis of the LDM used in this study, total N was approximately 50% NH_4^+ and 50%

organic-N. The organic-N, which is slowly mineralized to NH_4^+, was likely of very little concern as a source of toxicity (Agehara and Warncke, 2005; Paul and Beauchamp, 1993a). It is therefore not surprising that T_{FERT+B} and T_{FERT-B}, which contained N as urea, showed evidence of reduced emergence due to ammonia toxicity, while this effect was not evident in T_{LDM}.

Ammonia volatilization, where NH_4^+ is converted to NH_3, can also occur in the NH_4^+ fraction of the LDM and can be toxic to germinating seeds (Chambers et al., 1999); However there was not likely a large amount of ammonia volatilization that occurred in pots that received LDM, since there was a thin layer of top-soil applied on top of the manure to simulate incorporation, which reduces N-losses by volatilization.

3.3.2.2 *Leaf Area*

There was a significant experiment x treatment interaction observed between the winter and summer experiments for LA of harvested plants, and therefore data for this response variable were not pooled across experiments (Table 3.2). The interaction stemmed from the fact that in the winter study, T_0 had a significantly lower LA than all other treatments, while the LA of T_0 plots in the summer study did not differ from the other treatments. This inconsistency may be attributable to the greenhouse temperature difference in the winter versus the summer months. In the winter study, temperatures followed that of the set target day/night temperatures of 22/17^{o}C, whereas in the summer study, the actual temperatures deviated from the set targets. Specifically, throughout the summer experiment, the average day/night temperatures were 31/18^{o}C. This is a 10^0C temperature difference, which may be large enough to affect soil mineralization (Paul and Beauchamp, 1993b). In addition, soil used for the summer experiment was stored indoors at the

Elora Research Facility where mineralization also could have occurred with temperatures of approximately 20^{o}C. The combination of these factors likely led to T_0 plots having a greater supply of nutrients in the summer months compared to the winter months, and in turn, LA means separations deviated from those of the winter study.

Another difference between the winter and summer studies is that in the winter, mean LA of T_{LDM} and T_{FERT-B} were significantly less than that of T_{FERT+B}; However, in the summer study, T_{FERT+B} had a significantly lower LA than T_{LDM} (Table 3.2)**.** The inconsistency between experiments was likely due to the number of plants per box. T_0 and T_{LDM} in the summer study resulted in an average of 36 and 31% more plants per box than the T_{FERT+B} treatment, respectively.

Plants grown at a lower density have a tendency towards greater branching and leaf expansion in response to utilizing additional resources that come with increased space per plant (Donald, 1963). Higher density plantings then, in comparison, tend to have less LA per plant and fewer branches. Regarding the observation in this study that T_{FERT+B} had a significantly lower LA than T_{LDM}, we speculate that the plant density differed enough that, regardless of the nutrition, the low-density plot (T_{FERT+B}) was not able to compensate up to the LA achieved by the high-density plot (T_{LDM}). This issue was not evident in the winter study, since plant number per box differed by only 13% from the highest number in T_0 to the lowest number in T_{FERT-B} (Table 3.2).

LA on a per-plant basis was not significantly different between experiments, so the data was pooled Table 3.3. Trends of LAPP are different from that of the LA response variable likely due to the number of plants per box, which varied across treatments in a similar pattern. LAPP in T_{FERT+B} and T_{FERT-B} was significantly greater than that of T_0, which is not surprising since there were fewer plants for LA to be divided over (Table 3.3). LAPP of T_{LDM} did not differ from T_0 or T_{FERT-B}. Since plant number per box varied across treatments and this had a great effect on this response variable, it is not appropriate to interpret this data based on treatment effects alone.

3.3.2.3 *Shoot Dry Weight*

Results of mean SDW were not pooled across experiments due to a treatment x experiment interaction (Table 3.2). SDW in the winter study follow a similar pattern to LA, in that T_0 had significantly less above ground biomass on an area basis when compared to all other treatments. The winter SDW trends differ from the LA trends likely due to the effect that plant number per box has on the distribution of shoot biomass into LA versus stems and petioles. T_{LDM} had a significantly greater SDW than T_{FERT-B}, though it failed to differ from T_{FERT+B}. T_{FERT-B} and T_{FERT+B} did not differ (Table 3.2). This observation was the underlying reason that six more replications were conducted in the study, to further investigate the possibility that T_{LDM} and T_{FERT+B} have an effect on SDW that T_{FERT-B} does not. In the summer study however, T_{LDM} resulted in significantly greater SDW than all other treatments, and the remaining treatments did not differ, resulting in another treatment x experiment crossover interaction. Since T_{FERT+B} could not replicate the effect that T_{LDM} had in the winter study, it can be concluded that the effect was not likely due to the presence of B in the manure.

Shoot dry weight per plant was significantly affected by treatments, and treatment means were pooled together (Table 3.3) due to the absence of a treatment x experiment interaction (Table 3.2). As found in the results of LA and LAPP, trends of SDWPP are different from that of the SDW response variable due to the number of plants per box, which varied across treatments. An exception to this is the significant increase in LAPP in T_{LDM} compared to that of T_0, which can be attributed mostly to treatment effects, since plant number per box did not differ between these treatments. Since plant number per box varied across treatments and this had a great effect on this response variable, it is not appropriate to compare treatment effects on LAPP of T_{FERT+B} and T_{FERT-B} to that of T_0 and T_{LDM}.

3.3.2.4 *Normalized Difference Red-Edge Index*

NDRE treatment means were not pooled across experiments due to a significant treatment x experiment interaction (Table 3.2). NDRE in all nutrient treatments were significantly greater than those of T_0 in the winter study, whereas in the summer study, the treatments did not differ from one another in their effect on NDRE (Table 3.2)**.** Since NDRE in the greenhouse study was at the leaf level, this could only be looked at as an indication of nutrient status of the plants. The leaf colour differential indicated by NDRE results therefore confirm that T_0 was nutrient-deficient in the winter study. Deficiency was not as extreme in the summer study, possibly due to soil mineralization from warm temperatures.

3.3.3 Comparison of Field and Greenhouse Experiment Results

The effect of nutrient treatments on stand establishment in this study were measured by NDRE in the field (Figure 3.5) and by LA and SDW in the greenhouse (Table 3.2). Comparing

these results on an area basis, SDW was the greatest in T_{LDM} for both winter and summer studies, whereas in the field, NDRE values of T_{LDM} were less than that of T_{FERT+B} and T_{FERT-B}. Thus, the results from the greenhouse and field are in conflict.

The percent emergence was significantly affected by nutrient treatments in the summer greenhouse study (P = 0.0092), and in both winter and summer greenhouse studies, the trends were for T_{FERT+B} and T_{FERT-B} to have reduced plant emergence compared to that of T_{LDM} and T_0, indicating a fertilizer toxicity issue. In 2012 and 2013 field studies, percent emergence was not significantly affected by nutrient treatments (P = 0.17; P= 0.14), which suggests that the fertilizer toxicity seen in the greenhouse was absent in the field. Based on this, it is likely that the enhanced effect on SDW by T_{LDM} in the greenhouse was not a result of an additional nutrient or factor that was absent in T_{FERT+B} and T_{FERT-B} conditions, but rather due to T_{FERT+B} and T_{FERT-B} producing a reduced plant stand from fertilizer toxicity.

The fertilizer toxicity that was observed in the greenhouse was not observed in the field likely because the application and incorporation of the materials was different. In the greenhouse study, application of the fertilizer was in a more concentrated band, which was very close to seeding depth. By contrast fertilizers applied in the field were mixed by tillage, which reduced the potential for fertilizer toxicity.

3.4 General Conclusions

NDRE and yield data from field experiments failed to show any benefit to a pre-plant application of LDM that could not be reproduced by a liquid fertilizer of the same nutrient

content. Similarly, greenhouse experiments did not show evidence that pre-plant B at the level present in a LDM application improves early vigour and establishment.

SDW measurements in the greenhouse study suggested that T_{LDM} may have had a positive effect on early crop growth compared to that of T_{FERT+B} and T_{FERT-B} treatments; However we could not draw conclusions from this. Plots were not established with an equal plant population, in order to observe the full effect of nutrient treatments on early growth and development (from germination until the late vegetative stage), and the effect that nutrient treatments had on germination and emergence made the effect that they had on subsequent vigour unclear. T_{FERT+B} and T_{FERT-B} had reduced plant emergence, likely due to fertilizer toxicity, and the unequal plant number among treatments influenced SDW in individual plots. Enhanced SDW observed in T_{LDM} was therefore likely due to this reduction in emergence of T_{FERT+B} and T_{FERT-B} plots, especially since results were not consistent with that of the field experiments, where fertilizer toxicity was not apparent.

Table 3.1 Means of spring canola (*Brassica napus* L.) yield as affected by seeding rate in a field experiment at the Elora Research Station, ON in 2013.

Seeding Rate (seeds m^{-2})	**Yield † (kg ha^{-1})**
15	1469 *c**
40	1812 *b*
120	2064 *a*

* Seeding rate means followed by the same letter do not differ significantly ($P < 0.05$) according to a Tukey's HSD test.
† Yields were adjusted based on seed moisture corrected to 10%.

Table 3.2 Summary of Analysis of Variance (ANOVA) and means comparisons of harvest measurements and leaf Normalized Difference Red-Edge (NDRE) index values for spring canola (*Brassica napus* L.) harvested at a late vegetative stage as affected by starter nutrient treatments in a greenhouse setting at the University of Guelph in winter and summer 2013 experiments.

Treatment	Plants (box^{-1})	Leaf Area (cm^2)	Leaf Area $Plant^{-1}$ (cm^2)	Shoot Dry Weight (g)	Shoot Dry Weight $Plant^{-1}$ (g)	Leaf NDRE
Experiment 1: Winter 2013						
Control	12.1	700 *c* †	57.8 *b*	9.7 *c*	0.80 *b*	0.242 *b*
Liquid Dairy Manure	11.3	1087 *b*	97.3 *a*	12.9 *a*	1.15 *a*	0.275 *a*
Fertilizer+B	11.0	1215 *a*	111.0 *a*	12.5 *ab*	1.14 *a*	0.272 *a*
Fertilizer-B	10.5	1098 *b*	109.7 *a*	11.3 *b*	1.12 *a*	0.264 *a*
Experiment 2: Summer 2013						
Control	10.5 *a*	1016 *ab*	107.0 *b*	6.5 *b*	0.69 *b*	0.247
Liquid Dairy Manure	9.7 *ab*	1145 *a*	109.9 *ab*	8.0 *a*	0.77 *ab*	0.243
Fertilizer+B	6.7 *b*	966 *b*	148.6 *a*	6.2 *b*	0.96 *a*	0.237
Fertilizer-B	7.7 *ab*	1016 *ab*	139.3 *ab*	6.4 *b*	0.87 *ab*	0.246
Summary of ANOVA (Pr > F)						
Treatment, Winter 2013	NS ‡	**	**	**	**	**
Treatment, Summer 2013	**	*	*	**	*	NS
Experiment x Treatment	NS	**	NS	**	NS	**

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† Within an experiment, nutrient treatment means followed by the same letter do not differ significantly ($P < 0.05$) according to a Tukey's HSD test.
‡ NS = not significant.

Table 3.3 Treatment means for harvest measurements which were pooled across experiments for spring canola (*Brassica napus* L.) harvested at a late vegetative stage as affected by pre-plant nutrient treatments in a greenhouse setting at the University of Guelph in 2013.

Treatment	Plants (box^{-1})	Leaf Area $Plant^{-1}$ (cm^2)	Shoot Dry Weight $Plant^{-1}$ (g)
Control	10.9 *a**	82.4 *c*	0.74 *b*
Liquid Dairy Manure	10.9 *a*	103.6 *bc*	0.96 *a*
Fertilizer+B	8.8 *b*	129.8 *a*	1.05 *a*
Fertilizer-B	9.1 *ab*	124.5 *ab*	1.00 *a*

* Treatment means followed by the same letter do not differ significantly ($P < 0.05$) according to a Tukey's HSD test.

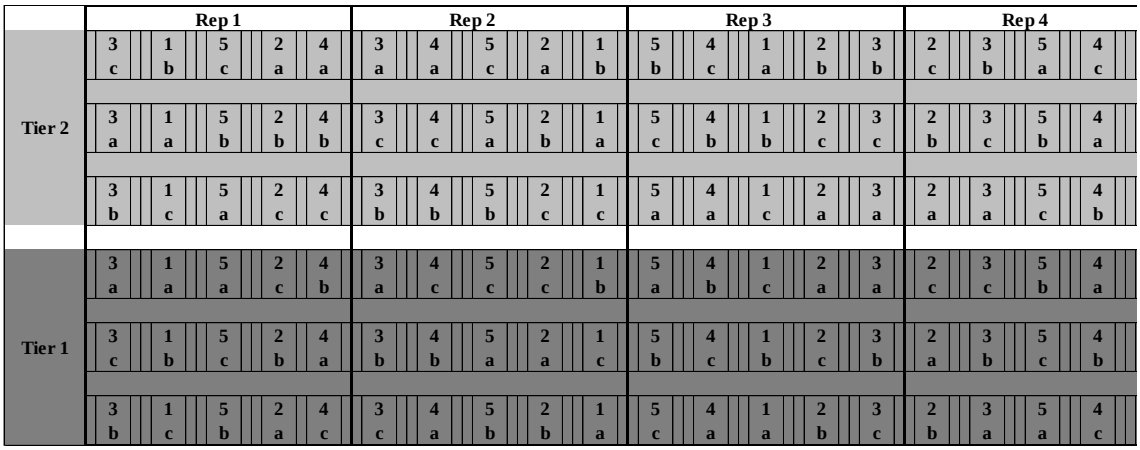

Figure 3.1 A schematic plot plan of the modified split-plot design, including two tiers with four replications. Pre-plant soil nutrient treatments (1 to 5) were applied in vertical strips and not re-randomized within tiers. Seeding rate treatments (a, b, c) were re-randomized within each tier.

Figure 3.2 Experimental plots of Spring canola (*Brassica napus* L.) in the vegetative stage at the Elora Research Station, ON in 2012.

Figure 3.3 Obtaining canopy reflectance data using the Unispec dual channel reflectance spectrometer at the Elora Research Station, ON.

Figure 3.4 Unispec SC reflectance spectrometer (top) and leaf area meter (bottom) used to take measurements during harvest of the winter and summer 2013 greenhouse experiments at the University of Guelph.

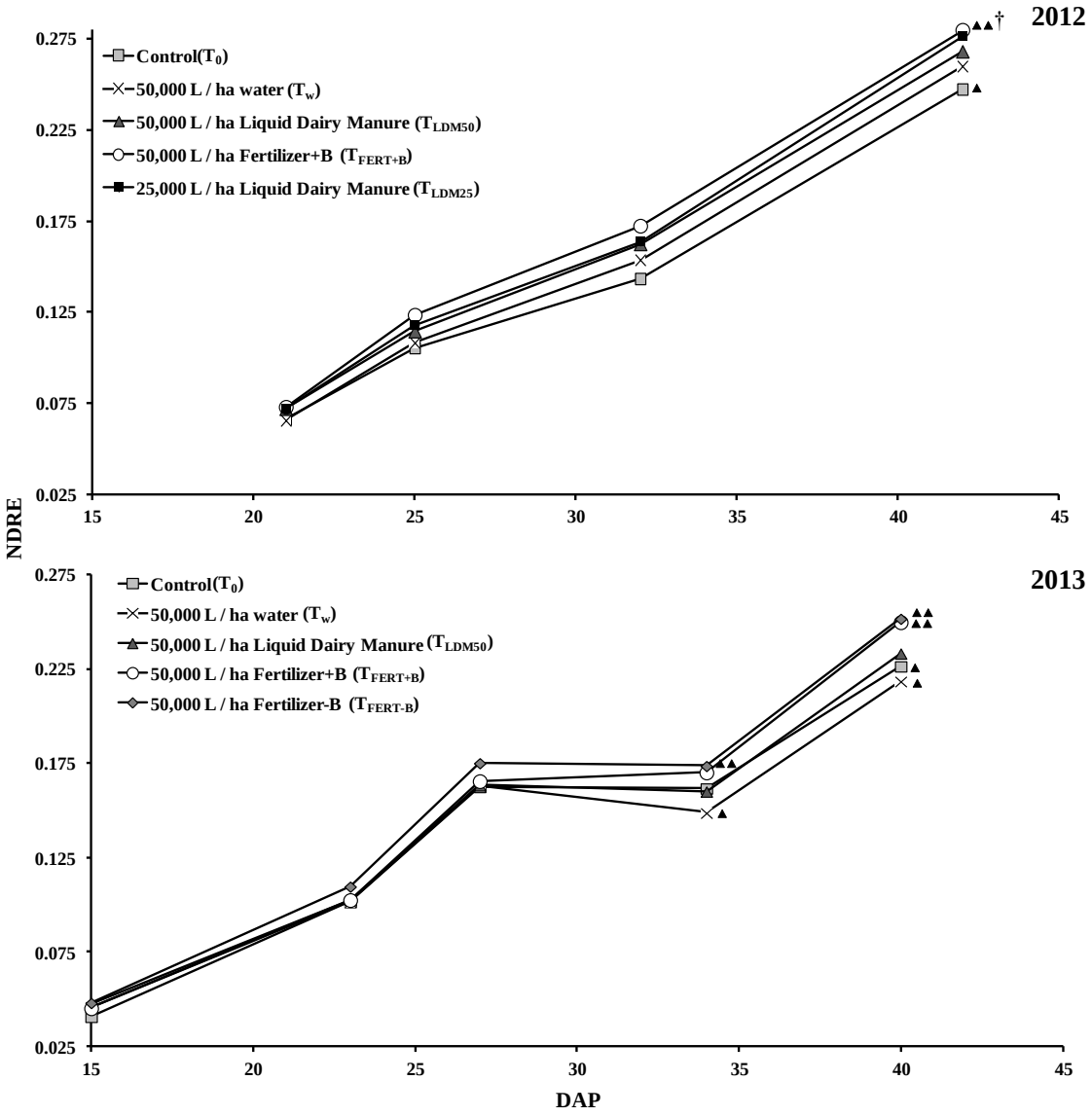

Figure 3.5 Mean NDRE values, averaged over seeding rates, of a spring canola (*Brassica napus* L.) canopy from 15 to 45 days after planting (DAP), as affected by starter nutrient treatments in 2012 and 2013 field seasons at the Elora Research Station, ON.

† Within each date and year, NDRE means labelled ▲▲ differ significantly than those labelled ▲ ($P < 0.05$) according to a Tukey's HSD test.

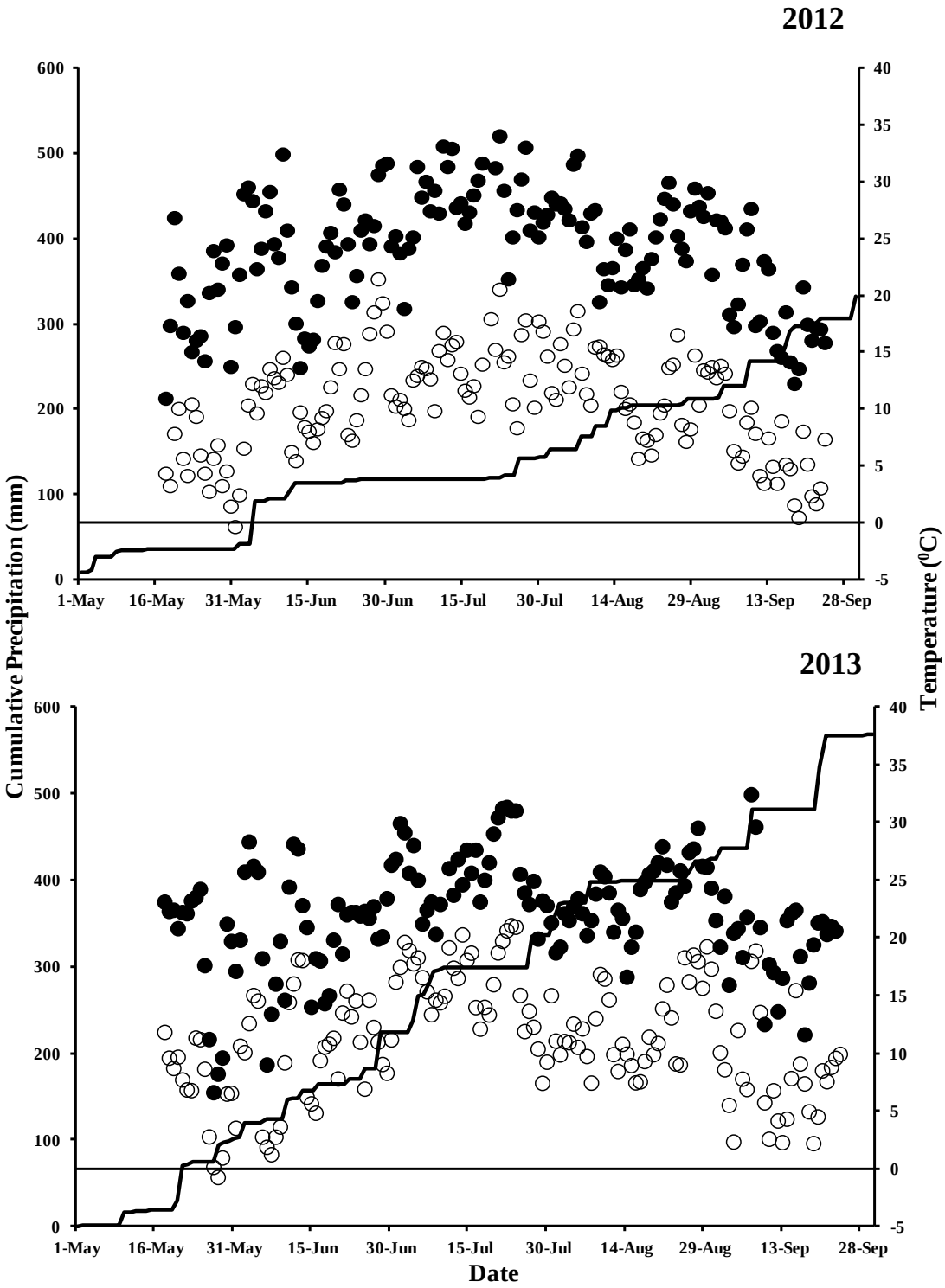

Figure 3.6 Climate data for Elora, ON in 2012 and 2013.
Daily high and low temperatures are shown by closed and open circles, respectively. Cumulative precipitation beginning May 1st is shown as a solid line. Date of seeding was May 25th in 2012 and June 9th in 2013.

CHAPTER 4

RESPONSE OF CANOLA TO A TRANSIENT WATER STRESS AS AFFECTED BY HIGH- VERSUS LOW-DENSITY PLANT POPULATIONS

4.1 Hypotheses and Research Objectives

4.1.1 Hypotheses

The hypothesis of this experiment is that increased assimilate sources and subsequent increased branching of individual plants under low-density populations result in an extended flowering time period and affords canola plants more flexibility for re-allocation of resources to additional sinks and for pod retention, thus increasing the tolerance of the crop to a transient water stress event.

This hypothesis was tested by creating canopies of high and low plant densities in a greenhouse and imposing a transient water stress event on the plants during flowering, the most critical period for determining yield potential. Greenhouse conditions other than the transient water stress were as optimal as possible, to limit confounding factors and determine whether effects of planting density could be observed under conditions where the limiting factor was water availability.

If low-density plants can utilize additional assimilates available per plant to retain pods that may otherwise be aborted from a water stress event, then low-density crops under water stress will have less yield loss compared to a high-density crop under water stress. In addition, abortions of early flowers and pods as a result of the water stress treatment will be compensated for by an extended flowering period on additional branches in low-density plantings. The extended period of reproductive development in low-density plantings will permit additional pods to be established on branch racemes after the stress has been relieved. By contrast, plants in

high-density conditions, having fewer branch racemes and a shorter flowering time, will be less able to recover and have an increased yield loss from the stress.

4.1.2 Research Objectives

1. To determine if the population density of a canola crop has an effect on the morphological and reproductive response of plants to a transient water stress.
2. To assess whether canola plants grown under low-density conditions have a greater resistance to yield loss than those grown under high-density conditions under the circumstances of a transient water stress event.

4.2 Materials and Methods

4.2.1 Experimental Design

This experiment was conducted from September 2013 until January 2014 in a greenhouse at the University of Guelph. The experiment was designed as two-factor factorial RCBD with four replications, two levels of density treatments and two levels of watering treatments. Density and watering treatments were applied in all combinations to each replication, making four plots per replication. Replications were established sequentially (7-10 days apart), beginning on September 12. Plots were made up of four 9-inch pots, surrounded by 14 border pots of the same size (Figure 4.1).

4.2.2 Greenhouse Conditions

The greenhouse had target day/night air temperature settings of 22/17^{o}C. A mixture of metal halide and high pressure sodium lamps provided a photosynthetically active photon flux

density of approximately 600 μmol m^{-2} s^{-1} to the tops of plants, which supplemented incoming sunlight for 16 hours per day. The greenhouse settings allowed a shade cloth to deploy, and lamps to turn off if temperatures in the greenhouse exceeded 35^{o}C.

Prior to, and following water stress treatment periods, all pots were watered daily as needed with a nutrient solution containing 0.5 g L^{-1} water of "All Purpose" 20:20:20 fertilizer (Plant Products Co. Ltd., Brampton, ON) (Ramsahoi, 2013). Insect control was achieved throughout the experiment by integrated pest management (BioBee Biological Systems Ltd., Sde Eliyaho, Israel), where predators were applied to plants based on weekly scouting records. To control a mild occurrence of powdery mildew during the seed-filling stage, two separate applications of Senator 70WP (Engage Agro Co., Guelph ON) and Folpan 50WP (Makhteshim Agan of North America Inc., Raleigh NC) at a concentration of 0.8 and 1.25 g of product L^{-1} of water were applied to leaves of affected plants.

4.2.3 Treatment Preparation and Application

Pots of each replication were filled uniformly with LA4 Sunshine mix (Sun Grow Horticulture Ltd, Vancouver BC) to an equal volume and weight. Water holding capacity (WHC) of the soil was determined by saturating the soil of one extra pot for each replication and leaving the pot to drain. Plastic wrap was used to create a seal around the top of each of these pots, in order to reduce water loss by evaporation. Once the soil was fully drained, a soil wet-weight was taken for each pot. Soil was then emptied into paper bags and dried to a constant weight at 80^{o}C in a forced air drier. The soil was mixed daily to ensure uniform drying. From the saturated and

dry weights, pot WHC was calculated for each replication, and an average was calculated for an estimate of pot WHC for the experiment.

The canola variety InVigour 5440 treated by the supplier with the insecticidal and fungicidal seed treatment Prosper (Bayer Crop Science, Guelph ON) (1000-sdwt = 5.25 g; germination = 95%), was seeded to plots using a 1.25 cm planting depth. Emergence was observed four days following planting for all replications. Density treatments consisted of target plant populations that were either high (90 plants m^{-2}) (D_{90}) or low (30 plants m^{-2}) (D_{30}). To create these conditions, plots were thinned four days after emergence to five and two plants per pot, respectively. Distance from center to center of each pot was 24 cm, and therefore the total area that plants in each pot occupied was 0.058 m^2. This resulted in actual high and low treatment populations of 86 and 34 plants m^{-2}.

Water stress treatments began in each replication two days after first flower, with first flower defined as the date when 50% of plants had at least one open flower (Lamb and Johnson, 2004). Low and high-density populations reached first flower simultaneously. First flower was deemed the beginning of the critical stress period, as it is noted that the flowering period is most sensitive to water stress (Ahmadi and Bahrani, 2009). At this time, one sample pot was removed from each plot for a destructive measure to estimate shoot biomass (Figure 4.2). The roots of this pot were also harvested, cleaned and weighed at this time in the first replication only. In this first replication, roots weighed approximately 20% of the shoot weight. Estimates of root biomass in additional replications were under the assumption that roots were also approximately 20% of shoot weight. These values were used to increase the accuracy of soil-water content calculations.

Target weights were then calculated for pots in each plot by adding together the estimated plot shoot and root biomass, the dry soil weight, the pot weight and the water weight at the desired target percentage of WHC.

The water-stress treatments (W_T) were imposed by weighing and watering each pot daily to simulate a gradual dry-down of pots in the stressed condition from a soil moisture of 80% WHC to 10% WHC over six days (Figure 4.3). After holding the plants at a soil moisture of 10% WHC for 24 hours, the pots were watered back up to 60% WHC, and the gradual dry-down was repeated until the pots were held again for 24 hours at 10% WHC. The reason that the water stress was applied gradually was to ensure that pots planted at high densities did not dry down at a significantly faster rate than pots planted at low densities. The entire stress period of each replication was 13 days long, from first flower through early seed development.

During the stress period of each replication, control treatment (W_C) pots were weighed and watered daily to a soil moisture content of 90% WHC, to ensure that plants did not experience water stress. Following the stress period, both control and water-stressed pots were watered daily as described above until maturity.

4.2.4 Harvest and Measurements

Border plants were removed from plots approximately seven days prior to harvest, to avoid seed shattering. Replications were harvested on December 11th, December 18th, December 27th and January 7th. The average growing period, from emergence to harvest, was 88 days.

At the time of harvest, viable pods on the main raceme and branch racemes of sample plants were counted. Viable pods were defined as those that had produced at least one seed (Angadi, 2003; Tesfamariam et al., 2010).

Pods, which were still attached to their racemes, were stored in a low-heat drying room for 5 days, and then threshed. Harvested seed was cleaned using an Agriculex CB-1 column blower (Agriculex, Guelph ON) and dried further in a forced-air drier at 60^{o}C for two days.

4.2.5 Calculation of Yield Components

After seeds were cleaned and dried, seed weights (g) of each plot were obtained separately for main racemes and branch racemes. These weights were averaged over the area that the plants occupied to get plot seed yield in g m^{-2} for main racemes and branch racemes. Main raceme and branch raceme seed weights were combined to calculate total yield per plot on an area basis.

Viable pods m^{-2} were calculated for main racemes and branch racemes by totaling the number of viable pods in each experimental unit over the occupied area of the plants. Viable pods on main racemes and branch racemes were combined to calculate total pods m^{-2} for each plot.

The 1000-seed weight (g) of plots for main racemes and branch racemes was obtained by taking the weight of two 100-seed samples and multiplying the sum of the samples by 5. The 1000-seed weight for each plot (KSW_{TOT}) was calculated as:

$$KSW_{TOT} = (KSW_{MR} \times \%SW_{MR}) + (KSW_{BR} \times \%SW_{BR}) / 100,$$

where KSW_{MR} and KSW_{BR} are the 1000-seed weight (g) of plots for main racemes and branch racemes, respectively, and $\%SW_{MR}$ and $\%SW_{BR}$ are the percentage of seed weight accounted for by the main raceme and branch racemes, respectively.

Seeds per pod was calculated for main racemes (SPP_{MR}), branch racemes (SPP_{BR}) and total plots (SPP_{TOT}) as:

$$SPP_{MR} = Y_{MR} / KSW_{MR} \times 1000 / POD_{MR},$$

$$SPP_{BR} = Y_{BR} / KSW_{BR} \times 1000 / POD_{BR},$$

$$SPP_{TOT} = Y_{TOT} / KSW_{TOT} \times 1000 / POD_{TOT},$$

where Y_{MR}, Y_{BR} and Y_{TOT} are yield in g m^{-2} for main racemes, branch racemes and total plots, respectively and POD_{MR}, POD_{BR} and POD_{TOT} are viable pods m^{-2} for main racemes, branches and total plots, respectively.

4.2.6 Statistical Analysis

All yield response variables were analyzed as a two-factor RCBD using SAS version 9.2. A mixed model procedure with a type I error rate (α) of 0.05 was used. An ANOVA was conducted and assumptions of the ANOVA were met according to tests in an analysis of residuals. A Tukey's HSD test was performed to obtain mean separations.

4.3 Results and Discussion

4.3.1 Interaction Effects of Density and Water Stress on Yield

There was not a significant density x water treatment interaction for seed yield (Table 4.1), indicating that evidence that density affects how yield responds to water stress was not found. Figure 4.4 shows pod yields of plants at maturity in each of the density x water treatment combinations for one replication. The hypothesis that low-density plots have an advantage over high-density plots in terms of yield retention under the conditions of a transient water stress during the flowering period was not statistically supported (Table 4.1).

4.3.2 Effects of Density on Yield and Its Components

Density did not have an effect on total yield when averaged across water treatments (Table 4.2). This is the result of plants under D_{30} conditions compensating for reduced plant stands by increasing branching and consequently increasing pod number per plant (Table 4.2). D_{30} conditions resulted in more pods m^{-2} on branch racemes compared to D_{90} conditions. This negative correlation between pods per plant and plants per unit area is consistent in the literature (Leach et al., 1999; Diepenbrock, 2000). Low-density plantings have minimal intra-plant competition, allowing for plants to establish a greater LA and subsequent carrying capacity (Angadi et al., 2003; McGregor, 1987; Leach et al., 1999). These canola plants go on to exhibit a high degree of morphological plasticity, producing more branches that hold a larger number of pods.

In agreement with the literature, plants in D_{90} conditions had reduced branching and consequently a significantly reduced number of pods produced on branch racemes (Diepenbrock,

2000; Tayo and Morgan, 1979; Rood and Major, 1984). Decreased branching in high-density conditions can be explained by increased intra-plant competition, resulting in limited leaf expansion and light interception and therefore a reduced source capacity of individual plants (Clarke, 1979). There were however, significantly more pods m^{-2} contributed from main racemes under D_{90} conditions. This is a result of the increased number of plants and therefore the larger number of main stem racemes per unit area. The effect of this on total yield was much less than that of the pods produced on additional branch racemes under D_{30} conditions. It should also be noted that reducing plant population by 67%, from 90 to 30 plants m^{-2}, reduced main raceme pods m^{-2} less than 50%. This disproportional decrease indicates that there were more pods produced on individual main racemes of plants under D_{30} conditions. That is, plants grown at low density produced not only more branch racemes, but also larger main racemes with more pods (Figure 4.5). Yield compensation by plants under D_{30} conditions was not accomplished solely by increasing pods m^{-2} on branch racemes, but also by increased pod numbers on main racemes.

On a total plot basis, seed number per pod and 1000-seed weight were not significantly affected by density treatments (Table 4.2). These findings support Olsson (1960) and Diepenbrock (2000) who report that pod number is the yield component most responsive to variations in plant density, while seeds per pod and 1000-seed weight are less affected**.**

The lack of effect that density had on average plot yields supports findings in the literature that canola in low-density conditions adjusts seed yield to high-density levels over a wide range of populations (Angadi et al.,2003; Potter et al., 1999; Mendham et al., 1981;

Christensen and Drabble, 1984; Morrison, 1990a). There are other reports in the literature that disagree with this finding as they observed that low-density plantings did not have the ability to compensate up to high-density yields, indicating that yield compensation may be conditional on environmental factors (Lythgoe et al., 2001; May et al., 1994; Ohlsson,1972; Clarke, 1979).

4.3.3 Effects of Water Stress on Yield and Its Components

The water stress imposed was severe enough that it significantly affected yield, though contrary to our original hypothesis, this reduction occurred almost entirely on the branch racemes and not main racemes (Table 4.2). A comparison of water stressed plants versus control plants in terms of leaf wilt and flowering in each density can be viewed in figures 4.6 and 4.7, respectively. The effect that water stress had on pods m^{-2} from main racemes is particularly evident in the comparison of percent yield loss attributed to by pods m^{-2} on main racemes in the D_{30} x W_S to that of D_{30} x W_C treatment combinations (Table 4.3). The yield loss was only 1%, indicating that plants under D_{30} conditions did not differ significantly in the pod number produced by main racemes whether they did or did not receive water stress. The yield loss of D_{90} x W_S compared to D_{90} x W_C is also low (4.1%), which again highlights that pods produced on main racemes under D_{90} conditions failed to differ in yield loss due to water stress.

The lack of effect of water stress on yield of main racemes may have been due to the timing of the stress, apical dominance exhibited by the main raceme, or a combination of both factors. Water stress conditions were imposed two days after first flower, though the lowest soil water content was not reached until six days after the stress treatment was initiated. The plants may have not experienced a significant water stress until most of the pods on the main racemes

were set. It is possible that a stress initiated at an earlier phenological stage may affect the main racemes more severely.

The main raceme exhibits apical dominance over branch racemes, which is why they are the first to flower and therefore the first to set pods and seeds (OECD, 2012). At the beginning of flowering, competition for assimilates is low and LA nears its maximum. The timing of main raceme flowering contributes to pods produced on the main raceme having a greater assimilate supply compared to subsequent branch racemes. In the literature, main racemes are consistently least affected by environmental stresses (Tayo and Morgan, 1975; Clarke, 1979; Clarke and Simpson, 1978). This suggests that our stress may have been imposed at a time before main raceme pods were set as intended, but it failed to have an effect on the number of pod abortions due to apical dominance exhibited by the main raceme.

The yield components of pods m^{-2} and 1000-seed weight were significantly affected by water stress, with W_C conditions having more pods m^{-2} and a lower 1000-seed weight compared to W_S conditions on branch racemes, main racemes, and consequently, on a whole-plot basis (Table 4.2). The water stress alone had less of an effect on the number of seeds per pod. In terms of how these components affected yield, increased 1000-seed weight compensated fully for the reduced main raceme pod number under Ws conditions and yield contribution by main racemes was therefore not significantly reduced by W_S. The reduction in pods on branch racemes under W_S conditions was much greater than that of main racemes, and increased 1000-seed weight was therefore unable to compensate for this yield loss. The reduction in pods per plant on branch racemes was therefore the main contributor to yield loss from water stress. This is in agreement

with the literature in that pods m^{-2} is the yield component most responsive to stresses at flowering **(**Clarke and Simpson 1978; Kondra, 1975; Degenhardt and Kondra, 1981; McGregor, 1987; Shahin and Valiollah, 2009). It is likely that the main mechanism of water stress that caused these reduced yields is reduced photosynthesis, which limits assimilate production and results in flower and pod abortions (Ahmadi and Bahrani, 2009).

4.3.4 The Interaction of Density and Water Stress on Yield Components

There was a significant density x water treatment interaction for seed number per pod on branch racemes and for total plots (Table 4.3). Plants under the D_{90} x W_S treatment combination had a significantly lower seed number per pod on branch racemes when compared to all other treatment combinations (D_{90} x W_C, D_{30} x W_C and D_{30} x W_S). On a total plot basis, plants under the D_{90} x W_S condition continued to have the lowest seed number per pod, though it was only significantly lower than that of the D_{30} x W_S condition (Table 4.1). Under the D_{30} x W_S treatment combination, for both branch racemes and total plots, the number of seeds per pod was not significantly different from D_{30} x W_C and D_{90} x W_C conditions (Table 4.1 and 4.3). These results indicate that water stress had little effect on the seed number per pod of plants in the D_{30} condition, whereas in the D_{90} condition seed number per pod was negatively affected by water stress and the effect was mostly confined to the branch racemes. Plants in the D_{90} x W_S condition lost 10.5% of total yield from decreased seed number per pod on branch racemes (compared to the control treatment of D_{90} x W_C), whereas plants in the D_{30} x W_S condition actually showed a slight yield compensation from seed number per pod (Table 4.3). This may indicate that plants under the D_{30} condition were better able to cope with the water stress at the time of seed set than

plants that were raised under the D_{90} conditions, which provides evidence in support of our original hypothesis.

Plants under low-density conditions have a greater LA per plant, which suggests more photosynthates available for translocation per plant (Diepenbrock, 2000; Tayo and Morgan, 1979; Rood and Major, 1984). In early reproductive development, leaves establish the structural sink potential of plants in terms of pod number per plant and seeds per pod (Major et al. 1978; Chapman et al. 1984). Clarke and Simpson (1978) state that increased LA and LA duration observed in low-density plantings could reduce seed abortion. The results of the current study support this, as low-density plants were less affected by a water stress in terms of seeds per pod.

Results also indicate a significant density x water treatment interaction for average 1000-seed weight on branch racemes (Table 4.3). The mean weight of individual seeds produced on branch racemes of plants in the D_{90} x W_S treatment combination was significantly higher compared to that of all remaining treatment combinations. This is not surprising, since D_{90} x W_S conditions resulted in plants with significantly fewer seeds per pod, as well as lower pods m^{-2}. The outcome was a lower number of seeds for each plant to fill. Therefore, plants under D_{90} x W_S conditions allocated photosynthates to a smaller number of seeds and had more photosynthate available per seed compared to plants in other treatment combinations. This resulted in a yield compensation of 10.8% for the D_{90} x Ws treatment combination, which almost exactly compensated for the yield loss associated with the reduced seeds per pod. The work of Leon and Becker (1995) corroborates this finding, as they reported a negative relationship between individual seed weight and number of seeds per pod. These notions are also consistent

with the observations of Diepenbrock (2000), who found that individual seed weight depends to a lesser extent on environmental conditions and more on the status of other yield components.

The mean 1000-seed weight produced on branch racemes of plants under the D_{30} x W_S treatment combination was significantly greater than that of the D_{30} x W_C and D_{90} x W_C treatment combinations (Table 4.3). This result was not surprising, since water stress significantly decreased pods m^{-2} on branch racemes, and therefore assimilate of individual plants was spread over a reduced sink size (Table 4.2).

4.4 General Conclusions

There was no significant density x water interaction effect on seed yield. Numerically, however, compared to the highest-yielding treatment combination of a high plant density in the absence of water stress, plots under high and low density that endured a transient water stress episode had average yield losses of 25.4 and 16.4%, respectively (Table 4.3). These numerical trends are in accordance with the hypothesis, since after a water-stress event, low-density populations retained 10% more yield compared to high-density populations.

Another factor that may have affected the outcome of this study is the timing of the water stress. Main racemes, along with higher positions of primary branch racemes, are the first to flower and set pods. Since we imposed the stress just following first flower, we expected that the stress would have a large impact on main raceme flower and pod abortion. Under this circumstance we predicted that later-flowering branch racemes on plants under low-density conditions would fully compensate for this loss, while high-density plants would not have the

branches or resources to carry out the same compensation. Since the water stress was imposed gradually, it is possible that it may have not reached extreme conditions before main raceme pod set was nearly complete. If this was the case, the stress would have failed to disrupt pod set on the main raceme and subsequent pod abortions on main racemes would not be observed, as was the case in this study. The literature challenges this way of thinking, since main racemes of canola plants are consistently less affected by environmental stresses, likely due to a combination of apical dominance and assimilate supply at the time of main raceme flowering. It is possible that water stress encompassing the time from first flower to pod set of the entire main raceme would still fail to affect main raceme pods as strongly as expected.

The current study supports the idea that low-density canola crops have the ability to compensate up to high-density yield levels under the condition of a transient drought stress at flowering. This challenges findings of Clarke and Simpson (1978) and McGregor (1987), who used similar reductions in plant density under field conditions, that low-density plant populations cannot compensate up to high-density yield levels under water stress. Additional work should be done in this area to assess and quantify the limits of low-density plant population yield compensation.

There are limitations in this greenhouse study that may have caused yields to respond to treatments differently from how they may respond in a typical field situation.
First, the soil medium was very different from field soil in terms of water movement and availability. Roots of plants were limited to the volume of the pots and therefore any effect that plant density may have had on the total root biomass as well as rooting depth in the field were

not simulated in the greenhouse. In addition to this, yields reported in this study are much larger than typical yields produced in field situations. To illustrate what can be produced from an average canola crop, Canadian producers saw a record national average yield of 2,200 kg ha^{-1} in 2013 (Statistics Canada, 2013). This is well below what a canola crop has the potential to produce, since individual growers have reported yields up to 4,500 kg ha^{-1}. Statistics Canada (2013) notes that these high yields in field conditions are rare and only occur when careful attention is paid to agronomic practices and all possible conditions are ideal. To put the yields obtained from the current study in perspective, the average total plot yields from plants under water-replete conditions was 6,250 kg ha^{-1} (Table 4.1). Even under water stress conditions, average plot yields were 4,830 kg ha^{-1}, approximately equivalent to record high field yields. Since the yields in this experiment were extremely high in comparison to typical yields produced in field conditions, it is assumed that the plants were highly branched with an extended reproductive period. This may have altered the way in which the crop responded to a water stress event and how yield components were subsequently affected. The results of this study may therefore not be typical of a field scenario where plants are smaller and the main raceme contributes to a larger portion of yields.

Table 4.1 Plot means of yield components of a spring canola (*Brassica napus* L.) crop as affected by density x water treatment interactions in a greenhouse setting at the University of Guelph, ON in 2013.

Treatment	pods m^{-2}	x	seeds pod^{-1}	x	1000-seed weight	=	seed yield ‡ (g m^{-2})
Density x Water							
90 plants m^{-2} x *Control*	10772	x	18.3 *ab* *	x	3.26	=	641
30 plants m^{-2} x *Control*	10319	x	17.9 *ab*	x	3.32	=	610
90 plants m^{-2} x *Stress*	7533	x	16.4 *b*	x	3.78	=	466
30 plants m^{-2} x *Stress*	7470	x	18.6 *a*	x	3.62	=	500
interaction p-value	0.68		**0.01** †		0.10		0.25

* For response variables that have a significant interaction, nutrient treatment means followed by the same letter do not differ significantly ($P < 0.05$) according to a Tukey's HSD test.
† P values < 0.05 indicate a significant density x water treatment interaction. For main effects see Table 4.2.
‡ Seed yield is measured yield and differs slightly from calculated yield in Table 4.3.

Table 4.2 Means comparisons of yield components and seed yield as contributed from main racemes (MR), branch racemes (BR) and total plots (TOT) for a spring canola (*Brassica napus* L.) crop as affected by density and watering treatments in a greenhouse setting at the University of Guelph, ON in 2013.

Treatment	----------Pods (m^{-2})----------			----------Seeds pod^{-1}----------			-----1000-seed weight (g)-----			--------Yield (g m^{-2})----------		
	MR	**BR**	**TOT**	**MR**	**BR**	**TOT**	**MR**	**BR**	**TOT**	**MR**	**BR**	**TOT**
Density												
90 plants m^{-2}	3650	5502	9152	18.1	16.7	17.4	3.61	3.46	3.52	237	317	553
30 plants m^{-2}	1902	6992	8894	18.1	18.4	18.3	3.74	3.39	3.47	128	427	555
p value <	**0.0001***	**0.01**	0.59	0.99	**0.002**	0.06	0.07	0.16	0.42	**0.0001**	**0.003**	0.96
Water												
Control	2907	7638	10545	17.9	18.2	18.1	3.48	3.22	3.29	180	446	625
Stress	2645	4856	7501	18.3	17.0	17.6	3.87	3.62	3.70	185	298	483
p value	**0.02**	**0.0003**	< **0.0001**	0.59	**0.01**	0.24	**0.0003**	< **0.0001**	**0.0001**	0.58	**0.0005**	**0.0005**

* P values in bold text represent treatment means separation significance at $P < 0.05$ according to a Tukey's HSD test.

Table 4.3 Yield calculation model of spring canola (*Brassica napus* L.), depicting density x water treatment interaction means of yield components as contributed to by main racemes or branches and how each yield component individually affects total plot yields, whether positively or negatively, in comparison to the optimal treatment combination of 90 plants m^{-2} x control.

Treatment	Main Racemes: pods m^{-2}	x	Main Racemes: seeds pod^{-1}	x	Main Racemes: 1000-seed weight (g)	+	Branch Racemes: pods m^{-2}	x	Branch Racemes: seeds pod^{-1}	x	Branch Racemes: 1000-seed weight (g)	=	calculated seed yield ‡ (g m^{-2})
Density x Water													
90 plants m^{-2} x *Control*	3860	x	18.3	x	3.39	+	6912	x	18.3 *a*†	x	3.19	*c* =	641
30 plants m^{-2} x *Control*	1954	x	17.4	x	3.58	+	8364	x	18.0 *a*	x	3.26	*c* =	613
90 plants m^{-2} x *Stress*	3441	x	17.9	x	3.83	+	4092	x	15.2 *b*	x	3.73	*a* =	467
30 plants m^{-2} x *Stress*	1850	x	18.8	x	3.91	+	5620	x	18.7 *a*	x	3.52	*b* =	505
p-value	0.11		0.3		0.47		0.94		**0.0009***		**0.01**		
	Yield Gain (+) or Loss (-) (%)												
30 plants m^{-2} x *Control*	-18.4		-1.8		+2.1		+13.2		-0.8		+1.4		-4.3
90 plants m^{-2} x *Stress*	-4.1		-0.9		+4.8		-25.6		-10.5		+10.8		-25.4
30 plants m^{-2} x *Stress*	-19.4		+0.9		+5.8		-11.7		+1.5		+6.5		-16.4

* P values < 0.05 indicate a significant treatment interaction.

† Of those response variables that have a significant interaction, nutrient treatment means followed by the same letter do not differ significantly (P < 0.05) according to a Tukey's HSD test.

‡ Calculated seed yield differs from measured seed yield in Table 4.1 by up to 1%. Calculated seed yield shown here is based on the weighted products of the means for each yield component across replications.

Figure 4.1 Depiction of the greenhouse double factorial randomized complete block design at the University of Guelph, ON in 2013. Replications were spread out over time, planted approximately one week apart.
Each plot is three pots wide and six pots long. Pots used for experimental measurements are the four pots in the center, completely surrounded by border plants.

Figure 4.2 Destructive samples of spring canola (*Brassica napus* L.) taken at the beginning of the water stress period for a comparison of shoot biomass in low-density (furthest left and right) and high-density (middle) treatments.

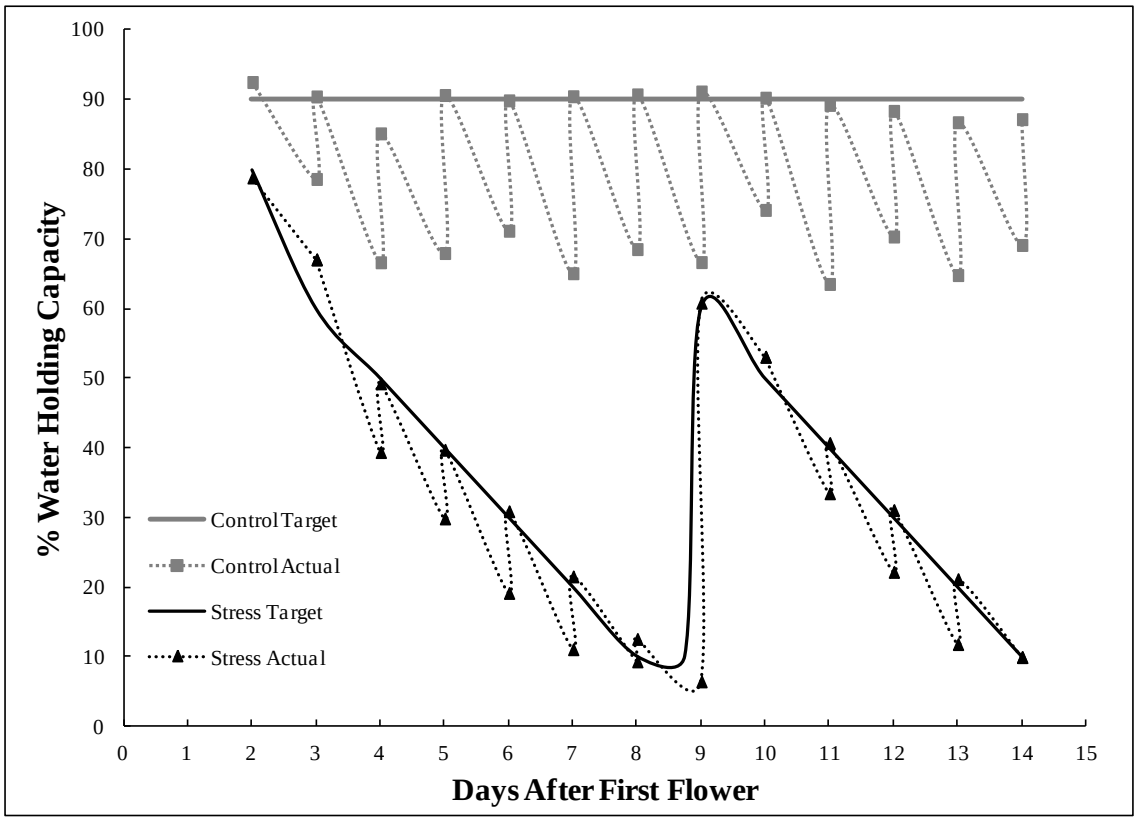

Figure 4.3 Average water status of individual pots after first flower of canola plants for control and water-stressed treatments. Pots were watered daily up to a target water holding capacity, represented by solid lines.
Symbols connected by dashed lines represent actual soil water content as measured gravimetrically before and after each watering. Data represent treatment means across four replications.

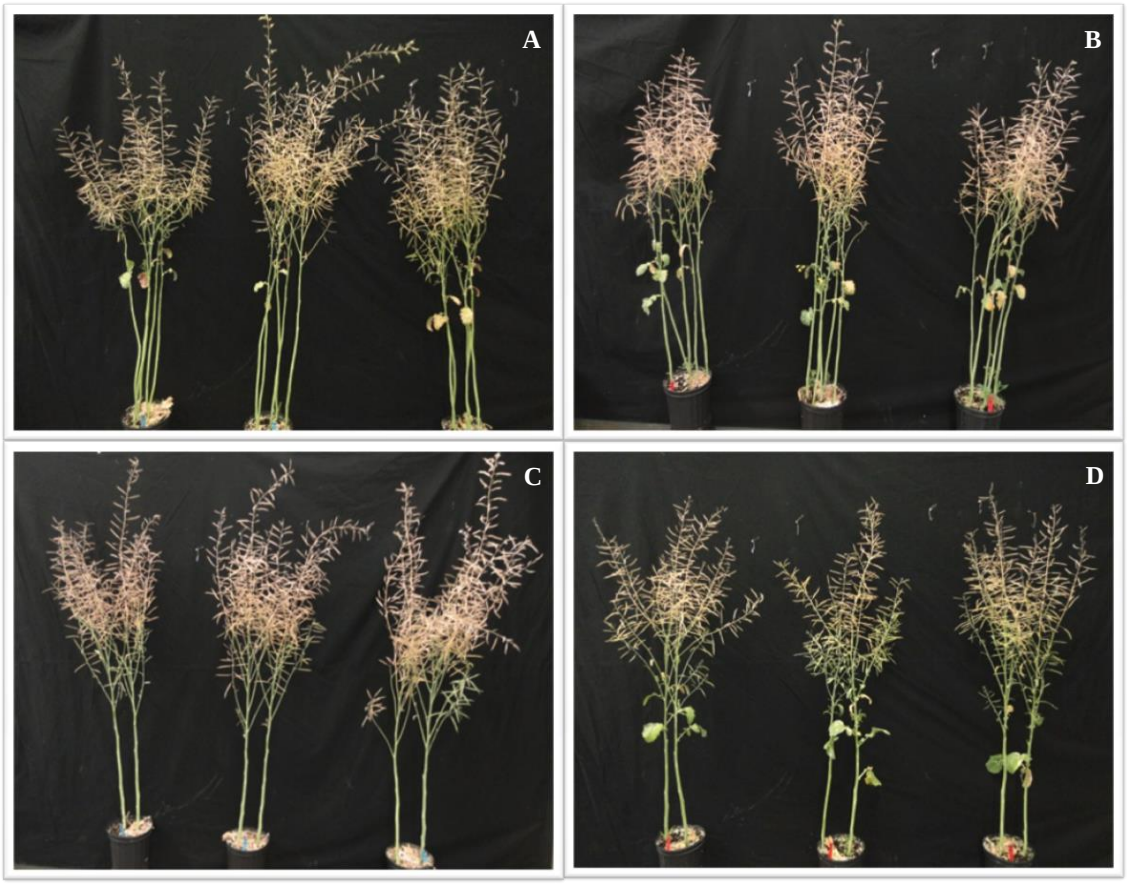

Figure 4.4 Spring Canola (*Brassica napus* L.) plants at full maturity: (A) High density x Control, (B) High density x Water stress, (C) Low density x Control, (D): Low density x Water stress.

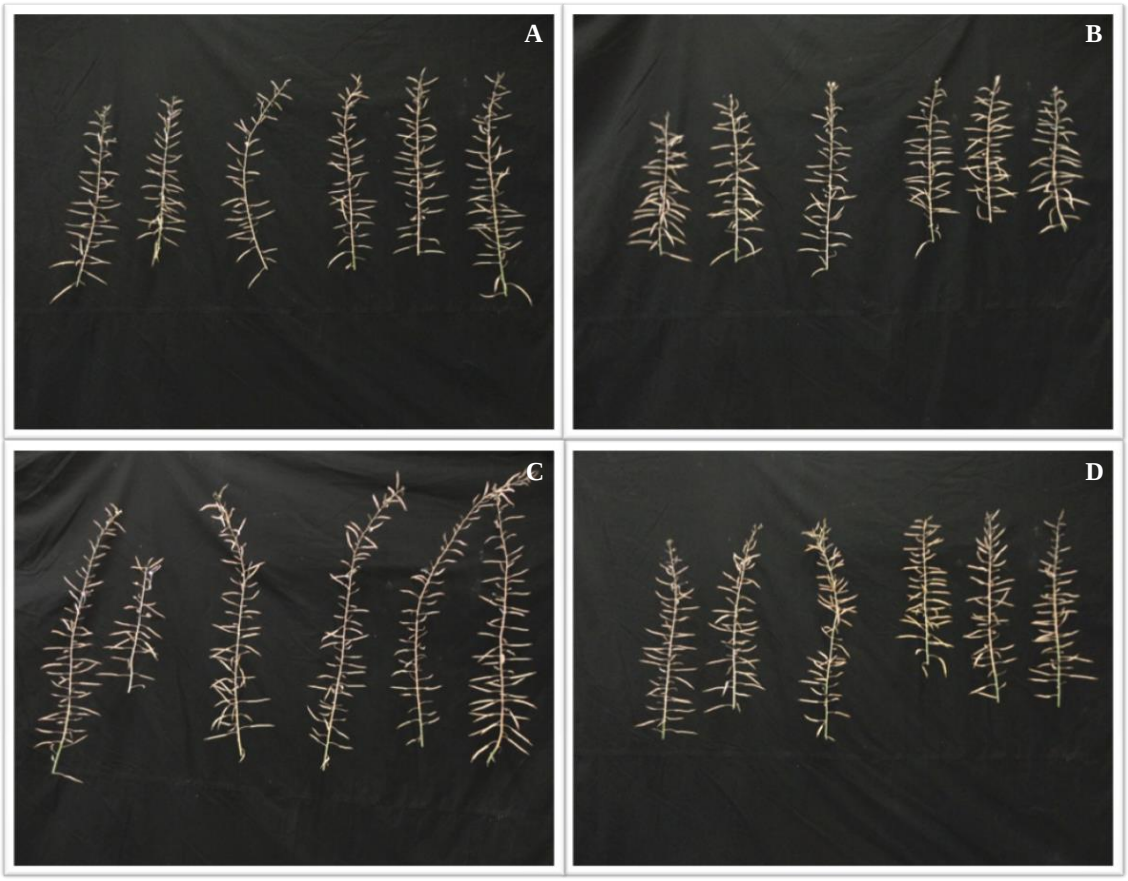

Figure 4.5 Main racemes of spring canola (*Brassica napus* L.) plants at full maturity: (A) High density x Control, (B) High density x Water stress, (C) Low density x Control, (D): Low density x Water stress.

Figure 4.6 Spring canola (*Brassica napus* L.) plants at the end of the water stress period: (A) High density x Control, (B) High density x Water stress, (C) Low density x Control, (D): Low density x Water stress.

Figure 4.7 Terminal flowers on racemes of spring canola (*Brassica napus* L.) plants following the water-stress period: (A) High density x Control, (B) High density x Water stress, (C) Low density x Control, (D): Low density x Water stress.

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

5.1 Effects of Manure and Boron Application on Early Vigour and Establishment of Canola

Measurements taken in field and greenhouse experiments to measure canopy establishment and vigour failed to show that LDM application has an effect that cannot be reproduced with a synthetic fertilizer application of the same macronutrient nutrient values, with or without the addition of B. These observations are in contrast to what was observed in previous preliminary field trials that observed increased establishment and yields for plants receiving a pre-plant LDM application when using similar methods to this study.

The major limitation in this study was that fertilizer solutions, which were prepared with the intent of creating an identical macronutrient supply to the one provided by the LDM source, may have provided more crop-available nutrients than the manure during the experimental period. Higher plant available nutrients from the fertilizer solutions than the manure was plausible because the manure analysis was interpreted according to total nutrient content and did not account for organic, unavailable nutrient fractions of the manure. A more accurately simulated nutrient content of the fertilizer solution could have been created if nutrients were mixed based on estimates of plant available nutrients in the manure for the duration of the vegetative period only. Although it may have benefitted this study to estimate what may be plant-available from the manure during vegetative growth, it would have been difficult to estimate accurately, since there are so many factors contributing to nutrient availability in the soil (Barber, 1995). Such methods would have failed to vigorously test the hypothesis that LDM contributes something to early vigour that is in addition to N, P, K and S nutrients.

One way that that we could have addressed this issue is by including a treatment that contains nutrients in the amounts estimated to be available in the first year of application, in addition to the treatment used in this study that simulated total nutrients. In this case, if no observable response difference occurred between the estimated available nutrient treatment and the LDM treatment, we could have concluded that it is unlikely that LDM has additional short-term benefits. On the other hand, if there was a benefit observed from LDM over the estimated available nutrient treatment, then comparisons with the treatment containing total nutrients would serve to evaluate whether additional macronutrients may be responsible for the effect.

In greenhouse studies testing for the manure effect and the effect of B on early establishment, effects could not be clearly observed since the effect of plant population of individual pots confounded the effect that treatments had on LA and SDW measurements. It would have been beneficial to include treatments where pots were over-seeded and thinned to a uniform population. The non-uniform plant stands were a result of reduced emergence in synthetic fertilizer treatments due to a nutrient toxicity, which should have been avoided by planting the seeds a number of days following treatment application or by more thoroughly mixing the nutrients in the soil.

Regardless of the limitations regarding non-uniform plant stands in the greenhouse and the over-estimated nutrient content of fertilizer solutions, this study showed that LDM did not have an effect on crop establishment and vigour as strong as was observed in preliminary studies. It is possible that the preliminary studies in question may have been conducted on soil that had a specific characteristic in which the manure may have been more beneficial, or that the manure

source used in the preliminary studies contained different fractions of nutrients and organic substances than the one used in the current study.

5.2 Response of Canola to a Transient Water Stress as Affected by High-Versus Low-Density Plant Populations

We did not find statistically significant evidence to support the hypothesis that the morphological changes associated with low plant densities would make a canola crop less susceptible to yield loss under a transient water stress during early flowering. These findings may have differed if the water stress was applied during an earlier or later phenological stage and therefore additional experiments could be conducted to further explore these notions. Also, the numerical trends did agree with our hypothesis that a low-density planting would be more resistant to yield loss under water stress. Perhaps additional replication or further attention to potential sources of experimental error would have clarified these trends.

We did observe that low-density plantings had the ability for yield-compensation up to high-density levels, even while undergoing a significant yield-limiting transient water stress. These findings challenge conclusions made in the literature by Clarke and Simpson (1978) and McGregor (1987), and therefore additional work should be done in this area to assess and quantify the limits of low-density yield compensation.

The conclusions of this study, in terms of application to field conditions, may be limited since yield produced in the greenhouse culture system were not typical of what is normally

produced in the field. It would therefore be advantageous for research in this area to proceed under field conditions.

5.3 Conclusion Summary

In conclusion, each of the experiments conducted highlighted the effect that plant population has on the morphology of canola, which should not be discounted by producers when considering other supporting agronomic management practices. Further steps may be taken to quantify and weigh local advantages and disadvantages for producers to plant a high- versus a low-density canola crop.

REFERENCES

A & L Canada Laboratories Inc. 2014. Manure analysis reference guide. Available at: http://www.alcanada.com/Agricultural-Manure.htm

Abraham, V. and J.M. DeMan. 1986. Hydrogenation of canola oil as affected by Chlorophyll. *Journal of American Oil Chemists' Society*. 63:1185-1188.

Adams, J. F. Yield responses to molybdenum by field and horticultural crops. In: U.C. Gupta, Molybdenum in Agriculture. (Ed.), U.K: Cambridge University Press; 1997: p. 182–201.

Agehara, S. and D.D. Warncke. 2005. Soil moisture and temperature effects on nitrogen release from organic nitrogen sources. *Soil Science Society of America Journal*. 69:1844-1855.

Ahmad, W., M.H. Zia, S.S. Malhi, A. Niaz and Saifullah. 2012. Boron deficiency in soils and crops: A review. In*:* A. Goyal. Crop Plant. (Ed.), ISBN: 978-953-51-0527-5, I, InTech. Available at: http://cdn.intechopen.com/pdfs/35614/InTech-Boron_deficiency_in_soils_and_crops_a_review.pdf

Ahmad, W., A. Niaz, S. Kanwal, Rahmatullah and M.K. Rasheed. 2009. Role of boron in plant growth: A review. *Journal of Agricultural Research*. 47:329-338.

Ahmadi, M. and M.J. Bahrani. 2009. Yield and yield components of rapeseed as influenced by water stress at different growth stages and nitrogen levels. *American-Eurasian Journal of Agriculture and Environmental Science*. 5(6):755-761.

Ahmadi, M.R., and R. Javidfar. Plant nutrition of canola produce of oilseeds. Anjman Press; 1998. 196 p.

Al-Barzinjy, M., O. Stolen, J.L. Christiansen and J.E. Jensen. 1999. Relationship between

plant density and yield for two spring cultivars of oilseed rape (*Brassica napus* L.). *Acta Agr. Scand. Sect. B. Soil Plant Science*. 49:129–133.

Al-Jaloud, A.A., G. Hussian, S. Karimula, and A.H. Ali-Hamidi. 1996. Effect of irrigation and nitrogen on yield and yield components of two rapeseed cultivars. *Agricultural Water Management*. 30:57–68.

Ali, M.H., S.M.H. Zaman and S.M.A. Hossain. 1996. Variation in yield, oil and protein content of rapeseed (Brassica campestris) in relation to levels of nitrogen, Sulphur and plant density. *Indian Journal of Agronomy*. 41:290–295.

Allen, E.J. and D.G. Morgan. 1972. A quantitative analysis of the effects of nitrogen on the growth, development and yield of oilseed rape. *Journal of Agricultural Science*. 78:315-324.

Altieri, M.A. 1999. The ecological role of biodiversity in agroecosystems. *Agriculture, Ecosystems and Environment*. 74:19-31.

Amtmann, A., J.P. Hammond, P. Armengaud, and P.J. White. 2006. Nutrient sensing and signaling in plants: potassium and phosphorus. *Advanced Botany Research*. 43:209-257.

Amtmann, A., S. Troufflard and P. Armengaud. 2008. The effect of potassium nutrition on pest and disease resistance in plants. *Physiologia Plantarum*. 133:682-691.

Angadi, S. V., H.W. Cutforth, B.G. Mcconkey and Y. Gan. 2003. Yield adjustment by canola grown at different plant populations under semiarid conditions. *Crop Science*. 43:1358-1366.

Arriaga, F.J. and B. Lowery. 2003. Soil physical properties and crop productivity of an eroded soil amended with cattle manure. *Soil Science*. 168(12):888-899.

Asare, E. and D.H. Scarisbrick. 1995. Rate of nitrogen and sulphur fertilization on yield,

yield components and seed quality on oilseed rape (*Brassica napus* L.). *Field Crops Research*. 44:41–46.

Bailey. L. D. and Grant, C. A. 1990. Evaluation of the effects of method, rate and composition of fluid fertilizer application for barley and canola grown on soils of the Canadian prairies. 1990 Fluid Fertilizer Foundation Symposium Proceedings, Scottsdale, Arizone. p. 266–271

Barber, S. A. 1995. Soil nutrient bioavailability: a mechanistic approach. John Wiley and Sons, New York, NY, U.S.A.

Beauchamp, E.G. 1986. Availability of nitrogen from three manures to corn in the field. *Canadian Journal of Soil Science*. 66:713-720.

Bengtsson, A. 1978. Influence of row distance on the cultivation of autumn rape. *Svensk Frotidning*. 47:93-96.

Bengtsson, A. and I. Ohlsson. 1974. Row spacing in oilseed crops. Proceedings of the 4th International Rapeseed Conference, Giessen, West Germany, June 4-8. p. 207-215.

Berger, K.C. 1962. Micronutrient deficiencies in the United States. *Agricultural and Food Chemistry*. 10(3):178-181.

Berglund, D.R., K. McKay and J. Knodel. 2007. Canola Production. North Dakota State University Extension Service, A-686 (revised). Available at: http://www.ag.ndsu.edu/pubs/plantsci/crops/a686.pdf.

Berti, A. and G. Mosca. 1987. Effetti della concimazione azotata sulla fruttificazione del colza invernale da olio (*Brassica napus* var. oleifera D.C.). *Rivista di Agronomia*. 2l: 45-49.

Bidwell, R.G.S. Plant physiology. 2nd ed. New York (NY): Macmillan Publishing Co.,

Inc.; 1979. 726 p.

Bolton, J., T.Z. Nowakowski and W.J. Lazurus. 1976. Sulphur-nitrogen interaction effects on the yield and composition of protein N, non-protein-N, and soluble carbohydrates in perennial ryegrass. *Journal of Science of Food and Agriculture*. 27:553.

Boyer, J. S. 1982. Plant productivity and environment. *Science*. 218:443–448.

Bremner, J.M and M.J. Krogmeier. 1989. Evidence that the adverse effect of urea fertilizer on seed germination in soil is due to ammonia formed through hydrolysis of urea by soil urease. *Proceedings of the National Academy of Sciences of the United States of America*. 86:8185-8188.

Brennan, R.F., M.G. Mason and G.H. Walton. 2000. Effect of nitrogen fertilizer on the concentrations of oil and protein in canola (*Brassica napus*) seed. *Journal of Plant Nutrition*. 23(3):339-348.

Brewster, J.L., K.K.S. Bhat and P.H. Nye. 1976. The possibility of predicting solute uptake and plant growth response from independently measured soil and plant characteristics. IV. The growth and uptake of rape in solutions of different phosphorus concentration. *Plant and Soil*. 44:279-293.

Bybordi, A. and G. Mamedov. 2010. Evaluation of application methods efficiency of zinc and iron for Canola (*Brassica napus* L.). *Notulae Scientia Biologicae*. 2(1):94-103.

Cakmak, I. 2002. Plant nutrition research priorities to meet human needs for food in sustainable ways. *Plant and Soil*. 247:3-24.

Camacho-Cristobal, J.J., J. Rexach and A. GonzÁlez-Fontes. 2008. Boron in plants: deficiency and toxicity. *Journal of Integrated Plant Biology*. 50:1247–1255.

Canadian Food Inspection Agency (CFIA). 2012. Specific Work Instructions:

Cruciferous crop. Available at: http://www.inspection.gc.ca/plants/seeds/inspection-procedures/cruciferous-crops/eng/1347390022952/1347391637966.

Canola Council of Canada (CCC). 2005. Factors Affecting Canola Survival from Seeding to 21 Days after Emergence. Available at: https://canola-council.merchantsecure.com/canola_resources/product26.aspx.

Canola Council of Canada (CCC). 2011. Effects of moisture: Rosette, Elongation and Flowering. Available at: http://www.canolacouncil.org/crop-production/canola-grower's-manual-contents/chapter-4-effects-of-moisture/effects-of-moisture.

Canola Council of Canada (CCC). 2012. Canola Grower's Manual. Available at: http://www.canolacouncil.org/crop-production/canola-grower's-manual-contents/.

Canola Council of Canada (CCC). 2013a. Economic impact of the canola industry. Available at: http://www.canolacouncil.org/markets-stats/industry-overview/economic-impact-of-the-canola-industry/.

Canola Council of Canada (CCC). 2013b. Canola can meet the demand. Available at: http://www.canolacouncil.org/canola-biodiesel/canola-biodiesel/canola-can-meet-the-demand/

Ceccoti, S.P. 1996. Plant nutrient sulphur: a review of nutrient balance, environment impact and fertilizers. *Fertilizer Research*. 43:117-125.

Celik, I., I. Ortas and S. Kilic. 2004. Effects of compost mycorrhizza, manure and fertilizer on some physical properties of a Chromoxerert soil. *Soil and Tillage Research*. 78:59-67.

Chambers, B.J., E.I. Lord, F.A. Nicholson and K.A. Smith. 1999. Predicting nitrogen

availability and losses following application of organic manures to arable land. *Soil Use and Management*. 15:137-143.

Champolivier, L. and A. Merrien. 1996. Effects of water stress applied at different growth stages to *Brassica napus* L. var. oleifera on yield, yield components and seed quality. *European Journal of Agronomy*. 5:153-160.

Chang, S. S., W.T. Huang, S. Lian, and W.L. Wu. 1992. Research on soil testing and leaf diagnosis as guides to fertilization recommendation for the citrus orchards in Taiwan. Annual Research Reports on Soils and Fertilizers 81. Taiwan: Provincial Department of Agriculture and Forestry. p.167-195.

Chapman, J.F., R.W. Daniels and D.H. Scarisbrick. 1984. Field studies on ^{14}C assimilate fixation and movement in oil-seed rape (*B. napus*). *Journal of Agricultural Sciences. (Cambridge)*. 102:23-31.

Chen, G., F.Z. Nian, F.S. Xu, and Y.H. Wang. 2005. Effect of boron and molybdenum on yield and quality of two rapeseed cultivars. *Plant Nutrition and Fertilizer Science*. 11:243–247.

Chiu, T. F. and S.S. Chang. 1985. Diagnosis and correction of boron deficiency in citrus orchard. In: Seminar on Leaf Diagnosis as a Guide to Orchard Fertilization. Technical Bulletin, No. 91. Food Fertilizer and Technology Center, Taipei, Taiwan. p. 1-12.

Christensen, J. V. and J.C. Drabble. 1984. Effect of row spacing and seeding rate on rapeseed yield in Northwest Alberta. *Canandian Journal of Plant Science*. 64:1011-1013.

Clarke, J.M. 1977. Growth relationships and yield of *Brassica napus*. Ph.D. Thesis. University of Saskatchewan, Saskatoon, Sask. 158 p.

Clarke, J.M. 1979. Intra-plant variation in number of seeds per pod and seed weight in Brassica napus 'Tower'. *Canadian Journal of Plant Science*. 59:959-962.

Clarke, J.M. and Simpson, G. M. 1978. Influence of irrigation and seeding rates on yield and yield components of Brassica napus cv. Tower. *Canadian Journal of Plant Science*. 58:731-737.

Cooke, I.J. 1963. Damage to plant roots caused by urea and anhydrous ammonia. *Nature*. 194:1262–1263.

Copeland, L.O., S. Nelson and R. Freed. 2001. Canola production in Michigan. MSU Extension, Bulletin E2766. Michigan State University, East Lansing.

Dabas, S. and I.S. Sheoran. 1984. Photosynthesis, respiration and stomatal conductance in two species of *Brassica* under water stress. *Indian Journal of Plant Physiology*. 27(2):177-183.

Deckers, J. and E. Steinnes. 2004. State of the art on soil-related geo-medical issues in the world. *Advanced Agronomy*. 84:1-35.

Degenhardt, D.F. and Z.P. Kondra. 1981. The influence of seeding date and seeding rate on seed yield and yield components of five genotypes of *Brassica napus*. *Canadian Journal of Plant Science*. 61(2):175-183.

Deora, A., B.D. Gossen, F. Walley and M.R. McDonald. 2011. Boron reduced development of clubroot in canola. *Canadian Journal of Plant Pathology*. 33(4):475-484.

Dewes, T., 1995. Nitrogen losses from manure heaps. *Biological Agriculture and Horticulture.* 11:309–317.

Diepenbrock, W. 2000. Yield analysis of winter oilseed rape (*Brassica napus* L.): A review. *Field Crops Research*. 67:35-49.

Donald, C.M. 1963. Competition among crops and pasture plants. *Advances in Agronomy.* 15:1-118.

Duczek L.J., L.L. Jones-Flory, S.L. Reed, K.L. Bailey and G.P. Lafond. 1996. Sporulation of Bipolaris sorokiniana on the crowns of crop plants grown in Saskatchewan. *Canadian Journal of Plant Science*. 76:861-867.

Dupriez, H., and P. De Leener. Land and Life: African Gardens and Orchards, Growing Vegetables and Fruits. London: Macmillan Publishers Ltd.; 1989. p. 63-72.

Earl, H.J. 2014. Personal Communication.

Eghball, B., B.J. Wienhold, J.E. Gilley and R.A. Eigenberg. 2002. Mineralization of manure nutrients. *Journal of Soil and Water Conservation.* 57(6):470-473.

Eghball, B. and J.F. Power. 1994. Beef cattle feedlot manure management. *Journal of Soil and Water Conservation*. 49(2):113-122.

Eghball, B. and J.F. Power. 1999. Phosphorus- and nitrogen-based manure and compost applications: corn production and soil phosphorus. *Soil Science Society of America Journal.* 63:895-901.

Eitel, J.U.H., D.S. Long, P.E. Gessler, E.R. Hunt Jr. and D.J. Brown. 2008. Sensitivity of ground-based remote sensing estimates of wheat chlorophyll content to variation in soil reflectance. *Soil Science Society of America*. 17:1715-1723.

Elrashidi, M. A. and G.A. O'Connor. 1982. Boron sorption and desorption in soils. *Soil Science Society of America*. 46:27-31.

Fismes, J., P.C. Vong, and A. Guckert. 1999. Use of labelled sulphur-35 for tracing sulphur transfers in developing pods of field-grown oilseed rape. *Communications in Soil Science and Plant Analysis*. 30:221–234.

Fismes, J., P.C. Vong, A. Guckert and E. Frossard. 2000. Influence of sulfur on apparent N-use efficiency, yield and quality of oilseed rape (*Brassica napus* L.) grown on a calcareous soil. *European Journal of Agronomy*. 12:127-141.

Foehse, D. and A. Jungk. 1983. Influence of phosphate and nitrate supply on root hair formation of rape, spinach and tomato plants. *Plant and Soil*. 74:359-368.

Friedrich, J., W. D. Smith and L.E. Schrader. 1977. Herbage yield and chemical composition of switchgrass as affected by N, S, and K fertilization. *Agronomy Journal*. 69:30-33.

Gallagher, J.N. and P.V. Biscoe. Radiation absorption, growth and yield of cereals. 1978. *The Journal of Agricultural Science*. 91(1):47-60.

Gan, Y., S.V. Angadi, H. Cutforth, V.V. Angadi and C.L. McDonald. 2004. Canola and mustard response to short periods of temperature and water stress at different developmental stages. *Canadian Journal of Plant Science*. 84:697-704.

Gan, Y., S.S. Malhi, S. Brandr, E. Katepa-Mupondwa and H.R. Kutcher. 2007. *Brassica juncea* canola in the Northern Great Plains: Responses to diverse environments and nitrogen fertilization. *Agronomy Journal*. 99:1208-1218.

Ganeshaiah, K.N. and R. Uma Shanker. 1992. Frequency distribution of seed number per fruit in plants: A consequence of the self- organizing process. *Current Science*. 62:359–365.

Gao, J., K.D. Thelen, D.H. Min, S. Smith, X. Hao and R. Gehl. 2010. Effects of manure and fertilizer applications on canola oil content and fatty acid composition. *Agronomy Journal*. 102(2):790-797.

Giddens, J. 1964. Micronutrients – the new dimension in agriculture. *Fertilizer*

Solutions Magazine. 7:18-21.

Ghobadi, M., M. Bakhshandeh, G. Fathi, M.H. Gharineh, K. Alami-Said, A. Naderi and M.E. Ghobadi. 2006. Short and long periods of water stress during different growth stages of canola (*Brassica napus* L.): Effect on yield, yield components, seed oil and protein content. *Journal of Agronomy*. 5(2):336-341.

Govahi, M. and M. Saffari. 2006. Effect of potassium and sulphur fertilizers on yield, yield components and seed quality of spring canola (*Brassica napus* L.). *Journal of Agronomy*. 5(4):577-582.

Government of Saskatchewan. 2012. Canola production: Importance of a good start. Available at: http://www.agriculture.gov.sk.ca/Default.aspx?DN=a866a31e-266f-49aa-a3db-44f31dda65ea.

Grant, C.A. and L.D. Bailey. 1993. Fertility management in canola production. *Canadian Journal of Plant Science*. 73:651-670.

Grant, C.A., A.M. Johnston and G.W. Clayton. 2003. Sulphur fertilizer and tillage effects on early season sulphur availability and N:S ratio in canola in Western Canada. *Canadian Journal of Soil Science*. 83:451-463.

Grant, C.A., D.A. Derksen, D.L. McLaren and R.B. Irvine. 2011. Nitrogen fertilizer and urease inhibitor effects on canola seed quality in a one-pass seeding and fertilizing system. *Field Crops Research*. 121:201-208.

Grant, C.A., S.S. Malhi and R.E. Karamanos. 2012. Sulfur management for rapeseed. *Field Crops Research.* 128:119-128.

Grinsted, M.J., M.J. Hedley, R.E. White and P.H. Nye. 1982. Plant-induced changes in the rhizosphere of rape (*Brassica napus* var. Emerald) seedlings. I. pH change and the

increase in P concentration in the soil solution. *New Phytologist*. 91:19.

Gupta, U.C. 1969. Effect and interaction of molybdenum and limestone on growth and molybdenum content of cauliflower, alfalfa and bromegrass on acid soils. *Soil Science Society of America Proceedings*. 33:929-932.

Gupta, U.C. 1972. Effects of boron and lime on boron concentration and growth of forage legumes under greenhouse conditions. *Communications in Soil Science and Plant Analysis*. 3:355-365.

Gupta, U.C. 1979. Boron nutrition of crops. *Advanced Agronomy*. 31:273-307.

Gupta, U.C. 1981. Molybdenum in soils, plants and animals. *Advanced Agronomy*. 34:73-113.

Gupta, U.C. Introduction. In: Boron and its role in crop production. U.C. Gupta, (Ed). ISBN 0-8493-6582-1. Florida USA: CRC Press Inc; 1993. 237 p.

Gupta, U.C., K. Wu and S. Liang. 2008. Micronutrients in soils, crops and livestock. *Earth Science Frontiers*. 15(5):110-125.

Hall, A.E. 1992. Breeding for heat tolerance. *Plant Breeding Review*. 10:129-168.

Halvorson, A.D., B.J. Wienhold and A.L. Black. 2001. Tillage and nitrogen fertilization influence grain and soil nitrogen in annual cropping system. *Agronomy Journal*. 93:836-841.

Hashem, A., M.N. Amin Majumdar, A. Hamid and M.M. Hossain. 1998. Drought stress effects on seed yield, yield attributed, growth, cell membrane stability and gas exchange of synthesized *Brassica napus* L. *Journal of Agronomy and Crop Science*. 180:129-136.

Haynes, R.J. and Naidu, R. 1998. Influence of lime, fertilizer and manure applications on

soil organic matter content and soil physical conditions: a review. *Nutrient Cycling in Agroecosystems*. 51:123-137.

Helps, M.B. 1971. Methods of sowing, seed rate and nitrogen level for oilseed rape. *Exp. Husb*. 20:69-72.

Hocking, P.J., J.A. Kirkgaard, J.F. Angus, A.H. Gibson and E.A. Koctz. 1997. Comparison of canola, Indian mustard and linola in two contrasting environments. 1. Effects of nitrogen fertilizer on dry-matter production, seed yield and seed quality. *Field Crops Research*. 49:107-125.

Hocking, P.J., J.A. Mead, A.J. Good and S.M. Diffey. 2003. The response of canola (*Brassica napus* L.) to tillage and fertilizer placement in contrasting environments in southern New South Wales. *Australian Journal of Experimental Agriculture*. 43:1323-1335.

Hoffland, E., G.R. Findenegg and J.A. Nelemans.1989a. Solubilization of rock phosphate by rape. I. Evaluation of the role of the nutrient uptake pattern. *Plant and Soil.* 113:155-160.

Hoffland, E., G.R. Findenegg and J.A. Nelemans. 1989b. Solubilization of rock phosphate by rape. II. Local root exudation of organic acids as a response to P-starvation. *Plant Soil*. 113:161-165.

Hussain, I., M. Ayyaz Khan and K. Ahmad. 2003. Effect of row spacing on grain yield and the yield components of wheat (Triticum aestivum L.). *Pakistan Journal of Agronomy*. 2(3):153-159.

Hu, D., R.W. Bell and Z. Xie. 1996. Zinc and phosphorous responses in transplanted oilseed rape. *Soil Science and Plant Nutrition*. 42:333–344.

Hu, Y. S., Y.H. Ma, Y.L. Sun and G. Guo. 1994. Effect of B application on the agronomic traits, yields and oil contents of a double-row rape (*Brassica napus* L.) cultivar. *Oil Crops* (China). 16:43-46.

Irvine, R.B. and D. Duncan. 2002. Seeding rate and row spacing effects on sclerotinia stem rot. Available at: http:// www.agr.gc.ca/pfra/sidcpub/sclerot.htm

Issarakraisila, M., Q. Ma, and D.W. Turner. 2007. Photosynthetic and growth responses of juvenile Chinese kale (*Brassica oleracea* var. Alboglabra) and Caisin (*Brassica rapa* subsp. parachinensis) to waterlogging and water deficit. *Scientia Horticulturae*. 111:107-113.

Jackson, G.D. 2000. Effects of nitrogen and sulfur on canola yield and nutrient uptake. *Agronomy Journal*. 92:644-649.

James, S.B. and N.J. Anderson. 1994. Dry matter accumulation, nitrogen, potassium, and plant density. *Journal of Agricultural Research*. 8(2):173-149.

Jan, A., N. Khan, L.A. Khan and B. Khattak. 2002. Chemical composition of canola as affected by nitrogen and sulphur. *Asian Journal of Plant Science*. 1:519–521.

Janzen, H.H. and J.R. Bettany. 1984. Sulfur nutrition of rapeseed. I. Influence of fertilizer nitrogen and sulfur rates. *Soil Science Society of America*. 48:100–107.

Dick, W.A., D. Kost, and L. Chen. 2008. Availability of sulfur to crops from soil and other sources. In: Jez, J. (eds). Sulfur: A Missing Link between Soils, Crops, and Nutrition. ASA-CSSSA-SSSA, 2008. p59-65.

Jurke, C.J. and W.G.D. Fernando. 2008. Effects of seeding rate and plant density on sclerotinia stem rot incidence in canola. *Archives of Phytopathology and Plant Protection*. 41(2):142-155.

Kaiser, B. N., K.L. Gridley, J. Ngaire Brady, T. Phillips and S.D. Tyerman. 2005. The role of molybdenum in agricultural plant production. *Annals of Botany*. 96:745-754.

Karamanos, R.E., T.B. Goh and T.A. Stonehouse. 2003. Canola response to boron in Canadian prarie soils. *Canadian Journal of Plant Science*. 83:249-259.

Karamanos, R.E., T.B. Goh and D.P. Poisson. 2005. Nitrogen, phosphorous and sulfur fertilization of hybrid canola. *Journal of Plant Nutrition*. 28:1145-1161.

Karamanos, R.E., T.B. Goh and D.N. Flaten. 2007. Nitrogen and sulphur fertilizer management for growing canola on sulphur sufficient soils. *Canadian Journal of Plant Science*. 87:201-210.

Kondra, Z. P. 1975. Effects of row spacing and seeding rate on rapeseed. *Canadian Journal of Plant Science*. 55:339-341.

Korte, L.L., J.H. Williams, J.E. Specht and R.C. Sorenson. 1983. Irrigation of soybean genotypes during reproductive ontogeny: II. Yield component responses. *Crop Science*. 23:528-533.

Kuchtova P. and J. Vasak. 1998. The effect of nitrogen and phosphorous fertilization and plant population on Brassica campestris. *Field Crops Research*. 63(11):93-103.

Kumar, A., J. Elston and P. Singh. 1994. Leaf area growth to two *Brassica* species in response to water stress. *Field Crops Research*. 8:594-602.

Kutcher, H.R., J.S. Warland and S.A. Brandt. 2010. Temperature and precipitation effects

of canola yields in Saskatchewan, Canada. *Agricultural and Forest Meteorology*. 150:161-165.

Lääniste, P., J. Jõudo, V. Eremeev and E. Mäeorg. 2008. Effect of sowing date and increasing sowing rates on plant density and yield of winter oilseed rape (*Brassica napus* L.) under Nordic climate conditions. *Soil and Plant Science*. 58:330-335.

Lamb, R.J. 1984. Effects of flea beetles, *phyllotreta* spp. (chrysomelidae: coleoptera), on the survival, growth, seed yield and quality of canola, rape and yellow mustard. *The Canadian Entomologist*. 116:269-280.

Lamb, K. and B. Johnson. 2004. Seed size and seedling depth influence on canola emergence and performance in the northern great plains. *Agronomy Journal*. 96: 454-461.

Lauzon, J. 2014. Personal Communication.

Leach J.E., H.J. Stevenson, A.J. Rainbow and L.A. Mullen. 1999. Effects of plant populations on the growth and yield of winter oilseed rape (*Brassica napus*), *Journal of Agricultural Science (Cambridge)*. 132:173-180.

Leon, J. and H.C. Becker. 1995. Rapeseed (*Brassica napus* L.) genetics. In: Diepenbrock, W. and Becker, H.C. (eds). Physiological Potentials for Yield Improvement of Annual Oil and Protein Crops. *Advances for Plant Breeding*. 17:54-81.

Li, X., Y. Feng, and L. Boersma. 1994. Partition of photosynthates between shoot and root in spring wheat (Triticum aestivum L.) as a function of soil water potential and root temperature. *Plant and Soil.* 164:43-50.

Land Resource Science Agrometeorology (LRSA). 2013. Weather Records for Elora Research Station and Guelph Turfgrass Institute. University of Guelph, ON. Available at: http://agmet.lrs.uoguelph.ca/wxrecord/.

Lythgoe B., R.M. Norton, M.E. Nicolas and D.J. Conner. 2001. Compensatory and competitive ability of two canola cultivars. In: proceeding of the eighth Australian Agronomy Conference. p. 1-8.

Mahan, J.R. and D.R. Upchurch. 1988. Maintenance of constant leaf temperature by plants. I. Hypothesis-limited homeothermy. *Environmental and Experimental Botany.* 28:351-357.

Major, D.J., J.B. Bole and W.A. Charnetski. 1978. Distribution of photosynthates after $^{14}CO_2$ assimilation by stems, leaves and pods of rape plants. *Canadian Journal of Plant Science*. 58:783-787.

Malakouti, M.J. 2007. Zinc is a neglected element in the life cycle of plants: A review. Middle East. *Russian Journal of Plant Science and Biotechnology*. 1:1- 12.

Malakouti, M.J. 2008. The effect of micronutrients in ensuring efficient use of macronutrients. *Turkish Journal of Agriculture and Forestry*. 32:215-220.

Malakouti, M.J. and M.M. Tehrani. Effects of micronutrient on the yield and quality of agricultural products: Micronutrient with macro-effects. Tehran, Iran: Tarbiat Modares University Press; 2005. p. 445.

Malhi, S.S. and K. S. Gill. 2004. Placement, rate and source of N, seedrow opener and seeding depth effects on canola production. *Canadian Journal of Plant Science*. 84:719-729.

Malhi, S.S. and R. Karamanos. 2013. Feasibility of B fertilization on canola and alfalfa in Canadian Prairies. Northeast Agricultural Research Foundation, , Saskatchewan. Available at: http://www.neag.ca/Portals/0/Research%20Results/Articles/Boron%20and%20Boron%2

0Fertilizers.pdf.

Mandal, K.G., K.M. Hati, A.K. Misra and K.K. Bandyopadhyay. 2010. Root biomass, crop response and water-yield relationship of mustard (*Brassica juncea* L.) grown under combinations of irrigation and nutrient application. *Irrigation Science*. 28(3):271-280.

Mandal, K.G. and A.C. Sinha. 2004. Nutrient management effects on light interception, photosynthesis, growth, dry-matter production and yield of Indian mustard (*Brassica juncea*). *Journal of Agronomy and Crop Science*. 190:119-129.

Marquard, R. and A. Gendy. 1989. Studies on the effect of nitrogen fertilization and growth regulators on seed-yield and some quality criteria of oilseed-rape (*Brassica napus* L.) *Fett Wissenschaft Technologie*. 9:353-357.

Marschner, H. Mineral plant nutrition of higher plants. 2nd edn, Academic Press; 1995. p. 889.

Marshall, G., I. N. Morrison, L. Friesen and W. Rother. 1989. Effects of 'volunteer' wheat and barley on the growth and yield of rapeseed. *Canadian Journal of Plant Science*. 69:445-453.

Martin, S.G., R.C. Van Acker and L.F. Friesen. 2001. Critical period of weed control in spring canola. *Weed Science*. 49:326-333.

Mason, M. 1971. Effects of urea, ammonium nitrate and superphosphate on establishment of cereals, linseed and rape. *Australian Journal of Experimental Agriculture and Animal Husbandry*. 11:662-669.

May, W.E., D.J. Hume and B.A. Hale. 1994. Effects of agronomic practices on free fatty acid levels in the oil of Ontario-grown spring canola. *Canadian Journal of Plant Science*. 74:267-274.

McFarlane, J.D., G.J. Judson, and J. Gouzos. 1990. Copper deficiency in ruminants in the southeast of South Austrialia. *Australian Journal of Experimental Agriculture*. 30:187-193.

McGrath, S.P. and F.J. Zhao. 1995. A risk assessment of sulphur deficiency in cereals using soil and atmospheric deposition data. *Soil Use and Management*. 11:110-114.

McGregor, D.I. 1981. Pattern of flower and pod development in rapeseed. *Canadian Journal of Plant Science*. 61:275-282.

McGregor, D.I. 1987. Effect of plant density on development and yield of rapeseed and its significance to recovery from hail injury. *Canadian Journal of Plant Science.* 67:43-51.

McGonigle, T.P., D.G. Evans, and M.H. Miller, M.H. 1990. Effect of soil disturbance on mycorrhizal colonization and phosphorus absorption by maize in growth chamber and field experiments. *New Phytologist*, 116(4):629-636.

McWilliam, S.C., J.A. Stafford, R.K. Scott, G. Norton, D.T. Stokes, and R.S. Bradley. 1995. The relationship between canopy structure and yield in oilseed rape. In: Murphy et al. (ed.) Proceedings of the 9th International Rapeseed Congress. Cambridge, UK: Dorset Press; 1995. (2): p. 491–493.

Mendham, N.J. and P.A. Salisbury. Physiology, crop development, growth and yield. In: Kimber, D.S. and D.I. McGregor. (Ed.). Brassica Oilseeds: Production and utilization. London: CAB International; 1995. p. 11-64.

Mendham, N. J., P.A. Shipway and R.K. Scott. 1981, The effect of seed size, autumn nitrogen and plant population density on the response to delayed sowing in winter oil-seed rape (Brassica napus). *Journal of Agricultural Science (Cambridge)* 96:417-428.

Mengel, K. and E.A. Kirkby. Principles of plant nutrition, 5th ed. Dordrecht: Kluwer Academic Publishers; 2001. p. 243-436.

Miller, M.H., T.E. Bates, D. Singh and A.J. Baweja. 1971. Response of corn to small amounts of fertilizer placed with seed: I. Greenhouse studies. *Agronomy Journal*. 63:365-368.

Minotti, P.L. and Sweet, R.D. Role of crop competition in limiting losses from weeds. In: D. Pimentel, ed. Handbook of Pest Management in Agricultre. Boca Raton, FL: CRC Press; 1981. p 31-367.

Mohammadi, K., A. Ghalavand, M. Aghaalikhani, H. Gholamreza, B. Shahmoradi, Y. Sohrabi. 2011. Effect of different methods of crop rotation and fertilizer on canola traits and soil microbial activity. *Australian Journal of Crop Science*. 5(10):1261-1268.

Molberg, E.S. 1961. Injurious effects of fertilizers applied with the seed on the emergence of flax. *Canadian Journal of Soil Science.* 41:35–43.

Morrison, M. J., P.B.E. McVetty and R. Scarth. 1990a. Effect of altering plant density on growth characteristics of summer rape. *Canadian Journal of Plant Science*. 70:139-149.

Morrison, M.J., P.B.E. McVetty and R. Scarth. 1990b. Effect of row spacing and seeding rates on summer rape in Southern Manitoba. *Canadian Journal of Plant Science.* 70:127-137.

Morrison, M.J. 1993. Heat stress during reproduction in summer rape. *Canadian Journal of Botany*. 71:303-308.

Morrison, M.J. and D.W. Stewart. 2002. Heat stress during flowering in Summer *Brassica*. *Crop Science*. 42:797-803.

Nabipour, A., S.S. Hemayati, M.H. Shirzadi and B. Moosavi. 2009. Study of effects of soluble and soil manganese sulfate on spring canola in Jiroft, Iran. *Plant Ecophysiology*. 3:123-127.

Niaz, A., M. Ibrahim, A. Nisar and S.A. Anwar. 2002. Boron contents of light and medium textured soils and cotton plants. *International Journal of Agriculture and Biology*. 4:534-536.

Niaz, A., A.M. Ranjha, Rahmatullah, A. Hannan and M. Waqas. 2007. Boron status of soils as affected by different soil characteristics–pH, $CaCO_3$, organic matter and clay contents. *Pakistan Journal of Agricultural Sciences*. 44:428-435.

Nielsen, D.C. 1997. Water use and yield of canola under dry land conditions in the Central Great Plains. *Journal of Production Agriculture*. 10:307-313.

Norouzi, M., M. Toorchi, Gh. Hosseini Salekdeh, S.A. Mohammadi, M.R. Neyshabouri and S. Aharizad. 2008. Effect of water deficit on growth, grain yield and osmotic adjustment in rapeseed. *Journal of Food, Agriculture and Environment*. 6(2):312-318.

Nuttall, W.F., H. Ukrainetz, J.W.G. Stewart and D.T. Spurr. 1987. The effect of nitrogen, sulphur and boron on yield and quality of rapeseed (*Brassica napus* L. and *B. campestris* L.). *Canadian Journal of Soil Science*. 67:545-559.

Nyborg, M. 1961. The effect of fertilizer on emergence of cereal grains, flax and rape. *Canadian Journal of Soil Science*. 41: 89–98.

Nyborg, M. and P.B. Hoyt. 1970. Boron deficiency in turnip rape grown on gray wooded soils. *Canadian Journal of Soil Science*. 50:87-88.

O'Donovan J.T. 1994. Canola (*Brassica rapa*) plant density influences Tartary buckwheat (*Fagopyrum tataricum*) interference, biomass, and seed yield. *Weed Science.* 42:385–389.

Oenema, O., Oudendag, D. and Velthof, G.L. 2007. Nutrient losses from manure management in the European Union. *Livestock Science*. 112:261-272.

Ohlsson, I. 1972. Spring rape and spring turnip rape seed sowing at close row spacing. *Svensk Frotidning*. 4l:25-27.

Ohlsson, I. 1976. Effects of row spacing and quality of spring rape and spring turnip rape. *Lantbrukshogskolans Medd*. 253:52.

Olsson, G. 1960. Some relations between number of seeds per pod, seed size and oil content and the effects of selection for these characters in *Brassica* and *Sinapsis*. *Hereditas*. 46:27-70.

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2013a. Zero in on canola seeding rates. Available at: http://www.omafra.gov.on.ca/english/crops/field/news/croptalk/2013/ct-0313a2.htm.

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2013b. Available nutrients and value for manure from various livestock types. Available at: http://www.omafra.gov.on.ca/english/crops/facts/13-043.pdf.

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2013c. Estimated yield, production and farm value of specified field crops, Ontario, 2011-2013. Available at: http://www.omafra.gov.on.ca/english/stats/crops/estimate_new.htm

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2002. Agronomy Guide for Field Crops. Publication 811.

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2009. Spring and Winter Canola: Planting and Crop Development. Available at: http://www.omafra.gov.on.ca/english/crops/pub811/6planting.htm

Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). 2010. Fertilizer Recommendation Tables – 2010 Revisions. Available at: http://www.omafra.gov.on.ca/english/crops/facts/fert-rec-tables-toc.htm.

Organization for Economic Co-Operation and Development (OECD). 2012. Oilseed rape, B *napus* var. *napus* f. *annual* and f. *biennis.* Available at: http://www.oecd.org/officialdocuments/publicdisplaydocumentpdf/?cote=env/jm/mono(2012)41&doclanguage=en.

Ozer, H. 2003. The effect of plant population densities on growth, yield and yield components of two spring rapeseed cultivars. *Plant Soil Environment*. 49:422-426.

Parr, A. J. and Loughman, B. C. Boron and membrane function in plants. In: D. A. Robb & W. S. Pierpoint (Eds.) Metals and Micronutrients: Uptake and utilization by plants. New York: Academic Press; 1983. p. 87-107.

Paul, J.W. and E.G. Beauchamp. 1993a. Nitrogen availability for corn in soils amended with urea, cattle slurry, and solid and composted manures. *Canadian Journal of Soil Science*. 73:253-266.

Paul, J.W. and E.G. Beauchamp. 1993b. Short-term nitrogen dynamics in soil amended with fresh and composted cattle manures. *Canadian Journal of Soil Science*. 74: 147-155.

Peacock, A.D., M.D. Mullen, D.B. Ringelberg, D.D. Tyler, D.B. Hendrick, P.M. Gale and D.C. White. 2001. Soil microbial community responses to dairy manure or ammonium nitrate applications. *Soil Biology and Biochemistry*. 33 (7):1011-1019.

Potter, T.D., J.R. Kay and I.R. Ludwig. 1999. Effect of row spacing and sowing rate on canola cultivars with varying early vigour. In: proceedings of the 10th GCIRC International Rapeseed Congress, Canberra, Australia, 26-29. 4 p.

Qian, P., and J.J. Schoenau. 2010. Effects of conventional and controlled release phosphorus fertilizer on crop emergence and growth response under controlled environment conditions. Effects of Conventional and Controlled Release. *Journal of Plant Nutrition*. 33:1253-1263.

Qian, P., R. Urton, J.J. Schoenau, T. King, C. Fatteicher and C. Grant. Effect of seed-placed ammonium sulfate and monoammonium phosphate on germination, emergence and early plant biomass production of Brassicae oilseed crops. In: U.G. Akpan. Oilseeds. Rijeka, Croatia: Janeza Trdine; 2012; p 53.

Rao, M.S.S. and N.J. Mendham. 1991. Comparison of chinoli (*Brassica campestric* subsp. *Oleifere*. subsp. *Chinensis*) and *B. napus* oilseed rape using different growth regulators, plant populations, densities and irrigation treatments. *Journal of Agricultural Science (Cambridge).* 117:177-187.

Ramsahoi, L. 2013. Alleviating heat stress in spring canola (*Brassica napus* L.) with foliar boron treatment. M.Sc. Thesis, University of Guelph, Ontario. 65pp.

Ramsahoi, L., and H.J. Earl. 2011. Foliar boron reduces heat stress-induced pod abortion in spring canola. Abstracts of the 2011 American Society of Agronomy meetings, San Antonio, TX.

Ramsey, B.R. and A.P.L. Callinan. 1994. Effects of nitrogen-fertilizer on canola production in north central Victoria. *Australian Journal of Experimental Agriculture*. 34:789-796.

Rashid, A., E. Rafique and N. Bughio. 1994. Diagnosing boron deficiency in rapeseed and mustard by plant analysis and soil testing. *Communications in Soil Science and Plant Analysis*. 25:2883-2897.

Rathke, G.W., O. Christen and W. Diepenbrock. 2005. Effects of nitrogen source and rate on productivity and quality of winter oilseed rape (*Brassica napus* L.) grown in different crop rotations. *Field Crops Research*. 94:103-113.

Rendig, V.V., C. Optua, and E.A. MacComb. 1976. Effects of sulfur deficiency on non-protein nitrogen, soluble sugars and N/S ratios in young corn (*Zea mays* L.) plants. *Plant and Soil*. 44:423-437.

Rengel, Z. and P.M. Damon. 2008. Crops and genotypes differ in efficiency of potassium uptake and use. *Physiologia Plantarum*. 133:624-636.

Robertson, M.J. and J.F. Holland. 2004. Production risk of canola in semiarid subtropics of Australia. *Australian Journal of Agricultural Research*. 55:525-538.

Romheld, V. and H. Marschner. Function of micronutrients in plants. In: J.J. Mortvedt, F.R. Cox and L.M. Schuman (eds.). Micronutrients in Agriculture, SSSA Book Series No. 4, 2nd ed. Madison, Wisconsin: Soil Science Society of America, Inc.; 1991. p. 297-328.

Rood, S.B. and D.J. Major. 1984. Influence of plant density, nitrogen, water supply and pod or leaf removal on growth of oilseed rape. *Field Crops Research*. 8:323-331.

Ros, G.H., E.J.M. Temminghoff and E. Hoffland. 2011. Nitrogen mineralization: a review and meta-analysis of the predictive values of soil tests. *European Journal of Soil Science*. 62:162-173.

Rose, T.J., Z. Rengel, Q. Ma, J.W. Bowden. 2008. Post-flowering supply of P, but not K,

is required for maximum canola seed yields. *European Journal of Agronomy*. 28:371-379.

Sadrasm V.O. and D. Calderini. 2009. Environmental Influences on Competitive Ability - Responses to environmental change. In: Crop Physiology: Applications for Genetic Improvement and Agronomy. p. 459. Available at: http://books.google.ca/books?id=LUSC9lT5Q8IC&pg=PA459&dq=Red+to+far+red+ratios+-+plant+intraspecific+competition&hl=en&sa=X&ei=TIQWU7v1Ko_pqAGylYEg&ved=0CDEQ6AEwAQ#v=onepage&q=Red%20to%20far%20red%20ratios%20-%20plant%20intraspecific%20competition&f=false.

Scherer, H.W. 2001. Review: Sulphur in crop production – invited paper. *European Journal of Agronomy*. 14:81-111.

Schwarz, G. and R.R. Mendel. 2006. Molybdenum cafactor biosynthesis and molybdenum enzymes. *Annual Review of Plant Biology*. 57:623–647.

Schöpp, W, M. Posch, S. Mylona and M. Johansson. 2003. Long-term development of acid deposition (1880–2030) in sensitive freshwater regions in Europe. *Hydrology and Earth System Sciences*. 7(4):436–446.

Schroder, J. 2005. Revisiting the agronomic benefits of manure: a correct assessment and exploitation of its fertilizer value spares the environment. *Bioresource Technology*. 96:253-261.

Scott, H. D., S.D. Beasley and L.F. Thompson. 1975. Effect of lime on boron transport and uptake by cotton. *Soil Science Society of America Proceedings*. 39:1116-1121.

Scott, R.K., E.A. Ogunremi, J.D. Ivins and N.J. Mendham. 1973. The effect of fertilizers

and harvest date on growth and yield of oilseed rape sown in autumn and spring. *Journal of Agricultural Sciences*. 81:287-293.

Sen Gupta, A., G.A. Berkowitz and P.A. Pier. 1989. Maintenance of photosynthesis at low leaf water potential in wheat. Role of potassium status and irrigation history. *Plant Physiology*. 89:1358-1365.

Shahin, Y. and R. Valiollah. 2009. Effects of row spacing and seeding rates on some agronomic traits of spring canola (*Brassica napus* L.) cultivars. *Journal of Central European of Agriculture*. 10(1):115-122.

Sheppard, S.C. and T.E. Bates. 1980. Yield and chemical composition of rape in response to nitrogen, phosphorus and potassium. *Canadian Journal of Soil Science*. 60:153-162.

Shorrocks, V. M. 1997. The occurrence and correction of boron deficiency. *Plant and Soil*. 193:121-148.

Sinaki, M.J., E. Majidi Heravan, A.H. Shirani Rad, G. Noormohammadi and G. Zarei. 2007. The effects of water deficit during growth stages of canola (*Brassica napus* L.). *American–Eurasian Journal of Agricultural and Environmental Science*. 2:417–422.

Singh, S. D, and M. Yusuf. 1978. Effect of water, nitrogen and row spacing on yield and oil content of Brown Sarson. *Canadian Journal of Plant Science*. 59:437-444.

Singh Grewal, H. and R.D. Graham. 1999. Residual effects of subsoil zinc and oilseed rape genotype on the grain yield and distribution of zinc in wheat. *Plant and Soil*. 207:29–36.

Singh Grewal, H., Z.G. Lu and R.D. Graham. 1997. Influence of subsoil zinc on dry matter production, seed yield and distribution of zinc in oilseed rape genotypes differing in zinc efficiency. *Plant and Soil*. 192:181–189.

Sinha, A.C., P.K. Jana and B.B. Mandal. 1990. Effect of micronutrients on rapeseed grown on acid soils of Eastern India. *Indian Journal of Agronomy*. 35:126-130.

Sommerfeldt, T.G., C. Chang and T. Entz. 1988. Long-term annual manure applications increase soil organic matter and nitrogen, and decrease carbon to nitrogen ratio. *Soil Science Society of America Journal*. 52:1668-1672.

Statistics Canada. 2009. Canola: a Canadian success story. Available at: http://www.statcan.gc.ca/pub/96-325-x/2007000/article/10778-eng.htm.

Statistics Canada. 2012. Canola seeded area, cencus 2006, and 2001. Available at: http://www.statcan.gc.ca/pub/96-325-x/2007000/t/10778t1-eng.htm.

Statistics Canada, 2013. Production of principal field crops, November 2013. Available at: http://www.statcan.gc.ca/daily-quotidien/131204/dq131204b-eng.htm.

Statistics Canada. 2014. Principal field crop areas, March 31, 2014. Available at: http://www.statcan.gc.ca/daily-quotidien/140424/dq140424a-eng.htm.

Stewart, B.A. and L.K. Porter. 1969. Nitrogen-sulphur relationships in wheat (*Triticum aestivum* L.) corn (*Zea Mays*), and beans (*Phaseolus vulgaris*). *Agronomy Journal*. 61:267-271.

Stoker, R. and K.E. Carter. 1984. Effect of irrigation and nitrogen on yield and quality of oilseed rape. *New Zealand Journal of Experimental Agriculture*. 12:219-224.

Tabatabai, M.A. and Y.M. Chae. 1991. Mineralization of sulfur in soils amended with organic wastes. *Journal of Environmental Quality*. 20:684-690.

Takkar, P.N. Requirement and response of crop cultivars to micronutrients in India – A review. In: P.J. Randall, E. Delhaize and R.A. Richards (eds.), Genetic Aspects of Plant Mineral Nutrition. Netherlands: Kluwer Academic Publishers; 1993. p. 341-348.

Takkar, P. N., I.M. Chibba and S.K. Mehta. Twenty years of coordinated research on micronutrient in soils and plants. Bhopal, India: Indian Institute of Soil Science; 1989. 314 p.

Tandy, S., S. Mundus, J. Yngvesson, T.C. de Bang, E. Lombi, J.K. Schjoerring and S. Husted. 2011. The use of DGT for prediction of plant available copper, zinc and phosphorus in agricultural soils. *Plant Soil*. 346:167-180.

Tautorus, C.L. and N.H. Low. 1994. Possible causes for decreased stability of canola oil produced from green seed. *Journal of American Oil Chemists' Society*. 71:1123-1128.

Taylor, A.J., C.J. Smith and I.B. Wilson. 1991. Effect of irrigation and nitrogen fertilizer on yield, oil content, nitrogen accumulation and water use of canola (*Brassica napus* L.). *Fertilizer Research*. 29:249-260.

Tayo, T.O. and D.G. Morgan. 1975. Quantitative analysis of the growth, development and distribution of flowers and pods in oil seed rape (*Brassica napus* L.). *Journal of Agricultural Science, Cambridge*. 85:103-110.

Tayo, T.O. and D.G. Morgan. 1979. Factors influencing flower and pod development in oil-seed rape (*Brassica napus* L.). *Journal of Agricultural Science*. 92:363-373.

Tesfamariam, E.H., J.G. Annandale and J.M. Steyn. 2010. Water stress effects on winter canola growth and yield. *Agronomy Journal*. 102(2):658-666.

Thomas, P. 1984. Canola growers manual. Canola Council of Canada, Winnipeg, MB. 1308 p.

Toews, W. and R.J. Soper. 1978. Effects of nitrogen source, method of placement and soil type on seedling emergence and barley crop yield. *Canadian Journal of Soil Science*. 58:311–320.

Tomar, J.S. and R.J. Soper. 1987. Fate of 15N-labeled urea in the growth chamber as affected by added organic matter and N placement. *Canadian Journal of Soil Science*. 67:639– 646.

Turkington, T.K, R.A.A. Morrall and R.K. Gugel. 1991. Use of petal infestation to forecast sclerotinia stem rot of canola: Evaluation of early bloom sampling, 1985 – 1990. *Canadian Journal of Plant Pathology*. 13:50–59.

Turkington, T.K. and R.A.A. Morrall. 1993. Use of petal infestation to forecast Sclerotinia stem rot of canola: The influence of inoculum variation over the flowering period and canopy density. *Phytopathology*. 83:682-689.

Uhvits, R. 1946. Effect of osmotic pressure on water absorption and germination of alfalfa seeds. *American Journal of Botany*. 33:278–285.

Velasco, L., C. Mollers and H.C. Becker. 1999. Estimation of seed weight, oil content and fatty acid composition in intact single seeds of rapeseed (*Brassica napus* L.) by near-infared reflectance spectroscopy. *Euphytica*. 106:79-85.

Vyn, T.J., X. Yin, T.W. Bruulsema, C.J.C. Jackson, I. Rajcan and S.M. Brouder. 2002. Potassium fertilization effects on isoflavone concentrations in soybean. *Journal of Agriculture and Food Chemistry*. 50:3501-3506.

Walker, K.C. and E.J. Booth. 1992. Sulphur research on oilseed rape in Scotland. *Sulphur Agriculture*. 16:15-19.

Wang, X., J. Xin, C.A. Grant and L.D. Bailey. 1995. Effects of placement of urea with a urease inhibitor on seedling emer- gence, N uptake and dry matter yield of wheat. *Canadian Journal of Plant Science*. 75:449-452.

Wang, Y. and W-H. Wu. 2010. Plant sensing and signaling in response to K^+ deficiency.

Molecular Plant. 3:280-287.

Warington, K. 1923. The effect of boric acid and borax on the broad bean and certain other plants. *Annals of Botany*. 37:629–672

Welch, R. M. 1995. Micronutrient nutrition of plants. *Critical Reviews in Plant Sciences.* 14:49-82.

Whelpdale, D.M. An overview of the atmospheric sulphur cycle. In: R.W.I. Howarth, J.W.B. Stewart and M.V. Ivanov. Sulphur Cycling on the Continents, SCOPE 48 (eds). Chichester, England: John Wile & Sons; 1992. p. 5-26.

White, P.J. and A.J. Karley. Potassium. In: R. Hell and R.R. Mendel. Plant cell Monographs 17, Cell Biology of Metals and Nutrients. Berlin: Springer; 2010. p. 199-224.

Withers, P.J.A., and F.M. O'Donnell. 1994. The response of double-low winter oilseed rape to fertiliser sulphur. *Journal of the Science of Food and Agriculture*. 66:930-101.

Wooding, F. J. 1985. Interior Alaska crops respond to boron applications. *Agroborealis*. 17:47-49.

Wright, G.C., C.J. Smith and M.R. Woodroffee. 1988. The effect of irrigation and nitrogen fertilizer on rapeseed (*Brassica napus*) production in South-Eastern Australia. I. Growth and seed yield. *Irrigation Science*. 9:1-13.

Wright, P.R., J.M. Morgan, R.S. Jessop and A. Cass. 1995. Comparative adaptation of canola (*Brassica napus*) and Indian mustard (*B. juncea*) to soil water deficits: yield and yield components. *Field Crops Research*. 42:1-13.

Yang, M., L. Shi, F-S. Xu, J-W. Lu and Y-H. Wang. 2009. Effects of B, Mo, Zn and their

interactions on seed yield of rapeseed (*Brassica napus* L.). *Pedosphere*. 19(1):53-59.

Yau, S.K., M.M. Nachit, J. Hamblin and J. Ryan. 1995. Phenotypic variation in boron-toxicity tolerance at seedling stage in durum wheat (*Triticum durum*). *Euphytica*. 83:185-191.

Zentner, R.P., D.W. Wall, C.N. Nagy, E.G. Smith, D.L. Young, P.R. Miller, C.A. Campbell, B.G. McConkey, S.A. Brandt, G.P. Lafond, A.M Johnston and D.A. Derksen. 2002. Economics of crop diversification and soil tillage opportunities in the Canadian prairies. *Agronomy Journal*. 94:216–230.

Zhao, F.H., E.J. Evans, P.E. Bilsborrow and J.K. Syers. 1993. Sulphur uptake and distribution in double and single low varieties of oilseed rape (*Brassica napus* L.). *Plant and Soil*. 150:69-76.

Zhao, F.J., P.E. Bilsborrow, E.J. Evans and S.P. McGrath. 1997. Nitrogen to sulphur ratio in rapeseed and in rapeseed protein and its use in diagnosing sulphur deficiency. *Journal of Plant Nutrition*. 20(5):549-558.

APPENDIX

A 1. Degrees of freedom (df) of effects tested in the modified split-plot design in canola field experiments at the Elora Research Station in 2012 and 2013.

df	Effect
3	Rep
4	Nutrient Treatment
12	Rep x Nutrient Treatment (used to test nutrient treatment main effect)
4	Tier(Rep)
16	Nutrient Treatment x Tier(Rep)
2	Seeding Rate Treatment
8	Seeding Rate Treatment x Tier(Rep) (used to test seeding rate main effect)
8	Nutrient Treatment x Seeding Rate
32	Nutrient Treatment x Tier(Rep) x Seeding Rate (used to test nutrient treatment x seeding rate interation)
30	Residual

A 2. Soil test analyses for fields used in experiments in 2012 and 2013.

Year	Sample Depth	OM	P		K	Mg	Ca	pH	CEC	Al	Nitrate-N
			Bicarb	Bray-P1							
	cm	%	ppm	ppm	ppm	ppm	ppm		meq/100g	ppm	ppm
2012	0-15	5	6	9	20	300	2240	6.9	15	852	45
	15-30	4.5	7	15	27	355	2390	7.5	15	832	11
2013	0-15	4.8	9	12	30	375	2420	7.2	16.1	837	30
	15-30	4.3	8	12	29	360	2420	7.2	15.9	835	22

A 3. Soil test analysis for field soil used in winter and summer 2013 greenhouse studies.

OM	P		K	Mg	Ca	pH	CEC	B	Cu	Fe	Mn	Zn	S	Al	Nitrate-N
	Bicarb	Bray-P1													
%	ppm	ppm	ppm	ppm	ppm		meq/100g	ppm	ppm	ppm	ppm	ppm	ppm	ppm	ppm
3.2	17	32	106	290	2230	7.6	14	0.6	1.2	59	290	4	28	693	32

A 4. Manure source analyses prior to application in field and greenhouse studies.

Experiment		N		P		K		OM	DM	C:N	S	B	Ca	Cu	Fe	Mg	Mn	Zn	Na	Al
		Total	NH_4	Total	P_2O_5	Total	K_2O													
		%	ppm	%	%	%	%	%	%		ppm	ppm	%	ppm	ppm	%	ppm	ppm	%	ppm
Field	2012	0.15	703	0.04	0.09	0.14	0.17	3.5	4.4	13:1	237.6	1.2	0.12	46.2	120.9	0.04	14.2	13.5	0.04	55
	2013	0.16	863	0.03	0.07	0.09	0.11	2.1	2.6	7:1	152.5	1.3	0.07	39.2	29.4	0.03	7.5	9	0.02	14
Greenhouse	Winter	0.19	1043	0.03	0.07	0.13	0.16	3.1	3.7	9:1	187.5	1.5	0.09	37.7	36.3	0.04	8.0	12.3	0.04	9.2
	Summer	0.15	640	0.03	0.07	0.09	0.11	3.1	4.0	12:1	162.5	1.4	0.10	48.9	46.3	0.03	11.2	14.3	0.03	37.6