Improving Weed Management in Flax (*Linum usitatissimum* L.)

A Thesis Submitted to the College of Graduate and Postdoctoral Studies In Partial Fulfilment of the Requirements For the Degree of Master of Science In the Department of Plant Sciences University of Saskatchewan Saskatoon, SK

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Abstract

Flax (*Linum usitatissimum* L.) is a high utility crop that has value in both food and industrial markets. However, because flax is a poor competitor the presence of weeds can negatively influence its growth and development. The objective of this thesis was to evaluate different approaches to help improve weed management in flax, including new herbicide options and investigating integrated weed management (IWM) methodologies. The goal of the IWM experiment was to identify if different combinations of seeding date (early May vs. late May), seeding rate (400 vs. 800 seeds m^{-2}), cultivar height (short vs. tall), and herbicide rate (0X vs. 1X) could improve the competitive ability of flax. Seeding a tall cultivar, at a high seeding rate, in early May, with an in-crop herbicide resulted in the greatest plant population. Factors that improved crop establishment ultimately had a positive influence on the competitive ability of flax by increasing aboveground crop biomass, crop yield, and reducing aboveground weed biomass. In addition, the goal of the herbicide screening trial was to evaluate the tolerance of flax to Group 14 (PPO inhibitors), Group 15 (VLCFA inhibitors), and Group 27 (HPPD inhibitors) herbicides in comparison to registered industry standards. Seven novel modes of action and three registered industry standards were compared to an untreated check to determine the effect that these treatments had on flax growth and development. Flax showed impressive tolerance to most herbicide treatments, with the exception of those containing flumioxazin. Treatments containing topramezone, pyroxasulfone, and fluthiacet-methyl were found to be generally safe. Overall, initial injury caused by these herbicides was transient and did not translate into any reduction in flax yield or thousand seed weight (TSW) at the majority of sites. Results of this study show that flax has acceptable tolerance of several novel herbicides. Moreover, these herbicides can be combined with several cultural factors to improve the competitive ability of flax and ultimately improve how weeds are managed in this crop.

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Dedication

I would like to dedicate this work to my parents, Cameron and Judy Petruic, and to my husband, Zachary. Mom and Dad: I owe this accomplishments to the life lessons you have taught to me. You have shown me that with hard work and a lot of faith, you can accomplish anything. Without many long nights and many prayers, this work would not have been possible. Zach: it was your confidence in me and your endless encouragement that made the difficult days of this process not so bad. You have been my biggest support system throughout this process; thank you for always believing in me.

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List of Abbreviations

ACCase	Acetyl CoA Carboxylase
ALS	Acetolactate Synthase
AMF	Arbuscular mycorrhizal fungi
ANOVA	Analysis of Variance
CPWC	Critical Period of Weed Control
CWSS	Canadian Weed Science Society
CYT450	Cytocrhome P450 monooxygenase
DAT	Days after treatment
HPPD	4 - hydroxphenylpyruvate dioxygenase
HR	Herbicide Resistant
IWM	Integrated Weed Management
LAI	Leaf Area Index
МСРА	2-methyl-4-chlorophenoxyacetic acid
MSD	Minimum Significant Difference
POST	Post emergence
PPO	Protoporphyrinogen oxidase
PRE	Pre emergence
PSII	Photosystem II
PQ	Plastoquinone
TSW	Thousand Seed Weight
VLCFA	Very Long Chain Fatty Acid
WSSA	Weed Science Society of America

1.0 Introduction

Considered one of the first domesticated crops, flax (*Linum usitatissimum* L.) has been cultivated for an estimated 8000 years (Vaisey-Genser and Morris, 2003). It is believed that the cultivated flax crop we grow today was derived from wild relatives of the *bienne* species, (synonymous with the species *Linum angustifolium* Huds.) which is widely distributed across middle eastern countries and shares many of the same genotypic and phenotypic characteristics of flax (Vaisey-Genser and Morris, 2003; Pavelek et al., 2015). However, because of the wide distribution of wild flax species, from Western Europe to North Africa to parts of China, it is difficult to identify the specific center of origin (Vaisey-Genser and Morris, 2003). Nevertheless, the center of origin for the cultivated crop is believed to be in the sub-continental area of India because of the high presence of biological diversity amongst the wild *Linum* populations (Vaisey-Genser and Morris, 2003). Through an increase in trade, flax migrated from eastern areas of Asia and Europe to more western locations.

The consumption of the pressed oils of flax crops, as well as its use in fibre production, make it a desirable plant for cultivation (Vaisey-Genser and Morris, 2003). The seed is approximately 40% oil (Kochhar, 2011), of which 52% is linoleic acid, an omega-3 fatty acid (Oomah, 2001). The qualities of the seed and seed oil give flax high utility as both an industrial oil and one destined for human consumption (Oomah, 2001; Goyal et al., 2014). The characteristics of its oil profile make it an excellent drying oil (Kochhar, 2011; Oomah, 2001), hence a large proportion of the oil's end use is in industrial goods such as linoleum floors, varnishes, and paint products (Pavelek et al., 2015). In fact, the crop currently has its highest utility in industrial markets (Pavelek et al., 2015; Kochhar, 2011). Its use is not limited to only the seed oils, however, as stem fibers have been used as replacements for fiberglass, as well as in the production of paper, textiles, and concrete (Morvan et al., 2003).

The seed and seed meal can be fed to animals and has shown to improve the quality of products such as meat and eggs (Pavelek et al., 2015). For instance, poultry that is fed pressed flax meal produce eggs with a higher omega – 3 fatty acid content (Kochhar, 2011).

Additionally, dairy cattle fed the pressed seed meal, produce milk and milk fat with a higher content of conjugated linoleic acids (Kochhar, 2011). These fatty acids can have a beneficial effect on human health, protecting against cancer, obesity and diabetes (Koba & Yanagita, 2014). Naturally, if consumption of animal products fed a flax diet can have a beneficial influence on human health, then consuming the seed and seed oil directly will have a positive effect as well. The abundance of polyunsaturated and omega – 3 fatty acids (Kaul et al., 2008), lignin's, proteins, antioxidants, and both soluble and insoluble fibers has shown to help the human body combat the onset of diseases such as cardiovascular disease and diabetes, as well as autoimmune disorders (Goyal et al., 2015) such as Crohn's and colitis (Pavelek et al., 2015).

Flax is a crop that commands a high market price and offers a large profit margin to those who choose to produce it, largely due to the low input costs associated with the crop. China, India, and the United States all play a significant role in flax production (Pavelek et al., 2015). However, Canada is the world leader in flax production and export, producing 816 200 tonnes in 2014/2015 (Flax Council of Canada, 2016). In 2014, flax exports from licensed facilities was 451 000 metric tonnes (Canadian Grain Commission, 2015) and was in the top 10 of crops exported from Canada. Most flax varieties have been developed to have either optimal oil seed production or optimal fibre production, but not both. Oilseed flax varieties do not develop the same quality of fibre as those varieties bred for fibre production (Richters, 2011). Many varieties are bred for oil production and quality (Diederichsen, 2001), yet with proper straw management, these varieties can produce fibres of similar quality to fibre flax varieties (Ulrich, 2009).

Despite its high utility, flax productivity can be dramatically influenced by the presence of weeds. Various studies have indicated that flax is a poorly competing crop species in comparison to other oilseed crops such as canola (Dew, 1972). This poor competitive ability can be attributed to the morphology of the flax as well as its initial slow growth. Numerous broadleaf and grassy weed species can be found in flax crops including, but not limited to, green foxtail (*Setaria viridis* L.), wild oat (*Avena fatua* L.), common lamb's quarters (*Chenopodium album* L.), Canada thistle (*Circium arvense* L.), and red root pigweed (*Amaranthus retroflexus* L.) (Leeson et al., 2005). While there are several options for herbicidal weed control, there is a scarcity of registered modes of action for safe in-crop use in flax. Currently only Group 1 (Acetyl CoA Carboxylase inhibitors), Group 4 (synthetic auxins), and Group 6 (Photosystem I

inhibitors) herbicides are registered for use in flax (Flax Council of Canada, 2016). This lack of diversity combined with herbicides classified as "higher risk" for the development of herbicide resistance (Beckie, 2006) has contributed to the evolution of herbicide resistant weedy species. Specifically, wild oat and green foxtail have both developed Group 1 herbicide resistance, resulting in limited options for weed control of these resistant populations (Beckie et al., 2013).

While utilizing different herbicides is not the only solution to deal with herbicide resistant weeds, identifying and utilizing novel modes of action can help to reduce selection pressure on these resistant populations. Thus, one of the goals of this research is to evaluate the tolerance of flax to Group 14 (PPO inhibitors), Group 15 (VLCFA inhibitors), and Group 27 (HPPD inhibitors) herbicides. It is our hopes that through this research novel, low risk modes of action that are not widely used in prairies can be integrated into current weed management systems and broaden the spectrum weeds controlled.

Nevertheless, solutions beyond finding new herbicides are required to improve weed management in flax. Instead, a diversified weed management methodology must be established. Adopting integrated weed management practices from multiple fields of study can not only help to reduce the competitive pressures produced by weeds, but ultimately improve the competitive ability and productivity of flax. Furthermore, it is not enough to identify optimal production practices to maximize crop productivity, but these practices must be easily adoptable by producers if they are ever to be utilized across thousands of hectares. Factors such as seeding date, seeding rate, and cultivar height have all shown to have an effect on crop competitive ability and productivity for numerous crops. As such, these cultural methods may also prove to improve flax competition with weedy species.

Therefore, the second goal of this research is to identify optimal combinations of cultural controls that maximize the competitive ability of flax. This research will also evaluate the combined effect of different seeding dates, seeding rates, and cultivar height on the growth and development of weedy species. As such, we hope to establish practical recommendations for production practices that not only maximize crop yield, but inhibit the competitive effect of weeds present. By evaluating how the combined effect of seeding date, seeding rate, crop height, and herbicide rate influence the development of weedy species, it is hopeful that these combinations of cultural factors can be used to better manage herbicide resistant wild oat as well.

2.0 Literature Review

2.1 Competition & Weed Control in Flax

A major challenge to growing flax (*Linum usitatissimum*) is its poor competitive ability with weedy species. Competition occurs when any essential resource, such as space, nutrients, or water, limits plant growth and development (Liu et al., 2009; Fahad et al., 2015). While there are a number of essential resources, numerous studies have established light as the key resource driving the competitive actions or reactions between different plants (Farrior, 2014; Pierik et al., 2003; Farrior et al., 2013; Ballaré, 1999; Holmgren et al., 1997). Light, and its availability, is a key aspect influencing plant growth and development (Grundel et al., 2014).

Plants phytochromes and chromophores sense various wavelengths of light to gather information about their surrounding environment and respond to it (Gundel et al., 2014; Ballaré, 1999). Using these photoreceptors, plants are able to detect various levels of back-scattered farred light from neighboring plants even before these neighbors are large enough to potentially compete with the crop (Ballaré, 1999). Thus, plants can respond proactively to impending competition from others. Plants with higher leaf areas are able to capture light more efficiently than those with lower leaf areas (Ballaré, 1999). Ballaré (1999) further states that when leaf area index (LAI) values are greater than 1 the amount of light intercepted by a plant's leaves declines dramatically, and as a result, so does its ability to respond to neighbors. Unfortunately, flax has LAI values that range between 3 and 4 (Zhang, 2013). As such this high leaf area index (i.e. low leaf area) could account for the poor competitive ability in flax.

Plants are able to interpret environmental changes through other mechanisms such as volatile compounds, root exudates, and through physical touching of leaves (Gundel et al., 2014). For instance, Pierik et al. (2003) found ethylene-insensitive tobacco plants were less competitive than their wild-type counterparts due to a delayed response to competition from neighbors. Additionally, relationships with soil fungi have also shown to influence competitive responses of plants. In a study evaluating the competitive ability of grassland species, Callaway et al. (2001) found plants having root associations with soil fungi were superior competitors. Roots can also

respond to aboveground cues, which is another aspect of competition that should not be ignored. Gundel et al. (2014) suggest that changes in light quality aboveground may even play an important role in root growth and function. Despite the fact that few studies have been able to distinguish what signals belowground competitive responses, root responses patterns to different aboveground scenarios have been reported in crops such as soybeans (*Glycine max* L. Merr.).

These studies show that capturing light is not the only important factor influencing aboveground competitive ability. No one environmental factor cannot operate without consequently influencing the value of other factors in the surrounding environment (Holmgren et al., 1996). Instead, being able to respond to other factors, such as the release of phytohormones emitted by neighbours, balancing defensive mechanisms, subterranean associations with fungi, and the response of roots to aboveground stimuli all play important roles in competitive plant responses. However, ecological hypotheses regarding crop competition do not fully explain the multitude of interactions within agricultural plant communities (Farrior et al., 2013). Better understanding of these relationships in an agronomic context, where competition is asymmetric, is important if we hope to improve the competitive ability of traditionally uncompetitive crops.

Flax is a slender plant that has the ability to produce apical branches (Diederichsen and Richards, 2003), and which takes until flowering/early boll formation to form a canopy (Zhang et al., 2014). This allows for significant time frame in which weeds can establish themselves. Flax also produces a small taproot with many slender lateral branches (Diederichsen and Richards, 2003) with the greatest density of roots occupying the top 20 cm of soil (Liu et al., 2011). The rooting system of flax is slower to develop when compared to other oil seeds, such as mustard or canola (Liu et al., 2011). Furthermore, these roots show little plasticity in response to environmental effects like droughts or floods (Liu et al., 2011). Zimdahl (2004) explained how competitive species possess the ability to capture light, and respond rapidly to their environment plant characteristics in an agricultural environment where plant densities are high, and there is immense competition for resource capture. Despite the fact that it is able to recover and regenerate from damage to its main stem (Diederichsen and Richards, 2003), flax is at a disadvantage in agricultural systems because of the initial slow growth of roots and shoots and a lower leaf area, both of which contribute to a lower responsiveness to environmental cues.

The Flax Council of Canada (2016) identify green foxtail (*Setaria viridis* L.) and wild oat (*Avena fatua* L.) as the two most abundant weed species in flax crops in the three Prairie Provinces. Despite the fact that there are herbicide resistant populations of both weed species, wild oat appears to be a more problematic species because of their resistance to herbicides, as well as cross resistance to multiple herbicide groups (Beckie et al., 2013). Based on the presence of multiple herbicide resistant populations, the need for improved control methods for wild oat crucial, especially in flax.

2.1.1 Challenges with Wild Oat Control in Flax

Wild oat is quickly becoming one of the most challenging weed species to control in flax and many other field crops (Willenborg et al., 2005; Kirkland, 1993) due to their many competitive characteristics. Wild oat has an innate ability to germinate from various depths and over a number of years, so predicting when seedling may emerge is difficult (Bell and Nalewaja, 1968). Additionally, wild oat can rapidly develop competitive root systems (Kirkland, 1993) that can be advantageous when competing with similar shoot systems above ground. Moreover, wild oat has a short lifecycle and often reach physiological maturity before the crop has matured (Bell and Nalewaja, 1968). Furthermore, wild oat are prolific seed producers with the ability to produce upwards of 2000 seeds per plant (Willenborg et al., 2005). This combination of highly competitive characteristics, is detrimental to crop growth and development.

Not only can the presence of wild oat affect crop yields, but their density, and the duration of competition with the crop also will influence the magnitude of yield reduction wild oat can cause. Bell and Nalewaja (1968) found that the final yield of flax continued to degrease as the population of wild oat increased. Populations of 8 plants m⁻² and 84 plants m⁻² and reduced flax yields by 29.5% and 84% respectively (Bell and Nalewaja, 1968). Kirkland (1993) found similar trends in the response of spring wheat (*Triticum aestivum* L.) to wild oat competition, with wild oat populations of 64 plants m⁻² reducing wheat yields by 28%. Moreover, various studies have shown that crops can grow in the presence of wild oat for a period of time before there is a negative effect on yield (Willenborg et al., 2005; Bell and Nalewaja, 1986; Kirkland, 1993). For example, Bell and Nalewaja (1968) found that removing wild oat at the 4 to 5 leaf stage resulted in yields that were not significantly different from the weed-free check. In spring

wheat, wild oat (64 plants m⁻²) that persisted beyond the 6-leaf stage did not significantly reduce yields (Kirkland, 1993).

In addition to competing vigorously for resources and space, wild oat also exhibits allelopathic effects. Schumacher et al., (1982) found that the exudates produced by wild oat at the 2-4 leaf stage can negatively impact spring wheat development and overall yield. Similarly, Perez and Ormeno-Nunez (1991) noted these root exudates inhibit the germination of spring wheat. Ray and Hastings (1992) found that flax is sensitive to not only the presence of wild oat, but to the allelopathic compounds produced by it. Hence, it is not enough for crops to grow vigorously both above and below ground when competing with wild oat. To compete with wild oat, the production, dissemination and germination of wild oat propagules must also be managed to avoid the allelopathic effects they can have on future crops (Sharma and Vanden Born, 1983).

Better understanding how a crop performs in the presence of wild oat competition is critical if we wish to gain control over this problematic weed species. It is not enough for a crop to only occupy space rapidly, or utilize resources more efficiently than wild oat. Additionally, crops must also be able to tolerate competition belowground from aggressive root systems and allelopathic root exudates. Dew (1972) classified flax as the poorest competitor with wild oat in comparison to wheat and barley. Therefore, if we are to improve the competitive ability of flax, we must understand its limitations, and attempt to exploit the positive characteristics to improve the crops competitive ability.

2.2 Integrated weed management

Integrated weed management (IWM) is a combination of alternative weed control techniques from different subject areas, such as genetic, cultural, biological, mechanical, and chemical weed control (Swanton and Weise, 1991). IWM is a systematic approach that encompasses crop rotation, chemical and mechanical weed control, competition, and soil management (Swanton and Murphy, 1996). Ultimately, the goal of IWM is to incorporate varied weed management techniques to improve crop competitive ability and thus, productivity (Swanton and Weise, 1991; Swanton and Murphy, 1996). A key attribute of integrated weed management is the fact that complete control or elimination of weeds is not the goal; elimination of a plant produces open area for a new weed to potentially establish itself (Zimdahl, 2004). Instead, Zimdahl (2004) explains how, by changing our mindset from controlling weeds to

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managing weeds, the concepts involved in integrated weed management may be easier to implement as well as more efficacious over a long period of time. Managing weed populations to minimize competition and yield reduction of the crop, as well as inhibiting the overall fitness of the weed itself is more important than complete eradication of weed populations from the field. This shift from elimination to management diversifies the strategies producers use when dealing with weeds in the field as well as improve the competitive ability of crops in agricultural systems.

IWM combines multiple weed management methods to manipulate the various environmental factors in a field to improve a crops competitive ability (Harker and O'Donovan, 2013). Factors, such as seeding date, seeding rate, and cultivar choice are some of the basic elements involved in integrated weed management (Harker, 2013). IWM is a multidisciplinary approach to weed control (Swanton and Weise, 1991). As such there are a multitude of weed management techniques that can be incorporated into IWM strategies. Practices such as crop rotation, using competitive cultivars, alternating crop seeding dates, adjusting row spacing, and inter-row tillage are all methods that can be easily adopted (Swanton and Weise, 1991; Harker, 2013; Chauhan et al., 2012). However, integrating new, more complex management techniques like biological control or cover-crops may be more challenging on a large scale for conventional producers (Sciegienka et al., 2011).

2.2.1 Biological Control

Biological control has been widely and successfully used to control invasive weed species such as leafy spurge (*Euphorbia esula* L.) and Canada thistle (*Cirsium arvense* L.) (Bond and Grundy, 2001; Lym, 2005; Sciegienka et al., 2011). However, their effectiveness is reduced when used independently of other weed control methods (Lym, 2005; Sciegienka et al., 2011). For example, in controlling leafy spurge, practices such as herbicides and burning, paired with biological agents (sheep and insects) has helped to manage invasive populations (Lym, 2005).

Pseudomonas syringae pv. *tagetis* is a bacterial pathogen that feeds on Canada thistle (Sciegienka et al., 2011). When acting on its own, this bacterium does not provide sufficient control of Canada thistle (Sciegienka et al., 2011). However, when used with a herbicide and another biological such as *Hadroplontus litura* (F.) there is a positive additive effect between these three factors, thus improving control of Canada thistle (Sciegienka et al., 2011). Biological

controls also can exist in the subterranean environment. Arbuscular mycorrhizal fungi (AMF) form associations with plant roots and can help with greater uptake of water and nutrients (Li et al. 2016). Callaway et al. (2001) found that *Centaurea nigra* (L.) plants which had formed an association with mycorrhizal fungi were larger than those that had not, and that there were active transfers of carbon between the two organisms. Li et al. (2016) further suggests promoting AMF diversity and growth as an effective weed suppression strategy through enhancing crop competitive ability.

2.2.2 Cover Crops

Cover crops are seeded in the field before or after the main crop (Liebman and Davis, 2000; Kruidhof et al., 2008) to improve soil quality and structure, improve nutrient availability, and reduce erosion. However, the use of cover crops has also been shown to help manage weeds (Haramoto and Gallandt, 2004; Liebamn and Davis, 2000; Kruidhof et al., 2008). Kruidhof et al. (2008) found that certain cover crop types, such as radish (*Raphanus sativus* L.) and winter rye (*Secale cereal* L.), were better at suppressing weeds emerging in the fall, while the residues of other species, such as alfalfa (*Medicago sativa* L.), were more effective at inhibiting the development of spring emerging weeds. The suppression is typically due to either more efficient light interception by the cover crop, or from the allelopathic effects of the various plant tissues (Kruidhof et al., 2008). In that regard, Haramoto and Gallandt (2004) have postulated that the glucosinolates in brassica cover crops have allelopoathic effects on sensitive plant species that can prevent seed germination, reduce emergence, and inhibit normal growth and development.

Before new weed management strategies are implemented, their effects and interactions with other weed management techniques must be consistent/predictable, and they should impact the crop and weed differently (Li et al., 2016). The interactions between the various methods used in an IWM system can be additive, antagonistic, or synergistic (Lym, 2005; Sciegienka et al., 2011). Understanding, and identifying, how different management tactics interact is important for developing effective IWM systems. Nevertheless, this has proved challenging. Brassica residues have been shown to inhibit not only the growth and development of weeds, but they can cause yield loss in sensitive crops such as flax (Haramoto and Gallandt, 2004). Similarly, even though AMF has the ability to be of benefit to crop species, they can also enhance moisture and nutrient uptake by weeds (Li et al., 2016). Nonetheless, the interactions

between these biological factors, in combination with herbicides, is an area that until recently has been largely unexplored in agricultural systems (Sciegienka et al., 2011).

2.2.3 Cultivar Selection

Selecting vigorous cultivars is important in regards to improving crop competitive ability with weeds. Several authors reinforce the importance of identifying and utilizing cultivars that grow rapidly as an important weed management tactic for suppressing weeds (Bond and Grundy, 2001; Swanton and Murphy, 1996; Chauhan et al., 2012; Jha et al., 2016). Moreover, Balyan et al. (1991) attribute differences in competitive ability to crop morphology, especially plant height. Plant height is a key factor associated when describing a competitive cultivar (Caton et al., 2001). Some researchers have associated the suppressive ability of tall cultivars with the innate ability to capture more light due to greater height and specific leaf angle (Caton et al., 2001; Jha et al., 2016). In spring wheat, taller cultivars were more effective at suppressing weeds than shorter cultivars (Jha et al., 2016). In soybeans, shorter cultivars were more effective at reducing weed growth in the early season, whereas taller cultivars showed greater weed suppression later into the growing season (Jha et al., 2016). Didon (2002) reported that the ability of barley (Hordeum vulgare L.) cultivars to respond to weeds triggered further growth responses, and those cultivars that emerged and grew rapidly were better able to withstand pressure from surrounding weeds (Sinapis alba L.). Similarly, winter wheat cultivars that grew taller, earlier in the growing season reduced wild oat dry matter, thus also reducing grain yield (Balyan et al., 1991). More recently, Lemerle et al. (2016) demonstrated that vigorous canola (Brassica napus L.) hybrids were more effective at suppressing volunteer wheat than were less vigorous cultivars.

Any traits that enables crops to capture more light will have a pronounced effect on crop yield and better weed management. Taller plants have a distinct advantage when competing for light and space, but variations between cultivars are related to the fact that a plant's competitive ability is tied to its ability to respond to neighbors and utilize light more efficiently. Understanding which cultivars perform better in the presence of competition can help with management decisions and how to best implement these competitive crops in an IWM system.

2.2.4 Crop Rotation

Crop rotations that utilize competitive crop cultivars can enhance weed management through improved functional diversity. Schreiber (1992) stated that crop rotations are essential

not only to weed management, but to the entirety of agriculture production systems. Crop rotations can influence numerous factors, such as nutrient availability and intensity of competition between crops and weeds, and as a result good rotations create an environment that is continually changing (Liebman and Dyck, 1993). It has been proposed that these rotations increase the diversity of weeds present, thus preventing one or two weed species from dominating in the field (Swanton and Weise, 1991; Doucet et al., 1999). However, Doucet et al. (1999) reported that over ten years, a rotation of corn (Zea maize L.), soybean and winter wheat alone did not increase species diversity. Instead they suggested using crop rotation as a component of weed management rather than the sole control measure (Doucet et al., 1999). In corn, control of giant foxtail (Setaria faberi Herrm.) was significantly improved by implementing a three-crop rotation of corn, soybean, and wheat (Schreiber, 1992). This threeyear experiment showed that adding crop diversity aided weed management by reducing total weed seed production (Schreiber, 1992). Similarly, Sarani et al. (2014) found that adopting rotations of wheat-canola, or wheat-safflower (Carthamus tinctorius L.), in combination with tillage increased crop yields and producer profitability by reducing the dominant weed populations. Integrating crops with different root architectures further offers opportunity to improve soil structure and quality (Doucet et al., 1999; Sarani et al., 2014) because healthier soils encourage diversity in soil microflora. These organisms can then form associations with plant roots which can then increase water and nutrient uptake, improving crop competitive ability and weed management as a result.

2.2.5 Seeding Rate

Crop rotations not only influence which cultivars are grown when, but the seeding date and seeding rates of these crops. Seeding at higher rates generally results in earlier canopy development (Vera et al., 2006; Ahmed et al., 2014), an important characteristic in regard to competition between plants. For example, Townley-Smith and Wright (1994) found seeding at a rate of 50 seeds m⁻² was more effective at suppressing weeds than was the growth habit of different field pea (*Pisum sativum* L.) cultivars. Similarly, increasing the seeding rate of hemp (*Cannabis sativa* L.) from 20 kg ha⁻¹ to 80 kg ha⁻¹ resulted in a reduction of weed biomass and thus, an increase in crop yield (Vera et al., 2006). In rice (*Oryza sativa* L.), Ahmed et al., (2014) showed that increasing the seeding rate resulted in a linear decline of weed biomass. A similar inverse relationship between crop density and weed biomass has also been observed in flax

(Stevenson and Wright, 1996). Thus, the benefits of establishing a greater number of crop plants per unit area are apparent and consistent amongst numerous crop types and species.

The interaction between crop density and weed suppression is not a simple correlative relationship, however. The law of constant final yield describes the growth of a plant stand as it relates to pant density (Weiner and Freckleton, 2010). At low densities (i.e. low seeding rates), there is little to no competition for light, space, or resources (Weiner and Freckleton, 2010). As the density of plants per unit area increases, so does the demand for resources, and at very high densities there is a risk of self-thinning (Weiner and Freckleton, 2010). However, densities high enough to result in self-thinning are not likely to occur in the scenario of an agroecosystem. While increased seeding rates can provide advantages to crops competing with weeds, it may only be advantageous until the point at which maximal biomass is achieved. Congruent with this point, seeding canola at higher than the recommended seeding rate had no additional benefit with regard to weed suppression and yield (Lemerle et al., 2016).

2.2.6 Seeding Date

Seeding date is another critical component influencing crop-weed competition. Weeds that emerge before the crop cause greater crop yield losses and produce more weed seeds (Willenborg et al., 2005). Early in the growing season the timing of weed emergence influences competition between the crop and weeds (O'Donovan et al., 1985; Liu et al., 2009). Fahad et al. (2015) found that when weeds and wheat emerged simultaneously, significant yield loss will occur. Likewise, O'Donovan et al. (1985) showed there was a significant relationship between the percent yield loss in a barley crop and wild oat emergence timing relative to that crop, with the greatest yield loss occurring when wild oat emerged before the crop. In spring wheat, wild oat that were not removed from plots reduced yields by as much as 39% (Kirkland, 1993). Wild oat allowed to compete with flax until crop maturity reduced crop yields by 74.9%, yielding on average 345.7 kg ha⁻¹ verses 1355.1 kg ha⁻¹ in the weed free plots (Bell and Nalewaja, 1968). Those weed species which emerge and establish before the crop can capture resources sooner, creating a competitive environment for the newly germinated crop (O'Donovan et al., 1985). Therefore, earlier seeding can enhance crop yields by reducing competition from early emerging weeds. Early seeding can also reduce weed seed production and thus, contribution to weed seed banks as well (Willenborg et al., 2005).

Seeding flax early did not have any significant impact on weed biomass based on Trump et al. (2008). However, these results were drawn from one year of study at only two locations. Seeding early does appear to have a positive effect on flax seed yield and seed oil content (Trump et al., 2008; Gallardo et al., 2014). Conversely, other studies conducted in Saskatchewan have shown that there is no yield benefit when seeding flax in early May compared to late May (Lafond et al., 2008), although seeding later did significantly increase plant density.

Improving weed management in flax requires improving wild oat control. Despite the challenges wild oat and other weedy species pose to flax yields, there are integrated management techniques that have been shown to dramatically improve crop yields as well as significantly reduce the fitness and fecundity of weedy species (Stevenson and Wright, 1996). By stacking multiple control methods, long term, sustainable wild oat management can be achieved. For instance, Harker et al. (2009) explored the additive effects of different agronomic techniques on the competitive ability of barley with wild oat. They found that growing a tall variety at a high seeding rate negatively affected the competitive ability of wild oat (Harker et al., 2009). Beckie et al. (2004) also suggested that increasing the seeding rate, varying the seeding date, and selecting competitive cultivars are effective strategies to improve crop competition with wild oat. Harker et al. (2016) reiterates the importance of high seeding rates and the use of competitive cultivars. Furthermore, Harker et al. (2016) recommended diversifying crop rotations with winter cereal crops silage crops such as alfalfa or barley into crop rotations, as effective management strategies for suppressing and controlling wild oat populations.

Nevertheless, this does not mean cultural practices will ultimately eliminate the usefulness of herbicides. The strength of IWM is its ability to integrate multiple weed management methodologies, including both cultural and chemical methods (Harker and O'Donovan, 2013). When managing problematic weeds, such as leafy spurge or Canada thistle, herbicides are a critical component of management systems and are able to improve the effectiveness of other management factors such as insects, bacteria, or fungi (Lym, 2005; Sciegienka et al., 2011). As well, crop competitive ability is improved with early weed control (Harker et al., 2003), which can be achieved through the use of residual herbicides applied prior to seeding. Furthermore, Sharma and Vanden Born (1983) found that using herbicides combined with competitive cultivars improved wild oat control in barley and wheat. While crop density

(i.e. seeding rate) significantly influence weed biomass and weed competition, Townley-Smith and Wright (1994) also found herbicides helped significantly reduce weed biomass where crop stands were thinner. Therefore, chemical weed controls (i.e. herbicides) should be used as a component of a diverse weed management program, but they should remain as a component and not the sole tactic of weed management systems.

2.3 Chemical weed control

Chemicals have been used to efficiently control weeds in crops since the 1940s (Reade and Cobb, 2002). For herbicides to be effective they must meet several criteria: be highly selective and non-toxic to other organisms, act quickly and be effective at low doses, rapidly degrade in the environment, and be cheap to produce (Reade and Cobb, 2002; Cobb and Reade, 2010). Through the adoption of herbicides, crop yield, crop quality, and producer profitability have all benefitted (Cobb and Reade, 2010). Herbicides act to inhibit a specific target site in the plant, typically an enzyme (Hall et al., 2009) that disrupts normal plant growth and development, causing plant death. The specific mechanism or chain of events that results in plant death is referred to as the mode of action; it is this mode of action that then allows for the classification of various pesticides (i.e. Group 1, Group 2, etc.) (Hall et al., 2009).

2.3.1 Herbicide options for weed control in flax

There is a lack of diversity within registered modes of action to control wild oat and other problematic weed species such as green foxtail, kochia (*Bassia scoparia* L.), wild buckwheat (*Fagopyrum esculentum* Moench.), lambs quarters (*Chenopodium album* L.), and wild mustard (*Sinapis arvensis* L.) in flax (Flax Council of Canada, 2016). Currently there are only 6 modes of action (1, 3, 6, 8, 14) registered for use in flax, with only Groups 1, 4, and 6 being registered for in-crop use (Flax Council of Canada, 2016). This has resulted in an increased use of a few herbicide products with similar modes of action, increasing selection pressure for weeds to develop resistance. In Alberta, Saskatchewan, and Manitoba there are multiple populations of herbicide weed species, such as green foxtail, wild oat, wild mustard, spiny sowthistle (*Sonchus asper* L.), false cleavers (*Galium* spp.) and kochia (Beckie et al., 2013).

Weeds that possess herbicide resistance were traditionally controlled by the herbicide, but now are able to withstand the label rate of the herbicide. Furthermore, the resistance trait is then inherited by the plants' progeny (Hall et al., 2009). Resistant plants can withstand herbicide

damage via target site changes, or increased metabolism to degrade a particular herbicide. A lack of diversity in weed management practices (i.e. an overreliance on herbicides) has resulted in a strong selection pressure for weeds to adapt and survive chemical control (Beckie, 2006). Unfortunately, once a weed develops herbicide resistance, it will always be resistant to that herbicide (Hall et al., 2009).

2.3.2 Herbicide resistance

Three enzymes have been identified as playing a large role in enhancing plant metabolic activity including aryl acylamidase, glutathione transferase, and cytochrome P450 monooxygenase (CYT450) (Preston, 2004). Glutathione transferase and CYT450 both play important roles in enhancing the metabolic activity of grassy weed species (Beckie et al., 2012). As well, enhanced CYT450 activity was found to function in the resistance mechanisms of five wild oat populations showing resistance to ACCase (Group 1) and ALS (Group 2) herbicides (Beckie et al., 2012). Resistance mechanisms conferred by increased aryl acylamidase and glutathione transferase activity are predictable in the sense that they inhibit a specific compound in a chemical family or group (Preston, 2004). However, the result of increasing the activity of CYT450 is less understood and predictable in a HR population (Wang et al., 2013; Preston, 2004). Nevertheless, enhanced activity of CYT450 have been associated with metabolic resistance in several problematic weed species, such as Lolium rigidum (Gaud.), Sinapis arvensis (L.), and Bromus tectorum (L.) (Wang et al., 2013). Preston (2004) showed that the resistance conferred through increased CYT450 activity will affect not only other herbicides in similar chemical groups/families, but herbicides with different modes of action entirely (i.e. crossresistance or multiple-resistance). This makes managing these resistant populations extremely complex.

Furthermore, resistance to herbicides is not limited to one of the two aforementioned options. Yu et al. (2013) suggest that HR weed populations, with the genes required to confer a target site mutation, also may possess genes that enhance their metabolic activity. As a result, HR populations have been found where both target site and metabolism have been altered, rendering certain herbicide groups ineffective. That mechanism is related to how some weed populations, specifically wild oat, have been identified to have resistance to multiple modes of action (Beckie et al., 2012). Consequently, wild oat with resistance to multiple herbicide groups

(Groups 1, 2 and 8) has evolved in various populations (Hall et al., 2009; Beckie et al., 2013). Furthermore, the ability of a single plant to self-fertilize and thus pass on the resistant trait, as well as produce a prolific amount of seed, allows the mutations which confer herbicide resistance to spread rapidly in wild oat populations (Beckie et al., 2012).

2.4 Potential new herbicide options

Without the use of in-crop herbicides, the weed free period is dramatically shortened. While there are several pre-plant herbicides registered for use, flax requires a longer duration of weed control during establishment. Even though there are two in-crop groups that are effective and have not yet selected for resistant populations (Beckie et al., 2013), that does not negate the need for additional control options. Furthermore, despite the fact that wild oat are the most problematic weed species in flax production, focus cannot solely be on controlling a single species. Therefore, the attention of researchers must focus not only on improving wild oat control, but managing other weedy species using novel modes of action.

Diversifying herbicide usage by incorporating novel modes of action that are currently not widely used is one strategy to improve the competitive ability of flax and thus, improve weed control. Herbicides classified as Group 14, 15 or 27 are showing potential to be used safely in flax production systems, as well as help to manage other problematic weed species. The nature of their selectivity and spectrum of weed control is what gives these products the potential to be used in flax. As such, these qualities and characteristics are discussed in greater detail below.

2.4.1 Group 14 - Protoporphyrinogen oxidase (PPO) inhibitors

Group 14 herbicides inhibit the activity of the protoporphyrinogen oxidase (PPO) enzyme, which functions in the synthesis of chlorophyll (Taiz and Zeiger, 2010) and heme (Birchfield, 1996). PPO can be found in both the mitochondrial and chloroplast membranes of plants (Birchfield, 1996). The inhibition of PPO results in a succession of reactions increasing the production of oxygen radicals, which are then able to damage and destroy cellular membranes (Biogot et al, 2007). These products are largely contact herbicides that are readily absorbed by leaves and roots, but translocation is limited (Ware and Witacre, 2004). As a result, PPO inhibitors have a dehydrating effect on plants, resulting in a loss of water and chlorophyll (Böger and Wakabayashi, 1995). Therefore, plants susceptible to this mode of action appear necrotic and brown when exposed to these peroxidizing herbicides (Böger and Wakabayashi, 1995; Ware and Witacre, 2004). In some instances, symptoms can also resemble the activity of auxinic herbicides, resulting in irregular growth patterns (Ware and Witacre, 2004). The activity of these herbicides is enhanced by light exposure (Böger and Wakabayashi, 1995; Ware and Witacre, 2004). When these inhibitors reach the target site, synthesis of chlorophyll is rapidly inhibited, followed by the degradation of chlorophyll, carotenoids, and cell membranes (Böger and Wakabayashi, 1995). Selectivity of these herbicides is based on the plant's ability to metabolize the active ingredient before it has an opportunity to reach and inhibit PPO.

These PPO inhibiting herbicides are divided into several different chemical families: Thiadiazoles, N-phenylphthalimides, and Triazolinones (Mallory-Smith et al., 2003; Senseman, 2007). Fluthiacet-methyl (Thiadiazole family), flumioxazin (N-phenylphthalimide family), and sulfentrazone (Triazolinone family) (Senseman, 2007) all have different chemical structures, but contain a central benzene ring with variable side branches (Böger and Wakabayashi, 1995). The variation of these chemical structures influences when the herbicides can be applied, and on what crops the herbicides are safely applied to. For instance, flumioxazin is applied pre-plant to the soil (Bigot et al., 2007) and is registered for control of broadleaf weeds in soybeans and cereal crops (Government of Saskatchewan, 2016; Ware and Witacre, 2004). Sulfentrazone is also a pre-plant applied herbicide registered to control both grassy and broadleaf weeds in cereal crops (Ware and Witacre, 2004; Government of Saskatchewan, 2016). In contrast fluthiacet-methyl is a post-emergence product that is registered to control broadleaf weeds in corn (Ware and Witacre, 2004).

2.4.2 Group 15 – Very long chain fatty acid (VLCFA) inhibitors

Group 15 herbicides function to inhibit the biosynthetic pathway of fatty acid synthesis, which converts long chain fatty acids (i.e. C16 - C18) into very long chain fatty acids (i.e. C20 - C30) (Tanetani et al., 2009). These herbicides, namely pyroxasulfone, are novel in the sense that few products are registered for safe use in the Prairie Provinces (Government of Saskatchewan, 2016; Ware and Witacre, 2004). VLCFAs are precursors of compounds that assemble to form cuticles on the exterior of plant tissues (Trenkamp et al., 2004). They also are used as fat storage in seeds, and as components of various cell membranes (Trenkamp et al., 2004). As a result, VLCFAs play a critical role in the formation of aerial and subterranean shoots, as well as cell and lipid production (Tanetani et al., 2009). For VLCFAs to be synthesized, a collection of elongating enzymes catalyzes a series of condensation, reduction, and dehydration reactions, resulting in the addition of carbon atoms to the fatty acid chain (Trenkamp et al., 2004). Pyroxasulfone, for example, inhibits plant growth by disrupting these numerous elongating enzymes, thus preventing the formation of >C20 fatty acids from shorter fatty acid chains (Busi et al., 2014).

Pyroxasulfone has been registered for use on corn, wheat, and soybeans and can effectively control both grassy and broadleaf weed species such as barnyard grass, green and yellow foxtail, common water hemp and redroot pigweed (Tanetani et al., 2009; Busi et al., 2014). In Alberta, Saskatchewan and Manitoba, pyroxasulfone is registered with carfentrazone (Group 14) as Focus[®] for use on wheat (spring or winter) corn and soybeans (Government of Saskatchewan, 2016; Government of Manitoba, 2016; Brook and Cutts, 2016), but is not registered for flax.

2.4.3 Group 27 - Hydroxphenylpyruvate dioxygenase (HPPD) inhibitors

HPPD functions in the carotenoid pathway and synthesizes prenylquinones, plastoquinone, and tocopherols (Grossmann and Ehrhardt, 2007), all of which are key components in the synthesis of carotenoids. Group 27 herbicides inhibit the function of HPPD by disrupting plastoquinone biosynthesis, thus inhibiting key steps in the carotenoid biosynthetic pathway including the production of α -tocopherol, and inhibiting normal electron transport. This inhibition negatively affects the integrity of the thylakoid membrane (Grossmann and Ehrhardt, 2007; Reade and Cobb, 2002). Not only is the function and synthesis of carotenoids disrupted, but the whole photosynthetic pathway, including the chloroplasts, are subjected to oxidative degradation (Grossmann and Ehrhardt, 2007; Pest Management Regulatory Agency, 2006). HPPD inhibitors affect actively growing tissues, with early symptoms being observed in the shoot apical meristem (Reade and Cobb, 2002; Cobb and Reade, 2010). When the carotenoid biosynthetic pathway is disrupted, tissues become bleached, followed by necrosis and eventually, plant death (Elmore et al., 2015; Reade and Cobb, 2002; Grossmann and Ehrhardt, 2007).

Selectivity of this herbicide is based on the sensitivity of the enzyme target, rate of herbicide uptake, and the metabolic rate at which the herbicide is broken down (Cobb and Reade, 2010). The tolerance of corn to Group 27 herbicides, namely topramezone, has largely been attributed to a rapid demethylation of topramezone, rendering it inactive (Elmore et al., 2015). In

addition, some herbicide safeners that are designed to protect cereal crops actually function to increase the activity of CYT450 (Elmore et al., 2015), further suggesting that the selectivity of tompramezone is largely based on the plant metabolism. Topramezone is currently registered to control broadleaf weeds in corn in Alberta, Saskatchewan and Manitoba (Government of Saskatchewan, 2016; Government of Manitoba, 2016; Brook and Cutts, 2016).

2.5 The potential for fluthiacet-methyl, flumioxazin, pyroxasulfone, and topramezone to be used in flax

All of the aforementioned herbicides have the potential to diversify chemical control in flax. PPO inhibitors (Group 14) offer efficient control of problematic weed species such as lamb's quarters, pigweed, green foxtail, and water hemp (*Amaranthus rudis* Sauer.) in broadleaf crops (Government of Saskatchewan, 2016; Ware and Witacre, 2004). VLCFA inhibiting herbicides also have the ability to control problematic broadleaves such as kochia, stinkweed (*Thlaspi arvense* L.), and round-leaved mallow (*Malva rotundifolia* L.), as well as grassy species such as wild oat, barnyard grass (*Echinochloa crus-galli* L. Beauv.), and brome species (Government of Saskatchewan, 2016). Furthermore, HPPD inhibitors exhibit the same broad spectrum weed control of both grassy and broad-leaved weeds, such as chickweed (*Stellaria media* L.), ragweed (*Ambrosia* spp.), and wild mustard (Government of Saskatchewan, 2016). Many of these products in these groups can be applied before and after seedling emergence, thus widening the window of weed free growth in the crop.

The selectivity of these modes of actions is largely based on the plants ability to metabolize the herbicide and withstand the damage it may cause. As has been stated earlier, flax has the ability to recover from early damage by forming several new branches from its base. Even though it is a sensitive plant, this plasticity could allow the crop to withstand and outgrow any initial injury. Lastly, these are three modes of action that are extremely novel to use on the prairies. With diversity being a central theme to integrated weed management as well as herbicide resistance management, finding and utilizing new modes of action and implementing them into a rotation is imperative for the longevity of their usefulness. Currently several of these herbicides are registered for safe use in other crops across the prairies, whereas others are only registered for use in the United States (Ware and Witacre, 2004). However, no research or testing has been done for these products in flax. Hence, there is a great need to evaluate the safety of

topramezone, flumioxazin, fluthiacet-methyl, and pyroxasulfone on flax in the Prairie Provinces. Furthermore, with the increasing existence of herbicide resistant weed populations there is a need to identify herbicides with novel modes of action that are safe to use on flax to minimize selection pressure on problematic weed species and improve overall crop productivity.

3.0 Integrated weed management improves flax competitive ability

3.1 Introduction

Integrated weed management (IWM) is a diverse, systematic approach to managing weeds (Swanton and Weise, 1991; Swanton and Murphy, 1996). The goal of IWM is to shift the balance of competition in favour of the crop species rather than complete elimination of weeds (Swanton and Weise, 1991; Swanton and Weise, 1996; Zimdahl, 2004). Adopting management strategies from multiple disciplines adds a new level of diversity into agricultural systems, which can result in more efficacious weed control over a long time-period (Zimdahl, 2004). For instance, well-designed crop rotations, effective chemical and mechanical weed controls, and growing competitive crops all play a role in IWM systems (Swanton and Murphy, 1996). Factors that can influence the surrounding environment to improve the competitive ability of a crop are imperative when developing IWM systems (Harker and O'Donovan, 2013). For example, seeding date, seeding rate, and cultivar choice are influential factors that are at the core of effective IWM strategies (Harker, 2013).

Flax (*Linum usitatissimum* L.) competes poorly with weeds and thus, factors that favour crop competitive ability will have great value in improving weed management in flax production systems. Increased seeding rates have been shown to improve not only crop yield, but crop competitive ability as well. Jha et al. (2017) demonstrated that higher seeding rates in corn (*Zea maize* L.), soybean (*Glycine max* L.), and wheat (*Triticum aestivum* L.) increased crop yield and reduced productivity and fitness of wild oat (*Avena fatua* L.) and wild mustard (*Sinapis arvensis* L.). Similar trends have also been observed in hemp (*Cannabis sativa* L.), where seeding at a 3x seeding rate increased crop biomass and yield while also reducing weed density and size (Vera et al., 2006). However, this response to seeding rate is not always consistent or predictable in all field crops. For example, in canola (*Brassica napus* L.) increasing the seeding rate above 40 plants m⁻² did not improve weed suppression (Lemerel et al., 2016). Furthermore, the response of flax to different seeding rates also has been variable. Optimal flax populations for maximum yield fall within 300 – 400 plants m⁻², with no significant yield benefit observed beyond that range (Lafond et al., 2008). However, in that same study Lafond et al. (2008) reported that the

recommended seeding rate is often too low to achieve optimum plant stands due to variation in soil characteristics, conditions at seeding, and environmental conditions from year to year. Furthermore, it has been observed that while increasing seeding rates can increase flax plant density (plants m⁻²), the yield response is not as dramatic with seeding rate increases (Lafond, 1993).

Seeding date can influence crop-weed competition through the duration of competition between the crop and weeds. This can be critical because it has been suggested that the weedfree period requirement of any crop can range from 25 - 35% of its entire lifecycle (Zimdahl, 2004). For flax, that requirement means the crop needs to be maintained weed free for up to 25 to 30 days (Saskatchewan Seed Growers Association, 2017) and as such, seeding early can help the crop avoid competition from early emerging weeds. This has been observed in other crops where, for example, wild oat caused the greatest yield losses when the weed emerged before the crop (O'Donovan et al., 1985; Thill et al., 1993). Willenborg et al. (2005) suggested that seeding a vigorous cultivar early can help to minimize competition from wild oat in oat (Avena sativa L.). Yet, there can be notable variations in seeding date responses in flax based on site latitude and year (Lafond et al., 2008). For instance, Lafond et al. (2008) found the most southern sites to have higher flax yields when seeded early (May 2nd to May 17th), whereas northern sites performed better when seeded later (May 19th to June 1st). However, that study focused on maximizing crop yield, and did not necessarily evaluate the effect of seeding date on weed management. Therefore, a greater understanding of how seeding date can influence the competitive ability of flax across various sites and site years is needed to improve the productivity of flax.

Crop height is a highly valued trait that plays an important role in crop canopy architecture and development, which in turn aids in overall competitive ability (Jha et al., 2017). Taller cultivars have been found to produce higher yields for crops such as rice (*Oryza sativa* L.), wheat, and corn (Zimdahl, 2004; Jha et al., 2017). Flax cultivars can range in height based on end use, with cultivars destined for the fibre market generally taller than oilseed cultivars (Diederichsen and Richards, 2003). CDC Sorrel is an oilseed cultivar and typically grows to $66.5 \text{cm} \pm 3.5 \text{cm}$ in height (Canadian Food Inspection Agency, 2016; Saskatchewan Seed Growers Association, 2017). Prairie Grande is a shorter oilseed cultivar than CDC Sorrel,
growing to 54.1cm \pm 5cm on average (Duguid et al., 2014; Saskatchewan Seed Growers Association, 2017). Overall, these two cultivars have similar traits, but the height differential between the two cultivars (12.4 cm \pm 1.5cm) may result in differential competitive ability with weeds. This height differential may be further affected by seeding rate, as Lafond (1993) reported higher seeding rates reduced flax crop height. Furthermore, the height of these cultivars can contribute to the vigour of the cultivar and as such, influence weed growth and development (Zimdahl, 2004).

Leeson et al. (2005) identified green foxtail (Setaria viridis L.), wild oat, and wild buckwheat (Polygonum convolvulus L.) as the three most abundant weed species in flax crops from 1970 to 2000 across the prairies. Of all the problematic species identified, wild oat was the only weed species with herbicide resistant populations consistently found across Alberta, Saskatchewan, and Manitoba (Beckie et al., 2007). Wild oat is the most economically detrimental weed on the prairies (O'Donovan et al., 1985) and is a strong competitor compared to many weed and crop species (i.e. flax) (Dew, 1972). Many populations of wild oat across Manitoba, Saskatchewan, and Alberta have evolved resistance to multiple herbicide modes of action (Beckie et al., 2013), making control with herbicides difficult in flax crops. In Saskatchewan, there has been a steady increase from the 1990s to 2003 in Group 1 herbicide use to control grassy species in oilseed crops, with 5% of producers applying a Group 1 every year from 1998 to 2003 (Beckie et al., 2006). Furthermore, producers across the Prairie Provinces have also reported using Group 1 herbicides in 50% of all their fields (Beckie et al., 2007). Such heavy Group 1 use has resulted in a greater selection pressure for resistance (Beckie et al., 2006). Field surveys have showed that the major form of herbicide resistance found in wild oat populations is to Group 1 herbicides (Beckie et al., 2007).

Thill et al. (1993) suggest that agronomic factors such as crop seeding rate, cultivar choice, and seeding date can all be used to collectively reduce competitive ability of wild oat. This is important in flax because there are few graminicides registered for safe use in flax (Flax Council of Canada, 2016). Therefore, there is a need to determine how herbicide resistant wild oat can be managed using integrated management tactics, all while minimizing the selection pressure exhibited on wild oat and other problematic weedy species. While there is a lack of current agronomic research in flax, previous work has helped identify optimal crop population

density, seeding dates, and seeding rates (Lafond et al., 2008; Stevenson and Wright, 1996). However, while these studies demonstrate the inconsistent nature of flax crop performance, the focus of many flax studies has been to determine which agronomic practices maximize crop yield. There is virtually no information on the competitive ability of flax with weeds and in particular, how to improve weed management in flax. The only major study in this area was that of Stevenson and Wright (1996), who found that without the use of a herbicide, weed biomass and flax yield were unaffected by different seeding rates. There remains a need to better understand how multiple agronomic practices can be integrated to improve the ability of flax to effectively compete with weeds, as this relationship is more complex and cannot necessarily be determined solely based on crop yield. Therefore, the objective of this research was to evaluate the combined effect of cultivar height, seeding rate, seeding date, and herbicide rate on the ability of flax to compete with wild oat and other weedy species. We hypothesize that seeding a tall cultivar, at a high seeding rate, early in the growing season, and using an in-crop herbicide will result in the lowest weed biomass, lowest weed seed production (i.e. fecundity), highest crop yield, and greatest thousand seed weight.

3.2 Materials & Methods

3.2.1 Experimental location and design

Field experiments were conducted in 2014, 2015, and 2016 at several locations across Alberta, Saskatchewan, and Manitoba. They study sites included one site in Alberta at the Ellerslie Research Farm (53°25'12.6" N -113°32'43.1" W) near Edmonton, AB in 2014 and 2015. Saskatchewan sites included the Kernen Crop Research Farm near Saskatoon, SK in 2014, 2015 and 2016 (52°09'10.3" N 106°32'41.5" W), the Goodale Research Farm near Saskatoon, SK in 2016 (52°03'48.6" N 106°29'59.7" W). The trial was run at two sites in Manitoba including the Ian Morrison Crop Research Farm in Carman, MB (49°29'18.1" N 98°02'20.9" W) in 2014 and 2015 and at Morden, MB (49° 11'22.2" N 98°05'22.9" W) in 2014, 2015, and 2016. Soil at Ellerslie is classified as a thin, Black Chernozem; at Kernen soil is a Black Chernozemic loam; at Goodale it is a Dark Brown Chernozemic loam; the soil at Carman is a Gleyed Black Chernozem; and Morden is classified with an Orthic and Regosolic Black Chernozemic soil. The Morden site had to be terminated in 2015 due to excessively high herbicide resistant green foxtail populations; thus, the trial was repeated in 2016.

3.2.2 Experimental Procedure

The experiment was conducted as a four-factor randomized complete block design (RCBD) with four replications and 16 treatments (Table 3.1). Treatments consisted of all factorial combinations of seeding date (early vs. late May), seeding rate (400 vs. 800 seeds m⁻²), crop height (tall vs. short cultivar), and herbicide rate (0x vs. 1x). Two flax cultivars varying in height were included in this trial: CDC Sorrel and Prairie Grande. CDC Sorrel is a tall variety that grows to 66.5 cm \pm 3.5 cm (Canadian Food Inspection Agency, 2016) while Prairie Grande is a short variety that typically grows to 54.1 cm \pm 5 cm (Duguid et al., 2014). All seed used for this experiment was untreated, breeder seed sourced from the Kernen Research Farm. Plot size at Ellerslie, Kernen, and Goodale was 2m wide x 8m long. Plots at Carman and Morden were 2.5m wide x 8m long and 3m wide x 8m long, respectively. Border plots were also seeded along both sides of the trial.

Table 3.1 Crop height, seeding rate, seeding date, and herbicide rate for the integrated weed management trail at the Kernen Crop Research Farm (Saskatoon, SK), Goodale Research Farm (Saskatoon, SK), Ian N. Morrison Research Farm (Carman, MB), Morden Research and Development Centre (Morden, MB), and the Ellerslie Research Station (Edmonton, AB) in 2014, 2015, and 2016

Treatment	Crop Height	Seeding rate (seeds m ⁻²)	Seeding Date	Herbicide Rate
1	Tall ^Y	400	Early May	0x
2	Tall	400	Early May	1x
3	Tall	400	Late May	0x
4	Tall	400	Late May	1x
5	Tall	800	Early May	0x
6	Tall	800	Early May	1x
7	Tall	800	Late May	0x
8	Tall	800	Late May	1x
9	Short ^Z	400	Early May	0x
10	Short	400	Early May	1x
11	Short	400	Late May	0x
12	Short	400	Late May	1x
13	Short	800	Early May	0x
14	Short	800	Early May	1x
15	Short	800	Late May	0x
16	Short	800	Late May	1x

Y CDC Sorrel

^Z Prairie Grande

The trial was established on cereal stubble in 2014 and 2015. At Kernen and Goodale, both flax and oats were seeded with a box drill, equipped with hoe openers a 23 cm row spacing. At Carman, a planter with 19 cm row spacing was used, while Morden used a self-propelled cone seeder with a row spacing of 25 cm and eight double offset disk openers capable of side banding fertilizer (Lafond et al., 2008). A Fabro self-propelled plot seeder with a 23 cm row spacing and eight double disc low disturbance openers capable of side banding fertilizer 5 cm beside and 5 cm below the seed was used at Ellerslie. Fertilizer at Kernen and Goodale was applied in two different forms. Nitrogen and sulfur were dribble banded as a liquid formulation (a 100-0-0-20 blend), while phosphorous was applied as a granular formulation with the seed. Fertilizer was applied based on soil test recommendations to attain a target crop yield of 2353 kg ha⁻¹ at all site-years. Actual rates of nitrogen and other fertilizers used at each site year can be found in Table 3.2.

Table 3.2 Agronomic information for actual rates of Nitrogen, Phosphorous, Potassium, and Sulfur applied for 3 years and five locations, integrated weed management study, 2014 to 2016

	A gronomia Variabla	Location							
	Agronomic variable	Kernen ^v	Goodale ^w	Ellerslie ^x	Morden ^Y	Carman ^z			
2014	N applied kg ha ⁻¹	90	-	80	139	-			
	P ₂ O ₅ applied kg ha ⁻¹ *	17	-	50	105	-			
	K applied kg ha ⁻¹	0	-	0	0	-			
	S applied kg ha ⁻¹	0	-	0	46	-			
2015	N applied kg ha ⁻¹	56	-	80	-	73			
	P ₂ O ₅ applied kg ha ⁻¹ *	17	-	50	-	25			
	K applied kg ha ⁻¹	0	-	0	-	0			
	S applied kg ha ⁻¹	0	-	0	-	8			
2016	N applied kg ha ⁻¹	56	67	-	139	-			
	P ₂ O ₅ applied kg ha ⁻¹ *	17	17	-	105	-			
	K applied kg ha ⁻¹	0	0	-	0	-			
	S applied kg ha ⁻¹	0	13	-	46	-			

^V 14 kg ha⁻¹ of 11-52-0 was applied with the seed in 2014, 2015, and 2016. Nitrogen was applied as a liquid formulation (28-0-0) before seeding: 90 kg N ha⁻¹ in 2014, 56 kg N ha⁻¹ in 2015 and 2016

^w 14 kg ha⁻¹ of 11-52-0 was applied with the seed in 2016. Nitrogen and sulfur were applied as a liquid

formulation (100-0-0-20) before seeding: 67 kg N ha⁻¹ and 13 kg S ha⁻¹ of in 2016

^x 46-0-0 and 0-46-0 were used at Ellerslie in 2014 and 2015

 $^{\rm Y}$ 232 kg ha $^{-1}$ of 60-20-0-20 was applied in both 2014 and 2016

 $^{\rm Z}$ In 2015 56 kg ha $^{\rm -1}$ of 16-20-0-14 and 139 kg ha $^{\rm -1}$ of 46-0-0 was applied

* rates of P were converted to P₂0₅ by multiplying rate of P by 2.29

Fertility information was not available for Carman in 2014

Prior to seeding, an application of glyphosate (900 g a.e. ha⁻¹) was applied to control early emerging weeds. Herbicide application at all sites was as described in Chapter 4. Before the flax was seeded, wild oat was cross-seeded at right angles to the flax across all plots at a rate of 100 seeds m⁻² and at a depth of 3 cm. The early seeded treatments were seeded the same day as the wild oat. These plots were seeded at both seeding rates, with both cultivars at a depth of 1 to 2 cm. Two weeks later, the remaining treatments were seeded. Because of poor germination of the wild oat at the Kernen Farm in 2014 and 2015, tame oats were cross-seeded into the plots at Kernen and Goodale in 2016 in place of wild oat to ensure even competition. Due to an excessively high population of herbicide resistant green foxtail, the Morden site was terminated in 2015. Other agronomic details related to dates of field operations can be found in Table 3.3.

Table 3.3 Field operation for integrated weed management trial at the Kernen Crop Research Farm (Saskatoon, SK), Goodale Research Farm (Saskatoon, SK), Ian N. Morrisson Research Farm (Carman, MB), Morden Research and Development Centre (Morden, MB), and the Ellerslie Research Station (Edmonton, AB) in 2014, 2015 and 2016

Site	Veen	Seeding	g Date	Herbicid	le Application	Harvest			
Sile	rear	Early	Late	Early	Late	Early	Late		
	2014	May 8	June 2	June 17	July 1	September 17	September 17		
Kernen	2015	May 11	June 8	June 10	July 2	October 8	October 8		
	2016	May 6	May 27	June 2	June 2	September 15	September 15		
Cormon	2014	May 22	May 28	June 19	June 27	September 17	September 17		
Carman	2015	May 21	June 10	June 18	June 30	September 1	September 28		
Fllovelie	2014	May 13	May 27	July 4	July 4	August 28	September 10		
Ellershe	2015	May 11	May 28	June 19	June 19	September 18	September 23		
	2014	April 24	May 7	June 9	June 17	October 11	October 11		
Morden	2015			S	ITE TERMINATE	D			
	2016	June 7	June 14	June 21	June 21	October 14	October 14		
Goodale	2016	May 6	May 25	June 9	June 9	September 20	September 20		

Treatments that required a herbicide application were sprayed with clethodim (50 g a.i. ha^{-1}) + Merge adjuvant (0.5% v/v) when the wild oats were between the 4 – 6 leaf stage, and the crop was approximately 15 cm tall (flax growth stage 5). Where broadleaf weeds were present, an Curtail–M® (75 g a.e. ha^{-1} clopyralid + 420 g a.e. ha^{-1} MCPA) or bromoxynil (277 g a.i. ha^{-1}) was applied to all plots. Clethodim treatments were applied in Saskatoon, Carman and Ellerslie using backpack applicators with a hand-boom. At Morden, treatments were applied with the same John Deere sprayer as was described in Chapter 4.

Flax and wild oat emergence counts were conducted 2-3 weeks after crop emergence. Crop density was assessed by counting the total number of seedlings present in 2, 1m paired rows in the front and back of each plot. Wild oat counts were performed using a 0.25 m² quadrat at the front and back of each plot. Aboveground biomass samples of flax and weedy species were taken when wild oat reached the soft dough stage. Plant shoot biomass was collected using 0.25 m² quadrats and clipping each plant at the base of its stem at the front and back of each plot. All biomass samples were placed in brown paper bags and oven dried at 130°C for 48 hours and then weighed. Seed from the oat biomass samples were hand threshed and counted to determine weed seed fecundity. Flax boll counts and plant height were taken during the mid-late boll stage of flax on five random plants in each plot. Flax bolls were counted individually, while heights were taken from the ground to the top of the plant using a meter stick. The trial was harvested when the crop was fully mature (flax growth stage 12).

Flax seed yield was collected from whole plots using a plot combine at both Saskatoon locations, Carman, and Morden. Yield was collected at Ellerslie by hand-harvesting plants on both sides of a 2-metre pole at two locations in each plot. A stationary thresher was then used to collect seed from the hand harvested samples in each plot. Yield samples were cleaned and weighed to determine true flax yield and thousand seed weight (TSW). In Saskatoon and Morden, 1000 seeds were counted and weighed from each cleaned yield sample. At Ellerslie and Carman, 250 seeds were counted from each yield sample. The seeds were then weighed and that value was multiplied by 4 to give the weight of 1000 seeds. This was done for four samples per plot and then averaged to determine the final TSW for these sites.

3.2.2 Data Analysis

Initially, an analysis of the data was done using the PROC MIXED procedure in SAS 9.4 (SAS Inst., 2016), with pre-planned contrasts used to make specific comparisons of interest. The fixed effects in the model were seeding date, crop height, seeding rate, and herbicide rate. The random effects were site, replication nested within site, and all site interactions with fixed factors. Differences between means were declared significant at P < 0.05. A PROC UNIVARIATE analysis was conducted to test the assumptions (homogeneous variances and normal distribution of residuals) of the analysis of variance for each variable (SAS Inst. 2016). In addition, residuals were observed using the RESIDUAL statement of PROC MIXED. After

determining the normality of the data set for specific variables, data was then analyzed using a normal, or log normal distribution in PROC GLIMMIX (Bowley, 2015; SAS Inst., 2016).

The WALD-Z test was used to determine if random by fixed effect interactions were significant across all site years. After conducting the covariance analysis using PROC GLIMMIX, it was found that site data could be combined from the 10 various site years (P>0.05). There was a significant (P=0.03) interaction between site-year x height x rate for crop population (Table 3.4), but due to the insignificant effects of all other random factors, and the significant site x rate interaction at all sites, data remained combined for the analysis of crop population. There was a very strong significant (P<0.001) 4-way interaction for flax yield, but after separating sites for individual analysis, problems with normality were experienced (data not shown). In addition, no significant 4-way interactions were observed at the individual site level (data not shown) and the Z-value for this effect was calculated to be negative (-4.13), which indicates a minimal contribution to the variation of the random effect (Kiernan et al., 2012). Therefore, data remained combined for the analysis of flax yield.

The normality of the residual data distribution ultimately determined what distribution would be used when analyzing the combined site data. Crop population, crop height, yield, and thousand seed weight (TSW) were all analyzed using PROC GLIMMIX with a Gaussian distribution because the residual data was normally distributed. Crop biomass, weed biomass, and fecundity had non-normally distributed residual data. As such these variables were analyzed using PROC GLIMMIX with a LOGNORMAL distribution and then back-transformed using an ODS OUTPUT statement (Bowley, 2015) to give us the mean and standard errors of the significant effects. A LSD test (P<0.05 for significance) was conducted to compare LSMEANS for those factors that had a significant effect or interactions for each measured variable.

	Flax Popu	lation	Flax Bior	nass	Weed Bi	omass	Fecundity	/	Crop Hei	ght	Flax Yiel	d	TSW	
	Z Value	Pr>Z	Z Value	Pr>Z	Z Value	Pr>Z	Z Value	Pr>Z	Z Value	Pr>Z	Z Value	$\Pr > Z$	Z Value	Pr>Z
site	1.11		0.95		1.37		0		1.44		1.17		1.98	*
rep(site)	3.16	**	2.72	**	1.66		0.3		3.32	***	3.09	*	1.03	
site*date	1.56		1.69		1.35		0.19		1.82		1.94		1.18	
site*rate	1.55		-0.48		-0.17				-0.66		0.36		-0.76	
site*hgt	1.66		0.92		0.51				0.04		0.89		1.51	
site*herb	0.07		1.47		1.58		0.39		-0.26		1.54		0.18	
site*rate*date	1.5		1.06		1.05		0.82		0.09		-0.57		-0.11	
site*hgt*date	1.01		0.63		0.76		0.83		1.77		1.38		1.85	
site*date*herb	-0.45		1.43		1.57		0.67		0.42		-1.04		0.54	
site*hgt*rate	2.18	*	-0.16		0.27		1.07		1.65		0.01		1.88	
site*rate*herb	0.88		1.14		0.19		0.6		1.62		0.82		0.48	
site*hgt*herb	0.06		0.98		0.25		0		1.2		1.52		1.06	
site*hgt*rate*date	-1.15		-0.85		0.02		0.1		0.39		1.23		-1.87	
site*rate*date*herb	0.11		-1.1		-1.35		0		0.13		1.07		0.37	
site*hgt*date*herb	0.16		0.03		-0.99		•	•	0.05		1.85		-1.44	
site*hgt*rate*herb	-1.62		-0.07		0.21		•	•	-1.39		-0.08		-1.28	
sit*hgt*rat*dat*herb	0.2		0.18		-1.28		0		-1.72		-4.13	***	0.81	
Residual	14.9	***	14.14	***	10.82	***	4.21	***	15.07	***	15.03	***	14.97	***

Table 3.4 Random effects (year and site) and their interactions with crop height, seeding rate, seeding date, and herbicide rate were assessed using the Wald Z Test (COVTEST). Data was combined over 10 site years at Saskatoon, SK, Carman, MB, Morden, MB, and Ellerslie, AB. The Z-values are presented based on the Wald Z Test for crop population (plants m⁻²), crop biomass (kg ha⁻¹), weed biomass (kg ha⁻¹), oat seed fecundity (seeds m⁻²), crop height (cm), crop yield (kg ha⁻¹), and thousand seed weight (g 1000 seeds⁻¹).

ndf = numerator degrees of freedom, ddf = denominator degrees of freedom

Rep = Replication, Hgt = Crop Height, Rate = Seeding Rate. Date = Seeding Date, Herb = Herbicide Rate

* Indicates significant difference p<0.05, ** Indicates strong significant difference p<0.01, ** Indicates very strong significant difference p<0.001

3.3 Results

3.3.1 Environmental Conditions

In 2014, 2015 and 2016 the mean temperatures of all sites were similar to the long-term average observed at each respective site (Table 3.5). The May to October precipitation was variable between site years; however, most sites received near- to above- normal precipitation with a couple of exceptions. The Kernen 2014 site received 61.1mm of precipitation (147% of normal) in May; whereas May 2015 received only 6.3mm of precipitation (15% of normal). For May-October precipitation, Kernen 2016 received above long-term normal moisture than the long-term average while 2014 and 2015 was near the long-term average of 255.2 mm. At the Goodale 2016 location, moisture events in May, June, and September were below the long-term average, whereas moisture in July and August was 193% and 159% higher than the long average respectively. The total May-October rainfall at Morden in 2014 and 2016 was 97 and 118% of the long-term average, respectively. Carman experienced similar rainfall events as Morden with 2014 being slightly drier than the long-term average and 2015 receiving above-normal precipitation. Ellerslie, AB experienced below normal precipitation with 2014 and 2015 receiving only 71% and 81% of the long-term average.

		May	June	July	August	September	October	Avg/Total
Location	Year				Mean Tem	perature (°C)		
	2014	10.1	14.1	18.3	17.9	12.4	6.7	13.3
IZ	2015	11.3	18.1	20.1	18.6	12.9	7.9	14.8
Kernen	2016	13.7	17.4	18.7	16.9	11.8	2.1	13.4
	Long Term ^z	11.8	16.1	19.0	18.2	12.0	4.4	13.6
Cardala	2016	13.7	17.4	18.7	16.9	11.8	2.1	13.4
Goodale	Long Term ^z	12.1	16.8	19.6	18.6	12.4	5.2	14.1
	2014	11.9	17.2	19.0	19.5	14.1	7.9	14.9
Morden	2016	12.8	16.9	17.6	16.9	12.8	3.9	13.5
	Long Term ^z	10.8	15.8	18.2	17.4	11.5	4.0	13.0
	2014	11.3	16.6	17.8	18.7	13.1	7.0	14.1
Carman	2015	10.7	17.5	19.9	18.3	15.8	7.2	14.9
	Long Term ^z	11.6	17.2	19.4	18.5	13.4	5.4	14.3
	2014	8.3	13.0	17.3	16.0	10.3	5.8	11.8
Ellerslie	2015	9.4	15.1	17.2	14.9	9.4	6.0	12.0
	Long Term ^z	10.2	14.1	16.2	15.2	10.2	3.8	11.6
					Precipita	tion (mm)		
	2014	61.1	94.8	44.5	18.5	10.7	14.1	243.7
V.	2015	6.3	20.2	15.1	58.2	50.8	32.7	253.3
Kernen	2016	41.6	49.7	58.6	70.2	24.1	40.8	285.0
	Long Term ^z	36.5	64.6	53.8	44.4	38.1	18.8	255.2
Cardala	2016	23	59.5	104	70	24.1	40.8	321.4
Goodale	Long Term ^z	34.3	63.3	53.9	44.3	38.1	12.0	245.9
	2014	31.7	105.3	43.9	94.0	14.1	6.1	295.0
Morden	2016	74.7	50.2	107.9	21.9	40.5	63.5	358.7
	Long Term ^z	51.7	77.4	63.8	51.2	35.3	24.9	304.3
	2014	30.9	116.7	27.6	122.4	46.3	6.0	349.9
Carman	2015	98.8	75.3	109.3	47.3	42.0	37.3	410.0
	Long Term ^z	69.6	96.4	78.6	74.8	49.0	43.4	411.8
	2014	34.7	46.4	86.0	17.8	33.0	10.2	228.1
Ellerslie	2015	17.3	40.5	95.1	25.3	66.3	12.5	257.0
	Long Term ^z	42.9	72.7	95.6	54.9	40.3	12.6	319.0

Table 3.5 Mean monthly temperature (°C) and precipitation data (mm) at the Kernen Crop Research Farm (Saskatoon), Goodale Research Farm (Saskatoon), Carman Research Farm, Ellerslie Research Farm, and Morden Research Farm in 2014, 2015 and 2016.

^zLong-term normals (1981 to 2010) <u>http://climate.weather.gc.ca/historical_data/search_historic_data_e.html</u>

3.3.2 Effect of seeding date, seeding rate, crop height and herbicide rate on flax density and biomass

Analysis of variance (ANOVA) revealed a significant effect of seeding date (early vs. late May), seeding rate (400 vs. 800 seeds m⁻²), cultivar height (short vs. tall) and herbicide rate (no herbicide vs. herbicide), as well as an interaction between these factors for 6 of the 7

measured variables (Table 3.6). However, seeding date had a significant effect on measured variables only when combined with one or more main factors.

There were two, three-way interactions influencing crop population: a cultivar height x seeding rate x seeding date interaction and a cultivar height x seeding date x herbicide rate interaction (Table 3.6; Figures 3.1a and 3.1b). Increasing the seeding rate from 400 to 800 seeds m^{-2} (P<0.05) increased plant density for both flax cultivars at both seeding dates. Seeding at 800 seeds m^{-2} significantly increased crop population, with the highest population (372 plants m^{-2}) observed for the tall cultivar (CDC Sorrel) planted in early May (Figure 3.1a). By comparison, the combined effect of a short cultivar (Prairie Grande) seeded in early May at the high seeding rate produced only 232 plants m^{-2} (P<0.001) on average. At the late seeding date CDC Sorrel (tall) had a significantly higher plant density than Prairie Grande (short) regardless of seeding rate, with the highest crop density (331 plants m^{-2}) observed when CDC Sorrel (tall) was seeded at 800 seeds m^{-2} . There was no difference in plant population when CDC Sorrel (tall) was seeded at 400 seeds m^{-2} and Prairie Grande (short) was seeded at 800 seeds m^{-2} and Prairie Grande (short) was seeded at 800 seeds m^{-2} and Prairie Grande (short) was produced the greatest crop population.

	Fla	x Pop	oulation		Fla	x Bio	omass		Wee	d Bio	mass		Fec	undi	ty	Cro	o Hei	ght		Flax	Yiel	ł		TSV	W		
Effect	ndf	ddf	F-value		ndf	ddf	F-value		ndf	ddf	F-value		ndf	ddf	F-value	ndf	ddf	F-value		ndf	ddf	F-value		ndf	ddf	F-value	;
Hgt	1	9	18.57	**	1	8	16.50	**	1	8	11.58	**	1	2	0.01	1	9	106.54	***	1	9	2.20		1	9	30.39	***
Date	1	9	0.24		1	8	0.25		1	7	0.03		1	1	0.03	1	9	3.10		1	8	0.75		1	8	4.37	
Hgt x Date	1	9	4.60		1	8	5.22	*	1	7	0.05		1	1	0.02	1	9	1.02		1	9	3.98		1	9	2.60	
Rate	1	9	18.07	**	1	8	7.37	*	1	8	10.57	**	1	2	0.02	1	9	8.17	**	1	9	15.48	**	1	9	0.10	
Hgt x Rate	1	9	12.67	**	1	8	3.53		1	2	1.27		1	2	0.00	1	9	2.22		1	9	3.05		1	9	1.03	
Rate x Date	1	9	0.58		1	8	0.58		1	7	0.94		1	1	0.01	1	9	4.93	*	1	9	0.00		1	8	4.08	
Hgt x Rate x Date	1	9	5.03	*	1	8	1.21		1	6	0.17		1	1	0.11	1	9	0.19		1	9	0.76		1	9	0.74	
Herb	1	9	3.48		1	7	6.15		1	6	6.60	*	1	1	1.12	1	9	0.90		1	8	9.46	**	1	7	0.01	
Hgt x Herb	1	9	4.49		1	8	0.21		1	5	6.52	*	1	1	0.25	1	9	1.59		1	9	3.01		1	9	1.22	
Date x Herb	1	9	0.90		1	7	0.04		1	5	0.00		1	1	0.14	1	8	1.35		1	8	0.24		1	8	0.05	
Hgt x Date x Herb	1	9	0.03		1	8	0.39		1	4	0.33		1	1	0.07	1	9	1.22		1	9	0.25		1	8	0.02	
Rate x Herb	1	9	2.26		1	8	0.18		1	3	3.03		1	1	0.53	1	9	0.61		1	9	0.86		1	9	0.39	
Hgt x Rate x Herb	1	9	6.07	*	1	8	0.00		1	1	0.07		1	2	1.14	1	9	2.69		1	9	9.59	**	1	9	1.79	
Rate x Date x Herb	1	9	0.21		1	8	0.05		1	2	0.58		1	1	0.01	1	9	0.54		1	9	0.98		1	9	0.51	
Hgt x Rate x Date x Herb	1	9	0.29		1	8	0.73		1	5	0.00		1	1	0.06	1	9	1.37		1	9	2.98		1	9	0.01	

Table 3.6 Analysis of variance results (P-values) for measured variables as affected by seeding date, seeding rate, crop height, and herbicide rate. Data was combined over 10 site years from Saskatoon, SK, Carman, MB, Morden, MB, and Edmonton, AB.

ndf = numerator degrees of freedom, ddf = denominator degrees of freedom

Hgt = Cultivar Height, Rate = Seeding Rate. Date = Seeding Date, Herb = Herbicide Rate * Indicates significant difference p<0.05 ** Indicates strong significant difference p<0.01 ** Indicates very strong significant difference p<0.001



Figure 3.1*a* The combined effect of seeding date, seeding rate and cultivar height on crop population (plants m^{-2}). A LS means comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.



A significant three-way interaction between seeding rate, cultivar height, and herbicide rate also impacted crop populations (Figure 3.1b). CDC Sorrel (tall) consistently had a greater plant density than Prairie Grande (short) both in the presence and absence of a herbicide treatment. Where herbicides were not applied, crop density was greater at the higher seeding rate for both cultivars (Figure 3.1b). In the absence of herbicides, however, the highest crop density (336 plants m⁻²) was observed when the tall cultivar was seeded at 800 seeds m⁻². Cultivar tended to have an impact here as CDC Sorrel (tall) often had greater plant establishment than Prairie Grande (short). Seeding CDC Sorrel (tall) at a higher rate, in early May, and utilizing an in-crop herbicide tended to optimize crop density. By contrast, a higher seeding rate was the only factor that improved crop establishment for Prairie Grande (short).

Seeding date and cultivar height interacted to affect crop biomass (Figure 3.2c), while a significant interaction was present for herbicide rate and cultivar height with regard to weed biomass (Figure 3.3b). Increasing the seeding rate from 400 to 800 seeds m⁻² increased (P=0.02) crop biomass by 116 kg ha⁻¹ (Figure 3.2a) and reduced (P=0.01) weed biomass by 64 kg ha⁻¹ on average (Figure 3.3a). Likewise, crop biomass was increased by 549 kg ha⁻¹ where a herbicide was applied (Figure 3.2b). With regard to cultivar height, CDC Sorrel (tall) produced more aboveground biomass than Prairie Grande (short) at both seeding dates, with the greatest biomass (1537 kg ha⁻¹) being produced by CDC Sorrel (tall) when seeded in early May (Figure 3.2c). Furthermore, CDC Sorrel (tall) reduced aboveground weed biomass significantly more than Prairie Grande (short) when a herbicide was applied (P=0.02) (Figure 3.3b). However, in the absence of herbicides, cultivar height did not have a significant effect on aboveground weed biomass production (Figure 3.3b).



Figure 3.2.*a* The effect of seeding rate on crop biomass (kg ha⁻¹) A LSmeans comparison from combined site years. An asterisk (*) between points indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

Figure 3.2.*b* The effect of herbicide rate on crop biomass (kg ha⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

Figure 3.2. The interactive effect of seeding date and cultivar height on crop biomass (kg ha⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a difference between the two means. * indicates P<0.01, ** indicates P<0.01.



Figure 3.3.*a* The effect of seeding rate weed biomass (kg ha⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between points indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

Figure 3.3.*b* The combined effect of herbicide rate and cultivar height on weed biomass (kg ha⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

With regard to cultivar height, we observed that CDC Sorrel (tall) was significantly taller than Prairie Grande (short), thus justifying our choice to use these two cultivars (Figure 3.4a). Furthermore, a significant interaction between seeding rate and seeding date was observed wherein seeding rate did not influence crop height when seeded in early May, but seeding at the later seeding date and at the lower seeding rate produced significantly taller plants (Figure 3.4b). However, the late seeding date and low seeding rate also resulted in lower crop biomass and greater weed biomass (Figures 3.2a, 3.2c, 3.3a). Therefore, despite the increase in crop height, the combination of low seeding rate and late seeding date did not improve competitive traits of flax.



Figure 3.4.a The effect of cultivar height on crop height (cm). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

Figure 3.4.b The combined effect of seeding date and seeding rate on crop height (cm). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

Cultivar height, seeding rate, and herbicide rate exhibited a significant 3-way interaction on crop yield (Figure 3.5). No significant differences were observed between the combinations of cultivar height and seeding rate when herbicides were used (P>0.05). However, CDC Sorrel (tall) had significantly greater yield than Prairie Grande (short) at both the low and high seeding rates in the absence of herbicides. CDC Sorrel (tall) seeded at low and high densities increased flax yields by 265 kg ha⁻¹ (72%) and 261 kg ha⁻¹ (75%), respectively. Although clear effects on yield were observed, there was no significant interaction between main factors for TSW, as cultivar height was the only factor that affected TSW (Figure 3.6). The TSW of the CDC Sorrel (tall) was 92% greater than that of Prairie Grande (short). This difference is likely a function of the inherent differences in seed size between the two cultivars.



Herbicide Rate

Figure 3.5 The combined effect of herbicide rate, seeding rate and cultivar height on crop yield (kg ha⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.01.



Figure 3.6 The effect of cultivar height on thousand seed weight (g 1000 seeds⁻¹). A LSmeans comparison from combined site years. An asterisk (*) between bars indicates a significant difference between the two means. * indicates P<0.05, ** indicates P<0.01, *** indicates P<0.001.

3.4 Discussion

The results of this study show that the competitive ability of flax can be improved by adopting integrated weed management tactics. Seeding rate (400 vs. 800 seeds m⁻²), cultivar height (tall vs. short), seeding date (early vs. late May), and herbicide rate (0X vs. 1X rate) did not exhibit a significant 4-way interaction for any of the measured variables, but these four factors did have a positive effect on the competitive ability of flax. For example, increasing the seeding rate from 400 to 800 seeds m⁻² resulted in a greater crop density for both cultivars. Uniform establishment results in a denser plant stand and produces crops that are better able to compete for light, water, and nutrients (Swanton and Weise, 1991; Swanton and Murphy, 1996). Moreover, Zhang et al. (2014) reported flax seed yield was positively associated with early season vigor and canopy development, which are two traits that also relate positively to competitive ability (Swanton and Murphy, 1996; Swanton and Weise, 1991). Likewise, an important finding in the current study was that increasing crop density resulted in higher crop biomass and lower weed biomass. This concurs with findings in corn, soybean, wheat, and cotton (*Gossypium* spp.), where increasing the seeding rate helped early crop canopy development and suppressed several problematic weed species (Jha et al., 2017).

The application of herbicides also had a significant impact on flax production in our study. In the absence of herbicides, increasing the seeding rate significantly increased yields for both cultivars, but when herbicides were applied, a higher seeding rate did not increase crop yield. This concurs with Stevenson and Wright (1996), who reported reduced weed biomass with increased flax seeding rates; however, seed yield was not improved by higher seeding rates when herbicides were applied in their study. It has been postulated that flax can compensate for variations in seeding rate through phenotypic plasticity. This includes reductions in TSW, adjustments to basal branching (Stevenson and Wright, 1996), or by producing additional bolls per plant (Zhang et al., 2014). This response to low seeding rates has been observed for several other crop species. Specifically, soybean and chickpea (*Cicer arietinum* L.) compensate for low seeding rates by producing more seeds pods per plant (Carpenter and Board, 1997; Nleya and Rickertsen, 2013). As well, sorghum (*Sorghum bicolor* L. Moench.) can increase the number of grains per panicle and increase seed weight when seeding rates are low (Berenguer and Faci, 2001). Hence, it is possible that such allometric adjustments occurred in the current study as

well, which would help to explain why no significant difference in yield was observed between cultivars when a herbicide was used, regardless of seeding rate. The use of an in-crop herbicide likely reduced competition from weeds, thus resulting in more resource availability for the flax crop.

In the current study, crop height responded to changes in seeding rate and seeding date. Treatments that were seeded in late May at a low seeding rate produced taller plants than those seeded in early May or at a high seeding rate. This response is not typical in comparison to other crop types. For example, the presence of competition induced shade avoidance responses in soybean, causing plants to grow taller, produce less branches, fewer pods, and reduce crop yields (Green-Tracewicz et al., 2011; Ballaré, 1999). It is possible that a thinner plant stand caused by a low seeding rate provided more open space for weeds to emerge, resulting in the crop rapidly elongating to intercept more light. The presence of early emerging weeds due to a later seeding date could cause more interspecific competition, potentially triggering shade avoidance responses. In a study conducted by Alex (1967), however, competition from cow cockle (Saponaria vaccaria L.) was found to reduce the height and branching of flax. Therefore, even though the combination of a low seeding rate and a late seeding date produced taller plants in the current study, it did not improve the competitive ability of flax. Moreover, treatments seeded in late May and at the low seeding rate consistently had lower crop establishment and biomass, which is consistent with other studies in flax (Bowden and Friesen, 1967; Gruenhagen and Nalewaja, 1969). Therefore, producers would be advised to avoid low seeding rates and late seeding dates when growing flax as it offers no benefit to competition with weeds.

Seeding date alone did not impact on any of the measured variables, but it did influence crop competitive ability when combined with other factors such as cultivar height and seeding rate. We observed that crop density was greatest when CDC Sorrel (tall) was seeded in early May at 800 seeds m⁻², which with contrasts with Lafond et al. (2008), who observed that the greatest flax density occurred at the later planting dates. They also found seeding date (early vs. late) to have no effect on flax seed yield overall. This response to seeding date was also supported by Holzapfel et al. (2016) who reported seeding early did not significantly increase crop density or yield. However, an important distinction must be made with regard to the context in which the growth and development of flax was evaluated. Lafond et al. (2008) and Holzapfel

et al. (2016) examined the effect of seeding date on flax productivity in an agronomic context, but did not evaluate the effect of seeding date on crop – weed competition. Their recommendation of a later seeding date is therefore not what would be recommended to improve the competitive ability of flax. Instead studies which have examined competition between flax and weeds recommend seeding earlier to shorten the duration of competition between the crop and weeds (Bowden and Friesen, 1967; Spratt et al., 1963).

We observed differences in competitive ability between the two cultivars CDC Sorrel (tall) and Prairie Grande (short). Treatments containing CDC Sorrel (tall) had better crop establishment, higher biomass production, and a greater reduction in weed biomass. While these cultivars were included in the study based on their differences in height, difference between cultivars for other traits may have been due to differences in vigour between the two cultivars. Specifically, CDC Sorrel (tall) was more responsive to changes in seeding date and herbicide rate during crop establishment, whereas the growth and development of Prairie Grande (short) was relatively unaffected. Early season vigour is a desirable trait for a competitive crop (Swanton and Murphy, 1996; Bajwa et al., 2017) and it is plausible that differences in competitive ability between these two cultivars were due to differences in early season vigour rather than height. This was unexpected as much of the literature on competition points to crop height as an important factor consistently influencing crop – weed competition (Peerzada et al., 2017; Jha et al., 2017; Caton et al., 2001; Balyan et al., 1991). Nevertheless, Lafond et al. (2008) observed that three flax cultivars responded differently to changes in seeding rate, with Norlin consistently producing greater plant establishment than AC McDuff and CDC Valour despite the similarities in height, yield, and TSW between these three cultivars (Rowland et al., 2002; Kenaschuk and Rashid, 1994; Kenaschuk and Hoes, 1985). Beckie et al. (2008) postulated this was due to greater early season vigour in Norlin compared with either McDuff or CDC Valour. It is possible that the phenotype (i.e. height) of CDC Sorrel (tall) and Prairie Grande (short) is not the most important quality influencing the competitive ability of the cultivar but rather, differences in early season vigour could be responsible for the differences in competitive ability we observed between the two cultivars.

We did not observe differences in yield between the two very different seeding rates included in the study when herbicides were applied. It is quite probable that herbicide application

significantly reduced weed competition in all plots, resulting in yields at 400 seeds m⁻² to be the same as those seeded at double that rate. Nevertheless, maximizing flax yield was not the ultimate goal of this research. Herbicides did influence yield but they also played a significant role in crop establishment and weed biomass reduction, two important components needed to shift the competitive balance in favour of flax (Pudelko et al., 2015). In the absence of herbicides, Stevenson and Wright (1996) found that unmanaged weeds reduced flax yields by 20%. They also stated that herbicides are a necessity for weed management in flax, but they should be used in combination with other practices that help to suppress weeds. For instance, seeding at a higher rate and removing wild buckwheat early in the growing season helped to improve crop density, reduce the competitive effects of wild buckwheat, and improve flax yields (Gruenhagen and Nalewaja, 1969). Hence, both seeding early and removing competition early in the growing season through the use of a herbicide can help to improve competitive ability in flax.

When there is a high degree of competition between crops and weeds, practices that reduce weed biomass translate directly into increased crop yield (Weiner et al., 2001). Based on this, our results show that seeding CDC Sorrel (tall cultivar) in early May at a high seeding rate with an in-crop herbicide maximized crop establishment, which then had a positive impact on crop biomass production and weed suppression. This agrees well with Kim et al. (2002), who suggested that utilizing competitive cultivars and seeding at a high rate improved the efficacy of herbicides and minimized weed competition. While we did not observe a significant interaction between seeding rate and herbicide rate, seeding at a higher density and using a herbicide did help to reduce weed biomass regardless of crop cultivar. Therefore, we can confidently recommend that increasing seeding rates and using an in-crop herbicide are two effective measures to reduce weed competition and ultimately, improve flax yield and competitive ability.

In the future, the goals of producers must expand beyond simply achieving maximum yields. Instead, they should take into consideration improved long-term weed management by enhancing the competitive ability of the crop. Management tactics like seeding dates and seeding rates are IWM strategies that can be quickly and easily adopted by producers to help develop an early, even flax stand. However, producers are well-advised to choose a cultivar that has shown good early season vigour, as well as one with good standability and tall stature. Results of our study clearly show that these characteristics in combination with early seeding can help

maximize crop light interception and minimize weed competition. In addition, managing weeds during the growing season through the use of an in-crop herbicide is a necessity to reduce weed biomass production. Taking proactive steps to give every competitive advantage to the crop will ultimately improve the longevity and effectiveness of weed management strategies implemented by flax producers.

3.5 Conclusion

Seeding date, seeding rate, herbicide rate, and cultivar height greatly impacted the competitive ability of flax. We found that seeding CDC Sorrel (tall), at a high seeding rate, in early May with a herbicide resulted in the greatest plant population. Factors that improved crop establishment also had a positive influence on crop biomass production and yield, as well as weed biomass reduction. Cultivar height was the only factor affecting TSW, with CDC Sorrel (tall) having a greater TSW than Prairie Grande (short). The overall superior performance of CDC Sorrel (tall) in this study was likely due to greater establishment and early season vigour, rather than the differences in height between the two cultivars. Therefore, seeding a tall, vigorous cultivar, in early May, at 800 seeds m⁻², and using an in-crop herbicide improved the competitive ability of flax which ultimately enhanced the productivity of the crop.

4.0 Identifying new herbicide options for flax production

4.1 Introduction

Canada is the world leader in flax (*Linum usitatissimum* L.) production and exports (Flax Council of Canada, 2016). Flax was grown on 641 400 hectares in 2014 across western Canada (Statistics Canada, 2016), and was one of the top ten crops exported from Canada with exports from licensed facilities totalling 451 000 metric tonnes (Canadian Grain Commission, 2015). It typically commands a high market price and offers a large profit margin to producers, largely due to the low input costs associated with the crop. However, the productivity and quality of flax can be negatively influenced by weed competition (Friesen, 1986). Flax is an uncompetitive species that initially grows more slowly than other crops (Diederichsen and Richards, 2003; Zhang et al., 2014; Liu et al., 2011) and so maintaining a weed-free environment during early stages of growth is critical to preserving crop yield potential.

Of the most troublesome and common weeds in Canada, seven of eight weedy species identified by the Weed Science Society of America (WSSA) are also regarded as the most problematic weeds in flax production (Zollinger, 2016; Flax Council of Canada, 2016). These species include wild oat (*Avena fatua* L.), wild buckwheat (*Polygonum convolvulus* L.), redroot pigweed (*Amaranthus retroflexus* L.), Canada thistle (*Circium arvense* L.), green foxtail (*Setaria viridis* L.), and common lambsquarters (*Chenopodium album* L.) (Zollinger, 2016; Flax Council of Canada, 2016; Leeson et al., 2005). These weeds have many traits in common including high amounts of seed production and rapid early season growth (Beckie, 2006). Despite strong competition and recurrent yield losses associated with these weedy species, there remains a lack of herbicides registered to control them in flax crops.

Currently, Groups 1, 3, 4, 6, 8 and 14 are registered for use in flax (Flax Council of Canada, 2016). Group 3 (cell division inhibitors), Group 8 (long chain fatty acid synthesis inhibitors), and Group 14 (protoporphryinogen oxidase inhibitors) herbicides are all registered as pre-plant (PRE) products and are applied before the crop is sown. While this appears to be a large number of herbicides, only Group 1 (ACCase inhibitors), Group 4 (synthetic auxins), and Group 6 (photosystem II inhibitors) products are registered as post-emergence (POST) products

for in-crop weed control. Thus, there is a lack of diversity with regard to in-crop herbicides. This is problematic because most of these products typically provide control of both grassy and broadleaf weeds such as wild oat, kochia (*Kochia scoparia* L.), and common lambsquarters.

The continued use homogeneous weed control tactics, including a heavy reliance on herbicides, has led to the development of herbicide resistance in many weed populations in western Canada (Beckie, 2006). Based on the results of the Prairie Weed Survey, Beckie et al. (2013) estimated that 7.7 million hectares in Alberta, Saskatchewan, and Manitoba contain weeds resistant to Group 1, 2, and 8 herbicides. Resistant species identified include wild oat, green foxtail, Persian darnel (*Lolium persicum* Boiss and Hohen), wild mustard (*Sinapis arvensis* L.), wild buckwheat, and chickweed (*Stellaria media* L. Vill.) (Beckie et al., 2013). Several of these species were previously identified as problematic in flax production, and herbicide resistance coupled with already limited in-crop weed control options creates the potential for large yield losses due to weeds.

As such, it is imperative that novel herbicides be identified and incorporated into flax production to help manage herbicide resistant weeds. By exploring modes of action that are not traditionally used in the Prairie Provinces, greater diversity in herbicides used can be implemented into crop production. Products such as fluthiacet-methyl (Group 14, POST), flumioxazin (Group 14, PRE), pyroxasulfone (Group 15, PRE), and topramezone (Group 27, POST) all control multiple weed species and provide a novel mode of action not widely utilized in crop production on the Prairies. Fluthiacet-methyl (PPO inhibitor) is traditionally used for incrop control of broadleaf weeds in corn (Zea maize L.) and soybean (Glycine max L.) (Senseman, 2007). Sorghum is similar to flax in that it initially grows slowly, has a limited number of herbicides registered for in-crop use, and is plagued by many weedy species. Reddy et al. (2014) found sorghum (Sorghum biocolor L.) to have an acceptable tolerance to fluthiacetmethyl applied both alone and in a tank mix. As well, fluthiacet-methyl was found provided acceptable control of Palmer amaranth (Amaranthus palmeri S. Wats.) and produced a 39% grain yield advantage compared to an untreated check (Reddy et al., 2014). Flumioxazin provides the same broadleaf weed control in soybeans, but is applied at a different time than fluthiacet-methyl (Senseman, 2007). Flumioxazin can be applied both as a PRE- or POST- product, but its use as a PRE- product is more common (Taylor-Lovell et al., 2002). Applied alone, flumioxazin has not

been found to have good control of weedy species, such as green foxtail (Taylor-Lovell et al., 2002). However, in a tank mix or sequential applications of flumioxazin followed by a POST-herbicide application has been found to have acceptable control of broadleaf species such as velvetleaf (*Abutilon theophrasti* Medic), common cocklebur (*Xanthium pensylvanicum* Wallr.), and common lambsquarters (Taylor-Lovell et al., 2002; Han et al., 2002).

Pyroxasulfone is another PRE- product used to control both annual grassy and broadleaf species in corn (UAP, 2012). Other crops such as wheat (*Triticum aestivum* L.), soybean, fababean (*Vicia faba* L.), and field pea (*Pisum sativum* L.) all have tolerance to pyroxasulfone, although tolerance varies based on rate of application (Tidemann et al., 2014). Pyroxasulfone has been found to provide effective control of several problematic weed species such as Italian ryegrass (*Lolium perenne* L.), redroot pigweed, lambsquarters, wild oat, and false cleavers (*Galium spurium* L.) (Tidemann et al., 2014). Topramezone is a POST- herbicide registered for safe use in corn (registered under the trade name Impact® by UAP, and Armezone® by BASF) that can control both grassy and broadleaf species, thus offering a broad spectrum of weed control (Government of Saskatchewan, 2016; Gitsopoulos et al., 2010). Weedy species controlled by topramezone are very similar to those controlled by pyroxasulfone (Gitsopoulos et al., 2010).

Selectivity of these herbicides is largely based on the plant's ability to metabolize the herbicide and thus avoid any damage that it may cause (Senseman, 2007). For example, tolerance of corn to topramezone has been attributed to the reduced sensitivity of the target enzyme as well as the ability of the crop to rapidly metabolize the herbicide (Grossmann and Ehrhardt, 2007). Similarly, the selectivity of pyroxasulfone has also been attributed to a plant's ability to metabolize the herbicide (Tanetani et al., 2013). Much like topramezone and pyroxasulfone, selectivity of fluthiacet-methyl and flumioxazin is based on differences in uptake, translocation, and degradation of the herbicide between plant species (Shimizu et al., 1995; Price et al., 2004). Based on the selectivity and the novel nature of these herbicides, as well as the ability of flax to recover from damage through the production of apical branches (Diederichsen and Richards, 2003), it is possible that these products have the potential to be safely used in flax production.

The lack of herbicides registered for safe use in flax, as well as the overreliance on Group 1 and 2 herbicides has the potential to place heavy selection pressure on weeds to evolve resistance mechanisms. Moreover, these groups offer limited control of weeds that have already evolved resistance (ex. Group 1 resistant wild oat). Therefore, the objective of this study was to evaluate the tolerance of flax to several novel herbicides that are not registered in flax crops. We hypothesized that flax would have acceptable crop tolerance to Groups 14, 15, and 27 herbicides when applied alone and in combination with currently registered modes of action.

4.2 Materials and Methods

4.2.1 Experimental location and design

An experiment was conducted in 2015 and 2016 at several locations across western Canada: the Kernen Research Farm (52°09'10.3" N 106°32'41.5" W) and Goodale Farm (52°03'48.6" N 106°29'59.7" W) located outside of Saskatoon, SK, Indian Head Research Farm (50°31'59.0" N 103°39'04.5" W) at Indian Head, SK, and Carman Research Farm (49°29'18.1" N 98°02'20.9" W) in Carman, MB. The soil at the Kernen and Carman sites is a Black Chernozemic loam while Goodale is located on a Dark Brown Chernozemic sandy loam. At Indian Head, the trial was conducted on a Regosolic Black Chernozemic clay soil (Saskatchewan Soil Survey, 1986). The pH and soil organic matter content at each location can be found in Table 4.1. The trial was seeded at a rate of 800 seeds m⁻², with the flax variety CDC Glas (Booker et al., 2014), which is a popular flax variety in western Canada (Flax Council of Canada, 2015). Plots at Kernen were 2m wide x 6m long, while plots at Carman were 2m wide x 8m long and plots in Indian Head were 3m wide x 10.7 m long. Border plots were seeded on both sides of the trial at all locations.

		Soil Description							
Site-year	Soil Type	pН	OM	Sand	Silt	Clay			
			(%)	(%)	(%)	(%)			
Kernen – 2015	Black Chernozem	7.9	2.4	19	36	45			
Carman – 2015	Black Chernozem	5.5	6.0	54	15	31			
Indian Head – 2015	Black Chernozem	7.4	3.4	13	21	66			
Kernen – 2016	Black Chernozem	7.9	2.4	19	36	45			
Carman – 2016	Black Chernozem	5.5	6.0	54	15	31			
Indian Head – 2016	Black Chernozem	7.4	3.4	14	21	66			
Goodale – 2016	Dark Brown Chernozem	7.0	1.9	37	40	23			

 Table 4.1 Soil classification and descriptions for each site-year.

4.2.2 Experimental procedure

Plots were established on fallow at Saskatoon and Carman and into standing cereal stubble at Indian Head. All plots were maintained weed-free throughout the entire growing season. Prior to seeding, a glyphosate burnoff at 675 g a.e. ha⁻¹ was applied. In cases where glyphosate resistant volunteer canola was present, carfentrazone (18 g ai ha⁻¹) or bromoxynil (280 g ai ha⁻¹) was tank-mixed to ensure adequate weed control. A tractor-mounted sprayer equipped with TurboTee Jet Airmix 100015 nozzles calibrated to deliver a volume of 100 L ha⁻¹ at 275 kPa was used to apply the pre-seed burnoff at both Saskatoon locations. Spraying equipment at Indian Head was a self-propelled sprayer equipped with 110015 nozzles calibrated to deliver treatments at a pressure of 276 kPa. For overspray of the entire trial, a sprayer equipped with Bubblejet 8002 tips was calibrated to deliver 11.25 L ha⁻¹ at 262 kPa was used at Carman. For treatment application at Carman, a hand-boom was used. This sprayer was equipped with 80015 XR sprayer tips to apply 1L per treatment at 276 kPa.

Fertilizer requirements at all sites were determined via pre-seeding soil tests at each site with a yield goal of 2197 kg ha⁻¹ of flax (approximately 33 bushels acre⁻¹). At the Kernen and Goodale locations, nitrogen and sulphur were dribbled banded on the crop in a liquid formulation (100-0-0-20), while potassium and phosphorous were applied as a granular with the seed. All fertilizer was applied as a side band at the time of seeding in Indian Head. At Carman fertilizer was applied across replications perpendicular to the direction that the plots were seeded. A double disc seeder was used to apply the fertilizer 5 - 7 cm deep before the crop was seeded. A

complete list of agronomic details, specifically actual rates of nitrogen and other fertilizers used at each site year can be found in Table 4.2.

. .	•		•							
		Location								
	Agronomic Variable	Kernen ^W	Goodale ^x	Indian Head ^Y	Carman ^Z					
2015	N applied kg ha ⁻¹	58	-	56	73					
	P ₂ O ₅ applied kg ha ⁻¹	18	-	39	25					
	K applied kg ha ⁻¹	0	-	8	0					
	S applied kg ha ⁻¹	0	-	8	8					
2016	N applied kg ha ⁻¹	58	69	-	73					
	P ₂ O ₅ applied kg ha ⁻¹	18	18	-	39					
	K applied kg ha ⁻¹	0	0	-	0					
	S applied kg ha ⁻¹	0	14	-	12					

Table 4.2 Agronomic information for actual rates of Nitrogen, Phosphorous, Potassium, and Sulfur applied for 2 years and five locations, evaluation of crop tolerance study, 2015 and 2016

^w14 kg ha⁻¹ of 11-52-0 was applied with the seed in 2014, 2015, and 2016. Nitrogen was applied as a liquid formulation before seeding: 90 kg N ha⁻¹ in 2014, 56 kg N ha⁻¹ in 2015 and 2016 ^x14 kg ha⁻¹ of 11-52-0 was applied with the seed in 2016. Nitrogen and sulfur were applied as a liquid formulation before seeding: 67 kg N ha⁻¹ and 13 kg S ha⁻¹ in 2016

^Y 55.6 kg ha⁻¹ of 100-30-15-15 was applied before seeding in both 2015 and 2016

^Z In 2015 56 kg ha⁻¹ of 16-20-0-14 and 139 kg ha⁻¹ of 46-0-0 was applied. In 2016 84 kg ha⁻¹ of 16-20-0-14 and 129 kg ha⁻¹ of 46-0-0 was applied

The trial was seeded in mid to late May, when the soil temperatures are typically 10°C or higher (Table 4.3). At Carman, a planter with 19 cm row spacing was used to seed the trial. At Saskatoon locations, the trial was seeded with a box drill, with hoe openers on a 23 cm row spacing, while Indian Head used a no-till drill with hoe openers on a 30 cm row spacing that side banded fertilizer 2.5 cm beside and 2.5 cm below the seed. A list of specific dates of field operations can be found in Table 4.3.

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Site	Year	Seeding Date	Pre-Seeding Application	Post-Emergence Application	Harvest
Kornon	2015	May 20	May 14	June 15	October 9
Kernen	2016	May 19	May 16	June 13	September 15
Common	2015	May 30	May 28	July 3	September 1
Carinan	2016	June 6	May 30	June 22	September 28
Indian Hood	2015	June 2	May 26/29	June 26	September 29
mutan Head	2016	June 13	June 10		
Goodale	2016	May 25	May 16	June 20	September 20

Table 4.3 Field operation for herbicide screening trial at the Kernen Crop Research Farm (Saskatoon, SK), Goodale Research Farm (Saskatoon, SK), Ian N. Morrison Research Farm (Carman, MB), and Indian Head Research Farm (Indian Head, SK)

The trial was designed as a randomized complete block with four replications, with a total of 18 treatments (Table 4.4). Treatments consisted of seven herbicides not currently registered for flax production including pyroxasulfone (PRE), pyroxasulfone + sulfentrazone (PRE), flumioxazin (PRE), topramezone (POST), topramezone + bromoxynil/MCPA (POST), fluthiacet-methyl (POST), and fluthiacet-methyl + MCPA (POST), all applied at 1X and 2X rates (Table 4.4). As well three registered industry standards were included for comparisons, and all treatments were compared to the untreated check. The industry standards used were MCPA (POST), bromoxynil + MCPA (POST), and sulfentrazone (PRE). The treatments were applied at various timings based on label recommendations. PRE- treatments were applied 5 – 7 days before seeing (Table 4.4). All remaining POST- treatments were applied when the flax was 5 – 10 cm tall (Table 4.4). All plots were hand weeded to keep plots weed-free throughout growing season. Due to unforeseen complications with the application equipment as well as localized weather events at Indian Head in 2016, the POST- treatments were not applied.

Trt. #	Herbicide common name	Herbicide Group	Conc. g/l or g/kg	Rate g a.i. ha ⁻¹	Timing	Surfactant/ Adjuvant	Adjuvant Rate (% v/v)
1	Control	-	-	-	-	-	-
2	Fluthiacet-Methyl	14	216	4	POST	NIS	0.25
3	Fluthiacet-Methyl	14	216	8	POST	NIS	0.25
4	Fluthiacet-Methyl	14	216	4	DOST	NIC	0.25
4	MCPA Ester	4	600	280	F051	1115	0.23
5	Fluthiacet-Methyl	14	216	8	DOST	NIC	0.25
5	MCPA Ester	4	600	280	F051	1115	0.23
6	Pyroxasulfone	15	850	125	PRE	-	-
7	Pyroxasulfone	15	850	250	PRE	-	-
0	Pyroxasulfone	15	850	125	DDE		
0	Sulfentrazone	14	480	140	FKE	-	-
0	Pyroxasulfone	15	850	250	DDE		
9	Sulfentrazone	14	480	280	FKE	-	-
10	Flumioxazin	14	511	107	PRE	-	-
11	Flumioxazin	14	511	214	PRE	-	-
12	Topramazone	27	336	13	POST	Merge ®	0.25
13	Topramazone	27	336	25	POST	Merge ®	0.25
14	Topramazone	27	336	13	DOST	Marga D	0.25
14	Bromoxynil	6	280	280	F051	Meige ®	0.23
15	Topramazone	27	336	25	DOST	Marga®	0.25
15	Bromoxynil	6	280	560	F051	Merge®	0.23
167	Bromoxynil	6	560	200	DOGT		
10-	MCPA ester	4	200	200	PUS1	-	-
17 ^z	MCPA Ester	4	600	415	POST	-	-
18 ^z	Sulfentrazone	14	480	140	PRE	-	-

Table 4.4 Herbicide common name, herbicide group, herbicide concentration, herbicide rate, timing,surfactant/adjuvant used, and adjuvant rate for the flax tolerance trial at Kernen, Goodale, IndianHead, and Carman in 2015 and 2016

^Z Industry Standards

POST = post-emergence

PRE = pre-emergence

NIS = non-ionic surfactant

Merge $\mathbb{B} = 50\%$ solvent / 50% surfactant blend. BASF Canada Inc.

The flax crop was monitored throughout the growing season for any signs of herbicide injury, including stand reduction, chlorosis, and stunting. Crop damage (phytotoxicity) was assessed using the Canadian Weed Science Society (CWSS) phytotoxicity rating scale (Table 4.5) with comparisons made to the untreated check plots in each replication. This rating scale is a percentage based scale ranging from 0 - 100%. In cases where crop tolerance is being evaluated,

the scale is largely focused on the range of 0 - 30%. Ratings between 0 - 10% are considered acceptable, ratings between 15 - 25% considered significant damage, and any rating above 30% is considered commercially unacceptable, or severe crop damage. The various phytotoxicity ratings were done during three time periods: 7 to 14, 21 to 28, and 56+ days after treatment (DAT).

Stand Reduction (%)	Injury	Interval Size	
0-10	Acceptable	2%	
11-30	Unacceptable	5%	
31-100	Severe	10%	

 Table 4.5 CWSS Scale for Percent Crop Injury

Estimate crop injury relative to the untreated check

if zero injury (i.e. stunting, chlorosis, or stand reduction), record as 0

In addition to phytotoxicity ratings, we evaluated flax crop density, crop height, boll counts, yield and thousand seed weight (TSW). Crop density was collected by counting the total number of seedlings on both sides of a meter stick in the front and back of each plot 2 - 3 weeks after crop emergence. Crop height and boll counts were taken once the crop had reached the midlate boll stage. Crop height was taken from 5 random plants in each plot by measuring from the base of the plant to the top. The bolls of 5 random plants were also enumerated in each plot. The crop was harvested with a small plot combine when seeds were ripe and the crop had started to senesce (flax growth stage 12). Yield samples were then used to determine TSW by counting and weighing 1000 individual seeds. At both Saskatoon locations, 1000 individual seeds were counted using a Data Count S 25+ seed counter and then weighed. At Carman, an Agriculex ESC-1 seed counter was used to count 250 seeds, which were then weighed and multiplied by 4. This was done for four samples per plot and then averaged to determine the final TKW.

4.2.3 Data analysis

Data was analyzed with analysis of variance (ANOVA) using the mixed model procedure of SAS 9.3 [PROC MIXED], with pre-planned contrasts used to make specific comparisons of interest (SAS Inst., 2016). The assumptions (homogenous variances and normal distributions of residuals) of the ANOVA were tested using PROC UNIVARIATE with the Shapiro-wilk test (SAS Inst., 2016). Fixed effects in the model were herbicide treatments while site, replication (nested within site) and their interactions with fixed effects were treated as random effects. These

random effects and their interactions with herbicide treatments (fixed effect) were assessed with a COVTEST to determine if site-years could be combined for analysis (SAS Inst., 2016).

For phytoxicity ratings, data that is collected as a proportion relative to a control is often skewed and thus needs to be analyzed using a different distribution in the PROC GLIMMIX procedure (Bowley, 2015). Therefore, a beta distribution with a log-link function was determined to be the best fit model for analysis (Bowley, 2015). To conduct the beta analysis, all data was converted from a percentage into a decimal fraction (Bowley, 2015). As well, because the beta distribution is between 0 and 1, any values equal to 0 were changed to 0.0001 and all values of 1 were changed to 0.9999 to account for the restrictions of the model (Bowley, 2015). Means were separated with an LSD test using a P<0.05 to declare significant differences. Data was back-transformed for presentation of results.

The aim of this experiment was to assess crop tolerance to these herbicides and as such, a DUNNETT'S test was used to compare the means of all treatments to the untreated check. This test was used to compare the means separation for crop population, crop height, boll counts, yield, and thousand seed weight. The calculated minimum significant difference (MSD) was used to determine if the mean of the treatment significantly differed from the untreated check.

4.3 Results

4.3.1 Environmental conditions

In 2015 and 2016, the temperature at each site was near normal when compared to the long-term average (Table 4.6). At Kernen, the precipitation in May was 94% lower and 14% higher than the long-term average in 2015 and 2016, respectively. Carman experienced precipitation events in May 42% and 55% higher than the long-term average in May in 2015 and 2016, respectively. Overall, for the remainder of the growing season, precipitation events were close to the long-term average for Carman. Precipitation events at Indian Head were lower in May and June than the long-term average and normalized over the remainder of the growing season in 2015. In 2016, rainfall was higher than the long-term average for May, July, September, and October. In general, Carman had higher rainfall than Indian Head, which had higher rainfall than Kernen in both site-years.

Location	Year	May	June	July	August	September	October	Avg/Total
					Mean Tem	perature (°C) ·		
	2015	11.3	18.1	20.1	18.6	12.9	7.9	14.8
Kernen	2016	13.7	17.4	18.7	16.9	11.8	2.1	13.4
	Long Term ^z	11.8	16.1	19.0	18.2	12.0	4.4	13.6
Coodela	2016	13.7	17.4	18.7	16.9	11.8	2.1	13.4
Goodale	Long Term ^z	12.1	16.8	19.6	18.6	12.4	5.2	14.1
	2015	10	16.2	18.1	17.0	12.2	6.6	13.4
Indian Head	2016	12.8	16.9	17.6	16.9	12.8	3.9	13.5
	Long Term ^z	10.8	15.8	18.2	17.4	11.5	4.0	13.0
	2015	10.7	17.5	19.9	18.3	15.8	7.2	14.9
Carman	2016	13.6	17.1	19.4	18.4	14.1	6.7	14.9
	Long Term ^z	11.6	17.2	19.4	18.5	13.4	5.4	14.3
					Precipita	tion (mm)		
	2015	6.3	20.2	15.1	58.2	50.8	32.7	253.3
Kernen	2016	45.0	51.0	80.5	66.0	24.1	40.8	285.0
	Long Term ^z	34.6	63.8	54.0	44.0	38.1	18.8	255.2
Goodala	2016	23	59.5	104	70	24.1	40.8	321.4
Goodale	Long Term ^z	34.3	63.3	53.9	44.3	38.1	12.0	245.9
	2015	15.6	38.3	94.6	58.8	67.8	39.0	314.1
Indian Head	2016	74.7	50.2	107.9	21.9	40.5	63.5	358.7
	Long Term ^z	51.7	77.4	63.8	51.2	35.3	24.9	304.3
	2015	98.8	75.3	109.3	47.3	42.0	37.3	410.0
Carman	2016	108.1	95.4	78.7	57.7	64.7	36.5	441.1
Currinun	Long Term ^z	69.6	96.4	78.6	74.8	49.0	43.4	411.8

Table 4.6 Mean monthly temperature (°C) and precipitation data (mm) at the Kernen Crop Research Farm (Saskatoon), Goodale Research Farm (Saskatoon), Indian Head Research Farm, and Carman Research Farm in 2015 and 2016.

^z Long-term normals (1981 to 2010) http://climate.weather.gc.ca/historical_data/search_historic_data_e.html

4.3.2 Crop tolerance

All data was analyzed separately based on site year due to a significant interaction between site-year and herbicide treatment (Table 4.7). Phytotoxicity varied between sites in all years (Figures 4.1, 4.2, and 4.3). At Carman, PRE- treatments caused the greatest damage in both site-years.

Table 4.7 Random effects (year and site) and their interaction with herbicide treatment were assessed using the Wald Z Test (COVTEST). Data was combined over 6 site-years at Saskatoon, SK, Carman, MB, and Indian Head, SK in 2015 and 2016 for the COVTEST analysis. The *P*-values are presented based on the Wald Z Test for crop population (plants m^{-2}), phytotoxicity (%), crop height (cm), bolls (plant⁻¹), yield (kg ha⁻¹), and TSK (g/1000s).

	Population		Phytotoxicity						Unight		Dolla		Viold		TSW	
			7-14 DAT		21-28 DAT		56+ DAT		neight		DOIIS		1 leiu		13 W	
Site	1.37		0.46		0.84		1.11		1.22		0.68		1.41		1.21	
Rep (Site)	2.33	**	2.42	***	1.80	*	1.70	*	1.72	*	1.17		2.44	**	1.17	
Site x Trt	2.86	**	5.99	***	5.63	***	6.17	***	4.01	***	0.73		2.18	*	1.33	
Residual	11.27	***	12.35	***	11.27	***	12.35	***	10.07	***	8.72	***	11.27	***	10.07	***

Trt = Treatment, DAT = Days after treatment, TSW = Thousand Seed Weight

*, **, *** indicates a significant difference atp<0.05, p<0.01, and p<0.001, respectively.

Treatments containing pyroxasulfone, pyroxasulfone + sulfentrazone, and flumioxazin caused unacceptable or severe crop damage (greater than the 30% threshold) 7 – 14 days after treatment (DAT) (Figure 4.1). Since these are soil applied herbicides, the primary form of injury was stand reduction, delayed emergence or stunting. Initial phytotoxicity ratings for pyroxasulfone (1X and 2X) were 35% and 69% respectively in 2015. However, damage was less severe in 2016 with pyroxasulfone causing 9% and 29% crop injury at the 1X and 2X rate, respectively. Applications of pyroxasulfone+sulfentrazone at both rates resulted in 42% and 73% crop damage in 2015. Again, damage was less severe in 2016 with treatments of pyroxasulfone+sulfentrazone resulting in injury ratings of 23% and 31% at the 1X and 2X rates respectively. Flumoxazin application resulted in 89% and 98% crop injury at the 1X and 2X rates rates, respectively. Unlike treatments containing pyroxasulfone, damage caused by flumioxazin was consistently severe, causing 84% and 92% at the 1X and 2X, rate respectively in 2016. In comparison to the industry standards, sulfentrazone applied alone caused a small amount of crop injury (<4%) 7 – 14 DAT in both site-years at Carman.



Figure 4.1 The mean rating of treatments on flax based on visual phytotoxicity ratings (0 - 100% scale) 7 – 14 days after treatment. Values derived from beta-analysis of visual ratings at individual site-years. Asterisk (*) represent a significant differentiation (P<0.05) from the untreated check (Phytotoxicity rating = 0). Error bars represent standard error of mean. The red line indicates 10% crop injury. The blue line represented 30% crop injury. Any rating below 10% is considered acceptable crop damage, between 10- 30% is considered unacceptable injury. Ratings above 30% are ranked as severe crop injury. Pyrox, PyrSft, Flum, and SFT applied PRE-planting. All remaining treatments applied POST. Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.
While treatments containing pyroxasulfone and flumioxazin caused severe initial injury at Carman in 2015 and 2016, not all products caused severe damage at the Saskatchewan locations (Figure 4.1). Treatments containing pyroxasulfone did not cause any significant initial injury at Kernen or Goodale in 2015 and 2016. Injury at Indian Head was similar to that of Carman with injury ratings ranging between 14 – 40% in 2015. Treatments containing flumioxazin had variable effects at the Saskatchewan locations. While not rated as severe, damage at Indian Head was 14% and 25%, respectively for the 1X and 2X rate. Flumioxazin appeared to cause no damage (0-2%) 7 DAT at Kernen in 2015 and Goodale in 2016. However, unacceptable damage in 2016 was caused by both rates of flumioxazin (12% and 17% respectively). Similar to Carman, sulfentrazone caused almost no crop injury (0-1%) at both Saskatoon locations in all years. By comparison, sulfentrazone caused similar injury at Indian Head as flumioxazin when applied at the 2X rate 7DAT (28%). However, injury at this location 7 DAT was more severe than the other site-years.

While some of the PRE- products caused unacceptable or severe injury initially, several POST- products showed acceptable crop safety at various sites in both years (Figure 4.1). Fluthiacet-methyl and topramezone did not cause any unacceptable crop injury 7 DAT when applied alone at Carman in 2015 and 2016, Goodale, and Kernen in 2015. In 2016, all treatments containing fluthiacet-methyl caused unacceptable crop injury that ranged between 15 and 61% 7 DAT at Kernen. Topramezone still had excellent crop safety (<10% crop injury) when applied alone at Kernen in 2016. Treatments that combined MCPA and fluthiacet-methyl caused both unacceptable and severe crop injury at all locations except Carman in 2015 where injury ratings 7 DAT were less than 4%. Crop damage at other locations ranged from 14 to 18% at Carman in 2016, 14 to 15% at Indian Head, 47 to 55% at Goodale, 27 to 34% and 52 to 61% at Kernen in 2015 and 2016, respectively. Damage caused by mixing topramezone and bromoxynil was not consistent among site-years. While no significant crop injury was observed 7 DAT at Carman and Kernen in 2015, unacceptable crop injury ranged from 14 to 81% at Carman, Kernen, and Goodale in 2016 and Indian Head in 2015. POST-emergence industry standard treatments, like several other treatments, had variable effects in different site-years. These treatments did not cause significant crop injury at Carman and Kernen in 2015. However unacceptable crop injury that ranged from 13 to 27% was observed at all other site-years 7 DAT.

By 21 to 28 DAT, the initial damage at all sites began to subside below the severe damage threshold for most treatments (Figure 4.2). Nevertheless, PRE- treatments of pyroxasulfone (1X and 2X rate), pyroxasulfone + sulfrentrazone (1X and 2X rate), and flumioxazin (1X and 2X rate) continued to cause severe crop damage at Carman 21 DAT in 2015 resulting in 27%, 41%, 32%, 53%, 88%, and 92% crop damage respectively. Conversely, crop recovery was more rapid in 2016 with only flumioxazin treatments continuing to cause severe crop damage (66 and 93% at the 1X and 2X rate respectively) 21 DAT (Figure 4.2). All other PRE- treatments had recovered to below the acceptable damage threshold. Similar crop injury was observed at Kernen in 2016 with pyroxasulfone (2X rate), pyroxasulfone + sulfentrazone (2X rate), and flumioxazin (1X and 2X rates) continuing to cause unacceptable or severe crop damage that ranged from 13 to 50% crop injury by 21 DAT. At the remaining sites (Kernen 2015, Indian Head 2015, and Goodale 2016) crop injury had subsided to below the acceptable threshold (i.e. below 10%) for all treatments containing pyroxasulfone and flumioxazin.



Figure 4.2 The mean rating of treatments on flax based on visual phytotoxicity ratings (0 - 100% scale) 21 - 28 days after treatment. Values derived from beta-analysis of visual ratings at individual site-years. Asterisk (*) represent a significant differentiation (P<0.05) from the untreated check (Phytotoxicity rating = 0). Error bars represent standard error of mean. The red line indicates 10% crop injury. The blue line represented 30% crop injury. Any rating below 10% is considered acceptable crop damage, between 10- 30% is considered unacceptable injury. Ratings above 30% are ranked as severe crop injury. Pyrox, PyrSft, Flum, and SFT applied PRE-planting. All remaining treatments applied POST. Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.

Similar recovery was observed 21 to 28 DAT for POST- treatments at all sites in all years (Figure 4.2). However, treatments containing fluthiacet-methyl + MCPA continued to cause unacceptable or severe injury at Indian Head, Goodale, and Kernen. Crop damage ranged from 13 to 60% across these site-years. Similarly, the combination of topramezone + bromoxynil continued to cause unacceptable crop damage at Goodale, Indian Head, and Carman in 2016. However, crop injury had subsided from the initial observations. By 21 DAT, crop injury ratings for all other treatments were below 10% at all site-years.

The trend of significant crop recovery continued past 56 DAT (Figure 4.3). At Kernen Goodale, and Carman all POST- treatments received acceptable crop injury ratings by 56 DAT. Damage from treatments containing fluthiacet-methyl + MCPA had subsided from 52% at 7 DAT to 10% by 56 DAT at Kernen in 2016, 34% 7 DAT to 6% by 56 DAT at Kernen in 2015, and 55% to 6% at Goodale in 2016. At Carman, the crop recovered from 18% damage initially, to only 2-4% crop injury by 56 DAT. The rates of recovery for topramezone + bromoxynil were equally as impressive in all site-years. At locations where topramezone + bromoxynil caused significant damage, recovery from initial damage ranged from 65 - 95%. Damage also subsided at Indian Head. However, crop damage from fluthiacet-methyl + MCPA and topramezone + bromoxynil (2X rate) was still found to range between 10 - 14% by 56 DAT. Similar trends were observed with the PRE- treatments. At all site-years, excluding Carman (2015, 2016), ratings for all PRE- treatments were below the acceptable threshold. The crop recovered an impressive 95% at Indian Head, 70% at Goodale, 32% at Kernen in 2015, and 95% at Kernen in 2016 to treatments containing flumioxazin. Recovery at Carman was not as dramatic. By 56 DAT the crop was still exhibiting severe injury which ranged between 26 to 82% as a result of flumioxazin applications.



Figure 4.3 The mean rating of treatments on flax based on visual phytotoxicity ratings (0 - 100% scale) 56+ days after treatment. Values derived from beta-analysis of visual ratings at individual site-years. Asterisk (*) represent a significant differentiation (P<0.05) from the untreated check (Phytotoxicity rating = 0). Error bars represent standard error of mean. The red line indicates 10% crop injury. The blue line represented 30% crop injury. Any rating below 10% is considered acceptable crop damage, between 10- 30% is considered unacceptable injury. Ratings above 30% are ranked as severe crop injury. Pyrox, PyrSft, Flum, and SFT applied PRE-planting. All remaining treatments applied POST. Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx = Tompramezone + Bromoxynil, BmxMCP = Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone

4.3.3 Crop development, yield, and quality

Crop density was not inhibited by any treatment at Indian Head in 2015 or Kernen in 2015 and 2016 (Figure 4.4). In turn, no treatment significantly reduced crop height at Kernen in 2015 or 2016 (Figure 4.5). However, applications of flumioxazin and pyroxasulfone increased crop height by more than 5 cm (Figure 4.5). While a 2X application of topramezone + bromoxynil significantly reduced crop density by 180 plants m⁻² at Goodale (Figure 4.4), this reduction in crop density did not influence crop height (Figure 4.5). Measurements for height were not taken at Indian Head in 2015. No treatment affected bolls plant ⁻¹ at Kernen in 2015 or Goodale in 2016 (Figure 4.6). However, pyroxasulfone (2X rate) and flumioxazon (2X rate) significantly increased the number of bolls plant⁻¹ at Kernen in 2016 (Figure 4.6). Measurements for bolls counts were not conducted at Indian Head or Carman.



Figure 4.4 The effect of herbicide treatment on crop density (plants m^{-2}). A Dunnett's comparison of the mean crop population of each treatment to the untreated check at each individual site. An asterisk (*) above bars indicate a significant deviation from the untreated check (P<0.05) by more than the indicated MSD. MSD = minimum significant difference. Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.



Figure 4.5 A Dunnett's comparison of the effect of treatment on crop height in comparison to the untreated check at each site. Values derived from measuring the height of 5 random plants in each plot, and averaging the values. An asterisk (*) above a bar indicates a significant deviation from the untreated check (p<0.05). Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.



Figure 4.6 Dunnett's comparison of the effect of all treatments on average number of bolls plant⁻¹ with the untreated check. Values were derived from counting the number of bolls on 5 random plants per plot and averaged. An asterisk (*) above the bar indicates a significant deviation from the mean of the untreated check (p<0.05). Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.

Despite any initial phytotoxic damage, reductions in crop density, height or bolls, no treatment significantly affected crop yields at Kernen (both years) or Goodale (Figure 4.7). While topramezone + bromoxynil (2X rate) reduced yield by 407 kg ha⁻¹ at Goodale, this reduction was not statistically significant when compared to the untreated check (MSD = 459 kg ha⁻¹). Yield at Indian Head in 2015 was more responsive to treatments (Figure 4.7). All treatments demonstrated a trend for reduced crop yield, while half of the treatments resulted in yield that differed from the untreated check (Figure 4.7). Pyroxasulfone + sulfentrazone (2X rate) resulted in the greatest (542 kg ha⁻¹) yield reduction (Figure 4.7).

At Carman, several PRE- products significantly reduced crop density. Pyroxasulfone (2X rate) and pyroxasulfone + sulfentrazone (1X rate) reduced plant populations by 246 and 186 plants m⁻² respectively in 2015 (Figure 4.4). Furthermore pyroxasulfone + sulfentrazone applied at the 2X rate as well as flumioxazin applied at the 1X and 2X rate reduced crop density by more than 60 plants m⁻² in both site-years (Figure 4.4). Flumioxazin was also found to reduce crop height by 17 cm in 2015 and 15 cm in 2016 (Figure 4.5). This reduction in crop population and crop height translated into a yield reduction of 218 kg ha⁻¹ in 2015 (Figure 4.7). While topramezone + bromoxynil did not affect crop density, it reduced crop height by 7 cm in 2015 (Figure 4.5). However, this reduction in height did not result in a significant yield reduction (Figure 4.7)



Figure 4.7 A Dunnett's comparison of the treatment effect on yield in comparisoin to the untreated check. Values were derived from yield of the entire plot. An asterisk (*) above the bar indicates a significant deviation from the untreated check (p<0.05). Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.

Two POST- products were also found to reduce crop density at the Manitoba location in 2016. Fluthiacet-methyl + MCPA applied at the 2X rate and bromoxynil + MCPA both reduced plant populations in 2016 on average by 54 plants m^{-2} (Figure 4.4). As well, fluthiacet-methyl + MCPA reduced crop height by 4cm (Figure 4.5). Measurements for boll per plant were not conducted at Carman in 2015 or 2016. While there were varied responses to treatments in regard to plant population and crop height, no treatment impacted yield in 2016 at the Carman location and overall yields were quite low in comparison to other site-years (Figure 4.7). For instance, the yield of the untreated check at Carman in 2015 (255 kg ha⁻¹) was 88% lower than the untreated check at Kernen in 2015 (2050 kg ha⁻¹), and 84% lower than Indian Head (1627 kg ha⁻¹). Similarly, in 2016 the yield of the untreated check (322 kg ha⁻¹) at Carman was 86% lower than the untreated check at Kernen (2243 kg ha⁻¹) and 85% lower than the yield at Goodale (2088 kg ha⁻¹).

Thousand seed weight, like several other variables, was relatively unresponsive to treatments in most site-years (Figure 4.8). However, TSW was affected at Indian Head in 2015 by applications of topramezone, flumioxazin, and fluthiacet-methyl + MCPA wherein these treatments reduced TSW by more than 0.168 g 1000 seeds⁻¹ (Figure 4.8).



Figure 4.8 A Dunnett's comparison of the treatment effect on flax thousand seed weight in comparison to the untreated check. An asterisk (*) above the bar indicates a significant deviation from the mean of the untreated check (p<0.05). Fluth = Fluthiacet-methyl, FluMCP = Fluthiacet-methyl + MCPA, Pyrox = Pyroxasulfone, PyrSft = Pyroxasulfone + Sulfentrazone, Flum = Flumioxazin, Top = Topramezone, TopBx= Tompramezone + Bromoxynil, BmxMCP= Bromoxynil + MCPA, MCPA = MCPA, SFT = Sulfentrazone.

4.4 Discussion

When evaluating new herbicides, crop tolerance is extremely important. Determining how certain herbicides affect crop density, growth, development, and yield over the course of a growing season is the first step to integrating novel herbicides into production. Furthermore, assessing how these herbicides behave across multiple environments is imperative to determine the consistency of herbicides. In this study, the damage caused by the tested products was transient and did not significantly reduce crop yield when site data were combined (data not shown), although there were significant differences between the treatment effects at various sites. Collectively, these differences were likely a result of variations in local environmental conditions, with spring moisture and soil moisture largely influencing the activity of certain herbicides. Specifically, treatments containing pyroxasulfone and flumioxazin caused greater crop phytotoxicity when spring moisture was high.

Flax appears to be tolerant to several novel herbicides, and this appeared consistent across multiple environments. Specifically, fluthiacet-methyl, pyroxasulfone, and topramezone present the greatest potential to be utilized in flax. Fluthiacet-methyl did not significantly reduce crop population, crop height, boll counts, crop yield or TSW (Figures 4.4, 4.5, 4.6, 4.7, 4.8). Tolerance of fluthiacet-methyl is usually a function of differential uptake, translocation, and degradation of the herbicide (Shimizu et al., 1995). Crops such as corn, soybeans, and sorghum have been shown to have impressive tolerance to this peroxidising herbicide (Shimizu et al., 1995; Reddy et al., 2014). Nevertheless, there is a lack of current literature on crop tolerance and spectrum of weeds controlled by fluthiacet-methyl (Reddy et al., 2014). As well no studies were found evaluating the effect of fluthiacet-methyl on flax growth and development, reinforcing the importance of this study.

The efficacy of fluthiacet-methyl can be enhanced through the addition of other herbicides into a tank-mix. In sorghum, weed control with fluthiacet-methyl was improved with the addition of 2,4-D (Reddy et al., 2014). Gaine et al., (2015) also found tank mixes of fluthiacet-methyl and mesotrione (Group 27) to provide effective control of glyphosate-resistant kochia and common waterhemp (*Amaranthus tuberculatus* Moq. J.D. Sauer) when plants were less than 10 cm tall. Gaine et al. (2015) also referenced several studies that found fluthiacetmethyl to have synergistic effects with other herbicides, such as 2,4-D and bromoxynil. These

tank mixes were found to improve the control of several broadleaved weed species such as common lambsquarters, velvetleaf, and common ragweed (Ambrosia artemisiifolia L.) (Gaine et al., 2015). While these tank mixes can improve weed control, they can also affect crop growth and development. The addition of 2,4-D was also found to increase crop phytotoxicity, which negatively affected crop yield (Reddy et al., 2014). This was similar to the results in our study where adding MCPA to a tank mix with fluthiacet-methyl resulted in significant crop injury, which in some instances further impeded normal growth and development of the crop. Similar increases in crop phytotoxicity have also been observed when MCPA was combined with other herbicides. In winter wheat tank mixes of Dicamba + MCPA + mesocarp caused a notable crop injury which in turn resulted in a reduction in crop height and crop yield (Sikkema et al., 2007). This result contrasts observations from our study; tank mixing MCPA with fluthiacet-methyl did not result in a significant yield reduction. Nevertheless, not all tank mixes with MCPA caused substantial crop damage or yield reductions in winter wheat (Sikkema et al., 2007). Specifically, applications of bromoxynil + MCPA had acceptable crop safety in all three winter wheat varieties and had no negative effect on crop height or yield. In contrast, various studies show that the addition of synthetic auxin herbicides (i.e. MCPA) can have antagonistic effects on the efficacy of herbicides such as imazamethabenz, dichlorfop methyl, and diclofop-methyl (Qureshi and Vanden Born, 1979; O'Sullivan et al., 1977; Liu et al., 1995). This antagonistic effect can reduce the phytotoxic effects these herbicides can have on weeds. However, Qureshi and Vanden Born (1979) did not find MCPA reduced the phytotoxic effects of diclofop-methyl on barley. As such, even though MCPA is registered for use in flax, combining MCPA with fluthiacet-methyl is not recommended due to the severe foliar injury it can cause.

Similar to fluthiacet-methyl topramezone showed promise to be used as an in-crop herbicide for flax when applied alone. Treatments containing topramezone often produced efficacy ratings lower than the industry standards, showing that topramezone is as safe as products currently registered for use in the crop. However, when combined with bromoxynil crop injury was observed, although this injury was not consistent across site-years, and the crop recovered with no significant reduction in yield or TSW. Additive interactions between Group 27 and Group 6 herbicides have also been observed between another Group 27 herbicide (pyrosulfotle) and several Photosystem II (PSII) inhibiting herbicides (Group 6) (Freigang et al., 2008). This synergistic relationship between Group 27 products and several Group 6 herbicides,

as explained by Freigang et al. (2008), supports the observations in our study. Freigang et al. (2008) further suggest that when using a Group 27 and a Group 6, a lower rate of Group 6 herbicide can be used. However, in our study we did not reduce the rate of bromoxynil when mixing it with topramezone and as such, the synergistic relationship between these two herbicides can explain why increased phytotoxicity was observed for treatments containing these products.

Synergistic interactions between Group 27 and 6 herbicides are complex, but have been explained. Group 27 herbicides interrupt the carotenoid biosynthetic pathway, which in turn disrupts the production of plastoquinones (PQ) (Freigang et al., 2008). PSII inhibiting herbicides prevent PQ from binding to the D1 protein of PSII by binding to the D1 site before PQ can. Therefore, when Group 27 herbicides reduce the production of PQ, there is less of that substrate present for Group 6 herbicides to compete for binding with the D1 protein. This results in an influx of reactive oxygen species in the plant which then cause severe cellular damage and eventual plant death (Freigang et al., 2008). Furthermore, this additive interaction has been observed between other HPPD and PSII inhibitors. Control of common water hemp was substantially improved through combining mesotrione (Group 27) and bromoxynil (Gaine et al., 2015).

Pyroxasulfone was one of the PRE-plant herbicides that did not cause severe crop injury in all site-years. However, flax at Carman (2015) did experience severe crop injury from this herbicide, which in turn produced significant reductions in crop density. Despite any initial injury or reduction in crop population, pyroxasulfone did not significantly reduce crop height, yield, or TSW at most sites. While pyroxasulfone can control a broad range of weed species, the efficacy of pyroxasulfone can be largely influenced by site-year and environmental conditions (Tidemann et al., 2014). For example, pyroxasulfone had the greatest control of Italian ryegrass during wet growing conditions, whereas control was more variable during drier growing years (Hulting et al., 2012). As well, fields with higher soil organic matter require a higher dose of pyroxasulfone to achieve effective weed control which can contribute to the variability in weed control between site-years (King and Garcia, 2008; Nurse et al., 2011). Unlike the addition of MCPA to fluthiacet-methyl or bromoxynil to topramezone, the addition of sulfentrazone to pyroxasulfone did not increase crop injury in our study. Similar effects were observed by

Niekamp et al. (1999) in soybean where tank mixes of sulfentrazone + chlorimuron and sulfentrazone + imazaquin provided greater control of velvetleaf, common ragweed, and common cocklebur than sulfentrazone alone. While these products did provide effective weed control, some crop injury was initially observed (11 - 19%).

Of all the products evaluated, flumioxazin showed the least benefit for use in flax. It caused severe crop injury at multiple sites and in some cases eliminated more than 90% of flax seedlings (Figure 4.1). Despite the severe injury observed, the only site that experienced reductions in crop populations or yield was the Carman (2015, 2016) location (Figure 4.4). Severe, unacceptable injury was observed when there was early spring moisture at that location. Flumioxazin efficacy has been shown to be highly influenced by soil organic matter, soil texture, and soil moisture (Sebastian et al., 2017). The herbicide binds tightly to soil colloids and will not readily disassociate without adequate soil moisture (Sebastian et al., 2017; Alister et al., 2008). Hence, in years of drought or low soil moisture, herbicides like flumioxazin will remain inert in the soil, which in turn results in little crop damage but also no weed control. Furthermore, flumioxazin has been found to be readily absorbed in soils with a high clay and organic matter content (Alister et al., 2008). The soil at Carman, MB had an organic matter content of 6% and consisted of 31% clay (Table 4.1). These soil characteristics paired with a high amount of soil moisture in the months of May and June (Table 4.6) could have created an environment with a high level of herbicide persistence in the soil, which would explain why flumioxazin and other PRE-herbicides caused such severe damage at Carman compared to other locations.

Environmental conditions can be particularly influential on the behaviour of certain herbicides, particularly PRE- herbicides. Under dry conditions, efficacy of PRE – herbicides declines (Jursík et al., 2015). While the majority of PRE- herbicides require moisture for activation and plant uptake (Stewart et al., 2010) under very high moisture conditions, there is also a risk of crop injury occurring from soil applied herbicides (Jursík et al., 2015). As well, the efficacy of acetochlor, dimethenamid, and oxyfluorfen was not influenced by changes in soil moisture conditions (Jursík et al., 2015). Conversely, cloransulam-methyl and flumetsulam/*S*-metolachlor require precipitation soon after application to achieve adequate herbicide activation (Stewart et al., 2010). The differences in environmental conditions, as well as soil conditions,

can help to explain why in our study PRE- herbicides such as pyroxasulfone, pyroxasulfone + sulfentrazone, and flumioxazin significantly reduced crop density, particularity when soil conditions were conducive for high herbicidal activity.

Fluthiacet-methyl, pyroxasulfone, and topramezone not only offer more diversity for chemical weed control. Products such as fluthiacet-methyl offer the opportunity to control weedy species that have developed resistance (Sarangi et al., 2015). Tank-mixes of fluthiacet-methyl + mesotrione have shown promise to control seedlings of glyphosate resistant waterhemp and kochia (Ganie et al., 2015). As well, pyroxasulfone has been shown to selectively control herbicide resistant ridged ryegrass (Lolium rigidum Gaud.) populations in various crop types in Australia (Walsh et al., 2011). Improved weed control in soybean has also been observed through tank-mixing pyroxasulfone + sulfentrazone (Belfry et al., 2015). Applied alone, pyroxasulfone and sulfentrazone did not adequately control certain weed species for the duration of the growing season (Belfry et al., 2015). However, when paired together pyroxasulfone + sulfentrazone had a synergistic effect on weed control in soybean across multiple soil types (Belfry et al., 2015). Furthermore, while pyroxasulfone, sulfentrazone, and flumioxazin controlled HPPD-resistant waterhemp populations, complete control of the weed was not achieved by any herbicide treatment (Hausman et al., 2013). This demonstrates the need for multiple weed control methods to manage problematic weeds. Nevertheless, these studies show novel modes of action can add a new level of chemical diversity for weed control in multiple crops. Furthermore, the tolerance that flax has exhibited to pyroxasulfone, topramezone, and fluthiacet-methyl combined with the capabilities of these groups to control herbicide resistant populations presents great promise moving forward in improving weed management in flax.

Utilizing these three modes of action will help to reduce the selection pressure for the development of herbicide resistance in at-risk weed species such as wild oat and green foxtail. Given the above, fluthiacet-methyl, pyroxasulfone, and topramezone can all be utilized in flax production for both PRE- and POST- weed control. Flumioxazin should be avoided by producers because of the severe crop damage it can cause during growing seasons with high moisture events. Pyroxasulfone can be safely tank mixed with sulfentrazone which then can add diversity to the herbicides used for PRE- weed management. However, tank mixing fluthiacet-methyl and

topramezone with other registered herbicides did increase crop phytotoxicity. Thus, these POSTtank mixes should be avoided until greater crop safety can be achieved.

4.5 Conclusion

Flax showed impressive tolerance to most herbicide treatments, with the exception of flumioxazin. In years with high spring moisture, flumioxazin consistently caused severe initial damage. While treatments containing fluthiacet-methyl did not cause severe crop injury, combining this herbicide with MCPA did cause initial, transient injury to the crop. Treatments containing topramezone and bromoxynil were found to be generally safe. Environmental conditions at the time of application could have resulted in the higher phytotoxicity observed at some site-years. Flax was also found to be tolerant to pyroxasulfone applied alone and in combination with sulfentrazone. Overall, early injury caused by these herbicides did not translate into any reduction in yield or quality at the majority of sites. Nevertheless, the severity of injury caused by flumioxazin makes it the least promising product for registration in flax. Therefore, acceptable crop tolerance was achieved when fluthiacet-methyl and topramezone were applied alone, and when pyroxasulfone was applied alone or in combination with sulfentrazone.

5.0 General Discussion

The goal of this thesis was to evaluate different approaches to help improve weed management in flax (*Linum usitatissimum* L.), including new herbicide options and investigating integrated weed management strategies. By combining the results of these two avenues of research, improved methods for weed control in flax were uncovered. We hypothesized that the results of this research would help to increase the competitive ability of flax and improve its agronomic value to producers in the prairies. Because we identified novel herbicides for flax, and found improved weed management tactics, we failed to reject our null hypothesis; we have, in fact, identified several management practices which can help to improve weed management in flax.

5.1 Cultural weed control in flax

The results of this research show that adjustments in seeding date, seeding rate, crop height, and herbicide rate all influence crop-weed competition in flax and ultimately, flax yield. As such, based on these results, we accept the first hypothesis that seeding a tall cultivar (CDC Sorrel) at a high seeding rate, in early May, with an in-crop herbicide application will improve the competitive ability of flax. By adjusting these four production factors, greater crop establishment was achieved, which in turn increased flax biomass production, reduced weed biomass, and ultimately increased crop yield. Hence, we successfully took four factors of integrated weed management and implemented them in such a way to improve both the competitive ability and seed yield of flax.

Our IWM study combined a total of 10 site-years of data across the three Prairie Provinces of Canada and examined several components that are central to IWM methodologies. The goal of this research was to improve the competitive ability of flax, reduce selection pressure on weedy species, and vary weed control methods by adjusting several different agronomic factors. By seeding at different dates, producers add diversity to planting patterns, which can benefit crops competitive ability. For weeds like wild oat (*Avena fatua* L.), seeding at different times can have inherently negative effects on the competitive ability of weeds (O'Donovan et al.,

2007). This is important as early emerging weeds can cause the greatest amount of crop yield loss (O'Donovan et al., 2007; Swanton et al., 2008). Seeding later can give weedy species an advantage over the crop if they are not properly controlled before planting. In a poorly competing crop like flax, this early and intense competition for resources can have an inherently negative effect on yield.

Seeding rate is another component influencing crop stand and therefore crop-weed competition. Adjusting seeding rates to develop a uniform crop establishment helps to create more aboveground biomass which results in a crop better able to compete for light, water and nutrients (Swanton and Weise, 1991; Swanton and Murphy, 1996). However, flax can be limited in the amount of light it intercepts due to a low leaf area index (Zhang, 2013). Increasing the seeding rate in flax helped to increase crop growth rate and nutrient acquisition as well as reducing weed biomass (Stevenson and Wright, 1996). As such increasing seeding rate could allow for greater capture of light by flax and potentially enable the crop to out-compete neighbouring weeds.

Along with adding variation to seeding dates and seeding rates, utilizing competitive cultivars is central to effective weed management systems. Characteristics of competitive cultivars typically include early emergence, early-season vigour, increased crop height, and rapid canopy development (Bajwa et al., 2017; Swanton and Weise, 1991; Swanton and Murphy, 1996; O'Donovan et al., 2007). Combining the use of competitive crops/cultivars at optimum seeding rates, so as to ensure early and vigorous emergence, can both reduce need for herbicide application as well as improve herbicide efficacy (O'Donovan et al., 2007). Therefore, adopting multiple practices that have an additive effect can help to improve the competitive ability of flax. By improving the competitive ability of the crop, the effectiveness of weed management systems is improved as well, which ultimately helps to add longevity to production systems.

5.2 New herbicide options for flax

It was hypothesized that flax would have acceptable tolerance to several novel modes of action, thus presenting new options for both PRE- and POST- weed control. The results of Experiment 2 were promising for several of the herbicides evaluated. Regardless of initial injury, crop development and yield were not impeded by treatments containing fluthiacet-methyl, pyroxasulfone, sulfentrazone, and topramezone. The only herbicide that caused severe and

unacceptable injury on the crop was flumioxazin, so it was therefore deemed unsuitable for use in flax. Hence, we partially accepted our second hypothesis that flax was able to tolerate four of the seven novel modes of action.

Protoporphyrinogen oxidase inhibitors (Group 14), very long chain fatty acid inhibitors (Group 15), and hydroxphenylpyruvate dioxygenase inhibitors (Group 27) are modes of action that are not widely registered for safe use in Saskatchewan or the other prairie provinces (Brook and Cutts, 2016; Government of Saskatchewan, 2016; Government of Manitoba, 2016). They encompass both PRE- and POST- emergent formulations that help to vary the timing of weed control. As well, these herbicides are largely registered for use in the United States in crops such as corn or soybean. Hence, their use in flax in Canada can be considered novel. Furthermore, the selectivity of these herbicides is largely associated with the ability of the crop to metabolize the active ingredients and withstand any injury the herbicide may cause. Flax can recover from main stem damage through the production of basal branches (Diederichsen and Richards, 2003). Therefore, this ability to outgrow damage could explain why flax is able to tolerate these modes of action. Nonetheless, the characteristics of these herbicides and the ability of flax to tolerate them renders fluthiacet-methyl, pyroxasulfone, and topramezeone promising candidates for use in flax.

Controlling weeds in the field is inherently more complicated than solely using herbicides, as communities of weed species will adapt to the amount and duration of selection pressure they are subjected to (Derksen et al., 2002). While chemical weed control plays an effective role in minimizing competition from weeds, this singular method of management does not provide longevity to weed management systems due to herbicide resistant (HR) weeds. Nevertheless, while herbicides can cause intense selection pressure on weed populations, cultural control methods can be equally as limiting when they remain the sole means of weed control. For instance, intense hand-weeding barnyard-grass (*Echinochola crus-galli* L. Beauv.) in rice (*Oryza sativa* L.) selected for individuals that mimicked the crop; hence a population of individuals that resisted control (Harker and O'Donovan, 2013). Therefore, a balance must be struck between the utilization of both chemical and cultural weed controls to establish an effective IWM system in flax.

The evolution of herbicide resistance will make control of these weedy species more difficult. While there is little evidence that herbicide resistance is positively associated with improved plant fitness (Vila-Aiub et al., 2009), HR weeds will have a distinct advantage in production systems where only singular methods of weed control are implemented. Therefore, careful management of these HR species is needed if weed management is to be improved in flax production. Tank mixing herbicides as well as utilizing novel modes of action are two recommended strategies when dealing with problematic weed populations (Beckie and Harker, 2017; Beckie, 2011).

5.3 Management Implications

Producers in Canada and around the world are facing a problem with HR-weeds that cannot be solved through the identification of new herbicidal modes of action. However, this problem can be combated through the implementation of diversified weed management strategies (Harker, 2013; Beckie and Harker, 2017). The underlying message of this thesis is that no one factor will ultimately improve weed management in flax. However, the combination of several factors can. Herbicides are an extremely effective tool for controlling weeds. Nevertheless, there is a need for a change in producer mindset; herbicides are an option for weed control, they are not the only option available. This research has shown that the combination of several cultural factors with novel modes of action can effectively improve the competitive ability of flax.

Minimizing early season competition from weeds is an important component to improving overall management of weeds throughout the growing season. Based on the results of this study, producers would be advised to seed flax in early May and to increase seeding rates. In addition, selecting cultivars that have exhibited early season vigour will help to encourage rapid germination and establishment at early seeding dates. By doing so, producers will increase the likeliness of developing a thick, even plant stand that is better able to compete with weeds in the spring. Moreover, using a PRE- herbicide such as pyroxasulfone or sulfentrazone can help to reduce early season competition from weeds and allow the crop to emerge and establish with minimal competition from weeds.

Combining cultural controls with herbicides is important to improve the effectiveness and longevity of weed management systems. Utilizing these novel herbicide groups helps to reduce

selection pressure on weeds which in turn helps to combat the evolution of herbicide resistance. For instance, using pyroxasulfone for PRE- seeding weed control and topramezone or fluthiacetmethyl for POST- emergence weed management can help to prolong the weed free period and minimize the detrimental effects weeds can have on flax growth and development throughout the growing season. However, before topramezone, pyroxasulfone, and fluthiacet-methyl are used by producers, a better understanding of what weeds they control and at what stage is needed. Additionally, these herbicides should be either tank mixed or used in rotation with other herbicides to ensure the durability and efficacy of these modes of action. However, safe tank mixes need to be identified as our research has shown crop damage can be severe when combining topramezone and fluthiacet-methyl with registered industry standards. Therefore, while these herbicides are promising options for flax producers, more research is needed to better understand the spectrum of weed control topramezone, pyroxasulfone, and fluthiacet-methyl have, and how to utilize these new herbicides effectively, as well as responsibly.

If adjustments in seeding dates, seeding rates, herbicide rates, and crop height/varietal choice can benefit a poorly competing crop such as flax, then these principles may also be applied to other crop species thus, diversifying current weed management systems. By developing systems that utilize multiple weed management techniques, the onset of HR weed species can potentially be delayed, adding longevity and value to these weed management strategies. Although flax has not received the same attention regarding IWM research, the concepts encompassed in IWM methodologies have been well researched in numerous crop types (Blackshaw et al., 1999; Fahad et al., 2015; Harker, 2013; Harker and O'Donovan, 2013; Harker et al., 2003; Harker et al., 2016; Jha et al., 2017; Lemerle et al., 2016; Liebman and Davis, 2000; Schreiber, 1992; Swanton and Weise, 1991; Swanton and Murphy, 1996). Therefore, despite the lack of research in flax, IWM does offer an opportunity to improve the competitive ability and overall productivity of flax.

5.4 Future Research

This thesis has shown that several cultural factors can be combined to improve the competitive ability of flax. We used two different flax cultivars because of their differences in height. However, the differences in competitive ability that was observed between CDC Sorrel and Prairie Grande is likely due to differences in vigour rather than differences in height. Hence,

there is a need to better understand and identify what is causing the differences in competitive ability between flax cultivars commonly grown in the prairies. Moving forward it will be important to test these differences in competitive ability among cultivated flax varieties. As well, through better understanding of the differences in vigour between cultivars, the actual effect of flax height on crop-weed competition can be more clearly understood.

Determining the differences in competitive ability between flax cultivars is important if we hope to add longevity to weed management strategies. However, defining the critical period of weed control in flax is equally as important if IWM systems are to be implemented to have maximum benefit. The critical period of weed control is defined by two components: a) the length of time the crop must remain weed-free to prevent crop yield loss, b) the amount of time weeds can remain in the crop before they begin to interfere with crop yield (Swanton and Weise, 1991; Swanton and Murphy, 1996). This time interval is defined based on the growth stage of the crop and can help producers make decisions on when to make herbicide applications or when to implement other weed control measures (Swanton and Murphy, 1996). For example, in white bean (*Phaseolus vulgaris* L.) the CPWC occurs between the second trifoliate and the first flower growth stages. The CPWC has not been defined in flax. Hence if IWM systems are to be implemented to have maximum benefit, identifying the time that flax is most vulnerable to weed competition is needed.

While herbicides are not the only solution to improving weed management in flax, they are an important tool that can greatly improve the efficacy of cultural weed management strategies. Fluthiacet-methyl and topramezone often caused less crop damage than current registered herbicides, such as MCPA or bromoxynil + MCPA. Pyroxasulfone also did not cause significant crop damage, demonstrating the same crop safety that was observed by treatments containing sulfentrazone. Flax also showed acceptable tolerance to mixtures of pyroxasulfone + sulfentrazone, resulting in a novel PRE- tank mix that could be utilized in-crop. Moving forward a better understanding of what weeds are controlled by these herbicides is needed. While the PRE- herbicides are able to control both grassy and broadleaf species, identifying which weeds fluthiacet-methyl and topramezone might control is needed for flax crops. The tank mixes we examined, specifically fluthiacet-methyl + MCPA and topramezone + bromoxynil, did cause unacceptable initial injury on the crop. However, exploring the effect of different adjuvants or

surfactants on crop safety could help to minimize initial crop injury caused by these products. Furthermore, understanding how to effectively utilize both PRE- and POST- herbicides in flax production systems is needed. Multi-factor competition studies that examine the combined effect of different herbicide timings (i.e. PRE vs. POST) with optimal flax seeding rates, seeding date, and competitive cultivars could help improve our understanding of how flax competes and provide additional weed management methodologies for flax producers.

5.5 Conclusions

The current study has shown that competitive ability and overall productivity of flax can be improved through choosing a competitive cultivar, seeding at higher rates, in early May, and using a herbicide for in-crop weed control. Furthermore, the novel herbicides used in this study offer producers the ability to utilize several modes of action, which in turn reduces selection pressure on weeds and offers more control of those species that have evolved resistance. By combining these cultural factors, both weed management and the overall productivity of flax can be improved. Moreover, producers cannot continue to measure the productivity of flax by yield alone. Good weed management is observed early in the growing season during the initial stages of development. Hence, production practices which encourage rapid growth, a dense crop stand, and high crop biomass production are needed if the benefits of an IWM system are to be noticed. Ultimately, those practices that benefit weed management will in turn have a positive effect on flax yield. Through improving the productivity of flax, the acceptance of this minor crop could expand across the prairies and allow producers to lengthen crop rotations, reduce selection pressure against weeds, increases biodiversity, and improve the health of their agroecosystem (Swanton and Murphy, 1996). As such, through this research we have identified a way to effectively add diversity to a typically non-diverse production system and improve how weeds are managed in flax.

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