University of Alberta

Volunteer spring triticale (× *Triticosecale* Wittmack) seed persistence and control

by

Lisa Lynn Raatz

A thesis submitted to the Faculty of Graduate Studies and Research
in partial fulfillment of the requirements for the degree of

Master of Science
in
Plant Science

Department of Agricultural, Food and Nutritional Science

©Lisa Lynn Raatz
Fall 2012
Edmonton, Alberta

Permission is hereby granted to the University of Alberta Libraries to reproduce single copies of this thesis and to lend or sell such copies for private, scholarly or scientific research purposes only. Where the thesis is converted to, or otherwise made available in digital form, the University of Alberta will advise potential users of the thesis of these terms.

The author reserves all other publication and other rights in association with the copyright in the thesis and, except as herein before provided, neither the thesis nor any substantial portion thereof may be printed or otherwise reproduced in any material form whatsoever without the author's prior written permission.

Abstract

Spring triticale is being evaluated as a platform crop for bio-industrial products on the Canadian prairies and may require genetic modification (GM). Seed lost at harvest may persist and result in volunteer GM triticale populations in following crops that could impact co-existence with conventional cereals. Field experiments were conducted from 2006-2010 to assess the persistence of spring triticale in the soil seed bank and evaluate the effect of herbicide timings within four following rotations on volunteer triticale survival and fecundity. Relative to buried seed, triticale on the soil surface persisted longest, although 99% was non-viable after 19 months. Shallow buried seed germinated readily and formed volunteer populations. The combination of pre-seed and crop-specific in-crop herbicides provided the most consistent control, reducing volunteer triticale densities by 72-100%. Competitive subsequent crops, such as glyphosate tolerant canola, in combination with pre-seed and in-crop herbicides, minimize volunteer triticale seed bank replenishment in Alberta.

Acknowledgements

I would like to thank my supervisor, Dr. Linda Hall, for her encouragement, support, patience, and mentorship and for generously allowing me to work as her technician and graduate student simultaneously. I would also like to acknowledge my thesis committee, Dr. Lloyd Dosdall, Dr. Stephen Strelkov, and Dr. Ellen Macdonald for their support and assistance throughout the process. I would like to thank Dr. Rong-Cai Yang, Dr. Marc McPherson, and Ryan Nielson for statistical and experimental design advice and encouragement.

I would like to express sincere gratitude to technical, support, summer, and managerial staff at University of Alberta and Government of Alberta, Agriculture and Rural Development, as well as fellow graduate students at University of Alberta: Judy Irving, Jaime Crowe, Melissa Hills, Keith Topinka, Ryan Dyck, Debby Topinka, Gabriella Mendoza, Cara Kozack, Robert Latimer, Carly Hanson, Greg Iwaasa, Lisa World, David Brown, Ryan Nielson, Dr. Marc McPherson, Dr. Jody Dexter, Dr. Amit Jhala, Boris Henriquez, Susan Jess, Cameron Stevenson, and Christine Murray for their expertise, assistance, and advice. I would especially like to thank Kristina Polziehn, Janet Montgomery, Ryan Nielson, Judy Irving, and Jaime Crowe, for their moral support and friendship when I needed it most.

I especially thank my parents, Walter and Frieda Raatz, my sister, Connie Raatz, and my brother and sister-in-law, Earl and Debra Raatz for loving, encouraging, and supporting me throughout the entire process.

Funding for this thesis was provided by Alberta Agricultural Research Institute (AARI); University of Alberta; Government of Alberta, Agriculture and Rural Development (ARD); and Agriculture and Agri-Food Canada (AAFC), Agricultural Bioproducts Innovation Program (ABIP).

Table of Contents

List of Tables

List of Figures

List of Abbreviations

AAFC	Agriculture and Agri-Food Canada
AARI	Alberta Agricultural Research Institute
ABA	Abscisic acid
ABIP	Agricultural Bioproducts Innovation Program
AC	Agriculture Canada
ACCase	Acetyl-coenzyme A carboxylase
ANOVA	Analysis of variance
AP	Adventitious presence
ARD	Government of Alberta, Agriculture and Rural Development
BC	Back-cross
ºC	Celsius
CDC	Crop Diversification Centre, Saskatoon
CFIA	Canadian Food Inspection Agency
DAP	Days after pollination
DKL	Dekalb, Monsanto
DS	Danisco Seed
EC	Electrical conductivity
EU	European Union
EX_{50}	Extinction of 50%
EX_{99}	Extinction of 99%
FAOSTAT	Food and Agriculture Organization of the United Nations, statistics
GA	Gibberellic acid

GM	Genetically modified/genetic modification
GURT	Genetic use restriction technologies
kPa	Kilopascal
LL	Liberty Link® (glufosinate tolerant) canola
LLP	Low level presence
LSMeans	Least square means
OM	Organic matter
PNT	Plants with novel traits
RR	Roundup Ready® (glyphosate tolerant) canola
rRNA	Ribosomal ribonucleic acid
SE	Standard error
TKW	Thousand kernel weight
UK	United Kingdom
USA	United States of America
XR	Extended range

Chapter 1. Introduction

1.1. Background

On the Canadian prairies, volunteer crops are weeds (Leeson et al., 2005). Volunteers are crop plants that emerge within the following crop that were not intentionally seeded, but are the progeny arising from natural seed losses or mechanical harvest losses from previous crops (CSSA, 2012). When volunteers survive management practices and produce seed which is harvested with the crop, they cause economic losses via yield reductions in a density dependent manner (O'Donovan et al., 2007; O'Donovan et al., 1989), as well as through reduced grain quality (dockage). Volunteer cereals can be controlled in crops such as pea or canola, but are difficult to selectively remove from cereals.

When volunteers are genetically modified (GM), there is heightened incentive for control. Harvested seed may unintentionally contain GM seed from the previous crop (adventitious presence, AP) (Demeke et al., 2006), which may have implications for trade and export markets (Kalaitzandonakes, 2011; Kershen and McHughen, 2005). GM flax within conventional flax shipments has caused trade disruptions for Canada and has raised concerns about conventional and GM crop co-existence (Jhala et al., 2011). The intended introduction of glyphosate tolerant wheat raised concerns about volunteer management (Lyon et al., 2002; Rainbolt et al., 2004; Harker et al., 2005; Harker et al., 2006; Gruber et al., 2008) and potential risks to conventional wheat markets (Wilson et al., 2008). While the

economic risks posed by GM crops to established commodity markets are substantial, they are beyond the scope of this thesis.

Environmental risks encompass the potential for GM crops to become weedy or invasive. The Canadian Food Inspection Agency (CFIA) regulates GM crops within Canada and collects biological data in order to assess the invasiveness potential of GM crops ([CFIA] Canadian Food Inspection Agency, 2010). Assessing risk requires the collection of biological information throughout the lifecycle of the plant, including seed survival through to fecundity (Parker and Kareiva, 1996). While the majority of GM crops to date have involved herbicide and insect tolerance, abiotic stress tolerance traits are also being developed and these may confer a competitive advantage in stress-prone environments (James, 2010; Park et al., 2010). If this is the case, plants with abiotic stress tolerance could represent a greater risk to the environment.

On the Canadian prairies, triticale (× *Triticosecale* Wittmack) is being considered as a platform crop for bio-energy production which may benefit from genetic modification (Canadian Triticale Biorefinery Initiative, 2011). While the development and commercialization of imidazolinone-tolerant wheat and the intended, but abandoned, development of glyphosate-tolerant wheat prompted research on volunteer wheat management (Rainbolt et al., 2004), there is little comparable information for managing volunteer triticale.

The propensity for a crop to form a volunteer population depends largely on seed bank dynamics. Seed banks are reservoirs of viable seeds that accumulate and deplete in and on the soil surface (Thompson, 1987; Leck et al., 1989). While

managing weed seed banks in agricultural environments has been a focus of research (e.g. Legere et al., 2011; Gallandt, 2006; Menalled et al., 2001), crop seed banks are less frequently studied. The ability of a species to form a viable seed bank depends upon many interrelated factors such as seed production and dispersal, dormancy, predation, mortality, seed depth and size, as well as environmental factors such as soil moisture and temperature (Baskin and Baskin, 1998). Understanding factors that influence triticale seed banks will allow us to predict seed longevity and determine suitable management strategies to control volunteer populations in anticipation of GM triticale.

1.2. Research Objectives

1.2.1. The persistence of triticale seed within the seed bank

With the anticipation that triticale will require genetic modification in order to be a suitable platform for some end-use bio-products, information about seed-mediated gene flow will be needed to allow a science-based risk assessment to be conducted. Wheat, one of the triticale progenitors, is well understood and was therefore used as a comparative species for the purpose of risk assessment. Prior to the introduction of GM triticale and before it can be grown on a larger landscape scale, the following questions need to be addressed:

- How long will viable triticale seed persist in the soil following crop harvest?
- Does triticale exhibit primary dormancy and does this contribute to seed persistence?

- Does triticale exhibit secondary dormancy?
    - Will deep seed burial induce secondary dormancy or alternatively, is seed burial an effective means of hastening seed bank depletion?

Therefore, the following experimental hypotheses were made and are addressed in Chapter 3:

1. Seed from triticale cultivars ‘AC Alta’, Blue Aleurone, ‘AC Ultima’, and ‘Tyndal’ will persist for less or more time than wheat seed from a comparative cultivar ‘AC Barrie’ in an artificial seed bank.

2. In an artificial seed bank, the experimental Blue Aleurone triticale line will persist for more or less time than one of its parent cultivars, ‘AC Alta’.

3. In an artificial seed bank, buried triticale seed from cultivars ‘AC Alta’, Blue Aleurone, ‘AC Ultima’, and ‘Tyndal’ will persist for less or more time than seed remaining on the soil surface.

4. Triticale seeds from cultivars ‘AC Alta’, Blue Aleurone, ‘AC Ultima’, and ‘Tyndal’ will require a longer or shorter period of time to after-ripen than a wheat comparative cultivar, ‘AC Barrie’.

1.2.2. Volunteer triticale control and fecundity

Volunteer triticale populations represent a substantial risk for contributing to seed-mediated gene flow because triticale is a competitive species and can produce seed, replenish the seed bank, and contribute to AP when the subsequent crop is harvested. In anticipation of GM triticale production, the following questions were addressed in order to establish best management practices to control triticale volunteers in subsequent crops:

- What herbicide application timings are most appropriate for the control of volunteer triticale?
- To what extent are volunteer triticale populations controlled in subsequent cropping systems?
- How much seed will volunteer triticale survivors produce in subsequent crops contributing to AP and seed bank replenishment?
- In the absence of competition, how productive and fecund is triticale?
- How will the following crop yield be affected by the presence of volunteer triticale?
- Which crop(s) are most suited to follow triticale production?

These questions are addressed in Chapter 4 and the following experimental hypotheses were made about volunteer triticale populations:

1. Within following crops of glyphosate and glufosinate tolerant canola, field pea and imidazolinone tolerant wheat, preseed applications of glyphosate or glufosinate, and/or following crop-specific incrop applications of glyphosate, glufosinate, imazamox/imazethapyr, and imazamox + 2, 4-D ester, densities and

resulting fecundity of volunteer triticale will be less than those within untreated controls.

2. Within following crops of glyphosate and glufosinate tolerant canola, field pea and imidazolinone tolerant wheat, preseed applications of glyphosate or glufosinate and crop-specific incrop applications of glyphosate, glufosinate, imazamox/imazethapyr, and imazamox + 2, 4-D ester, densities and resulting fecundity of volunteer triticale will be lower than those receiving preseeding or incrop applications alone.

3. Within following crops of glyphosate and glufosinate tolerant canola, field pea and imidazolinone tolerant wheat, preseed applications of glyphosate or glufosinate and crop-specific incrop applications of glyphosate, glufosinate, imazamox/imazethapyr, and imazamox + 2, 4-D ester, the crop yields will be the higher than those in the untreated controls.

4. When triticale cultivars ‘AC Alta’, ‘Pronghorn’, and ‘AC Ultima’ are grown in the absence of competition, their measures of productivity such as biomass, tillering, fecundity, and seed weights will be significantly different.

GM crops require a large investment prior to release. Information to identify market or environmental factors that may block the crop from release are critical to assist with decision making. This research will provide information to

crop developers and the CFIA on the biology of non-GM triticale within the seed bank and the control of volunteers in subsequent crops. This research will be used as a baseline for triticale persistence and control in central Alberta with which to compare GM triticale in the event that it is developed for the Canadian prairies.

1.3. Literature Cited

Baskin, C.C. and J.M. Baskin. 1998. Seeds: ecology, biogeography, and evolution of dormancy and germination. Academic Press, San Diego, California, USA.

[CFIA] Canadian Food Inspection Agency. 2010. Safety assessment process for novel foods and agricultural products of biotechnology. Available at www.inspection.gc.ca/english/sci/biotech/reg/proce.shtml (verified Mar. 15, 2011). Government of Canada, Canadian Food Inspection Agency, Ottawa, Ontario, Canada.

[CSSA] Crop Science Society of America. 2012. Glossary of crop science terms. Available at https://www.crops.org/publications/crops-glossary# (verified Mar. 6, 2012). Madison, Wisconsin, USA.

Canadian Triticale Biorefinery Initiative. 2011. Canadian triticale biorefinery intiative: biotech applications. Available at http://www.ctbi.ca/index.php?option=com_content&view=article&id=15&Itemid=119 (verified Mar. 12, 2012). CTBI, Edmonton, Alberta, Canada.

Demeke, T., D.J. Perry and W.R. Scowcroft. 2006. Adventitious presence of GMOs: scientific overview for Canadian grains. Can. J. Plant Sci. 86:1-23.

Gallandt, E.R. 2006. How can we target the weed seedbank? Weed Sci. 54:588-596.

Gruber, S., N. Colbach, A. Barbottin and C. Pekrun. 2008. Post-harvest gene escape and approaches for minimizing it. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources 3(15). doi: 10.1079/PAVSNNR20083015.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y.T. Gan, H.A. Frederick, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2005. Glyphosate-resistant wheat persistence in western Canadian cropping systems. Weed Sci. 53:846-859.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y. Gan, F.A. Holm, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2006. Persistence of glyphosate-resistant canola in western Canadian cropping systems. Agron. J. 98:107-119.

James, C. 2010. Global Status of Commercialized Biotech/GM Crops: 2010. ISAAA Brief No. 42. ISAAA, Ithaca, New York, USA.

Jhala, A.J., H. Bhatt, K. Topinka and L.M. Hall. 2011. Pollen-mediated gene flow in flax (*Linum usitatissimum* L.): Can genetically engineered and organic flax coexist? Heredity 106:557-566.

Kalaitzandonakes, N. 2011. The economic impacts of asynchronous authorizations and low level presence: An overview. International Food and Agricultural Trade Policy Council, Washington, D.C., USA.

Kershen, D.L. and A. McHughen. 2005. Adventitious presence: inadvertent commingling and coexistence among farming methods. CAST Commentary QTA 2005-1:1-4.

Leck, M.A., V.T. Parker and R.L. Simpson (eds.) 1989. Ecology of soil seed banks. Academic Press, Inc., San Diego, California, USA.

Leeson, J.Y., A.G. Thomas, L.M. Hall, C.A. Brenzil, T. Andrews, K.R. Brown and R.C. Van Acker. 2005. Prairie weed surveys of cereal, oilseed and pulse crops from the 1970s to the 2000s. Rep. Weed Survey Series Publication 05-1CD. Agriculture and Agri-Food Canada, Saskatoon Research Centre, Saskatoon, Saskatchewan, Canada.

Légère, A., F.C. Stevenson and D.L. Benoit. 2011. The selective memory of weed seedbanks after 18 years of conservation tillage. Weed Sci. 59:98-106.

Lyon, D.J., A.J. Bussan, J.O. Evans, C.A. Mallory-Smith and T.F. Peeper. 2002. Pest management implications of glyphosate-rsistant wheat (*Triticum aestivum*) in the western United States. Weed Tech. 16:680-690.

Menalled, F.D., M. Hammond and K.L. Gross. 2001. Weed aboveground and seedbank community responses to agricultural management systems. Ecol. Appl. 11:1586-1601.

O'Donovan, J.T., K.J. Kirkland and A.K. Sharma. 1989. Canola yield and profitability as influenced by volunteer wheat infestations. Can. J. Plant Sci. 69:1235-1244.

O'Donovan, J.T., K.N. Harker, G.W. Clayton, L.M. Hall, J. Cathcart, K.L. Sapsford, F.A. Holm and K. Hacault. 2007. Volunteer barley interference in spring wheat grown in a zero-tillage system. Weed Sci. 55:70-74.

Park, J.R., I. McFarlane, R.H. Phipps and G. Ceddia. 2010. The role of transgenic crops in sustainable development. Plant Biotech. J. 9:2-21.

Parker, I.M. and P. Kareiva. 1996. Assessing the risks of invasion for genetically engineered plants: Acceptable evidence and reasonable doubt. Biol. Conserv. 78:193-203.

Rainbolt, C.R., D.C. Thill and F.L. Young. 2004. Control of volunteer herbicide-resistant wheat and canola. Weed Tech. 18:711-718.

Thompson, K. 1987. Seeds and seed banks. New Phytol. (Suppl.) 106:23-34.

Wilson, W.W., E.A. DeVuyst, R.D. Taylor, W.W. Koo and B.L. Dahl. 2008. Implications of biotech traits with segregation costs and market segments: the case of Roundup Ready(R) Wheat. Eur. Rev. Agric. Econ. 35:51-73.

Chapter 2. Literature Review

This literature review will focus on two major components. Seed banks will be discussed in the context of agricultural environments, specifically focusing on factors affecting seed banks of volunteer crops. Secondly, triticale, its development within Canada, and the need for research about triticale volunteer populations within the seed bank will be discussed.

2.1. Seed banks

Seed banks are a reserve of germplasm that ensure genetic diversity within a population (Simpson et al., 1989) and buffer populations from stochastic events by spreading out risk, enabling species to perpetuate (Venable and Brown, 1988; Levin, 1990). The purpose of the seed bank is to replace adult plants that produce seed and replenish the seed bank (Baker, 1989). Seed banks are vital for regeneration, expansion, and maintenance of species, particularly those that have not been domesticated and do not rely on human cultivation.

Seed banks have spatial and temporal elements. Figure 2.1 (Nielson et al., 2009) illustrates the general flow of seeds within the seed bank and the most salient factors affecting seed input and exit. Seeds disperse from the parent plant, land on the soil surface and may become buried in the soil. Seeds may be non-dormant and germinate when conditions permit or they may experience dormancy or abiotic conditions that cause them to remain ungerminated, though viable, and may persist for some time. Seeds are removed from the seed bank in a number of

ways. They are susceptible to exhaustion and senescence, consumption by predators, and disease or decay, or they may germinate and produce a plant capable of replenishing the seed bank.

Seeds entering and exiting the soil are influenced by factors which ultimately influence seed bank density including seed rain and dispersal, past and current species abundance and fecundity, seed longevity, soil disturbance, predation, pathogens, edaphic conditions, climate and stochastic events (Pakeman et al., 1999; Leck et al., 1989). Isolating the effects of any one factor is difficult since many are correlated. Factors that influence seed banks in the context of weeds and volunteer crops will be discussed. Describing inter-related factors separately can give the erroneous impression of simplicity. Therefore, correlations will be emphasized with appropriate examples from the literature.

Agricultural research has focused on the factors that influence seed banks in order to minimize weed seed accumulation and maximize depletion. The seed bank density in agricultural settings is dependent on past and present crop management and the intensity of management practices such as seeding rate, crop and variety choice, herbicides, and crop rotation (Harker et al., 2009; Légère et al., 2011). Weeds are more productive on fertile soils, although the type and placement of fertilizers can reduce the seed bank replenishment by restricting weed access to nutrients (Blackshaw et al., 2004; Blackshaw et al., 2005). Cultivation has traditionally been used to reduce the size of the weed seed bank by stimulating germination and by destroying existing seedlings, reducing seed bank replenishment. A combination of tillage and herbicides may deplete the

weed seed bank by up to 97% (Schweizer and Zimdahl, 1984). Adoption of reduced tillage practices favour a shift in species composition towards weeds that are suited to thrive under low disturbance situations (Blackshaw et al., 2001; Legere et al., 2011). Crop rotation is also a major tool in reducing weed seed banks (Cardina et al., 2002) by altering the selective pressures on weed populations. With increased public sensitivity towards food production, current research focuses on integrated weed management by employing multiple tools to affect the weed seed bank.

There are generally two types of seed banks: transient, in which seeds die or germinate within one year, and persistent, in which a proportion of seeds remain viable from one year to the next (Thompson and Grime, 1979). There are many weed seed species that have persistent seed banks where seeds can remain viable for several years; however, most crop species have transient seed banks.

2.1.1. Crops in the seed bank

2.1.1.1. Domesticity traits

Crops are domesticated plants that have been manipulated by humans for use in human-created habitats and as such require human intervention for survival (Harlan, 1992). Historically, crops have been selected for domesticity traits that maximize yields and ease of production. Crop species are generally characterized by a number of domesticity traits including: seed retention at maturity, loss of seed dormancy, germination synchrony or loss of secondary dormancy, increased seed size, and reduction in seed dispersal or loss of shattering, among other traits

(Warwick and Stewart, 2005). For example, the spike morphology of wheat (*Triticum aestivum* L.) progenitors includes barbed awns, pointed glumes, and backward pointing hairs which ensure that some proportion of seeds penetrate leaf litter and wedge into cracks in the ground. These morphological characteristics are subdued in domestic wheat, where seeds cannot penetrate the ground naturally and are therefore readily vulnerable to predation (Davies and Hillman, 1992). The loss of dormancy and the loss of rachis fragility are the traits that most differentiate cultivated wheat and barley (*Hordeum vulgare* L.) from their wild progenitors (Davies and Hillman, 1992). Many of these domesticity traits have come at the fitness cost of increased seed deterioration through the loss of seed vigor and eventual loss of germinability (Anderson and Baker, 1983), which ultimately contributes to the transient nature of the crop seed bank.

2.1.1.2. Ferality or de-domestication

Feral plants are derived from crop plants that have become partly or completely undomesticated and can reproduce outside of human-managed habitats (Gressel, 2005a). For example, feral rye is closely related to domesticated rye (*Secale cereale* L.) (Burger and Ellstrand, 2005) and the winter form causes economic losses in winter wheat production systems of midwestern USA (White et al., 2006). Feral rye has reverted to pre-domestication phenotypes having a brittle rachis, smaller seeds enclosed in the floret, more tillers, and delayed flowering relative to cultivated rye (Burger et al., 2007). While feral rye appears to exhibit low primary dormancy, in low moisture conditions seeds can persist in the seed bank for several years (Stump and Westra, 2000). Stump and Westra

(2000) showed that low levels of secondary dormancy could be induced that contributed to seed bank persistence, and while most seeds exhibited rapid germination within the first season, a small proportion of seeds remained ungerminated and viable within the seed bank. These persistent seeds were able to germinate up to 5 years after seed bank establishment and produced seeds to replenish the seed bank.

Many circumstances have contributed to the development of feral rye within the USA. Because many *Secale* spp. cross-pollinate, it has been speculated that the origin of feral rye is a result of hybridization between domesticated rye and wild relatives resulting in a weedy form (Burger and Ellstrand, 2005). The dramatic decrease in rye production and decline in popularity of the crop since the early 1990's, reduced selection pressure for non-shattering traits on rangelands, few crop rotation choices, and fewer domesticated cultivars available has also meant that there was less genetic diversity with which to dilute feral populations. The use of rye for erosion control in an unmanaged setting, lack of seed purity regulations, and suspected spread of feral seeds on contaminated harvest equipment (Burger et al., 2007; Miller et al., 2004; FAOSTAT, 2012) have also contributed to the spread of feral rye populations in the USA.

2.1.2. Seeds enter the seed bank

Seeds leave the parent plant and drop to the soil, entering the seed bank via seed rain (Simpson et al., 1989). The amount of seed entering the seed bank will depend on the reproductive capacity or fecundity of a particular species, which is influenced by environmental conditions during the growing season and

competition from surrounding plants. Following seed rain, secondary dispersal occurs for a distance and density that is strongly influenced by seed characteristics (weight, size, and shape), plant height, the timing and duration of seed release, the rate at which seeds can be produced, and the fecundity of the plant (Bakker et al., 1996). Dispersal is largely aided by species-specific reliance on wind, water, fire, animals (Simpson et al., 1989), or farm machinery (Boyd and White, 2009; Shirtliffe and Entz, 2005).

2.1.2.1. Fecundity

The fecundity of a species determines the potential maximum seed bank input and is influenced by inter- and intra-specific plant competition, time of emergence, plasticity and biomass of a species, as well as genotypic and environmental factors. The fecundity of many agricultural weed species has been determined with and without competition. For example, in the absence of crop competition, *Matricaria perforata* Mérat produced 71,000 to 256,000 seeds plant^{-1} (Blackshaw and Harker, 1997), while persian darnel (*Lolium persicum* Boiss. & Hohen. ex Boiss.) produced 1,700 and 2,800 seeds plant^{-1},when competing with canola (*Brassica napus* L.) and spring wheat crops, respectively (Holman et al., 2006). Cardina and Sparrow (1996) showed that common lambsquarters (*Chenopodium album* L.) seed density in the seed bank was highly variable, ranging from 7,700 seeds m^{-2} in tillage to 242,500 seeds m^{-2} in no-tillage. While weed species can be extremely fecund, volunteer crops typically produce fewer, but larger seeds per plant. Volunteer canola growing in winter wheat produced 1 to 120 seeds plant^{-1} or about 10% of a typical canola crop plant (Gruber and

Claupein, 2007), while Beres et al. (2010) showed that on the Canadian prairies, wheat, spring triticale (× *Triticosecale* Wittmack), barley and rye (*Secale cereale* L.) produce 60, 60, 53, and 114 seeds plant^{-1}, respectively, when grown as a crop. The potential fecundity of weeds can be high; however, reproductive success is also dependent on seed dispersal in order to reduce sibling competition and improve chances of seedling establishment.

2.1.2.2. Seed dispersal

In the absence of human intervention, the majority of seeds are dispersed relatively close to the parent plant. De Cauwer et al. (2008) found that among four pappus-bearing, wind dispersed (anemachorous) species in the UK, between 81 and 97% of total seeds were disseminated within 4 m of the original plant stand, although some seeds of *Conyza canadensis* (L.) Cronquist were found at the outer limit of the study, 32 m. Rew et al. (1996) found that in two barachorous species (seeds having no specialized dispersal structures), *Bromus sterilis* L. and *Anthriscus sylvestris* L. Hoffm., 99 and 87% of seeds were dispersed within 1 m of the source, respectively, within a plant community. When removed from the shelter of the plant community and placed in an open field, 84% of *B. sterilis* seed was dispersed within 1 m of the parent plant where the seed shadow was strongly influenced by the direction and speed of the prevailing winds (Rew et al., 1996). Seed rain depends largely on gravity; however, most seed dispersal occurs after seeds fall to the soil surface.

In agricultural environments, long distance seed dispersal occurs mostly as a result of the movement of agricultural equipment and foraging activity of seed

predators. Seed dispersal is dependent on the timing of seed input into the seed bank and is dictated by species-specific phenologies. The timing of seed release will affect seed movement via machinery. When seeds are shed prior to harvest, they are not widely dispersed (Barroso et al., 2006; Colbach et al., 2000; Shirtliffe et al., 2000). However, when the timing of seed set coincides with harvest processes, seeds can be moved large distances (McCanny and Cavers, 1988; Humston et al., 2005). Rew et al. (1996) recovered 46% of *B. sterilis* seed within 1 m of the seed source following combine harvesting, 43% of seeds were recovered up to 53 m in the direction of combine travel, and 10% of seeds were recovered within 7 m from the seed source that would have left the back of the combine as it traveled through the weed patch. Not only are seeds dispersed along a field with harvest equipment, they are also moved large distances between fields on equipment during transport (Boyd and White, 2009).

Seed dispersal via animals is closely linked with seed predation (See section 2.1.4.1). Animals forage for seed, often removing them from the source and storing or caching them for consumption later (Stiles, 2000). This is particularly true for tree seeds in forest habitats (Chambers and MacMahon, 1994). Owing to recent concerns over the movement of genetically modified (GM) crop seeds, Cummings et al. (2008) investigated the viability of non-GM corn (*Zea mays* L.), barley, safflower (*Carthamus tinctorius* L.), and rice (*Oryza sativa* L.) seeds following passage through the digestive tracts of various birds, and showed that no intact crop seeds were recovered, although a small number of seeds were transported on muddy feet. Animals disperse seeds often without

consuming them, but the amount and distance of seed dispersal is difficult to quantify.

2.1.2.3. Crop seeds enter the seed bank

Crop seeds are added to the seed bank through intentional planting; seed contamination; natural seed shedding prior to harvest which is aided by factors such as crop lodging, hail, or insect herbivory; and mechanical "shatter" losses during harvest (Anderson and Soper, 2003; Clarke, 1985; Willenborg and Van Acker, 2008; Vera et al., 2012). Harvest losses of spring wheat (*Triticum aestivum* L.) can vary depending on the shatter-resistance of the cultivar, growing conditions during seed development and harvest, and the travel speed and type of harvest equipment used. Spring wheat lost naturally at harvest ranged from 120 to 820 seeds m^{-2}, while direct combining losses ranged from 30 to 415 seeds m^{-2} (Clarke, 1985). Anderson and Soper (2003) summarized winter wheat survey data in the UK where harvest losses were 2 to 6% of the harvested yield (240 to 700 seeds m^{-2}). McPherson et al. (2009) reported harvest losses of 231 to 1,069 seed m^{-2} of safflower, although viability ranged from 81 to 518 seeds m^{-2}. Harvest losses of canola were approximately 3,000 viable seeds m^{-2} or approximately 20 times the typical crop seeding rate (Gulden et al., 2003a), while losses in flax (*Linum usitatissimum* L.) were up to 1,986 seeds m^{-2} in windrows (Dexter et al., 2011). Because crops are grown in monoculture, harvest losses tend to be substantial and relatively uniform and represent large seed bank inputs.

Seed dispersal of crops occurs mostly via machinery at time of harvest. Dispersal and density of lost crop seed will vary with travel speed and the

combine used because some are designed to concentrate screenings into a narrow band and others have chaff spreaders in order disperse the screenings more broadly (Anderson and Soper, 2003). Boyd and White (2009) showed that between 194,000 and 397,000 broadleaved and grass seeds were found in various spots on harvest machinery following harvest and transport of equipment, illustrating the potential for long-distance movement of seeds. Crop seed can also move with various tillage implements. Rew and Cussans (1997) showed that seeds on the surface of the ground moved further than those that were buried to 10 cm and smaller seeds (*Brassica napus* L.) were moved significantly further than larger seeded crops (*Hordeum vulgare* L. and *Vicia faba* L.). However, more than 84% of all seeds were not moved more than 1 m from the source and none were moved more than 5 m in the forward direction of the tillage operation. While tillage operations do not move seeds large distances, harvest operations have a greater potential for dispersal.

2.1.3. Seeds within the seed bank

2.1.3.1. Seed buried within the soil

After seeds are shed and dispersed, they can become buried in the soil through a number of mechanisms. They can enter via soil pores or cracks that have been formed through freeze/thaw or dry/wet cycles, covered by plant litter or soil through wind and rain, from the activities of animals (Chambers and MacMahon, 1994), or from mechanized agricultural processes such as tillage, harrowing, or seeding. Using beads to simulate movement of seeds into the soil

matrix, Westerman et al. (2009) showed that seeds rapidly enter the soil through cracks on the soil surface, covered by soil after heavy rainfall or wind, covered with crop residue after harvest, or gradually incorporated over time where smaller seeds became incorporated into the soil more easily than larger seeds. The length of time between seed shed and seed burial as well as depth of burial are also influenced by soil texture, seed size, seed shape, and seed coat sculpture; the smallest seeds have a tendency to become buried naturally and are therefore less exposed to seed predation (Benvenuti, 2007).

Once seeds become buried in the soil, they either persist for a period of time or they exit the seed bank by germinating, or succumbing to disease, exhaustion, or mortality (Figure 2.1). Burial increases seed contact with the soil so that light, nitrate, temperature, and moisture conditions stimulate germination (See section 2.1.4.5) or seeds may persist because conditions are not suitable for germination or because they are dormant (See section 2.1.3.3). However, buried seeds are generally no longer susceptible to seed predators (See section 2.1.4.3). The relative transient or persistent nature of the seed bank is influenced by how rapidly and abundantly seeds are being deposited into the soil compared with the number and rate at which seeds are exiting through germination, predation, disease, exhaustion, or mortality.

2.1.3.2. Seed persistence

Seed persistence within the seed bank has been extensively researched for many species. Classical, albeit artificial, seed burial studies have investigated seed viability over time. Dr. Beal's long-term seed viability study found that three of

the 23 weed species initially buried were still viable after 120 years (Telewski and Zeevaart, 2002). On the USA Great Plains, an average of 19 of 41 weed species buried deeply still had some viable seeds after 17 years, where 4 species had between 61 and 95% germination (Burnside et al., 1996). Similarly, 17 species deeply buried in Alaska generally persisted longer than those buried shallowly and 12 species still had some viable seeds 19 years after burial (Conn et al., 2006).

The persistence of annual crop seeds in the seed bank depends on the number of viable seeds in the soil and the seed microsite (biotic and abiotic factors surrounding the seeds), such as soil moisture and burial depth (Boyd and Van Acker, 2004). In general, annual crop seeds do not to persist in the soil seed bank (Cavers and Benoit, 1989). Safflower and flax have been shown to have short persistence in western Canada, where no viable seeds were found after 2 years (McPherson et al., 2009; Dexter et al., 2011). Volunteer canola generally does not persist for long periods of time in the seed bank. Persistence ranged from 4 to 5 years following production in eastern Canada (Simard et al., 2002), not beyond 3.5 years in Australia (Baker and Preston, 2008), not beyond 3 years when volunteers were prevented from returning seed into the seed bank in western Canada (Harker et al., 2006), although in the absence of disturbance they persisted up to 11 years in the UK (Lutman et al., 2003). Anderson and Soper (2003) reviewed several classical burial studies that included cereal crops and concluded that cereal seeds rapidly disintegrated and did not persist beyond 2 years. In an artificial seed bank study where predation was prevented, volunteer

wheat on the soil surface did not persist beyond 3 years while those buried deeply did not persist beyond 1 year (Nielson et al., 2009). Similarly, when volunteers were prevented from replenishing the seed bank, Harker et al. (2005) showed that wheat seeds were not recruited in high numbers beyond 3 years, while feral rye seeds were rapidly depleted within 1 year of burial with <1% viable after 4 years (Stump and Westra, 2000). While crop seeds generally do not persist for long periods of time, a few seeds may remain viable and persist for several years.

Artificial seed bank studies must be viewed with caution because seeds buried within retrievable containers such as mesh bags do not reproduce conditions found within natural seed banks (Van Mourik et al., 2005). Persistence may be under-estimated when seeds are aggregated at high densities making them more susceptible to the spread of pathogenic fungi (Chee-Sanford et al., 2006; Gilbert, 2002; Van Mourik et al., 2005). However, the persistence of seeds on the soil surface or at shallow depths may be over-estimated by artificially enclosing seeds in exclusion cages or mesh bags which prevents vertebrate and invertebrate seed predation (Baraibar et al., 2012; Baraibar et al., 2009; Graziani et al., 2007; O'Rourke et al., 2006). Additionally, Seerey et al. (2011) showed that intact wheat seed heads persisted longer within the *in situ* seed bank than threshed wheat seeds. However, *in situ* seed bank studies that evaluate seedling emergence may be confounded by factors that cannot adequately be quantified such as initial seed numbers, seed return, or variability in seed dispersal (Saatkamp et al., 2009). The study of the persistence of seeds within seed banks is challenging because factors such as disease or seed predation are difficult to isolate and quantify.

Seed size influences species persistence within the seed bank. In their widely used classification of seed banks, Thompson and Grime (1979) noted that the transient seed banks in northern UK consisted of seeds that were larger and not prone to burial. Generally, large-seeded species have carbohydrate reserves to support germination even when conditions may not be ideal. Persistent seed banks are most often associated with species that have small, light-weight seeds, and strict requirements for germination while larger seeded species that have less stringent germination requirements often lack seed banks (Thompson, 1987).

Seeds remain viable and persist for periods of time in the seed bank because the microsite is unsuitable for germination or the seeds are dormant. Species-specific narrow requirements for germination including abiotic requirements for light, soil moisture and pH, oxygen (hypoxia in water-logged soils), and available nitrate may influence long-term persistence (Thompson, 2000). Seeds which do not germinate because environmental conditions are unfavourable are called ‘quiescent’ (Murdoch and Ellis, 2000). Alternately, seed dormancy ensures seed survival until conditions are more appropriate or to synchronize germination with a period or season most suitable to germination and survival and contributes to short term seed persistence (Thompson, 2000).

2.1.3.3. Dormancy

Seed dormancy is an adaptation of species that are exposed to environmental conditions which are adverse for germination or plant growth during some portion of the year (Forcella et al., 2000). Seed dormancy ensures the survival of a portion of the population through unpredictable environmental

events, it decreases intra-specific competition by spreading germination out in time, and prevents germination out of season (Finkelstein et al., 2008). Species exhibiting seed dormancy can escape adverse conditions until triggered to be released from dormancy by favourable environmental cues which are complex interactions of temperature, moisture, and light. Benech-Arnold et al. (2000) define dormancy as "an internal condition of the seed that impedes its germination under otherwise adequate hydric, thermal and gaseous conditions". There are two distinct phases of dormancy: primary and secondary (Figure 2.2). Primary or innate dormancy is the inherited dormancy that seeds develop during maturation while on or upon being released from the mother plant and is strongly influenced by genotype as well as the maternal environment. Secondary or induced dormancy is a state induced or re-induced in imbibed, previously non-dormant seeds that have been exposed to unfavourable temperature, moisture, light conditions, or nitrogen availability (Baskin and Baskin, 1998; Murdoch and Ellis, 2000; Finkelstein et al., 2008). Seeds may undergo dormancy cycling when conditions are not favourable, progressively moving in and out of dormancy until they either germinate or die (Finkelstein et al., 2008).

Primary dormancy is acquired as seeds mature (Finkelstein et al., 2008) and declines just before and after seeds are released from the mother plant. The period of time over which seeds air dry and are released from primary dormancy is called after-ripening and is a function of air temperature, seed moisture content, and time (Murdoch and Ellis, 2000). Conditions that facilitate after-ripening vary with species. Many cereal crops such as wheat, barley, and triticale require dry

after-ripening to lose primary dormancy (Schramm et al., 2010; Benech-Arnold, 2004; Biddulph et al., 2005; Biddulph et al., 2008; Biddulph et al., 2007), although temperature effects vary with winter and summer cereals (Benech-Arnold, 2004). The cues that trigger breaking of dormancy and the cues that trigger germination can be similar (Bradford, 2002) and it can be difficult to distinguish between the two events.

As after-ripening progresses, seeds lose their dormancy and are able to germinate over a widening range of conditions. However, during this process, if unfavourable environmental conditions occur preventing germination in non-dormant seeds (e.g. darkness due to seed burial), further changes in the environment such as temperature fluctuations or changes in light or moisture can cause seeds of some species to enter secondary dormancy (Probert, 2000). During this phase, the conditions under which seeds may germinate begin to narrow until seeds are unable to germinate under any set of environmental conditions (Baskin and Baskin, 1985). Kruk and Benech Arnold (1998) show that low winter temperatures terminate dormancy in *Polygonum aviculare* and that high summer temperatures can induce secondary dormancy. Annual dormancy cycling may take place from dormant to non-dormant in many weed species and it can account for some of the periodicity in germination within the seed bank (Foley, 2001).

Like primary dormancy, the termination and induction of secondary dormancy in weed seeds revolves around species-specific adaptations to survive unpredictable and inhospitable environmental conditions. Secondary dormancy can be induced by a number of abiotic factors including anaerobic soil conditions,

prolonged exposure to white or far-red light, nitrate or nitrite, as well as unfavourable temperatures (Murdoch and Ellis, 2000). Secondary dormancy induction is similar to a reverse of after-ripening because the suitable range of conditions for germination is decreased (Baskin and Baskin, 1998). Many species cycle through different depths of dormancy (Hilhorst, 2007), where seeds of the same species within the same environment can be found along a continuum rather than being strictly dormant or non-dormant.

Agronomic practices affect seed dormancy cycling in weed seeds and are reviewed in Dyer (1995). Altering soil fertility in agricultural environments through addition of nitrite and nitrate fertilizers can facilitate dormancy breaking and stimulate weed seeds to germinate. Additionally, crop rotations can affect the amount of crop residue, stubble, species composition, crop canopy, and shading from nearby plants which affect light and light interception (red: far-red light ratios) at the soil surface (Dyer, 1995; Allen et al., 2007). Tillage facilitates seed burial or exposes seeds to light which can induce or break dormancy in small seeded weed species requiring a light stimulus to germinate. Depth of seed burial can affect dormancy by altering seed exposure to light, although there are complex interactions with temperature fluctuations, seed scarification, oxygen and moisture availability which can induce or break dormancy (Dyer, 1995 and references within).

The underlying mechanisms involved in the control of primary and secondary dormancy have not been clearly elucidated, nor is it clear whether similar mechanisms function in both primary and secondary dormancy (Foley,

2001). Debeaujon et al. (2007) outline two major mechanisms of dormancy: embryo dormancy, where germination inhibition is imposed by the embryo, and seed-coat imposed dormancy. Both mechanisms are conferred genetically, but are influenced by environmental factors.

2.1.3.4. Dormancy in cereal crops

Persistent dormancy is an undesirable characteristic that would negatively impact crop end-uses (e.g., malting in barley) and depending upon the length of dormancy, it would prevent or delay production of a new crop the following season (Benech-Arnold, 2004). Non-dormancy in crops is desirable because it enables emergence patterns to be more predictable in terms of temperature, available moisture, and gases (Benech-Arnold et al., 2000). Through the domestication of cereal crops, dormancy has been aggressively selected against, resulting in seeds that are capable of germinating on the head prior to harvest, a phenomenon known as pre-harvest sprouting (Benech-Arnold, 2004). Cereal grains that lack or have low levels of primary dormancy during seed development are susceptible to pre-harvest sprouting. Pre-harvest sprouting or vivipary is the premature germination of seed within the head prior to harvest, triggered by long exposure to rain, dew, high humidity, and temperatures conducive to germination during grain filling (Biddulph et al., 2007; Paulsen and Auld, 2004). Cereal breeders attempt to find the right balance of selecting sufficient primary dormancy to prevent pre-harvest sprouting without inadvertently selecting for prolonged dormancy which would interfere with further crop uses. The tendency

towards pre-harvest sprouting in cereals is associated with a lack of primary dormancy (Biddulph et al., 2007).

Cultivated species have been selected for rapid, uniform germination to ensure good crop yields and, as such, dormancy is absent or very low (Warwick and Stewart, 2005). Generally, seed dormancy is considered to be absent for many crop species because it is lost rapidly after seeds are shed. However, because crops are derived from wild ancestors, dormancy may re-appear under unfavourable temperature or moisture conditions (Hilhorst and Toorop, 1997) (See section 2.1.1.2). Dormancy in cereal crops is imparted by two mechanisms: 1) physically from tissues surrounding the embryo to prevent germination until the embryo is fully developed and the endosperm has enough storage reserve to support germination, and/or 2) physiologically through hormonal and enzymatic activity within the embryo (Hilhorst, 2007; Biddulph et al., 2008). Dormancy in cereals is heritable and is influenced by the maternal environment. While physical and physiological mechanisms may prevent germination during primary dormancy, once physical dormancy is lost, it is not reversible and therefore does not play a role in secondary dormancy (Hilhorst, 2007).

Physical or seed coat dormancy is the major mechanism of dormancy in cereals (Simpson, 1990). The seed coat poses a physical barrier that: interferes with water uptake and gas exchange with the embryo (Paulsen and Auld, 2004;Pickett, 1989); prevents radicle extrusion; prevents the leakage of germination inhibitors from within the embryo; and prevents light filtration (Debeaujon et al., 2007). The seed coat serves to protect the embryo from damage

and restricts embryo growth prior to being fully developed. Physical dormancy in cereal grains tends to be short-lived because seed coats are water permeable and in freely available water, can reach critical moisture content for germination in about 3 hours (Paulsen and Auld, 2004). Immature embryos of wheat and barley germinated rapidly when removed from the seed coat and placed in water or other growing media (Kermode, 1990; Walker-Simmons, 1987; Biddulph et al., 2007), which provides some evidence that physiological dormancy plays a lesser role in dormancy in these crops. Diurnal temperature fluctuations (heating of the soil surface from solar radiation during the day and cooling at night) can act to break physical seed coat dormancy (Probert, 2000). The seed coat and spikelet tissues also produce phenolic chemical inhibitors to germination, such as abscisic acid (ABA), or contain pigments which have been associated with germination inhibition (Pickett, 1989; Himi et al., 2002).

Cereal crops may also have some physiological or embryo-imposed dormancy (Debeaujon et al., 2007; Simpson, 1990). Embryo-imposed dormancy is related to the sensitivity of the embryo during grain-filling stage to endogenous ABA, which inhibits germination (Biddulph et al., 2007; Finkelstein et al., 2008). ABA is a phytohormone that induces seed dormancy, embryo tolerance to desiccation during seed development, as well as other responses to environmental stress cues such as stomatal closure in leaves when plants are under drought conditions (Schramm et al., 2010). ABA content within the embryo remains low until 15 days after pollination, after which time concentrations increase and peak when the seeds reach physiological maturity, then decrease as seeds desiccate or

undergo after-ripening (Walker-Simmons, 1987). Embryo-sensitivity to ABA varies with wheat genotype (Schramm et al., 2010). Endogenous gibberellic acids (GA) also play a role by decreasing the mechanical resistance of tissue around the embryo and promoting embryo growth, working in opposition to ABA, although there can be variability in embryo sensitivity to both GA and ABA depending on the cultivar (Gubler et al., 2005). However, it is more likely that GA plays a role in germination and not in dormancy termination because it is present during seed development, but levels drop as seeds mature. Rather, it is expected that ABA levels regulate the induction of seed dormancy during seed development (Foley, 2001).

Seed dormancy is under complex genetic control with continuous phenotypic variation (Benech-Arnold, 2004). Kato et al. (2001) found that over 80% of phenotypic variation in seed dormancy in 119 double haploid wheat lines was controlled by three quantitative trait loci. Andreoli et al. (2006) showed that in Brazilian wheat lines, dormancy was expressed by two major genes and appeared recessive to dormancy (*aabb*). Some wheat cultivars have increased expression of dormancy correlated to *Vp-1* (*viviparous*) genes through increased sensitivity of the embryo to ABA (Nakamura and Toyama, 2001). There is also evidence that dormancy is strongly associated with red grain colour. The *R* genes are dominant maternally expressed genes which encode the depositition of red phlobaphene isoflavones in the seed coat (Debeaujon et al., 2007). Although there are instances where white wheat lines exhibit higher levels of dormancy (Andreoli et al., 2006; Flintham and Humphray, 1993), red wheat tends to have increased

levels of dormancy potentially associated with pigmentation (Groos et al., 2002; Paulsen and Auld, 2004), particularly when conditions are cool and wet (Torada and Amano, 2002). Flintham and Gale (1990) showed an additive effect of an increased level of dormancy in wheat by increasing the number of *R* alleles for red seed colour. The most dormant wheat lines are homozygous dominant for red colour, although the relationship between pigment and dormancy is still unknown (Flintham and Humphray, 1993), and may be linked to genes imparting dormancy (Torada and Amano, 2002). There is some speculation that pigment polymers link to the seed coat during seed maturation which results in thicker cell layers and increases the physical barrier to water, oxygen, and hormones (Finkelstein et al., 2008). Pigments also regulate the sensitivity to light and photoperiod by filtering light reaching the embryo (Finkelstein et al., 2008). These pigments decline during after-ripening and germination is permitted (Paulsen and Auld, 2004). There is on-going research in wheat and triticale breeding programs to determine genetic sources of dormancy (physical and physiological) in order to prolong primary dormancy and prevent pre-harvest sprouting.

Cereal dormancy is strongly influenced by the maternal environment during grain development (Biddulph et al., 2007; Benech-Arnold, 2004). Higher temperatures during a sensitive period of grain-filling resulted in lower dormancy in barley prior to harvest (Benech-Arnold, 2004). Winter wheat in Australia that is exposed to adequate moisture at lower temperatures and higher relative humidity during grain-filling tends to have decreased levels of primary dormancy, while plants that are moisture-stressed and are grain-filling under relatively high

temperatures (>30 ^{o}C for 14 days) will have a tendency towards increased dormancy (Biddulph et al., 2007). A sprouting-resistant wheat cultivar showed greater primary dormancy when grown under cooler conditions (15 ^{o}C) and diminished dormancy under warmer conditions (26 ^{o}C) (Reddy et al., 1985). Similarly, winter triticale grown under cooler conditions (9 ^{o}C) in Norway had enhanced primary dormancy and reduced dormancy when grown under warmer temperatures (15 or 21 ^{o}C) (Buraas and Skinnes, 1985), which is true for many grass species (Simpson, 1990). Spring triticale in Canada exposed to high rainfall during the harvest period showed little or no primary dormancy and the same lines exposed to low rainfall during harvest had increased dormancy (Salmon and Helm, 1985). Additionally, triticale germinated at high temperatures (30 ^{o}C) showed increased primary dormancy than at 18 ^{o}C (Salmon and Helm, 1985). Some wheat cultivars show greater levels of dormancy when exposed to high temperatures during grain-filling and because ABA is more effective at high temperatures, the embryos showed enhanced sensitivity to ABA (Walker-Simmons, 1988). Low temperature and short day length increased dormancy in barley (Schuurink et al., 1992).Winter wheat dormancy is not expressed at low temperatures (Walker-Simmons, 1988) and the thermal range allowing germination widens with after-ripening. Differences in the effect of temperature on dormancy may occur depending upon whether a cereal is a winter or spring type. Benech-Arnold (2004) explains that winter cereals, which typically do not exhibit dormancy at lower temperatures, could be expected to exhibit pre-harvest sprouting in years of cool, moist conditions during harvest. Likewise, summer

cereals, which normally do not exhibit dormancy at high temperatures, could be expected to exhibit pre-harvest sprouting when conditions are moist and warm during harvest. However, the soil environment following seed shed onto the soil surface may also influence the progression of after-ripening (Pickett, 1989). Alaru et al. (2008) found that germination on the spike before physiological maturity was controlled more by precipitation than by cultivar effects, although following physiological maturity, genotype played a larger role and could be used as a tool for making selections. Gutterman (2000) discussed the effects of maternal environment for a number of plant species during seed development and listed a number of factors which contribute to dormancy and germinability: position of seed on the mother plant, age of the mother plant, day length, temperature, light quality, photo-thermal environment, and altitude. Many of the examples showed that the maternal environment had contributed to formation of harder or thicker seed coats which delayed germination or caused physical dormancy (Gutterman, 2000 and references within).

Cereal crops tend not to persist in the seed bank largely because they have a very short period of primary dormancy and no secondary dormancy. There are few instances of secondary dormancy induction in cereal crops, although seed burial depth has been speculated to contribute to seed persistence in winter cereals that did not undergo after-ripening in Europe (Pickett, 1989). Leymarie et al. (2008) induced secondary dormancy or reinforced a deeper primary dormancy in barley by exposing seed to 30^{o}C for up to 72 hours, which increased embryo sensitivity to ABA even after temperatures returned to the range suitable for

germination. However, it is uncommon for cereals to exhibit secondary dormancy cycling which occurs for closely related grass species (Simpson, 1990). Seed burial and temperature have contributed to induction of secondary dormancy in canola in western Canada and can cause the seeds to persist for prolonged periods in the seed bank (Gulden et al., 2003b; Gulden et al., 2004a; Gulden et al., 2004b). Feral rye, a winter annual, does not exhibit primary dormancy although seed persistence was prolonged on the soil surface and was attributed to unsuitable microsite while seeds buried to 5 or 25 cm did not persist (Stump and Westra, 2000). Toole and Brown (1946) summarized Duvel's buried seed experiment that ranged from 1902 through 1941 and concluded that crop seeds, including wheat, oats (*Avena sativa* L.), barley and rye were not viable longer than 1 year in the soil when artificially buried 20, 56, and 107 cm below ground level. Crops other than some forage and pasture grasses and a few legumes have short longevity within the soil seed bank (Lewis, 1973; Rampton and Ching, 1970).

2.1.4. Seeds exit the seed bank

Seeds exit the seed bank through predation following seed loss onto the soil surface; through mortality by succumbing to disease, anoxia, decay, or exhaustion; or by germinating (Figure 2.1).

2.1.4.1. Seed predation

Seed predation can occur before (pre-dispersal) or after (post-dispersal) seeds are released from the parent plant. Predators of seeds include vertebrates

(rodents, birds, bats) and invertebrates (ants, beetles, moth and fly larvae). Pre-dispersal predation prevents seed from entering the seed bank, while post-dispersal seed predation directly affects the input, abundance, survival, composition, longevity, distribution, and heterogeneity of seeds in the seed bank (Louda, 1989). However, even in years and locations of high seed predation, long-term impact to a seed bank may be negligible (Crawley, 2000). Much of the research on seed predation in agricultural settings tends to focus on weed control using biological control agents or investigations of the impact of endemic predators on weed populations as part of integrated management.

2.1.4.2. Pre-dispersal predation

Pre-dispersal predation is generally restricted to specialized insects (Zhang et al., 1997), which have specific cues to feed on a relatively narrow host range (within one Family, Genus or Species) (Crawley, 2000). Most are found within the orders: Diptera, Lepidoptera, Coleoptera, and Hymenoptera (Crawley, 2000). Predators lay eggs directly on the host plant, usually in close proximity of flowering parts so that seed development and maturation coincide with the larval life cycle. A major seed predator of Canada thistle (*Cirsium arvense* L. Scop.), *Orellia ruficauda* Fabr. (Diptera: Tephritidae), lays its eggs directly into developing flower heads and the larvae consume the seed. Larvae affected 5 to 86% of flower heads in sites across Canada, destroying 20 to 80% of seeds from affected heads (Forsyth and Watson, 1985). Because of large spatial variation in the rate of predation, seed losses are often not large enough to reduce the reproductive potential of the affected species. In corn cropping systems, moth

larvae (*Coleophora lineapulvella* Lepidoptera: Coleophoridae), attacked relatively few inflorescences of *Amaranthus retroflexus* prior to seed release, although they damaged as many as 93% of seeds on affected flower heads (DeSousa et al., 2003). Management practices such as crop choice, row spacing, and use of tillage may create conditions that influence the attractiveness of a host plant, egg survival, or the ability of larvae to overwinter, indirectly contributing to the levels of predation (Nurse et al., 2003).

In crops, pre-dispersal seed predation tends to be intensively managed. Because crops are grown in monoculture and the food source is not limiting, predator populations can outbreak causing significant seed losses. Economic thresholds for many insect pest species are developed to prevent major yield losses in years of population outbreak and associated management practices are taken if threshold levels are reached. Cultural (crop rotations, plant breeding), chemical (insecticides), and biological control measures are employed to minimize seed destruction. For example, Orange Wheat Blossom Midge (*Sitodiplosis mosellana* Géhin, Diptera: Cecidomyiidae) larvae feed on developing seeds of wheat and triticale (Wright and Doane, 1987). On the Canadian Prairies, timed insecticide applications (Alberta Agriculture, Food and Rural Development, 2001), wheat cultivars bred for antibiotic resistance (cultivars that contain or produce toxic compounds which interfere with insect development) (Ding et al., 2000), and encouragement of natural enemies (Olfert et al., 2009) are all management practices employed to negate pre-dispersal cereal seed losses.

2.1.4.3. Post-dispersal predation

Post-dispersal predators tend to be generalist, relatively mobile seed feeders, often consuming and dispersing seeds from a wide variety of plant species (Crawley, 2000). Seed predation will vary with the density of the predator and seed source, habitat, seed size, environmental conditions, availability of alternate foods for predators (Crawley, 2000), depth of buried seed (Thompson, 1987), and timing of seed shed (Westerman et al., 2003). On the soil surface, smaller seeds tend to escape predation by vertebrates, unless they are found aggregated together or resources are scarce (Tew et al., 2000). Invertebrates feed on small, dense seeds because the size of the granivore restricts accessibility to larger seeds (Zhang et al., 1997; Lundgren, 2005; Lundgren and Rosentrater, 2007). Smaller seeds which are buried will generally escape predation by invertebrates, although White et al. (2007) showed that weed seeds buried to 1 cm were still accessible to a larger carabid beetle (*Harpalus*) species. Larger seeds that are buried may still be accessible to rodents (Thompson, 1987; Hulme, 1996). Generally, losses from predation are highest for seeds that are easily accessible on the soil surface (Westerman et al., 2006), as well as those that are most abundant (Louda, 1989).

Much of the research investigates the relative importance of predator type or species in various habitats and seasons, often assessing the amount of seed being removed. In cereal fields in the Netherlands, vertebrates (mostly mice) accounted for 30 to 88% of weed seed consumption relative to invertebrates (4 to 38%) (Westerman et al., 2003), although it is possible that invertebrates become

prey to vertebrate seed predators, confounding their respective relative importance. In UK winter wheat fields, Holmes and Froud-Williams (2005) showed that predators removed 100% of weed seeds on the soil surface within one week. Birds fed mainly within the cropped area, while non-avian predators fed mainly at field edges. In Western Australia, ants and other invertebrates were the major predators and seed removal was generally highest along field edges (Jacob et al., 2006), while Marino et al. (1997) showed no differences between field edges and the field centre in Michigan. Mauchline et al. (2005) showed that carabid beetles were the main weed seed predators, followed by mice and small birds in spring barley in the UK. However, predation levels declined as the season progressed. Menalled et al. (2000) showed much spatial variation in predation over large field scale trials in conventional tillage corn systems, where vertebrates appeared to be the main seed predators. In grasslands, vertebrates were the main seed predators of large-seeded species (Blaney and Kotanen, 2001). The relative importance of the predator type varies with localized habitats, season, and environment.

Management practices influence predation. Brust and House (1988) showed that two to three times as many seeds were consumed in zero-tillage systems than in conventional tillage in soybean crops in North Carolina, USA. Increased invertebrate predation occurred in reduced input than conventionally managed soybean and more predation took place shortly after weed seed rain in fall than any other time during the growing season (O'Rourke et al., 2006). Soil disturbance such as tillage and irrigation decrease predation because they disrupt

predator habitat and reduce accessibility to seed by incorporating them into the soil (Baraibar et al., 2009; Zhang et al., 1997; Menalled et al., 2007). Westerman et al. (2006) modeled predation where the main parameters were demand for seed, seed burial, and dispersal. The authors estimated weed seed predation in forage crops on triticale stubble to be 32-35% relative to corn, soybeans or alfalfa at 60-65%. Heggenstaller et al. (2006) showed crop-specific seasonal patterns of invertebrate weed seed predation. Triticale/red clover had the highest levels of predation in spring, low in summer, and intermediate in late fall, although the intensity of tillage, herbicide or fertilizer applications did not appear to influence predation. When seed densities are high as is the case following monocultures, limitations to seedling recruitment are largely dependent on microsite availability and population density will not generally be affected by predation. However, when seed inputs are low and distribution is patchy, predation will limit population size (Crawley, 2000). Swanton et al. (1999) showed that both pre- and post-dispersal seed predation were important at reducing weed seed populations. Micro-moth *Coleophora lineapulvella* reduced *Amaranthus* spp. seed bank inputs by up to 37% while post-dispersal predation from mice, isopods, millipedes, while Carabid beetles reduced barnyard grass populations by up to 3% per day following seed shed in no tillage systems.

There is little research on predation of crop seed and the impact on subsequent volunteers. It is generally accepted that plant species with large, nutritious seeds are especially vulnerable to seed predation and therefore, tend not to persist within the seed bank (Foster, 1986). Vertebrates such as mice and birds

are the dominant seed predators following cereal harvest. Frequent pest outbreaks of house mice occur in regions adjacent to winter wheat fields in New Zealand (Mutze, 2007) because the food source is not limiting during the growing season. Marino et al. (2005) noticed that weed seed predation by rodents decreased over time as other food resources such as cereal grain became available. Capture and release diet studies of field mice and voles showed that the majority of their diet came from bent heads of maturing rye and volunteer winter rye grain following harvest from June through August in Germany and the authors presumed that one rodent species fed on cached grain later into the fall (Abt and Bock, 1998). While not quantified, Cummings et al. (2008) alluded to post-harvest grain consumption of barley, safflower, corn, and rice by birds had contributed to the removal of potential volunteers in subsequent crops. Wood mice in arable lands of the UK, preferred to feed on sweet corn > winter wheat > canola > winter barley > wild oat > *Plantago major* (Jensen, 1993). Brust and House (1988) showed that winter wheat seeds were preferred by mice and larger carabid beetles (specifically *Harpalus caliginosus*) relative to weed seeds in zero-tillage soybean fields following harvest. The carabid beetles had difficulty feeding on wheat seeds shortly after seed deposition, but following weathering and absorption of moisture, they were better able to penetrate the pericarp and consume the endosperm. In conventional tillage, the carabids were the primary consumers of wheat seed, followed by ants, smaller carabids, and then mice. Overall predation was much lower in conventional tillage than in zero-tillage (Brust and House,

1988). Crop seeds lost through shattering are abundant on the soil surface and susceptible to predation by both vertebrates and larger invertebrates.

Post-dispersal seed predation is difficult to quantify. Firstly, most research assumes that seed removal by animals equates to seed consumption, however without the ability to track the ultimate seed fate, researchers may overestimate seed bank losses. VanderWall et al. (2005) pointed out that predation studies may over-estimate the proportion of seeds being consumed and that predators disperse and cache seeds, frequently taking them to microsites that are more conducive for germination. For example rodents in rainforests of French Guiana removed 23 to 96% of tree seeds, but of those removed only 9 to 43% were consumed. The remainder were stored or cached (Forget, 1996). With the increased public interest in GM crops, there will be increased pressure for much-needed research on crop seed dispersal and seed fate. Secondly, many studies attempt to quantify seed predation by using artificial scenarios: exclusion cages designed to eliminate vertebrate (birds vs. rodents) and invertebrate predators and baiting predators with known quantities or types of seeds. These simulations, while having the advantage of precision that comes from a controlled environment, forego the reality of *in situ* seed predation. Lastly, as with all biological systems, seed bank loss via predation is intricately complex and is both influenced by and impacts seed dispersal (See section 2.1.2.2), seed persistence (See section 2.1.3.2), and germination (See section 2.1.4.5). There is a relatively short window of time when seeds are vulnerable to predation following seed rain, but prior to incorporation into the soil.

2.1.4.4. Seed mortality and exhaustion

A number of biotic and abiotic factors can cause loss from the seed bank through mortality. In some cases, the seed bank may be comprised mostly of dead seeds. Forcella et al. (1992) showed that dead seeds greatly outnumbered viable or dormant seeds where their seed banks consisted of between 50 and 90% dead seeds for green foxtail (*Setaria viridis*) and redroot pigweed (*Amaranthus retroflexus*). Zorner et al. (1984) showed that between 80 and 94% of wild oat (*Avena fatua*) seed buried to 1cm were dead, 51 to 68% of those buried to 5 cm, while of those buried to 10 cm 39 to 52% were dead as they were exhumed over 24 months. In Australia, a high proportion of rigid ryegrass (*Lolium rigidum*) seeds on the soil surface decayed and were prone to rapid desiccation and metabolic failure (Chauhan et al., 2006a). Peachey and Mallory-Smith (2007) showed higher levels of seed mortality over winter in Oregon, USA at shallow versus greater depths for hairy nightshade (*Solanum sarrachoides*). While, seed mortality appears to be influenced by depth of burial, tillage regime (conservation vs. no-till) did not appear to significantly influence wild oat mortality (Gallandt et al., 2004). Biotic mechanisms include seed exhaustion, senescence, and disease, and abiotic influences include crushing or abrasion, fire, and hypoxia or anoxia usually as a result of flooding (Chambers and MacMahon, 1994). Mechanical abrasion and piercing of weed seed coats hastened seed mortality by exposure to opportunistic bacteria or fungi (Davis et al., 2008). Prolonged exposure of buried weed seeds to oxygen deficient (hypoxic) conditions may induce secondary dormancy (see section 2.1.3.3), but may also cause reduced seed respiration and

an accumulation of toxic metabolites (Benvenuti and Macchia, 1995; Hendry, 1993). Exposure to oxygen, however, is one of the major factors involved in the aging process and loss of seed viability (Hendry, 1993). Flood-induced anoxic conditions may result in seed death, although seeds age naturally and expire as oxygen free-radicals and lipid peroxidation metabolites build up in seed tissues (Benvenuti, 2007). Abiotic factors are more closely associated with the seed-microsite relationship which may hasten seed death.

Microbial pathogens contribute to removing seeds from the seed bank. Seed bank research is considerable and while researchers frequently cite seed loss through disease, very little research has been done to assess the role of pathogens in the seed bank. Kremer (1993) reviewed research conducted on soil microorganisms as they impact weed seeds. Most research investigated seed-borne pathogens or toxic metabolites produced by bacteria or fungi and their potential as biological control agents rather than the effects of soil borne pathogens on seed bank dynamics. Some species are more susceptible to endemic microbial decay, although species susceptibility is influenced by seed characteristics, soil microhabitat, and diversity of pathogens, their distribution or dose within the soil (Chee-Sanford et al., 2006). In velvetleaf, Kremer et al. (1984) showed that the seed coat is the major barrier to pathogenic decay; hard seed coats that were punctured and in the presence of seed borne fungi had over 60% seed decay and over 50% of seedlings were also diseased. The activity of seed rotting fungi (*Pythium* spp, *Rhizoctonia solani*, and *Fusarium* spp.) and seed-colonizing (*Pseudomonas* spp.) species in the soil are dependent on soil factors:

moisture and temperature, organic matter, pH, and texture, as well as exudates leaking from compromised seeds which may act to stimulate germination in pathogenic resting spores (Harman, 1983). As fungi disable the defence mechanisms of a seed through production of toxins or lipid degradation, the pathogen depletes proteins, sugars, and amino acids, enhancing fungal growth and removing the reserves required by the seed, reducing viability (Cherry, 1983; St. Angelo and Ory, 1983). Using non-dormant *Bromus tectorum* seed, Meyer et al. (2010) showed that a virulent pathotype of *Pyrenophora semeniperda*, a seed pathogen which causes necrosis by toxin production, grew more slowly than less virulent isolates and could allow seeds to escape mortality through rapid germination. Crop seeds can escape seed mortality through rapid germination, although the pathogen can still sporulate on seedlings and cause seedling death or reduced vigor (Medd and Campbell, 2005).

Microbial associations are better described for crop than weed species because, similar to pre-dispersal seed predation (see section 2.1.4.2), crop protection to maximize yield is one of the major goals in agriculture. Numerous seed-applied fungicides have been formulated to protect crop species from decay prior to or just following seedling emergence, such as seed treatments used to prevent seed rots or seedling blights caused by *Pythium* or *Phytophthora* spp. in wheat (Brook, 2007). In the absence of crop seed treatments, resistance to pathogens is mostly attributed to the seed coat (Halloin, 1983). However, the seed coat can be easily mechanically damaged during harvest and contribute to seed disease and decay (McGee, 1995). Pathogens also affect and destroy seeds

prior to seed dispersal, such as ergot (*Claviceps purpurea*) in triticale and wheat. Karnal bunt (*Tilletia indica* Mitra) affects wheat and triticale, however, Warham (1990) showed that 94% of completely bunted seeds still had viable embryos although the endosperm had been replaced with fungus that prevented the newly germinated seedling from establishing.

Environmental factors, particularly temperature and moisture, can influence seed respiration and aging/deterioration/exhaustion (Chambers and MacMahon, 1994). Seed deterioration is difficult to measure and dissociate from effects of seed borne fungi. However, increased temperature and relative humidity contribute to seed deterioration. At high relative humidity, moisture content in stored crop seeds is elevated causing seeds to deteriorate faster than stored dry seed (Anderson and Baker, 1983; Harman, 1983). Anderson and Baker (1983) also speculated on the role of genetic mutation, rRNA degradation, increased enzymatic activity in the endosperm, decreased protein synthesis, and decreased glucose utilization on seed aging and deterioration, although it is not known which are primary factors leading to seed death. Domestication of crops may inadvertently have made them more susceptible to disease and decay by selecting against undesirable traits that likely play a role in phytoprotection such as dormancy, thick or impermeable seed coat, flavanols and tannins, lectins, and various fatty acids (Halloin, 1983).

2.1.4.5. Germination and seedling recruitment

While seeds exit the seed bank through predation, loss of viability and disease, the primary cause of non-dormant seed depletion from the soil seed bank

is through germination (Cardina and Sparrow, 1996). Non-dormant seeds exit the seed bank via germination in response to genetically controlled and species-specific responses to light, temperature, moisture, oxygen, nitrate, and chemical or gaseous stimulants (Simpson et al., 1989; Nonogaki et al., 2007; Nonogaki et al., 2010; Boyd and Acker, 2004; Hilhorst and Karssen, 2000). Cues that stimulate germination may also be similar to those that break dormancy. Germination and emergence are constrained by seed dispersal and the microsite or biotic and abiotic factors such as soil compaction and moisture directly surrounding the seeds (Boyd and Van Acker, 2004). Even though germination may occur, emergence from the soil surface or seedling recruitment may be prevented by burial depth relative to seed energy reserves, soil compaction and texture, poor seedling vigor, or a change in environmental conditions (Nonogaki et al., 2010; Boyd and Van Acker, 2003). Seed depth influences seedling emergence from the soil seed bank, where deep seed burial can have negative consequences for seedling emergence depending upon seed size. Limited seed reserves, particularly with smaller seeds or at greater soil depths, may become exhausted as the hypocotyl or coleoptile elongates, preventing or delaying seedlings from reaching the soil surface and ultimately leading to seedling death (Forcella et al., 2000; Benvenuti et al., 2001) termed fatal germination (Grundy et al., 2003). Grundy et al. (1996) found that small seeded species will emerge from shallow soils better than larger seeded species; larger seeded species require more time to imbibe water and risk dehydration on or just under the soil surface (Buhler, 1995). In a survey of tilled fields in western Canada, Van Acker et al.

(2004) showed that volunteer wheat seedlings could be recruited from greater depths than small seeded weeds such as green foxtail [*Setaria viridis* (L.) Beauv.]. Under greenhouse conditions when moisture was not limiting, wheat seedlings had >70% recruitment from the soil surface as well as shallow (1-2 cm) and deep (6-7 cm) burial, catchweed bedstraw (*Galium aparine* L.) had poor emergence on the soil surface and emerged when buried shallowly or as deep as 4 cm, while field pennycress (*Thlaspi arvense* L.) seedling recruitment occurred on the soil surface but significantly less when buried (Boyd and Van Acker, 2003). The same study indicated, however, that when soil moisture fluctuated at the shallow depths, wheat emergence from the surface was significantly lower (55%) than when buried (85%) (Boyd and Van Acker, 2003). Oriental mustard (*Sisymbrium orientale* Torn.) seedlings emerged on the soil surface and emergence was stimulated by nitrate when moisture was not limiting, but high salinity conditions showed depressed germination in Australia (Chauhan et al., 2006b).

Seedling emergence is influenced by soil characteristics: water, daily and seasonal temperature fluctuations, oxygen levels, light quality, seed burial depth, fertility, salinity, compaction, tillage, and surface residue, as well as factors contributing to seed dormancy and its complexities (Forcella et al., 2000; Grundy and Mead, 2000; Vleeshouwers and Bouwmeester, 2001). Seedling emergence is also influenced by competition from surrounding plants and their relative densities (Anderson and Nielsen, 1996). Chemicals produced by surrounding plants may also have allelopathic properties which inhibit germination or are deleterious to seedling growth (Hilhorst and Karssen, 2000). There is evidence

that root exudates from crops such as wheat, barley, rye and rice (*Oryza sativa* L.) can have inhibitory effects on the germination of some weed species (Belz, 2007). In agricultural settings, seedling emergence periodicities have been evaluated and modeled for various weeds in order to predict when management practices such as tillage or herbicides would be most effective; temperature and soil moisture are the primary factors involved in germination (Lawson et al., 2006; Hacault and Van Acker, 2006; Blackshaw et al., 2002; Boyd and Acker, 2004; Grundy and Mead, 2000; Ogg and Dawson, 1984; Oryokot et al., 1997). For weed species within a cropping environment, the seedling stage represents the most vulnerable stage to impose control measures and is the optimal stage for removal to maximize crop yields and profits (Harker et al., 2008; Harker et al., 2001; O'Donovan et al., 2007; Martin et al., 2001; Upadhyay et al., 2006; May et al., 2003; Sikkema et al., 2005). The seedling stage is not only vulnerable to management strategies, but natural mortality via abiotic (drought, flooding, uprooting via washout, freezing, salt, surface disturbances) and biotic (grazing of roots or shoots, genetic defect, inter- and intra-specific competition, disease) factors are difficult to elucidate (Fenner, 1987).

2.1.4.6. Volunteer crops

Volunteers are those plants that germinate in subsequent seasons after a crop has been grown, and are offspring from crop seed that has prematurely dropped to the ground naturally prior to harvest (through shattering, lodging, animal herbivory, hail, wind) or as a result of the mechanical harvest process (Willenborg and Van Acker, 2008; Anderson and Soper, 2003; Clarke, 1985;

Gressel, 2005b). For annual crops, seed enters the seed bank in fall, when the crop plants have reached physiological maturity and the seed is ripe. Volunteerism is a concern in agricultural settings because these plants compete with the following crops resulting in lower yields, contamination of the seed lot, reduced seed quality, or they serve as alternate hosts for insect pests or pathogens (Warwick and Stewart, 2005).

Large harvest seed loss and the frequency of a crop in rotation have contributed to volunteer crops being significant weeds in western Canada. Volunteer wheat ranked 19th in overall relative abundance (an index based on frequency, uniformity, and density) in the 2001 Alberta Weed Survey, with an average density of 6 plants m^{-2} within fields where it was found following incrop herbicides (Leeson et al., 2002). Across the Canadian prairies, volunteer wheat has doubled in relative abundance since the 1970s where high field densities have increased from 61 to 280 plants m^{-2} (Leeson et al., 2005). Volunteer canola has also increased in relative abundance, where high field densities ranged from 52 plants m^{-2} in 1970s to 143 plants m^{-2} in 2000s (Leeson et al., 2005). Leeson et al. (2005) compiled the relative rankings of weeds from weed surveys across the Canadian prairie provinces and of the volunteer crops, wheat ranked highest at 12th, while canola, barley, flax, oats, and rye ranked 14th, 25th, 26th, 46th, and 115th, respectively. Lawson et al. (2006) showed that the majority of volunteer canola seedlings emerged after crop seeding and could be as high as 2,015 seedlings m^{-2} under high-disturbance direct seeding and as few as 6 seedlings m^{-2} in conventional tillage in Manitoba, Canada. Volunteer winter wheat seedlings

emerged primarily in fall on the central Great Plains, USA but had continuous emergence throughout the following season where recruitment was highest in no-till and within a corn follow crop (Anderson and Nielsen, 1996).

Uncontrolled volunteers compete with the subsequent crop for light and nutrients causing yield losses, as well as produce seed that is harvested along with the crop. O'Donovan et al. (2007) showed that densities as low as 3 plants m^{-2} of volunteer barley could cause yield losses in wheat, although competition losses were also a function of crop density. Volunteer crops may produce seed and be harvested along with the following crop causing adventitious presence (AP), the unintentional and unwanted inclusion of other materials, including seeds, within harvested crop seed (Kershen and McHughen, 2005). Feral rye seed harvested along with following winter wheat crop on the Great Plains comprised up to 73% of the total harvested portion when left unmanaged (Stump and Westra, 2000). AP contributes to down-grading of seed due to unwanted impurities within a seed lot, which is undesirable when using seed lots for planting, as well as seed lots intended for bread or pasta production and other end-uses.

2.1.5. Summary

Cereal crop seeds within the agricultural seed bank do not persist for long periods of time, generally not longer than 5 years in the soil. Although variability is high and it is difficult to measure the magnitude of loss, crop seeds can be subject to predation following harvest as well as disease and mortality. Major seed predators include vertebrates (particularly mice and birds), although larger Carabid beetles also feed on cereal seeds. Volunteer cereals may be susceptible to

disease through exposure to seed borne or endemic soil pathogens. Cereal crops, in general, do not have prolonged periods of primary dormancy which would contribute to seed bank persistence and there is no evidence to suggest that cereal crops undergo secondary dormancy or dormancy cycling. Cereal seeds that do persist tend to do so because they are quiescent because conditions are not conducive to stimulate germination. However, most seeds rapidly exit the seed bank by germinating when soil moisture and temperature conditions are favourable. Seedlings may be unable to emerge from the soil surface, if buried at greater depths or when soil compaction or other environmental factors prevent emergence. Seedlings may be killed by freezing, frost, flooding, lack of moisture, or through grazing and disease; however, those seedlings that survive comprise the volunteer population and compete directly with the following crop. The seedling stage is most vulnerable to management practices, such as tillage, herbicides, or competitive cropping rotations. However, volunteers that survive or escape control measures can produce seed that become harvested with the following crop or lost to the soil surface and replenish the seed bank (Figure 2.1). Seeds enter and exit the seed bank in dynamic and stochastic processes which are influenced by complex and interconnected mechanisms. While information exists for cereals such as wheat, barley, rye, oat, rice within the seed bank as they behave as crops and volunteers, less information exists about the seed bank dynamics of the relatively new crop, triticale.

2.2. Triticale

Triticale (× *Triticosecale* Wittmack) is an amphidiploid resulting from an intergeneric cross between a wheat (*Triticum* sp.) female receptor and rye (*Secale* sp.) pollen (Kavanagh et al., 2010). Most of the triticale grown today descends from crosses involving common spring wheat (*Triticum aestivum* L., 2n=42=AABBDD) or durum wheat (*Triticum durum* Desf., 2n=28=AABB) and diploid rye (*Secale cereal* L., 2n=14=RR). The majority of modern cultivars are hexaploid triticales (2n=42=AABBRR) sharing a common ancestor, 'Armadillo' bred in 1967 (Ammar et al., 2004). In triticale, wheat traits dominate and as a result, triticale morphologically and chemically resembles wheat more than rye (Varughese et al., 1996). A complete description of triticale genesis is beyond the scope of this review.

2.2.1. Triticale production

In 2010, triticale was grown in 34 countries on over 3.9 million ha (Table 2.1). The majority of triticale grown worldwide is the winter form which is produced predominantly in Poland, Belarus, Germany, France, Australia and China, constituting over 75% of world triticale production in 2010. Canada produced 22,200 ha (or 0.6%) in 2010 and ranked 20th for area under triticale production, although this position has declined since 2002 when Canada reached a maximum acreage of 87,000 ha (FAOSTAT, 2012). In 2010, wheat was produced on over 216 million ha in 122 countries where 8.3 million ha (or 3.8%) were grown in Canada (FAOSTAT, 2012).

There are two major types of triticale: spring types which are seeded in spring and do not require a period of vernalization to produce reproductive structures and winter types that have an obligatory cold requirement after germination to enter a reproductive phase (Salmon et al., 2004). In Canada, triticale is used primarily as forage, animal feed, or for livestock grazing (Salmon et al., 2004; Mergoum et al., 2004). Canadian farmers also grow winter triticale for silage because it can be harvested before or after other crops allowing farmers to use machinery and time more efficiently (Wolf, 1989). Most triticale grain production is used as feed for pigs and poultry, but is also being used as feed for ruminant livestock (Mergoum et al., 2004). Triticale is a minor crop on the Canadian prairies.

Early triticales were initially intended as a human food crop in stress prone environments (Mergoum et al., 2004). However, because of poor bread-making qualities, such as low gluten content; poor gluten strength; a tendency towards pre-harvest sprouting; dark grain colour; and shriveled seeds, adoption for human consumption even in marginal environments remains limited (Mergoum et al., 2004; Oettler, 2005; Salmon et al., 2004). Currently, triticale remains a minor crop worldwide that has limited use on marginal lands for on-farm consumption (Oettler, 2005).

Triticale is capable of producing more biomass and grain yield compared with other cereals. This is particularly true in stress-prone ecologies where soil moisture is limiting, in areas with extreme temperatures and soil pH levels, and soil salinity. Triticale produces grain yields comparative to that of wheat and rye

under optimal growing conditions and out-competes wheat and rye when conditions are less favourable (Oettler, 2005). Spring triticale was shown to have similar or higher grain yields than wheat, barley, or rye and was generally more competitive with weeds relative to other cereal crops in Alberta, Canada (Beres et al., 2010; Goyal et al., 2011). In Canada, triticale is typically grown in the Prairie Provinces and is best suited to the brown soil zone of Southern Alberta and Saskatchewan, which receive lower levels of precipitation during the growing season (Alberta Agriculture, Food and Rural Development, 2005; Salmon et al., 2004).

2.2.2. Future of triticale

There has been growing interest in Canada to develop triticale as a platform for plant-based energy and industrial end-uses such as a feedstock for bioethanol production, monomer and polymer production, and biorefining for chemical production and will require genetic modification (GM) (Goyal et al., 2011; Canadian Triticale Biorefinery Initiative, 2011). Other potential triticale development opportunities include glucose syrup, straw for pulp and paper production, biodegradable detergents, high strength fibres, proteins, simple sugars, industrial enzymes, as well as byproduct usage such as seed hulls for industrial proteins (Eudes, 2006; Canadian Triticale Biorefinery Initiative, 2011). In the USA, numerous lignocellulose-based potential energy crops such as grasses, trees, and herbaceous plants that can be grown on marginal lands are also being considered for biofuel feedstocks and may require GM to enhance environmental tolerance (e.g., drought tolerance) or energy conversion (e.g.,

reduced lignin) (DiTomaso et al., 2007). The Canadian Triticale Biorefinery Initiative is focusing breeding efforts on triticale because it is a high yielding biomass crop with high starch levels, produces well in a wide range of environmental conditions, is not currently widely grown in Canada, and is generally not part of the human food system (Eudes, 2006). Triticale has also been successfully genetically transformed to tolerate the herbicide glufosinate ammonium (Zimny et al., 1995); herbicide tolerance genes may be used as a marker for other genes. Suitable triticale lines and germplasm are being evaluated for their agronomic and processing suitability on and for the Canadian prairies (Goyal et al., 2011; McLeod et al., 2010; Wang et al., 1997). Because many of these uses may require GM, studies are being conducted to assess the risks associated with pollen- and seed-mediated gene flow. Should many of these products become feasible, triticale production area within Canada is predicted to increase to 400,000 ha by 2015 (Canadian Triticale Biorefinery Initiative, 2010). While in Canada, the Canadian Food Inspection Agency (CFIA) regulates plants with novel traits (PNT) which encompasses both traits selected following mutagenesis and GM traits, most of the world regulates only GM traits and products. Within this thesis the term GM refers to genetic modification through the insertion of novel genes. Much of the debate around GM crop production revolves around coexistence and how to ensure that non-GM crops are kept free of unwanted transgenes. Since the first GM crops were commercialized in 1996, the production area of GM crops worldwide has been increasing (James, 2010).

2.2.3. Gene flow in triticale

Because triticale is being considered as a platform for novel bio-products which involve the use of GM, Canada requires an environmental assessment to quantify the potential risks to food or feed safety as well as to the environment. The Canadian Food Inspection Agency (CFIA) compiles biological information about the potential for GM crops to become weedy or invasive in natural habitats, outcross with wild or weedy relatives, become a plant pest, and impact non-target species and biodiversity ([CFIA] Canadian Food Inspection Agency, 2010). The USA uses trait-based weed risk assessment evaluations to determine whether GM species have the potential to be weedy or invasive by determining: invasiveness potential under various climate scenarios, potential for cross-hybridization with related taxa, susceptibility of native and managed ecosystems to invasion, and development of potential management protocols (DiTomaso et al., 2007); the potential risks of GM plants are compared to the risks posed in equivalent scenarios by non-GM comparators (Wolt, 2009). In Canada, data for biology documents is being or has been collected for a number of GM and potential GM crops such as sugar beet (*Beta vulgaris* L.), canola (*Brassica napus* L. and *B. rapa* L.), Camelina (*Camelina sativa* (L.) Cranz), wheat (*Triticum aestivum* L. and *T. turgidum* ssp. *durum*), among others ([CFIA] Canadian Food Inspection Agency, 2011). Baseline pollen- and seed-mediated gene flow data can be collected for conventional triticale prior to genetic modification to use in comparison with future modified lines.

2.2.3.1. Pollen movement

Pollen movement is a potential means by which introduced transgenes in triticale can be transferred to related species that can successfully out-cross with triticale such as conventional triticale, related cultivated and feral crop species, as well as wild and weedy relatives. Gene flow is defined as "the successful transfer of genetic information between different individuals, populations, and generations (to progeny) and across spatial dimensions" (Gealy et al., 2007) and generally occurs at very low frequencies in the primarily self-pollinating triticale (Kavanagh et al., 2010). Pollen movement has been shown to occur between crops and their wild relatives (Seefeldt et al., 1998; Warwick et al., 2003), as well as between related crop cultivars (Hucl et al., 2004; Hall et al., 2000). Transgene movement via pollen has serious implications for the coexistence of conventional and GM crops. Successful hybridization depends upon a number of factors: the sympatry of species or the synchrony of pollen production by the donor and the stigma receptivity; distribution and abundance of donor and receptor species; the pollen vector (insects or wind); the genetic compatibility of the species; floral structures; pollen viability and longevity; duration of anthesis; distance from and size of the pollen source; physical barriers; topography; surrounding vegetation; environmental conditions at the time of anthesis such as wind speed and direction; the geographical and temporal proximity of the species; and ultimately, the viability of the hybrid seeds (Conner et al., 2003; Kavanagh et al., 2010; Beckie and Hall, 2008; Gustafson et al., 2005). In general, rye florets are open at anthesis (chasmogamous) allowing for out-crossing, wheat florets are closed at anthesis

(cleistogamous) and are primarily self-pollinating, and triticale florets are usually cleistogamous although some out-crossing can occur (Kavanagh et al., 2010). Triticale is generally treated as a self-pollinated species by plant breeders (Oettler, 2005).

The potential for inter- and intra-specific out-crossing will need to be evaluated for GM triticale. Kavanagh et al. (2010) identified a number of species which could potentially hybridize with triticale in Canada: cultivated and feral rye and wheat species, barley (although it is extant on the prairies, artificially created hybrids have required embryo rescue), and weedy species, intermediate wheat grass [*Agropyron* intermedium (Host) Beauv.], jointed goatgrass (*Aegilops cylindrica* Host), quackgrass [*Elymus repens* (L.) Gould], pubescent wheatgrass [*Agropyron trichophorum* (Link) K. Richt.], and sea lyme grass [*Leymus arenarius*(L.) Hochst.]. Under greenhouse conditions where reciprocal crosses of triticale, rye, and wheat were made intentionally, Hills et al. (2007b) showed that triticale cultivars were self-compatible, but out-crossing between triticale and rye was low and hybrids resulting from crosses of triticale as the pollen donor and rye or wheat were usually sterile. The European Union has implemented a threshold limit of 0.9% AP of approved GM in non-GM material before labeling, and subsequently segregation is required on materials intended for food or feed products (European Union, 2003). Kavanagh (2012, in press) showed the maximum intra-specific out-crossing in triticale directly adjacent to the pollen source was 5%, but declined rapidly to less than 0.15% at 50 m from the pollen source; 50% of out-crossing occurred within 3 m of the pollen source. By

modeling data from several empirical pollen-mediated small-scale gene flow studies in wheat, Gustafson et al. (2005) predicted that the highest levels of out-crossing (>1%) occur within 1 m of the pollen source and decline rapidly with distance from the pollen source (<0.015% at 100 m); the model predicted pollen-mediated gene flow in wheat should fall below 0.1% at 30 m from the pollen source. In a multi-location study involving different cultivars grown on a commercial scale, Gaines et al. (2007) showed that maximum observed pollen-mediated gene flow of imidazolinone-resistant winter wheat was 5.3% at 0.23 m and the maximum pollen-mediated gene flow at the furthest distance detected (61 m) was 0.25%; the authors suggest that for cultivars that head out earlier than the pollen receptor, the 0.9% threshold would be reached at a distance of 41.1 m, but for later heading cultivars the threshold would be reached at 0.7 m. Pollen movement from a 16 ha field of imidazolinone tolerant wheat into an adjacent wheat field measured 0.2% along a common border (0.5 m) and declined exponentially with distance where pollen movement was 0.06% at 10 m (Beckie et al., 2011). While pollen-mediated gene flow for self-pollinating crops occurs at relatively low levels directly adjacent to the source, it declines with distance.

There are a number of measures that seed breeders employ to limit off-types and to ensure genetic purity of seed lines. The exposure of conventional crops to novel transgenes in GM triticale can be limited by implementing: isolation distances, limiting GM production area, removal of a same species trap strip crop adjacent to the pollen donor where AP is highest, harvest blending to dilute the ratio of off-types, as well as using new emerging technologies such as

genetic use restriction technologies (GURT) that impose sterility in second generation seeds and will limit the release of transgenes into the environment and food and feed systems (Gealy et al., 2007; Hills et al., 2007a; Gustafson et al., 2005). However, upon genetic transformation, release of transgenes into the environment will approach, but never reach 0% (Wolt et al., 2004). Part of risk mitigation is to reduce release of seed into the environment following harvest of the transgenic crop. Control of volunteer transgenic triticale can be assessed prior to genetic transformation by determining control in conventional cultivars.

2.2.3.2. Seed movement

While much of GM regulation revolves around pollen-mediated gene flow, seed-mediated gene flow of GM crops into non-GM crop materials is likely the greater source of AP for primarily self-pollinating small seeded crops where there are high amounts of seed loss and subsequently, large numbers of volunteers (Beckie and Hall, 2008). Potential sources of AP include impure seed lots intended for planting, volunteers within following crops, and admixture during planting, harvesting, transport, and storage (Beckie and Hall, 2008). Kalaitzandonakes (2011) reviewed the implications of low level presence (LLP) or "the accidental presence of small amounts of biotech events that have undergone full safety assessment and have received regulatory approval for all possible uses in one or more countries but are still unauthorized in others due to regulatory asynchronicity or expiration of their approvals" along the grain commodity supply chain. Low level presence could occur through commingling of grain as it is shipped from various farms to country and terminal elevators, feed

manufacturers, grain processors, port elevators, through to export and end uses of feed and seed; tolerance for LLP may be higher where food, feed or safety concerns are minimal, but when LLP risks are high the costs associated with seed segregation may be high and cause trade disruptions (Kalaitzandonakes, 2011). While the EU does not specify thresholds for GM seed within conventional or organic seed, it is probable that thresholds will be lower than 0.9% (Beckie and Hall, 2008; Devos et al., 2009).

In Canada, USA, and Japan, non-GM products can contain up to 5% GM material before they must be labeled as containing GM (Ramessar et al., 2010). Canada segregates cereal seed by assigning a seed grade classification which is based on the amount of foreign materials within a given seed sample. For example: triticale grade No. 1 Canada cannot exceed 1% cereal grains other than wheat or 0.5% of matter other than cereal grains (Canadian Grain Commission, 2011). For seed purity in breeding, it is recognized that AP can be minimized but not eliminated (Ramessar et al., 2010). There are a number of measures that are currently used to ensure cultivar and crop purity for seed production can be applied to limit AP of GM materials within non-GM crops: using certified seed; spatial isolation from the same crop species; using pollen barriers; intentionally seeding at different times to achieve asynchronous flowering; limit GM volunteers by extending crop rotations; meticulous cleaning of equipment used for seeding, transport, harvest, storage, and processing; controlling volunteers and wild or weedy relatives; using post-harvest tillage; keeping thorough records of field history; voluntary clustering fields with similar production goals;

communicating with neighbouring farmers; and in some instances, regional segregation of land for specific purposes (e.g. organic production only) (Devos et al., 2009; Ramessar et al., 2010).

Following production of a GM crop, seed losses are expected at harvest and will manifest as volunteers in the following crop. Should these volunteers be left uncontrolled, they can set seed and contribute to AP or replenish the seed bank. While GM triticale has not been developed, GM glyphosate tolerant wheat was being tested in the early 2000's with the intention of making it available for production in Canada and the USA (Rainbolt et al., 2004; Lyon et al., 2002). Control of herbicide tolerant volunteers is a major concern within a cropping system particularly when the following crops are also herbicide tolerant. Rainbolt et al. (2004) showed that volunteer spring glyphosate- and imidazolinone-tolerant wheat could be controlled with label rates of quizalofop and clethodim alone or mixed with glyphosate. However, the study assessed volunteer control within a preseed or fallow situation and the fecundity of survivors or AP at harvest could not be quantified. Similarly, in a preseeding situation, Lyon et al. (2002) showed that volunteer spring wheat was more effectively controlled by glyphosate than winter wheat and that glyphosate was more effective than using ACCase inhibiting herbicides (quizalofop, fluazifop, clethodim, or sethoxydim) which required an additional 2 to 4 weeks to adequately control winter wheat and are generally more costly. Volunteer wheat was effectively controlled with label rates of preseeding clethodim and quizalofop-P alone and with several broadleaf tank mix partners (Blackshaw et al., 2006). While herbicides can effectively control

volunteer cereals, some plants may escape or survive application and still produce seed.

Despite control measures, volunteer cereals can still contribute to AP. O'Donovan et al. (2007) modeled imidazolinone tolerant wheat crop losses from volunteer barley where AP was estimated to be as high as 7,663 kg ha^{-1} when wheat densities were low and volunteer barley densities were high. In volunteer flax, preseeding and incrop glyphosate reduced AP to near zero, although flax that survived or emerged after preseeding glyphosate still produced 233 seeds m^{-2} (Jhala et al., 2010). De Corby et al. (2007) demonstrated that volunteer wheat could escape tillage and/or preseeding herbicides and produce up to 54 seeds m^{-2} in the following flax crop. When volunteers are GM, minimizing AP is a high priority to ensure coexistence with conventional crops.

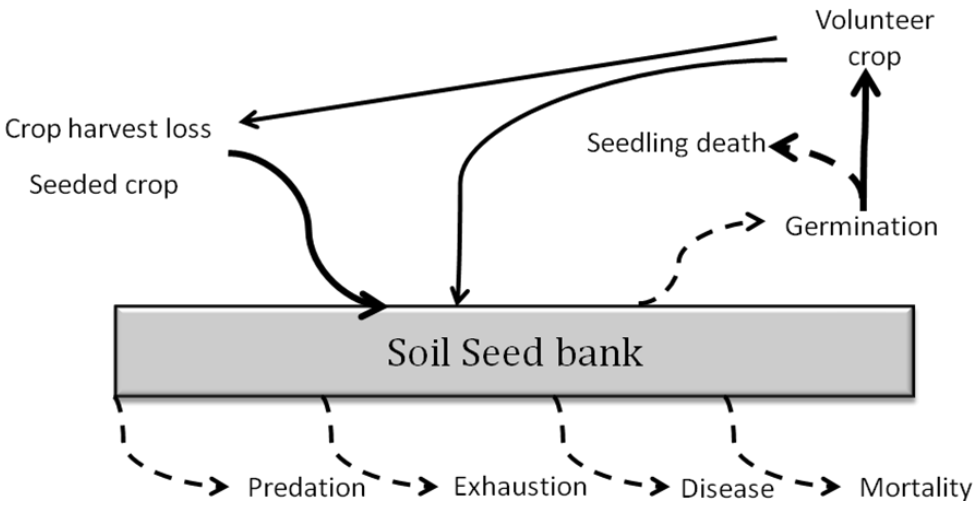

Figure 2.1. Crop seeds in the soil seed bank. Seeds enter the soil seed bank at harvest and through shatter and harvest losses and can remain on the surface exposed to seed predators or become buried. Seeds exit the seed bank through predation, disease, exhaustion, and mortality and through germination. Germinated seeds become volunteers in subsequent crops. Figure adapted from Nielson et al., 2009.

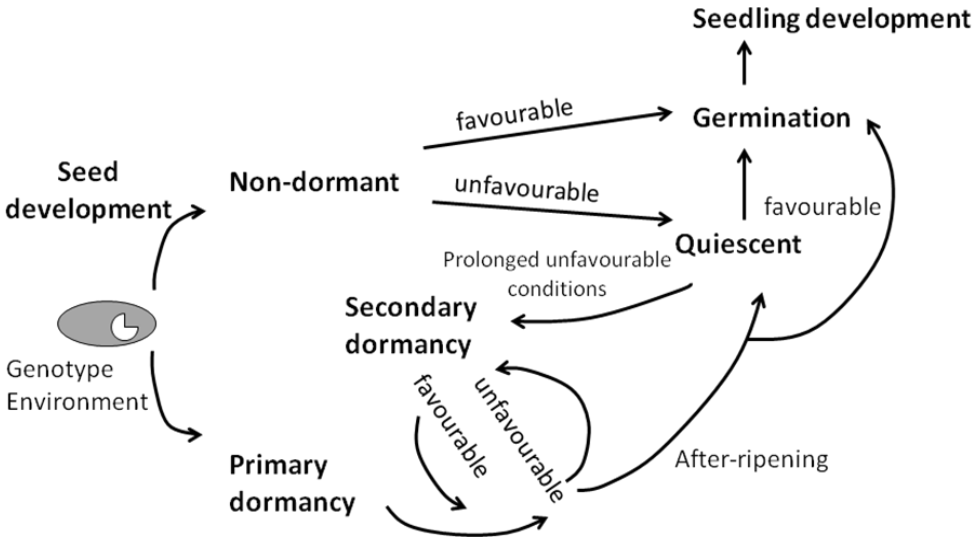

Figure 2.2. Seed dormancy cycling. Following maturity, seeds may have primary dormancy or be non-dormant. If dormant, they may after-ripen and gain the ability to germinate or may enter secondary dormancy in a cyclic environmentally-dependent fashion. Non-dormant seeds require a microsite with appropriate environmental conditions to permit germination. Figure adapted from Foley, 2001.

Table 2.1. Triticale area harvested globally in 2010. (Information taken from FAOSTAT, 2012).

Country	Triticale harvested area (ha)
Poland	1,258,700
Belarus	425,103
Germany	404,400
France	382,000
Australia	334,200
China	200,000
Russian Federation	140,700
Hungary	119,500
Lithuania	110,800
Spain	64,000
Austria	47,795
Brazil	46,602
Czech Republic	45,900
Romania	40,677
Sweden	37,400
Denmark	36,500
Serbia	36,274
Turkey	26,844
Portugal	24,500
Canada	22,200
Chile	20,963
United Kingdom	17,000
Bosnia and Herzegovina	11,299
Latvia	10,700
Switzerland	10,299
Tunisia	9,900
Bugaria	9,800
Slovakia	9,800
Belgium	6,666
Luxembourg	4,780
Estonia	3,900
Sovenia	3,477
Netherlands	2,676
Mexico	723
Total	3,926,078

2.3 Literature cited

[CFIA] Canadian Food Inspection Agency. 2011. Companion documents for Directive 94-08, assessment criteria for determining environmental safety of plants with novel traits. Available at http://www.inspection.gc.ca/plants/plants-with-novel-traits/applicants/directive-94-08/eng/1304475469806/1304475550733 (verified November 15, 2011). Plant Biosafety Office, Government of Canada, Ottawa, Ontario, Canada.

[CFIA] Canadian Food Inspection Agency. 2010. Safety assessment process for novel foods and agricultural products of biotechnology. Available at www.inspection.gc.ca/english/sci/biotech/reg/proce.shtml (verified November 15, 2011). Government of Canada, Ottawa, Ontario, Canada.

Abt, K.F. and W.F. Bock. 1998. Seasonal variations of diet composition in farmland field mice *Apodemus* spp. and bank voles *Clethrionomys glareolus*. Acta Theriol. 43:379-389.

Alaru, M., U. Laur and E. Lauringson. 2008. Pre-harvest sprouting tolerance of different winter triticale cultivars in the Baltic Sea area. Acta Agriculturae Scandinavica Section B, Plant Soil Sci. 58:11-16.

Alberta Agriculture, Food and Rural Development. 2005. Triticale production and utilization manual 2005: spring and winter triticale for grain, forage and value-added. Alberta Agriculture, Food and Rural Development, Edmonton, Alberta, Canada.

Alberta Agriculture, Food and Rural Development. 2001. Wheat midge. Available at http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/agdex2507 (verified March 12, 2012). Alberta Agriculture, Food and Rural Development, Edmonton, Alberta, Canada.

Allen, P.S., R. Benech-Arnold, D. Batlla and K.J. Bradford. 2007. Modeling of seed dormancy. p. 73-112. *In* K.J. Bradford and H. Nonogaki (eds.) Seed development, dormancy and germination. Blackwell Publishing Limited, Ames, Iowa.

Ammar, K., M. Mergoum and S. Rajaram. 2004. The history and evolution of triticale. p. 1-9. *In* M. Mergoum and H. Gomez-Macpherson (eds.) Triticale improvement and production. FAO Plant Production and Protection Paper: 179 ed. Food and Agriculture Organization of the United States, Rome, Italy.

Anderson, J.D. and J.E. Baker. 1983. Deterioration of seeds during aging. Phytopathol. 73:321-325.

Anderson, R.L. and G. Soper. 2003. Review of volunteer wheat (*Triticum aestivum*) seedling emergence and seed longevity in soil. Weed Tech. 17:620-626.

Anderson, R.L. and D.C. Nielsen. 1996. Emergence pattern of five weeds in the central Great Plains. Weed Tech. 10:744-749.

Andreoli, C., M.C. Bassoi and D. Brunetta. 2006. Genetic control of seed dormancy and preharvest sprouting in wheat. Scientia Agricola 63:564-566.

Baker, H.G. 1989. Some aspects of the natural history of seed banks. p. 9-21. *In* M.A. Leck, V.T. Parker and R.L. Simpson (eds.) Ecology of soil seed bank. 1st ed. Academic Press, San Diego, California, USA.

Baker, J. and C. Preston. 2008. Canola (*Brassica napus* L.) seedbank declines rapidly in farmer-managed fields in South Australia. Aust. J. Agric. Res. 59:780-784.

Bakker, J.P., P. Poschlod, R.J. Strykstra, R.M. Bekker and K. Thompson. 1996. Seed banks and seed dispersal: important topics in restoration ecology. Acta Bot. Neerl. 45:461-490.

Baraibar, B., D. Daedlow, F. De Mol and B. Gerowitt. 2012. Density dependence of weed seed predation by invertebrates and vertebrates in winter wheat. Weed Res. 52:79-87.

Baraibar, B., P.R. Westerman and J. Recasens. 2009. Effects of tillage and irrigation in cereal fields on weed seed removal by seed predators. J. Appl. Ecol. 46:380-387.

Barroso, J., L. Navarrete, M.J.S. Del Arco, C. Fernandez-Quintanilla, P.J.W. Lutman, N.H. Perry and R.I. Hull. 2006. Dispersal of *Avena fatua* and *Avena sterilis* patches by natural dissemination, soil tillage and combine harvesters. Weed Res. 46:118-128.

Baskin, J.M. and C.C. Baskin. 1989. Physiology of dormancy and germination in relation to seed bank ecology. p. 53-66. *In* M.A. Leck, V.T. Parker and R.L. Simpson (eds.) Ecology of soil seed banks. Academic Press, San Diego, California, USA.

Baskin, J.M. and C.C. Baskin. 1998. Seeds: ecology, biogeography, and evolution of dormancy and germination. Academic Press, San Diego, CA, USA.

Baskin, J.M. and C.C. Baskin. 1985. The annual dormancy cycle in buried weed seeds: a continuum. Bioscience 35:492-498.

Beckie, H.J. and L.M. Hall. 2008. Simple to complex: modelling crop pollen-mediated gene flow. Plant Sci. 175:615-628.

Beckie, H.J., S.I. Warwick, C.A. Sauder, L.M. Hall, K.N. Harker and C. Lozinski. 2011. Pollen-mediated gene flow in commercial fields of spring wheat in Western Canada. Crop Sci. 51:306-313.

Belz, R.G. 2007. Allelopathy in crop/weed interactions-an update. Pest Manag. Sci. 63:308-326.

Benech-Arnold, R.L. 2004. Inception, maintenance, and termination of dormancy in grain crops: physiology, genetics, and environmental control. p. 169-198. *In* R.L. Benech-Arnold and R.A. Sanchez (eds.) Handbook of seed physiology: applications to agriculture. 1st ed. The Haworth Press, New York, USA.

Benech-Arnold, R.L., R.A. Sanchez, F. Forcella, B.C. Kruk and C.M. Ghersa. 2000. Environmental control of dormancy in weed seed banks in soil. Field Crops Res. 67:105-122.

Benvenuti, S. 2007. Natural weed seed burial: effect of soil texture, rain and seed characteristics. Seed Sci. Res. 17:211-219.

Benvenuti, S. and M. Macchia. 1995. Effect of hypoxia on buried weed seed germination. Weed Res. 35:343-351.

Benvenuti, S., M. Macchia and S. Miele. 2001. Quantitative analysis of emergence of seedlings from buried weed seeds with increasing soil depth. Weed Sci. 49:528-535.

Beres, B.L., R.E. Blackshaw, R.J. Graf, E. Bremer, K.N. Harker and G.W. Clayton. 2010. Weed-competitive ability of spring and winter cereals in the northern Great Plains. Weed Tech. 24:108–116.

Biddulph, T.B., J.A. Plummer, T.L. Setter and D.J. Mares. 2008. Seasonal conditions influence dormancy and preharvest sprouting tolerance of wheat (*Triticum aestivum* L.) in the field. Field Crops Res. 107:116-128.

Biddulph, T.B., J.A. Plummer, T.L. Setter and D.J. Mares. 2007. Influence of high temperature and terminal moisture stress on dormancy in wheat (*Triticum aestivum* L.). Field Crops Res. 103:139-153.

Biddulph, T.B., T.L. Setter, J.A. Plummer and D.J. Mares. 2005. Drought and high temperature increases preharvest sprouting tolerance in a genotype without grain dormancy. Euphytica: International Journal of Plant Breeding 143:277-283.

Blackshaw, R.E. and K.N. Harker. 1997. Scentless chamomile (*Matricaria perforata*) growth, development, and seed production. Weed Sci. 45:701-705.

Blackshaw, R.E., L.J. Molnar and F.J. Larney. 2005. Fertilizer, manure and compost effects on weed growth and competition with winter wheat in western Canada. Crop Protection 24:971-980.

Blackshaw, R.E., L.J. Molnar and H.H. Janzen. 2004. Nitrogen fertilizer timing and application method affects weed growth and competition with spring wheat. Weed Sci. 52:614-622.

Blackshaw, R.E., R.N. Brandt and T. Entz. 2002. Soil temperature and soil water effects on henbit emergence. Weed Sci. 50:494-497.

Blackshaw, R.E., K.N. Harker, G.W. Clayton and J.T. O'Donovan. 2006. Broadleaf herbicide effects on clethodim and quizalofop-P efficacy on volunteer wheat (*Triticum aestivum*). Weed Tech. 20:221-226.

Blackshaw, R.E., F.J. Larney, C.W. Lindwall, P.R. Watson and D.A. Derksen. 2001. Tillage intensity and crop rotation affect weed community dynamics in a winter wheat cropping system. Can. J. Plant Sci. 81:805-813.

Blaney, C.S. and P.M. Kotanen. 2001. Post-dispersal losses to seed predators: an experimental comparison of native and exotic old field plants. Can. J. Bot. 79:284-292.

Boyd, N. and R. Van Acker. 2004. Seed and microsite limitations to emergence of four annual weed species. Weed Sci. 52:571-577.

Boyd, N. and R. Van Acker. 2004. Seed germination of common weed species as affected by oxygen concentration, light, and osmotic potential. Weed Sci. 52:589-596.

Boyd, N.S. and R.C. Van Acker. 2003. The effects of depth and fluctuating soil moisture on the emergence of eight annual and six perennial plant species. Weed Sci. 51:725-730.

Boyd, N.S. and S. White. 2009. Impact of wild blueberry harvesters on weed weed dispersal within and between fields. Weed Sci. 57:541-546.

Bradford, K.J. 2002. Applications of hydrothermal time to quantifying and modeling seed germination and dormancy. Weed Sci. 50:248-260.

Brook, H. (ed.) 2007. Crop protection 2008. Alberta Agriculture and Food, Agdex 606-1, Edmonton, Alberta, Canada.

Brust, G.E. and G.J. House. 1988. Weed seed destruction by arthropods and rodents in low-input soybean agroecosystems. Am. J. Alternative Agric. 3:19-25.

Buhler, D.D. 1995. Infleunce of tillage systems on weed population dynamics and management in Corn and Soybean in the central USA. Crop Sci. 35:1247-1258.

Buraas, T. and H. Skinnes. 1985. Development of seed dormancy in barley, wheat and triticale under controlled conditions. Acta Agric. Scand. 35:233-244.

Burger, J.C. and N.C. Ellstrand. 2005. Feral rye-evolutionary origins of a weed. p. 175-192. *In* J. Gressel (ed.) Crop ferality and volunteerism: a threat to food security in the transgenic era? Taylor & Francis, Taylor & Francis Group, Boca Raton, FL, USA.

Burger, J.C., J.M. Holt and N.C. Ellstrand. 2007. Rapid phenotypic divergence of feral rye from domesticated cereal rye. Weed Sci. 55:204-211.

Burnside, O.C., R.G. Wilson, S. Weisberg and K.G. Hubbard. 1996. Seed longevity of 41 weed species buried 17 years in eastern and western Nebraska. Weed Sci. 44:74-86.

Canadian Grain Commission. 2011. Official grain grading guide: triticale-chapter 8. Available at http://www.grainscanada.gc.ca/oggg-gocg/08/oggg-gocg-8-eng.htm (verified Mar. 12, 2012).

Canadian Triticale Biorefinery Initiative. 2010. Canadian triticale biorefinery initiative-home. Available at http://www.ctbi.ca (verified Mar. 12, 2012). CTBI, Edmonton, Alberta, Canada.

Canadian Triticale Biorefinery Initiative. 2011. Canadian triticale biorefinery intiative: biotech applications. Available at http://www.ctbi.ca/index.php?option=com_content&view=article&id=15&Itemid=119 (verified Mar. 12, 2012). CTBI, Edmonton, Alberta, Canada.

Cardina, J. and D.H. Sparrow. 1996. A comparison of methods to predict weed seedling populations from the soil seedbank. Weed Sci. 44:46-51.

Cardina, J., C.P. Herms and D.J. Doohan. 2002. Crop rotation and tillage system effects on weed seedbanks. Weed Sci. 50:448-460.

Cavers, P.B. and D.L. Benoit. 1989. Seed banks in arable land. p. 309-328. *In* M.A. Leck, V.T. Parker and R.L. Simpson (eds.) Ecology of soil seed bank. 1st ed. Academic Press, San Diego, California, USA.

Chambers, J.C. and J.A. MacMahon. 1994. A day in the life of a seed: movements and fates of seeds and their implications for natural and managed systems. Annu. Rev. Ecol. Syst.263-292.

Chauhan, B.S., G. Gill and C. Preston. 2006a. Influence of environmental factors on seed germination and seedling emergence of rigid ryegrass (*Lolium rigidum*). Weed Sci. 54:1004-1012.

Chauhan, B.S., G. Gill and C. Preston. 2006b. Influence of environmental factors on seed germination and seedling emergence of Oriental mustard (*Sisymbrium orientale*). Weed Sci. 54:1025-1031.

Chee-Sanford, J.C., M.M. Williams, A.S. Davis and G.K. Sims. 2006. Do microorganisms influence seed-bank dynamics? Weed Sci. 54:575-587.

Cherry, J.P. 1983. Protein degradation during seed deterioration *Aspergillus parasiticus* or *Aspergillus oryzae* infection of *Arachis hypogaea*, peanuts. Phytopathol. 73:317-321.

Clarke, J.M. 1985. Harvesting losses of spring wheat in windrower/combine and direct combine harvesting systems. Agron. J. 77:13-17.

Colbach, N., F. Forcella and G.A. Johnson. 2000. Spatial and temporal stability of weed populations over five years. Weed Sci. 48:366-377.

Conn, J.S., K.L. Beattie and A. Blanchard. 2006. Seed viability and dormancy of 17 weed species after 19.7 years of burial in Alaska. Weed Sci. 54:464-470.

Conner, A.J., T.R. Glare and J.P. Nap. 2003. The release of genetically modified crops into the environment - Part II. Overview of ecological risk assessment. Plant J. 33:19-46.

Crawley, M.J. 2000. Seed predators and plant population dynamics. p. 167-182. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Cummings, J.L., L.W. Handley, B. MacBryde, S.K. Tupper, S.J. Werner and Z.J. Byram. 2008. Dispersal of viable row-crop seeds of commercial agriculture

by farmland birds: implication for genetically modified crops. Environmental Biosafety Research 7:241-252.

Davies, M.S. and G.C. Hillman. 1992. Domestication of cereals. p. 199-224. *In* G.P. Chapman (ed.) Grass evolution and domestication. Cambridge University Press, Cambridge, UK.

Davis, A.S., K.A. Renner, J. Iannuzzi and B.J. Schutte. 2008. Chemical and physical defense of weed seeds in relation to soil seedbank persistence. Weed Sci. 56:676-684.

De Cauwer, B., D. Reheul, I. Nijs and A. Milbau. 2008. Management of newly established field margins on nutrient-rich soil to reduce weed spread and seed rain into adjacent crops. Weed Res. 48:102-112.

De Corby, K.A., R.C. Van Acker, A. Brûlé-Babel and L.F. Friesen. 2007. Emergence timing and recruitment of volunteer spring wheat. Weed Sci. 55:60-69.

Debeaujon, I., L. Lepiniec, L. Pourcel and J. Routaboul. 2007. Seed coat development and dormancy. p. 25-49. *In* K.J. Bradford and H. Nonogaki (eds.) Seed development, dormancy and germination. Blackwell Publishing Limited, Ames, Iowa, USA.

DeSousa, N., J.T. Griffiths and C.J. Swanton. 2003. Predispersal seed predation of redroot pigweed (*Amaranthus retroflexus*). Weed Sci. 51:60-68.

Devos Y., Demont M.,Sanvido O. 2009. Coexistence of genetically modified (GM) and non-GM crops in the European Union: A review. Agron. Sustain. Dev. 29:11-30.

Dexter, J.E., A.J. Jhala, R.C. Yang, M.J. Hills and Weselake, R.J. and Hall, L.M. 2011. Harvest loss and seed bank longevity of flax (*Linum usitatissimum* L.): Implications for seed-mediated gene flow. Weed Sci. 59:61-67.

Ding, H., R.J. Lamb and N. Ames. 2000. Inducible production of phenolic acids in wheat and antibiotic resistance to *Sitodiplosis mosellana*. J. Chem. Ecol. 26:969-985.

DiTomaso, J.M., J.N. Barney and A.M. Fox. 2007. Biofuel feedstocks: The risk of future invasions. Rep. QTA2007-1. CAST, Ames, Iowa, USA.

Dyer, W.E. 1995. Exploiting weed seed dormancy and germination requirements through agronomic practices. Weed Sci. 43:498-503.

Eudes, F. 2006. Canadian triticale biorefinery initiative. p. 85-88. *In* W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G. Oettler and D. Salmon (eds.) Proc. 6th International Triticale Symp., Stellenbosch, South Africa. 3-7 September 2006 Faculty of AgriSciences, Stellenbosch, South Africa.

European Union. 2003. Regulation (EC) No 1830/2003 of the European parliament and of the council. Available at http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2003:268:0024:0028:EN:PDF (verified Mar. 12, 2012). Official Journal of the European Union 1830/2003:L 268/24-L 268/28. Brussels, Belgium.

FAOSTAT. 2012. Food and Agricultural Organization Statistical Database. Available at http://faostat.fao.org/site/567/DesktopDefault.aspx?PageID=567#ancor (accessed Feb 11, 2012). Rome, Italy.

Fenner, M. 1987. Seedlings. New Phytol. 106:35-47.

Finkelstein, R., C. Steber, T. Ariizumi and W. Reeves. 2008. Molecular aspects of seed dormancy. Ann. Rev. Plant Biology 59:387-415.

Flintham, J.E. and S.J. Humphray. 1993. Red coat genes and wheat dormancy. Asp. Appl. Biology 36:135-141.

Flintham, J.E. and M.D. Gale. 1990. Breeding "sprout-proof" wheats: risks and resources. Asp. Appl. Biology 25:69-78.

Foley, M. 2001. Seed dormancy: An update on terminology, physiological genetics, and quantitative trait loci regulating germinability. Weed Sci. 49:305-317.

Forcella, F., R.L. Benech Arnold, R. Sanchez and C.M. Ghersa. 2000. Modeling seedling emergence. Field Crops Res. 67:123-139.

Forcella, F., R.G. Wilson, K.A. Renner, J. Dekker, R.G. Harvey, D.A. Alm, D.D. Buhler and J. Cardina. 1992. Weed seedbanks of the U.S. corn belt: magnitude, variation, emergence, and application. Weed Sci. 40:636-644.

Forget, P.M. 1996. Removal of seeds of *Carapa procera* (Meliaceae) by rodents and their fate in rainforest in French Guiana. J. Trop. Ecol. 12:751-761.

Forsyth, S.F. and A.K. Watson. 1985. Predispersal seed predation of Canada thistle. Can. Entomol. 117:1075-1081.

Foster, S.A. 1986. On the adaptive value of large seeds for tropical moist forest trees: a review and synthesis. Bot. Rev. 52:260-299.

Gaines, T.A., P.F. Byrne, P. Westra, S.J. Nissen, W.B. Henry, D.L. Shaner and P.L. Chapman. 2007. An empirically derived model of field-scale gene flow in winter wheat. Crop Sci. 47:2308-2316.

Gallandt, E.R., E.P. Fuerst and A.C. Kennedy. 2004. Effect of tillage, fungicide seed treatment, and soil fumigation on seed bank dynamics of wild oat (*Avena fatua*). Weed Sci. 52:597-604.

Gealy, D.R., K.J. Bradford, L.M. Hall, R. Hellmich, A. Raybould, J.D. Wolt and D. Zilberman. 2007. Implications of gene flow in the scale-up and commercial use of biotechnology-derived crops: Economic and policy considerations. CAST Issue Paper 37:1-24.

Gilbert, G.S. 2002. Evolutionary ecology of plant diseases in natural ecosystems. Annu. Rev. Phytopathol. 40:13-43.

Goyal, A., B.L. Beres, H.S. Randhawa, A. Navabi, D.F. Salmon and F. Eudes. 2011. Yield stability analysis of broadly adaptive triticale germplasm in southern and central Alberta, Canada, for industrial end-use stability. Can. J. Plant Sci. 91:125-135.

Graziani, F., E. Pannacci, G. Covarelli and F. Tei. 2007. Post-dispersal weed seed predation by vertebrates and invertebrates in organic and conventional cereal fields. p. 196. *In* E. Fløistad (ed.) European Weed Research Society, 14th EWRS Symposium, Hamar, Norway, 17-21 June 2007. Doorwerth, Netherlands.

Gressel, J. 2005a. Introduction-the challenges of ferality. p. 1-7. *In* J. Gressel (ed.) Crop ferality and volunteerism: A threat to food security in the transgenic era? Taylor & Francis, Taylor & Francis Group, Boca Raton, Florida, USA.

Gressel, J. 2005b. Crop ferality and volunteerism. Taylor & Francis, Taylor & Francis Group, Boca Raton, Florida, USA.

Groos, C., G. Gay, M.R. Perretant, L. Gervais, M. Bernard, F. Dedryver and G. Charmet. 2002. Study of the relationship between pre-harvest sprouting and grain color by quantitative trait loci analysis in a white x red grain bread-wheat cross. Theor. Appl. Genet. 104:39-47.

Gruber, S. and W. Claupein. 2007. Fecundity of volunteer oilseed rape and estimation of potential gene dispersal by a practice-related model. Agric. Ecosyst. Environ. 119:401-408.

Grundy, A.C. and A. Mead. 2000. Modeling weed emergence as a function of meteorological records. Weed Sci. 48:594-603.

Grundy, A.C., A. Mead and S. Burston. 2003. Modelling the emergence response of weed seeds to burial depth: interactions with seed density, weight and shape. J. Appl. Ecol. 40:757-770.

Grundy, A.C., A. Mead and W. Bond. 1996. Modelling the effect of weed-seed distribution in the soil profile on seedling emergence. Weed Res. 36:375-384.

Gubler, F., A.A. Millar and J.V. Jacobsen. 2005. Dormancy release, ABA and pre-harvest sprouting. Curr. Opin. Plant Biol. 8:183-187.

Gulden, R.H., A.G. Thomas and S.J. Shirtliffe. 2004a. Secondary dormancy, temperature, and burial depth regulate seedbank dynamics in canola. Weed Sci. 52:382-388.

Gulden, R.H., A.G. Thomas and S.J. Shirtliffe. 2004b. Relative contribution of genotype, seed size and environment to secondary seed dormancy potential in Canadian spring oilseed rape (*Brassica napus*). Weed Res. 44:97-106.

Gulden, R.H., S.J. Shirtliffe and A.G. Thomas. 2003a. Harvest losses of canola (*Brassica napus*) cause large seedbank inputs. Weed Sci. 51:83-86.

Gulden, R.H., S.J. Shirtliffe and A.G. Thomas. 2003b. Secondary seed dormancy prolongs persistence of volunteer canola in western Canada. Weed Sci. 51:904-913.

Gustafson, D.I., M.J. Horak, C.B. Rempel, S.G. Metz, D.R. Gigax and P. Hucl. 2005. An empirical model for pollen-mediated gene flow in wheat. Crop Sci. 45:1286-1294.

Gutterman, Y. 2000. Maternal effects on seeds during development. p. 59-84. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Hacault, K.M. and R.C. Van Acker. 2006. Emergence timing and control of dandelion (*Taraxacum officinale*) in spring wheat. Weed Sci. 54:172-181.

Hall, L., K. Topinka, J. Huffman, L. Davis and A. Good. 2000. Pollen flow between herbicide-resistant Brassica Napus is the cause of multiple-resistant *B. napus* volunteers. Weed Sci. 48:688-694.

Halloin, J.M. 1983. Deterioration resistance mechanisms in seeds. Phytopathol. 73:335-339.

Harker, K.N., R.E. Blackshaw and G.W. Clayton. 2001. Timing weed removal in field pea (*Pisum sativum* L.). Weed Tech. 15:277-283.

Harker, K.N., T.K. Turkington, G.W. Clayton, J.T. O'Donovan and R.B. Irvine. 2009. Integrating cropping systems with cultural techniques augments wild oat (*Avena fatua*) management in barley. Weed Sci. 57:326-337.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y. Gan, F.A. Holm, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2006. Persistence of glyphosate-resistant canola in western Canadian cropping systems. Agron. J. 98:107-119.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y.T. Gan, H.A. Frederick, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2005. Glyphosate-resistant wheat persistence in western Canadian cropping systems. Weed Sci. 53:846-859.

Harker, K.N., J.T. O'Donovan, G.W. Clayton and J. Mayko. 2008. Field-scale time of weed removal in canola. Weed Tech. 22:747-749.

Harlan, J.R. 1992. Origins and processes of domestication. p. 159-175. *In* G.P. Chapman (ed.) Grass evolution and domestication. Cambridge University Press, Cambridge, UK.

Harman, G.E. 1983. Mechanisms of seed infection and pathogenesis. Phytopathol. 73:326-329.

Heggenstaller, A.H., F.D. Menalled, M. Liebman and P.R. Westerman. 2006. Seasonal patterns in post-dispersal seed predation of *Abutilon theophrasti* and *Setaria faberi* in three cropping systems. J. Appl. Ecol. 43:999-1010.

Hendry, G.A.F. 1993. Oxygen, free radical processes and seed longevity. Seed Science Research 3:141-153.

Hilhorst, H.W.M. and C.M. Karssen. 2000. Effect of chemical environment on seed germination. p. 293-309. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Hilhorst, H.W.M. and P.E. Toorop. 1997. Review on dormancy, germinability, and germination in crop and weed seeds. Adv. Agron.61:111-165.

Hilhorst, H.W.M. 2007. Definitions and hypotheses of seed dormancy. p. 50-71. *In* K.J. Bradford and H. Nonogaki (eds.) Seed development, dormancy and germination. Blackwell Publishing Limited, Ames, Iowa, USA.

Hills, M.J., L. Hall, P.G. Arnison and A.G. Good. 2007a. Genetic use restriction technologies (GURTs): strategies to impede transgene movement. Trends Plant Sci. 12:177-183.

Hills, M.J., L.M. Hall, D.F. Messenger, R.J. Graf and B.L. Beres. 2007b. Evaluation of crossability between triticale (X *Triticosecale* Wittmack) and common wheat, durum wheat and rye. Environ. Biosafety Res. 6:249-257.

Himi, E., D.J. Mares, A. Yanagisawa and K. Noda. 2002. Effect of grain colour gene (R) on grain dormancy and sensitivity of the embryo to abscisic acid (ABA) in wheat. J. Exp. Bot. 53:1569-1574.

Holman, J.D., A.J. Bussan, B.D. Maxwell, P.R. Miller and J.A. Mickelson. 2006. Persian darnel (*Lolium persicum*) fecundity response to spring wheat, canola, and sunflower interference. Weed Tech. 20:430-437.

Holmes, R.J. and R.J. Froud-Williams. 2005. Post-dispersal weed seed predation by avian and non-avian predators. Agric. Ecosyst. Environ. 105:23-27.

Hucl, P., M.A. Matus-Cadiz, A.S. Sahota, A. Middleton, D. Mooney and J.L. Maruschak. 2004. Sources of off-types in pedigreed seed of common spring wheat. Can. J. Plant Sci. 84:519-523.

Hulme, P.E. 1996. Herbivory, plant regeneration, and species coexistence. J. Ecol. 84:609-615.

Humston, R., D.A. Mortensen and O.N. Bjornstad. 2005. Anthropogenic forcing on the spatial dynamics of an agricultural weed: the case of the common sunflower. J. Appl. Ecol. 42:863-872.

Jacob, H., D. Minkey, R. Gallagher and C. Borger. 2006. Variation in postdispersal weed seed predation in a crop field. Weed Sci. 54:148-155.

James, C. 2010. Global Status of Commercialized Biotech/GM Crops: 2010. ISAAA Brief No. 42. ISAAA, Ithaca, New York, USA.

Jensen, S.P. 1993. Temporal changes in food preferences of wood mice (*Apodemus sylvaticus* L.). Oecologia 94:76-82.

Jhala, A.J., L.L. Raatz, J.E. Dexter and L.M. Hall. 2010. Adventitious presence: Volunteer flax (*Linum usitatissimum* L.) in herbicide resistant canola (*Brassica napus* L.). Weed Tech. 24:244-252.

Kalaitzandonakes, N. 2011. The economic impacts of asynchronous authorizations and low level presence: An overview. International Food and Agricultural Trade Policy Council, Washington, D.C., USA.

Kato, K., W. Nakamura, T. Tabiki, H. Miura and S. Sawada. 2001. Detection of loci controlling seed dormancy on group 4 chromosomes of wheat and comparative mapping with rice and barley genomes. Theor. Appl. Genet. 102:980-985.

Kavanagh, V.B., J.C. Hall and L.M. Hall. 2010. Potential hybridization of genetically engineered triticale with wild and weedy relatives in Canada. Crop Sci. 50:1128-1140.

Kermode, A.R. 1990. Regulatory mechanisms involved in the transition from seed development to germination. Crit. Rev. Plant Sci. 9:155-195.

Kershen, D.L. and A. McHughen. 2005. Adventitious presence: Inadvertent commingling and coexistence among farming methods. CAST Commentary QTA 2005-1:1-4.

Kremer, R.J. 1993. Management of weed seed banks with microorganisms. Ecol. Appl. 3:42-52.

Kremer, R.J., L.B.J. Hughes and R.J. Aldrich. 1984. Examination of microorganisms and deterioration resistance mechanisms associated with velvetleaf seed. Agron. J. 76:745-749.

Kruk, B.C. and R.L. Benech Arnold. 1998. Functional and quantitative analysis of seed thermal responses in prostrate knotweed (*Polygonum aviculare*) and common purslane (*Portulaca oleracea*). Weed Sci. 46:83-90.

Lawson, A.N., L.F. Friesen and R.C. Van Acker. 2006. Emergence timing of volunteer canola in spring wheat fields in Manitoba. Weed Sci. 54:873-882.

Leck, M.A., V.T. Parker and R.L. Simpson (eds.) 1989. Ecology of soil seed banks. Academic Press, Inc., San Diego, California, USA.

Leeson, J.Y., A.G. Thomas and L.M. Hall. 2002. Alberta weed survey of cereal, oilseed and pulse crops in 2001. Weed Survey Series Publication 02-1. Agriculture and Agri-Food Canada, Saskatoon, Saskatchewan, Canada.

Leeson, J.Y., A.G. Thomas, L.M. Hall, C.A. Brenzil, T. Andrews, K.R. Brown and R.C. Van Acker. 2005. Prairie weed surveys of cereal, oilseed and pulse crops from the 1970s to the 2000s. Rep. Weed Survey Series Publication 05-1CD. Agriculture and Agri-Food Canada, Saskatoon Research Centre, Saskatoon, Saskatchewan, Canada.

Légère, A., F.C. Stevenson and D.L. Benoit. 2011. The selective memory of weed seedbanks after 18 years of conservation tillage. Weed Sci. 59:98-106.

Levin, D.A. 1990. The seed bank as a source of genetic novelty in plants. Am. Nat. 135:563-572.

Lewis, J. 1973. Longevity of crop and weed seeds: survival after 20 years in soil. Weed Res. 13:179-191.

Leymarie, J., R. Benech-Arnold, F. Corbineau, M. Robayo-Romero and E. Gendreau. 2008. Involvement of ABA in induction of secondary dormancy in barley (*Hordeum vulgare* L.) seeds. Plant Cell Physiol. 49:1830-1838.

Louda, S.M. 1989. Predation in the dynamics of seed regeneration. p. 25-52. *In* M.A. Leck, V.T. Parker and R.L. Simpson (eds.) Ecology of soil seed banks. Academic Press, San Diego, California, USA.

Lundgren, J. 2005. Ground beetles as weed control agents: effects of farm management on granivory. Am. Entomol. 51:224-226.

Lundgren, J.G. and K.A. Rosentrater. 2007. The strength of seeds and their destruction by granivorous insects. Arthropod-Plant Interactions 1:93-99.

Lutman, P.J.W., S.E. Freeman and C. Pekrun. 2003. The long-term persistence of seeds of oilseed rape (*Brassica napus*) in arable fields. J. Agric. Sci. 141:231-240.

Lyon, D.J., A.J. Bussan, J.O. Evans, C.A. Mallory-Smith and T.F. Peeper. 2002. Pest management implications of glyphosate-resistant wheat (*Triticum aestivum*) in the western United States. Weed Tech. 16:680-690.

Marino, P.C., K.L. Gross and D.A. Landis. 1997. Weed seed loss due to predation in Michigan maize fields. Agric. Ecosyst. Environ. 66:189-196.

Marino, P.C., P.R. Westerman, C. Pinkert and W. van der Werf. 2005. Influence of seed density and aggregation on post-dispersal weed seed predation in cereal fields. Agric. Ecosyst. Environ. 106:17-25.

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

Mauchline, A.L., S.J. Watson, V.K. Brown and R.J. Froud Williams. 2005. Post-dispersal seed predation of non-target weeds in arable crops. Weed Res. 45:157-164.

May, W., G. Lafond, E. Johnson, T. Hogg, A. Johnston, B. Nybo, N. Harker and G. Clayton. 2003. An assessment of the concept of early time of weed removal in field pea using natural weed populations. Can. J. Plant Sci. 83:423-431.

McCanny, S.J. and P.B. Cavers. 1988. Spread of proso millet (*Panicum miliaceum* L.) in Ontario, Canada. II. Dispersal by combines. Weed Res. 28:67-72.

McGee, D.C. 1995. Epidemiological approach to disease management through seed technology. Annu. Rev. Phytopathol.33:445-466.

McLeod, J.G., J.B. Thomas, P.D. Brown, C.L. Vera, W.E. May, D.F. Salmon and K. Sosulski. 2010. Changes in ethanol production potential due to species, cultivar and location on the Canadian prairie. Can. J. Plant Sci. 90:163-171.

McPherson, M.A., R.C. Yang, A.G. Good, R.L. Nielson and L.M. Hall. 2009. Potential for seed-mediated gene flow in agroecosystems from transgenic safflower (*Carthamus tinctorius* L.) intended for plant molecular farming. Transgenic Res. 18:281-299.

Medd, R.W. and M.A. Campbell. 2005. Grass seed infection following inundation with *Pyrenophora semeniperda*. Biocontrol Sci. Technol. 15:21-36.

Menalled, F.D., P.C. Marino, K.A. Renner and D.A. Landis. 2000. Post-dispersal weed seed predation in Michigan crop fields as a function of agricultural landscape structure. Agric. Ecosyst. Environ. 77:193-202.

Menalled, F.D., R.G. Smith, J.T. Dauer and T.B. Fox. 2007. Impact of agricultural management on carabid communities and weed seed predation. Agric. Ecosyst. Environ. 118:49-54.

Mergoum, M., W.H. Pfeiffer, R.J. Pena, K. Ammar and S. Rajaram. 2004. Triticale crop improvement: The CIMMYT programme. p. 11-26. *In* M. Mergoum and H. Gomez-Macpherson (eds.) Triticale improvement and production. FAO Plant Production and Protection Paper: 179 ed. Food and Agriculture Organization of the United States, Rome, Italy.

Meyer, S.E., T.E. Stewart and S. Clement. 2010. The quick and the deadly: growth vs virulence in a seed bank pathogen. New Phytol. 187:209-216.

Miller, S.D., C.M. Alford and W.L. Stump. 2004. Feral rye: A serious threat to high quality wheat. University of Wyoming, Cooperative Extension Service. Laramie, Wyoming, USA.

Murdoch, A.J. and R.H. Ellis. 2000. Dormancy, viability and longevity. p. 183-214. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Mutze, G. 2007. Does high growth rate of juvenile house mice with prolonged access to ripening grain and free water drive population outbreaks? N. Z. J. Zool. 34:195-202.

Nakamura, S. and T. Toyama. 2001. Isolation of a VP1 homologue from wheat and analysis of its expression in embryos of dormant and non-dormant cultivars. J. Exp. Bot. 52:875-876.

Nielson, R.L., R. Yang, L.M. Hall, K.N. Harker, M.A. McPherson and J.T. O'Donovan. 2009. Seed-mediated gene flow in wheat: Seed bank longevity in western Canada. Weed Sci. 57:124-132.

Nonogaki, H., J.D. Bewley and G.W. Bassel. 2010. Germination—Still a mystery. Plant Sci. 179:574-581.

Nonogaki, H., F. Chen and K.J. Bradford. 2007. Mechanisms and genes involved in germination *sensu stricto*. p. 265-304. *In* K.J. Bradford and H. Nonogaki (eds.) Seed development, dormancy and germination. Blackwell Publishing Limited, Ames, Iowa, USA.

Nurse, R.E., B.D. Booth and C.J. Swanton. 2003. Predispersal seed predation of *Amaranthus retroflexus* and *Chenopodium album* growing in soyabean fields. Weed Res. 43:260-268.

O'Donovan, J.T., K.N. Harker, G.W. Clayton, L.M. Hall, J. Cathcart, K.L. Sapsford, F.A. Holm and K. Hacault. 2007. Volunteer barley interference in spring wheat grown in a zero-tillage system. Weed Sci. 55:70-74.

Oettler, G. 2005. The fortune of a botanical curiosity - Triticale: past, present and future. J. Agric. Sci. 143:329-346.

Ogg, A.G. and J.H. Dawson. 1984. Time of emergence of 8 weed species. Weed Sci. 32:327-335.

Olfert, O., S. Hartley and R.H. Elliott. 2009. Non-native insects in agriculture: strategies to manage the economic and environmental impact of wheat midge, *Sitodiplosis mosellana*, in Saskatchewan. Biol. Invasions 11:127-133.

O'Rourke, M.E., M.E. Rice, M. Liebman and A.H. Heggenstaller. 2006. Post-dispersal weed seed predation by invertebrates in conventional and low-external-input crop rotation systems. Agric. Ecosyst. Environ. 116:280-288.

Oryokot, J.O.E., S.D. Murphy, A.G. Thomas and C.J. Swanton. 1997. Temperature- and moisture-dependent models of seed germination and

shoot elongation in green and redroot pigweed (*Amaranthus powellii*, *A. retroflexus*). Weed Sci. 45:488-496.

Pakeman, R.J., R.P. Cummins, G.R. Miller and D.B. Roy. 1999. Potential climatic control of seedbank density. Seed Science Research 9:101-110.

Paulsen, G.M. and A.S. Auld. 2004. Preharvest sprouting of cereals. p. 199-219. *In* R.L. Benech-Arnold and R.A. Sanchez (eds.) Handbook of seed physiology: applications to agriculture. 1st ed. The Haworth Press, New York, USA.

Peachey, R.E. and C. Mallory-Smith. 2007. Influence of winter seed position and recovery date on hairy nightshade (*Solanum sarrachoides*) recruitment and seed germination, dormancy, and mortality. Weed Sci. 55:49-59.

Pickett, A.A. 1989. A review of seed dormancy in self-sown wheat and barley. Plant Varieties and Seeds 2:131-146.

Probert, R.J. 2000. The role of temperature in the regulation of seed dormancy and germination. p. 261-292. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, UK.

Rainbolt, C.R., D.C. Thill and F.L. Young. 2004. Control of volunteer herbicide-resistant wheat and canola. Weed Tech. 18:711-718.

Ramessar, K., T. Capell, R.M. Twyman and P. Christou. 2010. Going to ridiculous lengths-European coexistence regulations for GM crops. Nat. Biotechnol. 28:133-136.

Rampton, H.H. and T.M. Ching. 1970. Persistence of crop seeds in soils. Agron. J. 62:272-277.

Reddy, L.V., R.J. Metzger and T.M. Ching. 1985. Effect of temperature on seed dormancy of wheat. Crop Sci. 25:455-458.

Rew, L.J. and G.W. Cussans. 1997. Horizontal movement of seeds following tine and plough cultivation: implications for spatial dynamics of weed infestations. Weed Res. 37:247-256.

Rew, L.J., R. Froud-Williams and N.D. Boatman. 1996. Dispersal of *Bromus sterilis* and *Anthriscus sylvestris* seed within arable field margins. Agric. Ecosyst. Environ. 59:107-114.

Saatkamp, A., P. Poschlod, T. Dutoit and L. Affre. 2009. The seed bank longevity index revisited: limited reliability evident from a burial experiment and database analyses. Annals of Botany 104:715-724.

Salmon, D.F., M. Mergoum and H. Gomez-Macpherson. 2004. Triticale production and management. p. 27-36. *In* M. Mergoum and H. Gomez-Macpherson (eds.) Triticale improvement and production. FAO Plant Production and Protection Paper: 179 ed. Food and Agriculture Organization of the United States, Rome, Italy.

Salmon, D.F. and J.H. Helm. 1985. Preharvest and postharvest dormancy in spring triticale. Agron. J. 77:649-652.

Schramm, E.C., K.G. Campbell, C.M. Steber, J.C. Abellera and L.C. Strader. 2010. Isolation of ABA-responsive mutants in allohexaploid bread wheat (*Triticum aestivum* L.): Drawing connections to grain dormancy, preharvest sprouting, and drought tolerance. Plant Sci. 179:620-629.

Schuurink, R.C., J.M.M.v. Beckum and F. Heidekamp. 1992. Modulation of grain dormancy in barley by variation of plant growth conditions. Hereditas 117:137-143.

Schweizer, E.E. and R.L. Zimdahl. 1984. Weed seed decline in irrigated soil after rotation of crops and herbicides. Weed Sci. 32:84-89.

Seefeldt, S.S., R. Zemetra, F.L. Young and S.S. Jones. 1998. Production of herbicide-resistant jointed goatgrass (*Aegilops cylindrica*) x wheat (*Triticum aestivum*) hybrids in the field by natural hybridization. Weed Sci. 46:632-634.

Seerey, N., S.J. Shirtliffe and P. Hucl. 2011. Seeds from unthreshed wheat (*Triticum aestivum* L.) spikes have reduced field emergence compared with threshed seed regardless of cultivar. Can. J. Plant Sci. 91:583-585.

Shirtliffe, S.J. and M.H. Entz. 2005. Chaff collection reduces seed dispersal of wild oat (*Avena fatua*) by a combine harvester. Weed Sci. 53:465-470.

Shirtliffe, S.J., M.H. Entz and R.C. Van Acker. 2000. *Avena fatua* development and seed shatter as related to thermal time. Weed Sci. 48:555-560.

Sikkema, P., Deen W. and S. Vyas. 2005. Weed control in pea with reduced rates of imazethapyr applied pre emergence and post emergence. Weed Tech. 19:14-18.

Simard, M., A. Legere, D. Pageau, J. Lajeunesse and S. Warwick. 2002. The frequency and persistence of volunteer canola (*Brassica napus*) in Québec cropping systems. Weed Tech. 16:433-439.

Simpson, R.L., M.A. Leck and V.T. Parker. 1989. Seed banks: General concepts and methodological issues. p. 3-8. *In* M.A. Leck, V.T. Parker and R.L. Simpson (eds.) Ecology of soil seed banks. Academic Press, Inc., San Diego, California, USA.

Simpson, G.M. 1990. Seed dormancy in grasses. Cambridge University Press, Cambridge, UK.

St. Angelo, A.J. and R.L. Ory. 1983. Lipid degradation during seed deterioration. Phytopathol. 73:315-317.

Stiles, E.W. 2000. Animals as seed dispersers. p. 111-124. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Stump, W.L. and P. Westra. 2000. The seedbank dynamics of feral rye (*Secale cereale*). Weed Tech. 14:7-14.

Swanton, C.J., J.T. Griffiths, H.E. Cromar and B.D. Booth. 1999. Pre- and post-dispersal weed seed predation and its implications to agriculture. p. 829-834. *In* Proc. of an international conference, 1999 Brighton crop protection conference: weeds, Brighton, UK, 15-18 Nov. 1999. Brighton, UK.

Telewski, F.W. and J.A.D. Zeevaart. 2002. The 120-yr period for Dr. Beal's seed viability experiment. Am. J. Bot. 89:1285-1288.

Tew, T.E., I.A. Todd and D.W. Macdonald. 2000. Arable habitat use by wood mice (*Apodemus sylvaticus*). 2. Microhabitat. J. Zool. 250:305-311.

Thompson, K. 2000. The functional ecology of soil seed banks. p. 215-235. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Thompson, K. 1987. Seeds and seed banks. New Phytol. (Suppl.) 106:23-34.

Thompson, K. and J.P. Grime. 1979. Seasonal variation in the seed banks of herbaceous species in ten contrasting habitats. J. Ecol. 67:893-921.

Toole, E.H. and E. Brown. 1946. Final results of the Duvel buried seed experiment. J. Ag. Res. 72:201-210.

Torada, A. and Y. Amano. 2002. Effect of seed coat color on seed dormancy in different environments. Euphytica: Netherlands J. Plant Breeding 126:99-105.

Upadhyay, B.M., E.G. Smith, G.W. Clayton, K.N. Harker and R.E. Blackshaw. 2006. Economics of integrated weed management in herbicide-resistant canola. Weed Sci. 54:138-147.

Van Acker, R.C., W.J. Bullied and M.J. Du Croix Sissons. 2004. Tillage index predicts weed seedling recruitment depth. Can. J. Plant Sci. 84:319-326.

Van Mourik, T.A., T.J. Stomph and A.J. Murdoch. 2005. Why high seed densities within buried mesh bags may overestimate depletion rates of soil seed banks. J. Appl. Ecol. 42:299-305.

VanderWall, S.B., K.M. Kuhn and M.J. Beck. 2005. Seed removal, seed predation, and secondary dispersal. Ecology 86:801-806.

Varughese, G., W.H. Pfeiffer and R.J. Pena. 1996. Triticale: A successful alternative crop (Part 1). Cereal Foods World 41:474-482.

Venable, D.L. and J.S. Brown. 1988. The selective interactions of dispersal, dormancy, and seed size as adaptations for reducing risk in variable environments. Am. Nat. 131:360-384.

Vera, C.L., S.D. Duguid, S.L. Fox, K.Y. Rashid, J.C.P. Dribnenki, and F.R. Clarke. 2012. Comparative effect of lodging on seed yield of flax and wheat. Can. J. Plant Sci. 92: 39-43.

Vleeshouwers, L.M. and H.J. Bouwmeester. 2001. A simulation model for seasonal changes in dormancy and germination of weed seeds. Seed Sci. Res. 11:77-92.

Walker-Simmons, M. 1988. Enhancement of ABA responsiveness in wheat embryos by high temperature. Plant, Cell and Environment 11:769-775.

Walker-Simmons, M. 1987. ABA levels and sensitivity in developing wheat embryos of sprouting resistant and susceptible cultivars. Plant Physiol. 84:61-66.

Wang, S., K.C. Thomas, W.M. Ingledew, K. Sosulski and F.W. Sosulski. 1997. Rye and triticale as feedstock for fuel ethanol production. Cereal Chem. 74:621-625.

Warham, E.J. 1990. Effect of Tilletia indica infection on viability, germination, and vigor of wheat seed. Plant Dis. 74:130-132.

Warwick, S.I. and C.N. Stewart Jr. 2005. Crops come from wild plants - how domestication, transgenes, and linkage together shape ferality. p. 9-25. *In* J.B. Gressel (ed.) Crop ferality and volunteerism. Taylor & Francis, Taylor & Francis Group, Boca Raton, Florida, USA.

Warwick, S.I., M.-. Simard, A. Légère, H.J. Beckie, L. Braun, B. Zhu, P. Mason, G. Séguin-Swartz and C.N. Stewart. 2003. Hybridization between transgenic *Brassica napus* L. and its wild relatives: *Brassica rapa* L., *Raphanus raphanistrum* L., *Sinapis arvensis* L., and *Erucastrum gallicum* (Willd.) O.E. Schulz. Theor. Appl. Genet. 107:528-539.

Westerman, P.R., M. Liebman and P.M. Dixon. 2009. Burial rates of surrogate seeds in arable fields. Weed Res. 49:142-152.

Westerman, P.R., M. Liebman, A.H. Heggenstaller and F. Forcella. 2006. Integrating measurements of seed availability and removal to estimate weed seed losses due to predation. Weed Sci. 54:566-574.

Westerman, P.R., J.S. Wes, M.J. Kropff and W. Van der Werf. 2003. Annual losses of weed seeds due to predation in organic cereal fields. J. Appl. Ecol. 40:824-836.

White, A.D., D.J. Lyon, C. Mallory-Smith, C.R. Medlin and J.P. Yenish. 2006. Feral rye (*Secale cereale*) in agricultural production systems. Weed Tech. 20:815-823.

White, S.S., K.A. Renner, F.D. Menalled and D.A. Landis. 2007. Feeding preferences of weed seed predators and effect on weed emergence. Weed Sci. 55:606-612.

Willenborg, C.J. and R.C. Van Acker. 2008. The biology and ecology of hexaploid wheat (*Triticum aestivum* L.) and its implications for trait confinement. Can. J. Plant Sci. 88:997-1013.

Wolf, E.C. 1989. Experiences around the world. p. 53-61. *In* National Research Council (ed.) Triticale: A promising addition to the world's cereal grains. 1st ed. National Academy Press, Washington, D.C., USA.

Wolt, J.D. 2009. Advancing environmental risk assessment for transgenic biofeedstock crops. Available at www.biotechnologyforbiofuels.com/content/2/1/27 Biotech. for Biofuels 2:27.

Wolt, J.D., Y.Y. Shyy, P.J. Christensen, K.S. Dorman and M. Misra. 2004. Quantitative exposure assessment for confinement of maize biogenic systems. Environ. Biosafety Res. 3:183-196.

Wright, A.T. and J. Doane. 1987. Wheat midge infestation of spring cereals in northeastern Saskatchewan. Can. J. Plant Sci. 67:117-120.

Zhang, J., F.A. Drummond, M. Liebman and A. Hartke. 1997. Insect predation of seeds and plant population dynamics. Technical Bulletin 163, Maine Agricultural and Forest Experiment Station. Department of Applied Ecology and Environmental Sciences, University of Maine, Orono, Maine, USA.

Zimny, J., D. Becker, R. Brettschneider and H. Lorz. 1995. Fertile, transgenic triticale (X *Triticosecale* Wittmack). Mol. Breed. 1:155-164.

Zorner, P.S., R.L. Zimdahl and E.E. Schweizer. 1984. Sources of viable seed loss in buried dormant and non-dormant populations of wild oat (*Avena fatua* L.) seed in Colorado. Weed Res. 24:143-150.

Chapter 3. Persistence of Triticale (× *Triticosecale* Wittmack) Seed in the Soil Seed Bank[1]

3.1. Introduction

Triticale (× *Triticosecale* Wittmack), an intergeneric hybrid of wheat (*Triticum* sp.) and rye (*Secale* sp.), is being developed for the production of renewable bio-products in Canada (Eudes, 2006). Triticale produces high levels of biomass-derived ethanol (McLeod et al., 2010) and cultivars are being genetically engineered to synthesize bio-products such as bio-polymers, bio-fuels, and bio-chemicals (Canadian Triticale Biorefinery Initiative, 2011). In 2009, triticale was grown on over 4 million ha in 29 countries, but on only 12, 000 ha in Canada. By comparison wheat was grown on >220 million ha in 92 countries with >9 million ha in Canada (FAOSTAT, 2010). The use of triticale in bio-products is predicted to increase triticale production area within Canada to 400, 000 ha by 2015 (Canadian Triticale Biorefinery Initiative, 2011). However, prior to production in Canada, genetically modified (GM) crops must receive regulatory approval for food and environmental safety.

In addition to food and feed safety approval, release of GM spring triticale requires assessing the potential for gene flow. In a principally self-pollinated species the potential for seed-mediated gene flow may be more significant than the potential for pollen-mediated gene flow. Seed loss during harvest or transport

[1] *A version of this chapter has been accepted for publication. Raatz, L.L., R.-C. Yang, B.L. Beres, and L.M. Hall. 2012. Crop Science. 52: 1868-1880.*

may result in admixture during planting, harvesting, transportation, and storage or enter the seed bank and produce volunteers (Beckie et al., 2010). Volunteers can increase the potential for gene flow through outcrossing or by producing seed that may replenish the seed bank or result in low level presence within harvested conventional crop seed. Low level presence (LLP) refers to the inadvertent and undesirable inclusion of small amounts of GM events that have received regulatory approval in one or more countries, but that are still unauthorized in others (Kalaitzandonakes, 2011). Loss of GM seed poses a two-fold risk: economic damage to established cereal markets through reduced value or product rejection due to LLP and environmental harm if the crop becomes weedy or invasive (Canadian Food Inspection Agency, 2004). The ability of seeds to enter and persist in the seed bank is critical when evaluating the potential for seed mediated gene flow.

Domesticated cereals have been selected for traits that maximize yields and ease of production. Traits associated with domesticity include seed retention at maturity, reduced seed dispersal, reduced dormancy (primary and secondary dormancy), synchronous germination, short-lived seeds, narrow germination requirements, and fewer larger, uniform-sized seeds (Warwick and Stewart, 2005). Primary dormancy is defined as a condition where developing or developed seeds are prevented from germinating and is inherited during seed maturation while on or upon being released from the mother plant (Baskin and Baskin, 1998); and is also referred to as ‘pre-harvest sprouting tolerance’ (Biddulph et al., 2007). Secondary dormancy is defined as a state induced when

previously non-dormant seeds are exposed to unfavorable conditions (unsuitable temperature, moisture, light, nitrates) and germination is prevented; germination continues to be prevented even after unfavorable conditions are removed until species-specific environmental cues to break dormancy (Baskin and Baskin, 1998; Murdoch and Ellis, 2000; Finkelstein et al., 2008). Dormancy extends the persistence of seeds in the seed bank.

Cereals are selected for some short-term primary dormancy (Baskin and Baskin, 1998) or pre-harvest sprouting tolerance (Biddulph et al., 2007). Seeds are released from primary dormancy during a process of 'after-ripening' which is a period of time when seeds air dry and undergo a decrease in moisture content (Murdoch and Ellis, 2000). Like wheat, spring triticale is reported to have relatively low levels of primary dormancy and is susceptible to pre-harvest sprouting prior to harvest when conditions are wet during grain maturation and at harvest (Biddulph et al., 2007; Paulsen and Auld, 2004; Skovmand et al., 1984; Biddulph et al., 2008). However, winter wheat, triticale and rye cultivars can have varying degrees of primary dormancy (Weidner et al., 1999).

Cereal crop volunteers are common in subsequent crops in western Canada (Leeson et al., 2005), but the lack of secondary dormancy and a persistent seed bank usually prevents them from persisting where rotations are practiced. For example, spring wheat shows no secondary dormancy and does not form a persistent seed bank (De Corby et al., 2007; Nielson et al., 2009). Harker et al. (2005) showed that volunteer wheat seedlings were recruited in the year following seed bank establishment when moisture was not limiting and seed bank

replenishment was prevented, although some volunteers were detected 3 years after trial initiation. Secondary dormancy was shown to be induced in winter wheat when seed was stored at high humidity and storage temperature decreased, while secondary dormancy was induced in winter triticale at declining temperatures without humid conditions (Buraas and Skinnes, 1985); however, secondary dormancy has not been quantified in spring forms of wheat, triticale, or rye.

Wheat seeds are added to the seed bank through intentional planting, seed contamination, seed loss via natural shatter (aided by factors such as hail, crop lodging, disease, or insect herbivory), and mechanical harvest (Willenborg and Van Acker, 2008). Cereal seeds lost prior to and during harvest are the primary source of the seed bank. Triticale has similar cultural practices for seeding and harvesting and similar yields to wheat, although under dryland conditions in the brown soil zone of western Canada, seed production can be higher with greater overall stability across environments (Alberta Agriculture, Food and Rural Development, 2005; Beres et al., 2010; Goyal et al., 2011; McKenzie et al., 2007). Although there is no documentation of harvest loss specific to triticale, because seed size and harvest practices are similar to wheat, seed harvest losses from direct combining practices may also be similar. Anderson and Soper (2003) cited unpublished UK field surveys which report that winter wheat harvest losses averaged between 2 to 6% of seed yields, or between 240 and 700 seeds m^{-2} added to the soil surface following harvest. Spring wheat lost naturally at harvest ranged from 120 to 820 seeds m^{-2}, while direct combining losses ranged from

approximately 30 to 415 seeds m^{-2} (Clarke, 1985). When harvest was delayed for over 30 days following seed maturity, natural wheat seed losses were as high as 13% of total yields in irrigated fields (Clarke, 1981). While harvest losses and distributions vary with the type and speed of harvest equipment and environmental conditions (Komatsuzaki and Endo, 1996; Anderson and Soper, 2003; Pickett, 1993; Clarke, 1985), cereal crops have the potential to initiate a large seed bank.

Within the seed bank, cereal seeds are subject to predation (Brust and House, 1988), disease and mortality. Cereal seeds may become quiescent (i.e. unable to germinate because conditions are not conducive) or dormant or they may germinate readily (Anderson and Soper, 2003). Volunteers recruited in fall are subject to death by frost while spring germination results in volunteer populations that can contribute to admixture in subsequent crops. Primary dormancy in cereals may reduce fatal fall germination resulting in more spring volunteers or a more persistent seed bank. The presence of secondary dormancy, in addition to primary dormancy, has the potential to extend the longevity of seeds in the seedbank beyond the subsequent growing season.

Knowledge of seed dormancy and seed bank dynamics is critical to determine the potential for seed-mediated gene flow in GM triticale. We examined seed dormancy and seed persistence of four spring triticale cultivars in comparison with a spring wheat cultivar by: 1) assessing seed germinability during grain development from post-anthesis until completion of after-ripening,

and 2) assessing seed viability over time following placement of seeds on the soil surface, as well as shallow and deep seed burial.

3.2. Materials and Methods

3.2.1. Plant Materials

All cereal cultivars were developed on and for the Canadian Prairies. While both winter and spring triticale cultivars are suitable for bio-product development, only spring cultivars were used in this study because they are more commonly grown for seed in Canada. With the exception of the unregistered Blue Aleurone line, cultivars were chosen because they were the most commonly grown and available at the time the experiments were conducted and because of their suitability for bio-industrial uses. AC Alta is a later maturing spring triticale cultivar, resistant to lodging and suitable for food, feed, and industrial uses (McLeod et al., 1996a; McLeod et al., 1996b). The unregistered Blue Aleurone line was developed in Lethbridge by Agriculture and Agri-Food Canada and is the BC_4 of AC Alta crossed to 'Purendo-38', an experimental wheat cultivar containing a blue aleurone as a visual marker, previously described by Hucl (1996). Tyndal spring triticale is a relatively new triticale cultivar used for greenfeed or silage production (Salmon et al., 2007). AC Ultima is an early-maturing spring triticale with improved lodging resistance and improved tolerance to pre-harvest sprouting, suited for industrial uses (McLeod et al., 2000). Across multiple locations from Alberta regional variety trials, the three registered spring triticale cultivars mature in 110 to 120 d and all have good seed retention, but fair

to poor tolerance to pre-harvest sprouting or little to no primary dormancy (Government of Alberta Agriculture and Rural Development, 2010). AC Barrie is a high-yielding hard red spring wheat cultivar moderately resistant to pre-harvest sprouting, requiring 108 d to mature (DePauw et al., 1997).

3.2.1.1. Triticale After-Ripening

Experiments were established in 2009 and 2010 to quantify the time dependent germinability of spring triticale compared to AC Barrie wheat during the after-ripening period beginning post-anthesis until post-harvest. In both years, experiments were conducted at Edmonton Research Station, Alberta, Canada (53^{o}29'19"N, 113^{o}34'8"W), an Eluviated Black Chernozemic soil (Udic Boroll). Four triticale cultivars and AC Barrie wheat were direct-seeded using a Fabro air seeder with atom-jet openers at a depth of 2.5 cm, on 11 May and 14 May, in 2009 and 2010, respectively. Seeding rates ranged from 330 to 360 seeds m^{-2} (targeting 310 plants m^{-2}) for triticale and 300 to 320 seeds m^{-2} (targeting 275 plants m^{-2}) for wheat. Soil available nutrient status was determined from a composite 0 to 15 cm soil sample. Approximately 20 soil samples (roughly 50 cm^3 each) were taken randomly from within each trial area, combined, stirred, and analyzed. Available nitrate, phosphate, potassium and sulfate levels in both years were optimal and therefore no fertilizer was added in 2009 and 22 kg ha^{-1} of N (46-0-0) was unintentionally side-banded in 2010. Plots consisted of 8 rows spaced 20 cm apart and trimmed to an 8 m row length. Trials were maintained weed-free by hand-hoeing. The cultivar-blocks were not replicated but seeds were harvested as three subsamples from each spring cereal cultivar plot, beginning

post-anthesis. Fifty primary seeds, the large basal florets within a spikelet, were removed from the central third of the main tiller spike from randomly selected plants. Only seeds that did not exhibit pre-harvest sprouting were selected. At each sample date, cultivars were staged using the Zadoks scale (Zadoks et al., 1974) from primary seeds taken from six randomly selected plants across the entire plot length. In 2009, sampling began on 31 July when cereal cultivars were generally at the early- to mid-milk seed stage or approximately 7 days after pollination (DAP). For the purposes of this experiment, we define 0 DAP as completion of anthesis or Zadoks 69. In 2010, sampling began on 30 July, shortly after anthesis was completed or at 0 DAP. Sampling and crop staging were conducted weekly until harvest, on 11 Sept. 2009 and 2 Oct. 2010, when cereals had reached harvest maturity (seed moisture content of 14% or less). Following harvest, seed germinations were performed for three samples of 50 seeds removed from the harvested portion of each cereal plot. Sampling and germinations continued for four weeks post-harvest in 2009 and for two weeks post-harvest in 2010.

Germination was assessed by placing 50 seeds into 24 x 16 x 4 cm acrylic germination boxes (Hoffman Manufacturing, Inc.) on top of absorbent non-toxic 15 x 23 cm Anchor steel blue blotter paper (Hoffman Manufacturing, Inc.) designed to retain moisture, and covered with non-toxic white filter paper 15 x 23 cm No. 601 Whatman #1 equivalent (Hoffman Manufacturing, Inc.). A total water volume of 40 mL was added to each germination box which included a 0.2% solution of Helix Xtra® (thiamethoxam + difenoconazole + metalaxyl-M +

fludioxonil) to reduce fungal growth (Nielson et al., 2009; McPherson et al., 2009). As much as possible, seeds removed from cereal spikes were kept inside palea and lemma structures (including awns if present) until post-harvest germinations. Seeds were maintained in the dark at ambient room temperature (approximately 20^{o}C) for 7 days. Germination was defined as radicle shoot >1 mm and seeds that germinated were recorded and discarded. Germination assessments began three days after sampling and continued daily until the next sampling date. Total germinations were recorded weekly.

3.2.1.2. Triticale Seed Bank Persistence

Seeds of all cereal cultivars were grown at Agriculture and Agri-Food Canada's Lethbridge Research Centre, in Lethbridge, Alberta, Canada in the same year that seeds were buried in order to minimize differences in seed attributed to maternal effects, and to emulate the physiological state of seed lost at harvest. Burial experiments were established at two sites in Alberta, Canada in both 2007 and 2008 to quantify the viability and longevity of spring seeded triticale cultivars compared to wheat seed at various burial depths. Field trials were initiated at the University of Alberta Ellerslie Research Station (53^{o}25'24"N, 113^{o}43'52"W) and the Agriculture and Agri-Food Canada (AAFC) Lethbridge Research Centre (49^{o}41'2"N, 112^{o}37'41"W) in fall of 2007 and at the University of Alberta Edmonton Research Station (53^{o}29'19"N, 113^{o}34'8"W) and AAFC Lethbridge Research Centre in the fall of 2008. The Lethbridge site is an Orthic Dark Brown Chernozemic soil, Ellerslie and Edmonton are Eluviated Black Chernozemic soils. Soil characteristics were determined for a composite 0 to 15 cm soil sample

prior to site establishment (Table 3.1). Prior to seed burial at Ellerslie and Lethbridge in 2007, the annual precipitation was 61 and 73% of 30 year averages for the two sites, respectively. In 2008, annual precipitation was 103 and 59% that of 30 year averages at Lethbridge and Edmonton, respectively. Sites were maintained weed-free using chemical fallow at least one growing season prior to trial establishment.

The experiments were established as a split plot design with three replicates, where the burial depth was the main plot and cereal cultivar was the sub-plot. Two hundred seeds each of four spring triticale cultivars (AC Alta, Blue Aleurone, Tyndal, and AC Ultima) and one spring wheat cultivar (AC Barrie) were randomly placed into compartments of 47 x 13 cm nylon mesh bags along with a plastic bead unique to each cultivar for identification purposes. Initial germination rates were determined (Table 3.2) and seeds placed in fall on 3 Oct. 2007 (Lethbridge), 5 Oct. 2007 (Ellerslie), 23 Oct. 2008 (Lethbridge), 24 Oct.2008 (Edmonton) (Table 3.3), approximating the time when crop seeds would likely be shed at harvest. Bags were either placed on the soil surface, or buried at 2 or 12 cm to simulate harvest loss on the soil surface, and shallow or deep fall tillage operations, respectively (Li et al., 2007; Burnside et al., 1996). While deep tillage operations (e.g. moldboard plow) are not common on the Canadian prairies, volunteer wheat can emerge from greater depths than small seeded weeds (Van Acker et al., 2004). Additionally, deep tillage has been shown to contribute to the induction of dormancy in volunteer canola (*Brassica napus* L.) (Gulden et al., 2004) and may also induce dormancy in volunteer wheat (Cussans, 1978).

Each treatment was enclosed within a 230 x 80 x 15 cm galvanized steel cage (65 mm- mesh) to reduce vertebrate seed predation.

Bags containing seeds were withdrawn five times during the following growing season at approximately four-week intervals beginning in May through September (Table 3.3) and seed germination and viability evaluated. Seeds that had germinated (radicle >1mm) or decayed were considered to have been removed from the seed bank. Ungerminated intact seeds were placed in 24 x 16 x 4 cm acrylic germination boxes (Hoffman Manufacturing, Inc.) and germination tested as previously described. Boxes were stored in the dark for five days at ambient temperature (approximately 20^{o}C). Seeds that germinated were recorded as being viable. Seeds that did not germinate were bisected longitudinally and the two halves were placed embryo side down in a Petri dish containing Whatman #1 filter paper and 2, 3, 5-triphenyltetrazolium chloride (0.15%) for 1 hour in the dark at ambient temperature (Grabe, 1970; Nielson et al., 2009). Seeds that tested positive for respiration were considered viable (Porter et al., 1947).

3.2.2. Data Analysis

3.2.2.1. Triticale After-Ripening

Germination data were $\log_{10}$ transformed to meet the assumptions of normality and homogeneity of variances for ANOVA. Proc MIXED of Statistical Analysis Software (SAS Institute Inc., 2007) was used to determine whether there were significant differences between years. Year and cultivar were considered to be fixed effects, subsample was considered a random effect, and crop stage at the

time of sampling was used as a covariate to account for differences in seed development between years and cultivars. Year, cultivar, and the crop stage were significant (p<0.0001); however, the interaction between year and cultivar were not (p=0.1048). Because rainfall in 2009 and 2010 were very different and only two site years of data were collected, years were analyzed separately. Crop stage was no longer significant by year and was therefore removed as a covariate. Weekly germination data were subject to ANOVA using Proc MIXED with Bonferroni-adjusted mean separations to communicate differences between the cultivars at every week of sampling. Because crop stage was variable between cultivars at every sampling date, germination data are reported by calendar date instead of DAP to assess differences between cohorts as the season progressed. Week 0 data in 2009 and weeks 0 and 1 data in 2010 did not meet assumptions of normality or homogeneity of variances and ANOVA was not performed for these weeks. Because cultivars were not replicated, a more stringent p<0.01 was used to test for significance.

3.2.2.2. Triticale Seed Bank Persistence

The number of viable triticale seeds in the seed bank was calculated as the sum of germinable and tetrazolium positive seeds. Viable seeds (y) from each of two years, two sites, 3 depths and 5 cultivars were regressed over time using a nonlinear regression mixed model (Proc NLMIXED) with Statistical Analysis Software (SAS Institute Inc., 2007). The binomial distribution (~binomial (n, P)) was used to approximate the dependent variable where n is the total number of buried seeds in each sample and P is the probability of viable seeds estimated as a

ratio of viable over total seeds recovered at each extraction date ($P = y/n$). Based on previous seed longevity studies that indicate seed survival in the agricultural seed bank declines exponentially (McPherson et al., 2009; Nielson et al., 2009; Conn et al., 2006), the data were fitted to a simple exponential decay curve given in Eq. 1.

$$P = ae^{-bd} \quad (1)$$

where P is the probability of viable seeds, a is the intercept which was set at 1, b is the slope of the curve and the parameter describing the rate of viability decline and d is the number of days after seed burial.

The rate of decline (b) generated by regression was used as a new parameter and analyzed with ANOVA, using a mixed model (SAS Institute Inc., 2007) within four environments: Ellerslie 2007; Lethbridge 2007; Edmonton 2008; Lethbridge 2008, where the main plot is depth and the split plot is cultivar. Depth and cultivar were considered to be fixed effects while environment, replicate(environment) and depth × replicate(environment) were random effects. Data were compared using two pre-planned non-orthogonal contrast statements to determine whether there were significant differences in longevity at each burial depth between wheat and the triticale cultivars and, because they are genotypically similar, AC Alta and Blue Aleurone.

The slope (b) or rate of decline parameter was inverse square root transformed to meet the assumptions of normality and homogeneity of variances for ANOVA. Surface placed seeds in Lethbridge 2008 could not be examined because of missing data: burial cage lids buckled and surface placed seeds were

predated by mice (observed, although not quantified). After testing whether depth, cultivar or their interaction were significant, the initial data set was regressed again with combined data, where appropriate. The time to 50% and 99% seed extinction was estimated (EX_{50} and EX_{99}; Eqs. 2 and 3, respectively),

$$EX_{50} = [\ln(0.5) - \ln(a)]/ -b \quad (2)$$

$$EX_{99} = [\ln(0.01) - \ln(a)]/ -b \quad (3)$$

where a is the intercept (set at 1) and b is the slope of the curve and the parameter describing the rate of decline. Estimates of seed viability the spring (May) and fall (Sept) following seed burial were also made.

3.3 Results

3.3.1. Triticale After-Ripening

There were significant differences between years in the time to maturity. Triticale cultivars required three fewer weeks to reach harvest maturity in 2009 compared to 2010. In 2009, Edmonton experienced drought, and therefore cereals were generally physiologically more advanced than 2010 on the same calendar dates. Greater precipitation was received in 2010, which was generally cooler than 2009 (Figure 3.1 and 3.2). Germinations and crop staging began on 31 July in 2009 and on 30 July in 2010. In both years, AC Barrie was more mature than triticale at every week of sampling, while AC Alta was consistently later

developed than all other cereals at every week (Figure 3.3). Differences in maturity were accounted for by including crop stage as a covariate in the analysis.

There were significant differences in germinations between years (p<0.0001) and therefore years were analyzed separately. In 2009, germination remained <56% for all cultivars until harvest (49 to 56 DAP), an indication that cultivars have some primary dormancy preventing germinating prior to harvest (Figure 3.4 A, Table 3.4). *In situ* pre-harvest sprouting was not observed. Prior to harvest, Tyndal had a higher germination than the other cultivars in 4 of 6 weeks (Table 3.4). This ability to readily germinate indicates low primary dormancy in Tyndal. Prior to harvest, AC Barrie and AC Ultima had significantly lower germinations than Tyndal or AC Alta at week 3; AC Barrie had the lowest germination compared to the other cultivars in 4 of 6 weeks prior to harvest. Following harvest, germinations increased rapidly for all triticale cultivars and AC Barrie. Previously, Hagemann and Ciha (1987) reported that warm environments during the after-ripening period accelerated loss of primary dormancy in winter wheat. Immediately following harvest, AC Barrie, and to a lesser extent Blue Aleurone and AC Alta, had low germination which increased over time. After-ripening in these cultivars was not reached until week 9 or three weeks after harvest, similar to observations in 2007 and 2008 (See footnotes in Table 3.2).

Due to the relatively cool and wet conditions in 2010, all triticale cultivars, particularly Tyndal, exhibited *in situ* pre-harvest sprouting several weeks prior to harvest (data not shown). All cereals, with the exception of AC Ultima, had

germinations >75% prior to harvest at week 9 (Figure 3.4 B, Table 3.4). Tyndal had a significantly higher germination at weeks 4 and 5 (approximately 28 to 35 DAP) compared to other cereals indicating comparatively lower primary dormancy at this early stage. Himi et al. (2002) reported that wheat cultivars with low dormancy may germinate as early as 30 to 40 DAP. AC Ultima showed significantly higher levels of dormancy with germinations of 38 and 46% by 8 and 9 weeks (approximately 56 to 63 DAP) relative to all other cereals where germinations ranged from 75 to 98%.

3.3.2. Triticale Seed Bank Persistence

Seeds buried at 12 cm were rapidly degraded and were generally not viable in the spring. As observed in this study, seeds may germinate when buried deeply, but they are unable to emerge ('fatal germination') and are removed from the viable seed bank (Grundy et al., 2003). Seeds buried at 2 cm simulate optimal seeding depth for triticale and seeds readily germinated when soil and temperature conditions were favorable. However, seedlings that germinated in the fall did not survive winter. The rate of seed extinction was significant for seed burial depth ($p<0.0001$), cultivar ($p<0.0001$) and the interaction between the two terms ($p=0.0032$) (Table 3.5).

Seeds placed on the soil surface remained viable and germinable for a longer period of time than those buried to 2 cm (Figure 3.5 A and B). The soil surface is susceptible to intermittent aridity and less likely to provide consistent conditions for germination. Because the Lethbridge 2008 surface treatments were lost, seed viability loss on the soil surface was based on data from the three

remaining environments. Estimates for 50% reduction in seed viability or extinction (EX_{50}) for seeds placed on the soil surface ranged from 74 to 117 days for Blue Aleurone and AC Barrie, respectively (Table 3.5). Estimates for 99% seed extinction (EX_{99}) ranged from 490 to 774 days for Blue Aleurone and AC Barrie, respectively. AC Barrie wheat persisted significantly longer on the soil surface than the four triticale cultivars (p <0.0001), although AC Alta and Blue Aleurone were not significantly different (p=0.3445) (Table 3.5, Figure 3.5 A).

Seeds buried to 2 cm tended to germinate and exit the seed bank as volunteers either in the fall or the following spring. Shallow buried seeds at Lethbridge tended to germinate and emerge in fall and be killed by winter temperatures while in the remaining three environments, emergence mostly took place the spring following seed burial (data not shown). For seeds buried at 2 cm, EX_{50} estimates ranged from 28 to 49 days for AC Alta and AC Barrie, respectively while EX_{99} estimates ranged from 187 to 327 days for AC Alta and AC Barrie, respectively (Table 3.5). Similar to seeds placed on the soil surface, AC Barrie at 2 cm tended to persist significantly longer than the triticale cultivars combined (p<0.0001), although Blue Aleurone persisted significantly longer than AC Alta (p=0.0050) (Table 3.5, Figure 3.5 B).

Seeds buried deeply at 12 cm tended to germinate, but not emerge from the soil, and decayed rapidly; few intact seeds were recovered the following season. Estimates for EX_{50} ranged from 17 to 29 days for AC Alta and AC Barrie, respectively. EX_{99} estimates ranged from 111 to 195 days following seed burial for AC Alta and AC Barrie, respectively (Table 3.5, Figure 3.5 C). However, at

12 cm there were no significant differences in seed persistence between AC Barrie and the triticale cultivars (p=0.3021) or between AC Alta and the Blue Aleurone line (p=0.3269).

3.4. Discussion

Triticale cultivars varied in primary dormancy (or pre-harvest sprouting tolerance) prior to harvest, but all cultivars rapidly after-ripened following harvest. In this study, cultivar differences were more obvious in cool moist conditions. Salmon and Helm (1985) previously reported that triticale cultivars varied in pre-harvest dormancy and exhibited lower dormancy under drier, warmer conditions. Cereals with very low or no dormancy may be susceptible to pre-harvest sprouting in temperate regions with higher levels of rainfall prior to or following harvest (Biddulph et al., 2007; Paulsen and Auld, 2004), a phenomenon which reduces grain quality, seed viability and yield (Biddulph et al., 2007). Relative to Canadian prairie triticale cultivars, AC Ultima has a high Hagberg Falling Number, a standardized test that measures the amount of starch in a cereal seed sample. Higher numbers indicate a longer time for a steel ball to drop through a slurry of cereal seed starches; higher numbers are associated with pre-harvest sprouting tolerance (McLeod, et al., 2000). As expected, AC Ultima had lower germination prior to harvest compared to the other cultivars, although this difference was not apparent following harvest. In this instance, primary dormancy appeared to be short-lived and provided little protection from germination given appropriate conditions following harvest. Triticale has been previously reported to

be more susceptible to pre-harvest sprouting than wheat (Alberta Agriculture, Food and Rural Development, 2005; Paulsen and Auld, 2004), although wheat cultivars vary (Biddulph et al., 2005; Himi et al., 2002) as do triticale cultivars (Salmon and Helm, 1985).

Time and intensity of tillage has a significant influence on the depth of burial of volunteer cereals and their subsequent fate. Within reduced tillage regimes, weed seeds and volunteer cereals accumulate on the soil surface and at shallow soil depths, while fewer seeds are found at greater depths (Torresen et al., 2003; Légère et al., 2011). In the absence of primary dormancy or inducible secondary dormancy, seeds remaining on the soil surface are less likely to experience prolonged conditions favoring germination and therefore may persist longer.

Conclusions from this study and others using artificial seed banks must be viewed with caution. Artificial seed bank studies are problematic because mesh bags do not reproduce conditions found in natural seed banks (Van Mourik et al., 2005). Exclusion cages and mesh bags typically prevent seed bank depletion from vertebrate seed predation (rodents and birds) (Baraibar et al., 2009; Graziani et al., 2007; O'Rourke et al., 2006). By eliminating seed predation, persistence on the soil surface is likely being over-estimated in artificial seed banks. However, seed aggregation in artificial seed banks may also create conditions for spread of pathogenic fungi (Chee-Sanford et al., 2006; Gilbert, 2002; Van Mourik et al., 2005) and subsequently under-estimate seed longevity. Additionally, intact cereal spikes within the seed bank persist for a longer period of time than threshed seeds

(Seerey, et al., 2011). These factors are difficult to evaluate because there are no studies of crop or weed seed persistence that quantify the relative importance of seed predators and disease in western Canadian agricultural systems.

The majority of seeds in transient seed banks exit through germination, which is largely influenced by soil moisture and temperature (Davis et al., 2005). Seed burial to 2 cm stimulates germination by improving seed to soil contact, causing rapid reduction in seed banks. Seeds buried at shallow depths in Lethbridge rapidly exited the seed bank by germinating prior to spring when moisture and temperature conditions were favorable. Volunteers that germinate before spring are subjected to fatal winter temperatures. Seeds buried at shallow depths in the northern locations of Ellerslie and Edmonton generally over-wintered as seeds and germinated in early spring (data not shown). Seeds buried to 2 cm at Edmonton in 2008 may have persisted longer because of inadequate moisture conditions during a drought in 2009. Similar to this study, Harker et al. (2005) showed that volunteer wheat seeds persisted for longer periods of time when moisture conditions were limiting. Seeds buried shallowly may remain quiescent (ungerminated due to unfavorable environmental conditions) (Murdoch and Ellis, 2000), although not dormant, until temperature and moisture are appropriate for germination.

Triticale seed persistence on the soil surface and when buried is comparable with other domesticated crop species such as flax (*Linum usitatissimum* L.) (Dexter et al., 2011) and safflower (*Carthamus tinctorius* L.) (McPherson et al., 2009). Triticale behaves similarly to wheat in the soil seed

bank. In a similar artificial seed bank experiment using various seed sizes of spring wheat left on the surface or shallow and deep burial, Nielson et al. (2009) showed that seeds remaining on the soil surface tended to persist significantly longer than those buried within the soil and those buried deeply rapidly exited the seed bank through fatal germination. Wheat deposited on the soil surface had an estimated time to 50% reduction in seed viability (EX_{50}) of between 57 and 152 days after seed burial (Nielson et al., 2009). Similarly, in this study AC Barrie wheat seed deposited on the soil surface had an EX_{50} value of 117 days while triticale ranged from 73 to 83 days after seed burial. Nielson et al. (2009) showed that buried wheat seeds tended not to persist for long periods of time within the shallow seed bank. Buried wheat seeds had EX_{50} values that ranged from 37 to 88 days after seed burial (Nielson et al., 2009). In this experiment, AC Barrie wheat buried to 2 cm had an EX_{50} value of 49 days, while triticale ranged from 28 to 34 days. Within the deep seed bank, Nielson et al. (2009) showed that EX_{50} values for wheat ranged from 20 to 47 days while EX_{99} values ranged from 175 to 352 days following burial. In this study, wheat had an EX_{50} value of 29 days while triticale ranged from 17 to 26 days and the EX_{99} for wheat was 195 days while triticale ranged from 111 to 172 days. Nielson et al. (2009) showed that less than 1% of wheat seeds were estimated to persist beyond 3 years and maximum seed viability the following spring for surface, shallow, and deeply buried seeds was 43, 7, and 2%, respectively. Likewise, Harker et al. (2005) showed that glyphosate-resistant wheat populations did not persist beyond three years when seed return from volunteers was prevented. While the triticale cultivars in this

study consistently persisted in the seed bank for a shorter time than the wheat cultivar, the reasons are not known since the seed size and morphology are similar. Triticale behaves similarly to wheat on the soil surface and when buried, and does not appear to persist within the seed bank any longer than wheat.

Triticale is no more likely to become persistent or weedy in agricultural systems than wheat. The results from this study indicate that seed persistence in the seed bank does not preclude triticale from further development as a bio-product crop. While no information exists on triticale or wheat in ruderal environments, volunteer herbicide-resistant canola is frequently found along transport routes, although it has not formed self-sustaining or feral populations on the northern Great Plains (Beckie and Owen, 2007). Populations of feral rye (*Secale cereale* L.), a winter form, are causing economic losses in winter wheat production systems of midwestern US (White et al., 2006). Similar to triticale and wheat, feral rye does not exhibit primary dormancy; however, unlike wheat or triticale, feral rye does show low levels of inducible secondary dormancy which contributes to the seed persistence (up to 5 years) in the US (Stump and Westra, 2000). Although the use of rye for erosion control in an unmanaged setting, lack of seed purity regulations, and suspected spread of feral seeds on contaminated harvest equipment (Burger et al., 2007; Miller et al., 2004) have contributed to the spread of feral rye populations in the US, ferality has not been observed with wheat, rye or triticale in Canada.

Some of the seed lost at harvest will survive the winter, germinate and form volunteer populations. These volunteers may be controlled prior to seeding,

but those that survive pre-seed tillage or herbicide application or germinate later will form volunteer populations that cannot be controlled in other cereals, including conventional wheat, triticale, barley (*Hordeum vulgare* L.) and oats (*Avena sativa* L.). The admixture of seed from volunteers would be unwelcome in many markets even if the GM traits were approved by the exporting and importing country and LLP could be maintained at levels below thresholds. The short term persistence of seed in the seed bank, coupled with appropriate non-cereal rotations and monitoring and mitigation strategies may facilitate the use of triticale for bio-product development. Co-existence of approved GM triticale with conventional cereal crops would require volunteer control for one to two years following the crop and rotation with non-cereal crops as well as herbicidal control to mitigate risk in reduced tillage cropping systems.

Table 3.1. Soil characteristics for Ellerslie, Lethbridge and Edmonton at the time of initiation of seed bank experiments, 2007 and 2008.

					Soil Texture			Soil Fertility			
Location	Year	Soil pH	Soil OM	Soil EC	Sand	Silt	Clay	Nitrate	P	K	Sulfate
			$g\ kg^{-1}$	$dS\ m^{-1}$	-------$g\ kg^{-1}$-------			----------$kg\ ha^{-1}$----------			
Ellerslie	2007	5.8	109	0.25	320	410	270	28	41	353	10
Lethbridge	2007	8.0	33	1.07	360	390	250	27	84	911	173
Edmonton	2008	5.6	120	0.34	130	400	470	43	38	524	19
Lethbridge	2008	8.0	27	1.6	420	330	250	86	55	650	389

Table 3.2. Initial germinations of triticale cultivars and wheat cultivar, 'AC Barrie', one week prior to seed burial in 2007 and 2008.

Year	Cultivar	Germination %
2007	AC Alta	98
	AC Barrie[†]	71
	Blue Aleurone	99
	Tyndal	95
	AC Ultima	97
2008	AC Alta	96
	AC Barrie[‡]	76
	Blue Aleurone	89
	Tyndal	97
	AC Ultima	97

[†]AC Barrie germination was 31% immediately following harvest and 98% when retested after burial.
[‡]AC Barrie germination was 46% immediately following harvest and 99% when retested after burial.

Table 3.3. Seed burial dates and withdrawal (calendar) dates and days after burial for Ellerslie, Lethbridge and Edmonton in 2007 and 2008.

Location	Year	Burial date	Date 1 (D)[†]	Date 2 (D)	Date 3 (D)	Date 4 (D)	Date 5 (D)	Date 6 (D)	Date 7 (D)	Date 8 (D)	Date 9 (D)	Date 10 (D)
			----------2008----------					----------2009----------				
Ellerslie	2007	5 Oct	9 May (218)	2 Jun (242)	2 Jul (272)	30 Jul (300)	27 Aug (328)	7 May (581)	4 Jun (609)	2 Jul (637)	31 Jul (666)	3 Sept (700)
Lethbridge	2007	3 Oct	12 May (223)	10 Jun (252)	8 Jul (280)	5 Aug (308)	2 Sept (336)	4 May (580)	3 Jun (610)	6 Jul (643)	6 Aug (674)	8 Sept (707)
			----------2009----------					----------2010----------				
Edmonton	2008	24 Oct	6 May (195)	4 Jun (224)	2 Jul (252)	31Jul (281)	3 Sept (315)	3 May (557)	3 Jun (588)	5 Jul (620)	3 Aug (649)	1 Sept (678)
Lethbridge	2008	23 Oct	4 May (194)	3 Jun (224)	6 Jul (257)	6 Aug (288)	8 Sept (321)	11 May (566)	10 Jun (596)	9 Jul (625)	18 Aug (665)	16 Sept (694)

[†]D, number of days after seed burial

Table 3.4. Percent germinations (least square means ± standard error) for four triticale cultivars and one wheat cultivar (AC Barrie) and results of analysis of variance at Edmonton in 2009 and 2010.

		Germination (%)[‡]					
		Week (Calendar Date)					
		1	2	3	4	5	
Year	Cultivar	(7 Aug)	(14 Aug)	(21 Aug)	(28 Aug)	(4 Sept)	
2009	Cultivar	ns[†]	***	***	ns	ns	
	'AC Alta'	2.0 ± 1.6 a	6.2 ± 1.3 b	47.2 ± 1.1 a	38.0 ± 1.4 a	33.5 ± 1.2 a	
	'AC Barrie'	3.6 ± 1.3 a	14.5 ± 1.3 ab	11.2 ± 1.1 c	9.4 ± 1.5 a	17.2 ± 1.2 a	
	Blue Aleurone	0 ± 0 a	4.3 ± 1.3 b	30.4 ± 1.1 ab	31.4 ± 1.4 a	23.3 ± 1.2 a	
	'Tyndal'	7.1 ± 1.3 a	41.2 ± 1.3 a	42.2 ± 1.1 a	53.4 ± 1.4 a	34.8 ± 1.2 a	
	'AC Ultima'	5.7 ± 1.4 a	27.3 ± 1.3 a	22.4 ± 1.1 b	17.1 ± 1.4 a	26.8 ± 1.2 a	
		(6 Aug)	(13 Aug)	(20 Aug)	(27 Aug)	(3 Sept)	
2010	Cultivar	.	**	***	**	**	
	'AC Alta'	.	2.4 ± 1.3 c	7.3 ± 1.3 ab	22.4 ± 1.2 b	42.8 ± 1.2 ab	
	'AC Barrie'	.	9.9 ± 1.2 ab	26.4 ± 1.3 a	24.4 ± 1.2 b	30.9 ± 1.2 abc	
	Blue Aleurone	.	2.9 ± 1.2 c	4.2 ± 1.3 b	16.8 ± 1.2 b	15.9 ± 1.2 c	
	'Tyndal'	.	14.4 ± 1.2 a	22.2 ± 1.3 a	49.3 ± 1.2 a	55.3 ± 1.2 a	
	'AC Ultima'	.	4.5 ± 1.2 bc	2.5 ± 1.3 b	19.1 ± 1.2 b	18.4 ± 1.2 bc	

		Germination (%)[‡]					
		Week (Calendar Date)					
		6	7	8	9	10	11
Year	Cultivar	(11 Sept)	(18 Sept)	(25 Sept)	(2 Oct)	(9 Oct)	
2009	Cultivar	***	**	ns	ns	ns	
	'AC Alta'	23.8 ± 1.2 a	84.0 ± 1.1 ab	91.2 ± 1.0 a	97.3 ± 1.0 a	95.3 ± 1.0 a	
	'AC Barrie'	8.3 ± 1.2 b	69.4 ± 1.1 b	85.9 ± 1.0 a	100.0 ± 1.0 a	98.0 ± 1.0 a	
	Blue Aleurone	32.6 ± 1.2 a	79.1 ± 1.1 ab	79.0 ± 1.0 a	96.0 ± 1.0 a	98.0 ± 1.0 a	
	'Tyndal'	38.6 ± 1.2 a	91.3 ± 1.1 a	93.3 ± 1.0 a	98.7 ± 1.0 a	97.3 ± 1.0 a	
	'AC Ultima'	56.3 ± 1.2 a	95.3 ± 1.1 a	96.6 ± 1.0 a	100.0 ± 1.0 a	98.7 ± 1.0 a	
		(10 Sept)	(17 Sept)	(24 Sept)	(1 Oct)	(8 Oct)	(15 Oct)
2010	Cultivar	**	ns	***	***	ns	ns
	'AC Alta'	68.6 ± 1.2 a	58.2 ± 1.1 a	98.0 ± 1.1 a	83.8 ± 1.1 a	88.0 ± 1.0 a	88.6 ± 1.0 a
	'AC Barrie'	44.7 ± 1.2 ab	86.0 ± 1.1 a	83.3 ± 1.1 a	77.9 ± 1.1 a	95.3 ± 1.0 a	96.6 ± 1.0 a
	Blue Aleurone	37.7 ± 1.2 ab	63.9 ± 1.1 a	88.5 ± 1.1 a	74.5 ± 1.1 a	83.3 ± 1.0 a	89.2 ± 1.0 a
	'Tyndal'	62.6 ± 1.2 a	75.8 ± 1.1 a	92.2 ± 1.1 a	85.1 ± 1.1 a	90.6 ± 1.0 a	85.2 ± 1.0 a
	'AC Ultima'	22.4 ± 1.2 b	41.3 ± 1.1 a	37.9 ± 1.1 b	45.5 ± 1.1 b	89.3 ± 1.0 a	91.2 ± 1.0 a

[**] Significant at $p<0.01$.
[***] Significant at $p<0.001$.
[†] ns, not significant at $p<0.01$.
[‡]Germinations were $\log_{10}$ transformed for ANOVA and LSMeans ± SE were back-transformed. Germination for each cultivar was tested weekly starting approximately 10 days following anthesis in 2009 and starting at the completion of anthesis in 2010. Cultivars were harvested between weeks 6 and 7 in 2009 and between weeks 9 and 10 in 2010 when seed reached ≤ 14% grain moisture.

[§]Least square means for different cultivars followed by the same letter are not significantly different for each week in 2009 and 2010 (Saxton, 1998). Mean separations are based on Bonferroni-adjusted *p*-values, $p < 0.005$.

Table 3.5. Summary of results of seed burial experiments conducted in 2007 at Ellerslie and Lethbridge and 2008 at Edmonton and Lethbridge, including slope (*b*) from regression analysis of the exponential loss of seed viability ($P = ae^{-bd}$ where P is the probablility of viable seeds, a is the intercept, b is the slope of the curve and the rate of seed viability decline, and d is the number of days after seed burial), estimated days to 50% and 99% reduction in seed viability (EX_{50} and EX_{99}), and estimated frequency of viable seed the following spring and fall.

		Slope estimate[†]			Frequency of viable seed[‡]	
Depth	Cultivar	*b*	EX_{50}	EX_{99}	Spring (May)	Fall (Sept)
--cm--			--------d--------			
0	AC Alta	0.0089 ± 0.0001	77.5 ± 1.0	514.7 ± 6.6	0.1514 ± 0.0037	0.0504 ± 0.0019
	AC Barrie	0.0060 ± 0.0001	116.5 ± 1.3	773.7 ± 8.3	0.2848 ± 0.0038	0.1369 ± 0.0029
	Blue Aleurone	0.0094 ± 0.0001	73.7 ± 1.0	489.9 ± 6.4	0.1376 ± 0.0036	0.0433 ± 0.0018
	Tyndal	0.0084 ± 0.0001	82.7 ± 1.0	549.2 ± 6.7	0.1705 ± 0.0037	0.0608 ± 0.0021
	AC Ultima	0.0084 ± 0.0001	82.5 ± 1.0	548.2 ± 6.8	0.1699 ± 0.0037	0.0605 ± 0.0021
Contrasts		*p*-value				
AC Barrie vs. all triticale		<0.0001				
AC Alta vs. Blue Aleurone		0.3445				
2	AC Alta	0.0247 ± 0.0008	28.1 ± 0.9	186.6 ± 5.9	0.0055 ± 0.0009	0.0003 ± 0.0001
	AC Barrie	0.0141 ± 0.0002	49.2 ± 0.8	327.1 ± 5.1	0.0513 ± 0.0024	0.0091 ± 0.0029
	Blue Aleurone[§]	0.0207 ± 0.0006	33.5 ± 0.9	222.2 ± 6.1	0.0126 ± 0.0015	0.0010 ± 0.0002
	Tyndal	0.0219 ± 0.0006	31.7 ± 0.8	210.7 ± 5.4	0.0099 ± 0.0012	0.0007 ± 0.0001
	AC Ultima	0.0243 ± 0.0008	28.5 ± 0.9	189.3 ± 6.1	0.0059 ± 0.0010	0.0003 ± 0.0001
Contrasts		*p*-value				
AC Barrie vs. all triticale		<0.0001				
AC Alta vs. Blue Aleurone		0.0050				
12	AC Alta	0.0417 ± 0.0034	16.6 ± 1.4	110.5 ± 9.1	0.0002 ± 0.0001	<0.0001 ± <0.0001
	AC Barrie	0.0236 ± 0.0007	29.4 ± 0.8	195.2 ± 5.6	0.0069 ± 0.0010	0.0004 ± 0.0001
	Blue Aleurone	0.0270 ± 0.0010	25.7 ± 0.9	170.6 ± 6.1	0.0034 ± 0.0007	0.0001 ± <0.0001
	Tyndal	0.0353 ± 0.0023	19.6 ± 1.3	130.4 ± 8.4	0.0006 ± 0.0003	<0.0001 ± <0.0001
	AC Ultima	0.0267 ± 0.0010	25.9 ± 1.0	172.3 ± 6.4	0.0035 ± 0.0007	0.0001 ± <0.0001
Contrasts		*p*-value				
AC Barrie vs. all triticale		0.3021				
AC Alta vs. Blue Aleurone		0.3269				

[†]Parameter estimate ± SE for *b* (the rate of seed viability decline), where intercept *a* = 1
[‡]Frequency of viable seed ± SE estimates for spring (day 211, roughly 1 May to 23 May) and fall (day 334, roughly 1 Sept to 23 Sept) following initial seed burial.
[§]Rapid decrease in seed viability at Ellerslie 2007 prevented regression for this cultivar. Results for this cultivar were generated using the three remaining environments.

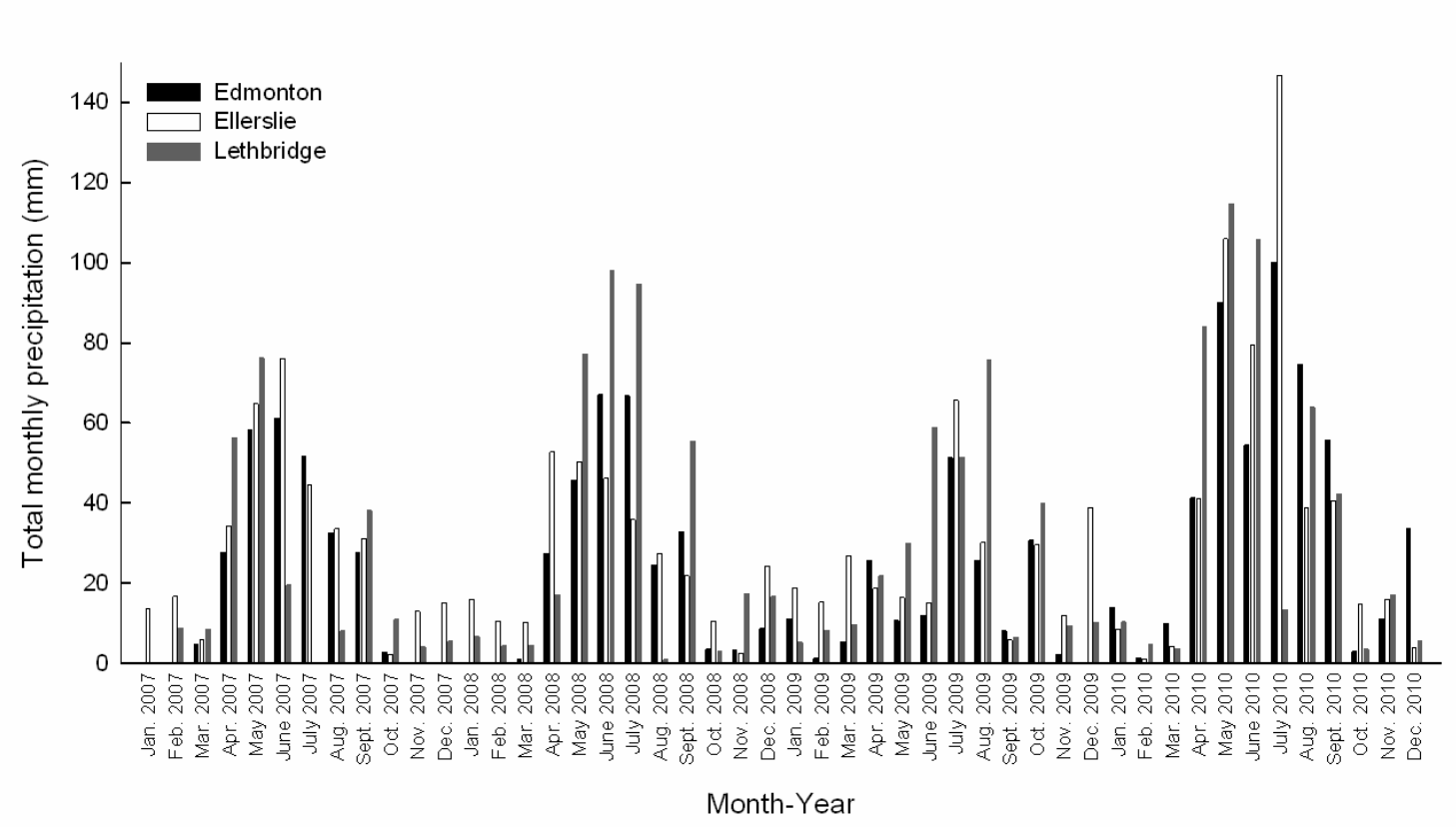

Figure 3.1. Total monthly precipitation (mm) for Edmonton, Ellerslie, and Lethbridge from 2007 to 2010.

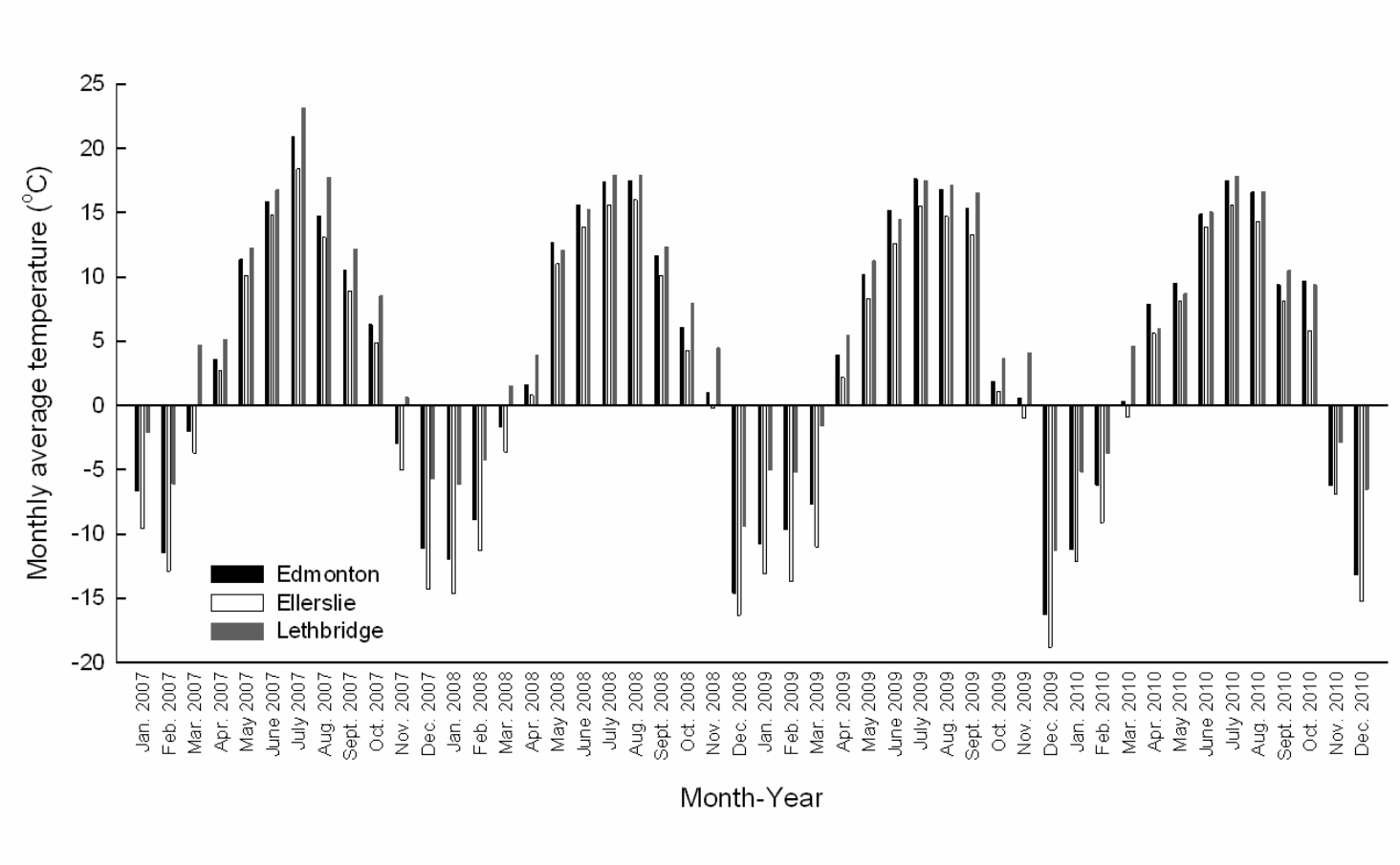

Figure 3.2. Average monthly temperatures (^{o}C) for Edmonton, Ellerslie, and Lethbridge from 2007 to 2010.

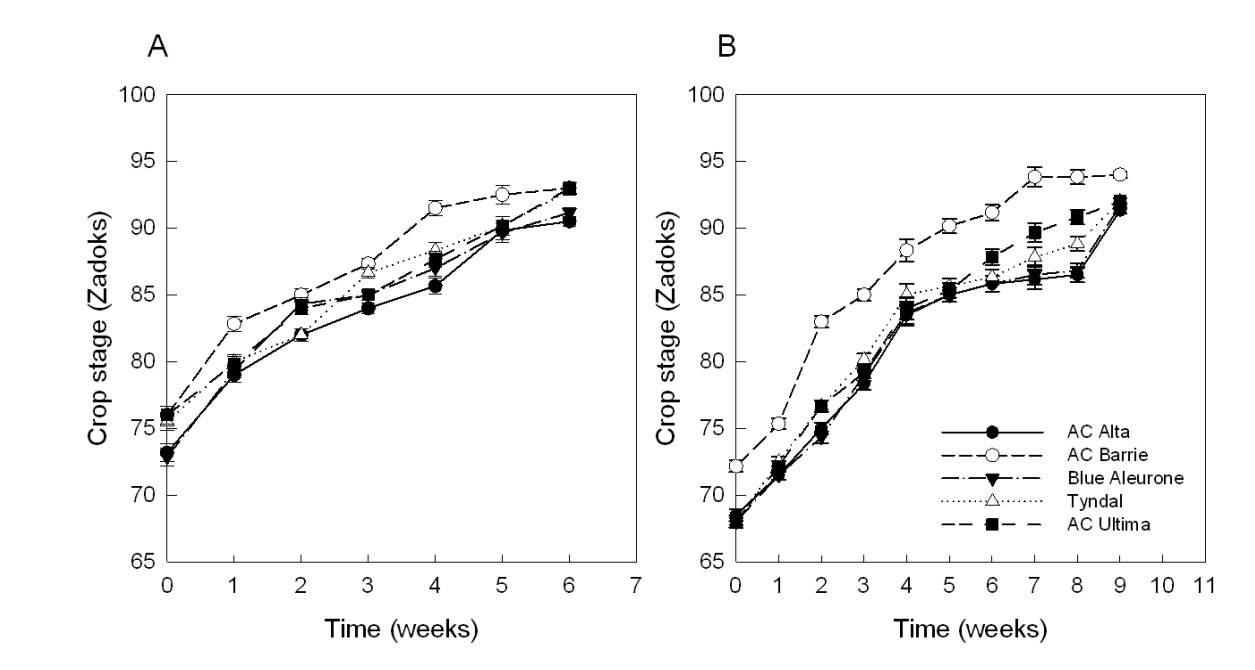

Figure 3.3. Crop stage (Zadoks et al., 1974) for spring triticale: AC Alta, Blue Aleurone, Tyndal, AC Ultima and spring wheat (AC Barrie) A. in 2009 beginning on 31 July until combine harvest on 11 Sept, after 6 weeks and B. 2010 starting on 30 July until combine harvest on 2 Oct, after 9 weeks. Symbols are the least square mean weekly crop stage ± standard error of the mean. Standard error bars may be obscured by symbols.

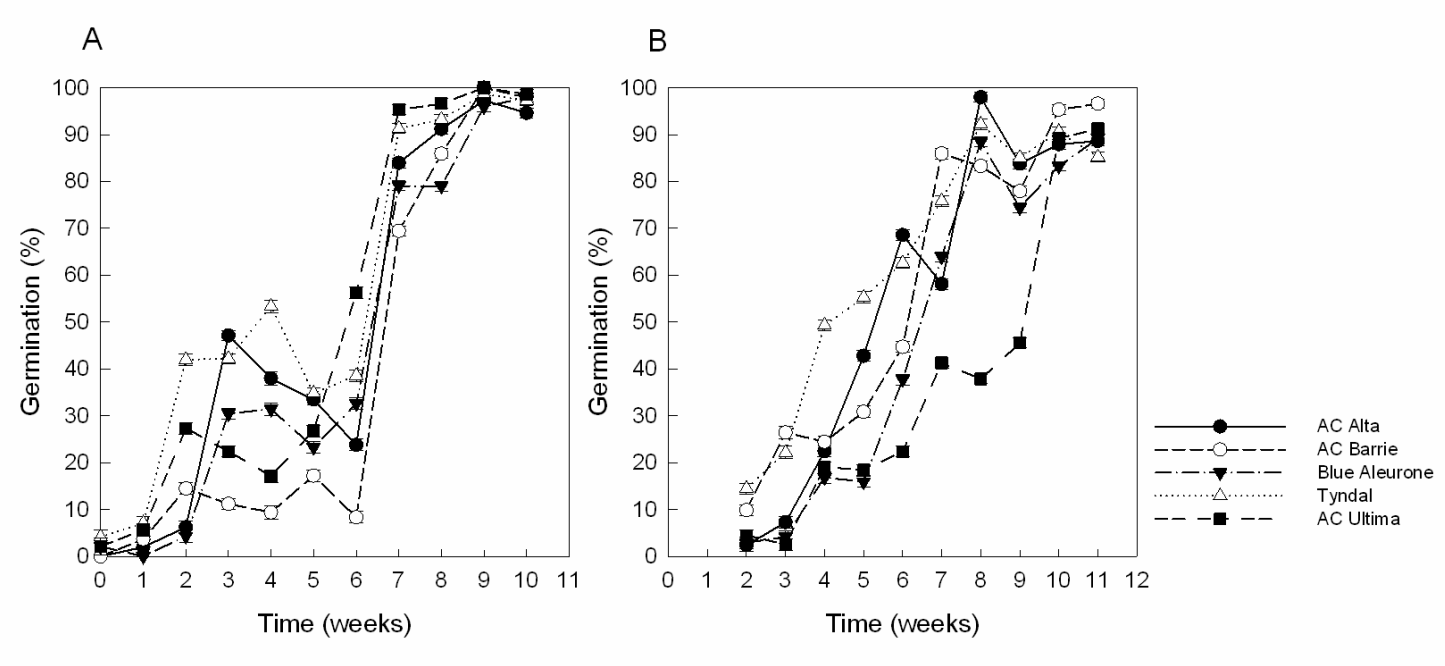

Figure 3.4. Germination of spring triticale cultivars: AC Alta, Blue Aleurone, Tyndal, and AC Ultima and wheat: AC Barrie A. in 2009 from 31 July until 9 Oct. (machine harvested 11 Sept., after 6 weeks) and B. in 2010 from 30 July until 15 Oct. (machine harvested 2 Oct., after 9 weeks). Symbols are the back-transformed least square mean weekly germination for each cultivar ± standard error of the mean. Standard error bars may be obscured by symbols.

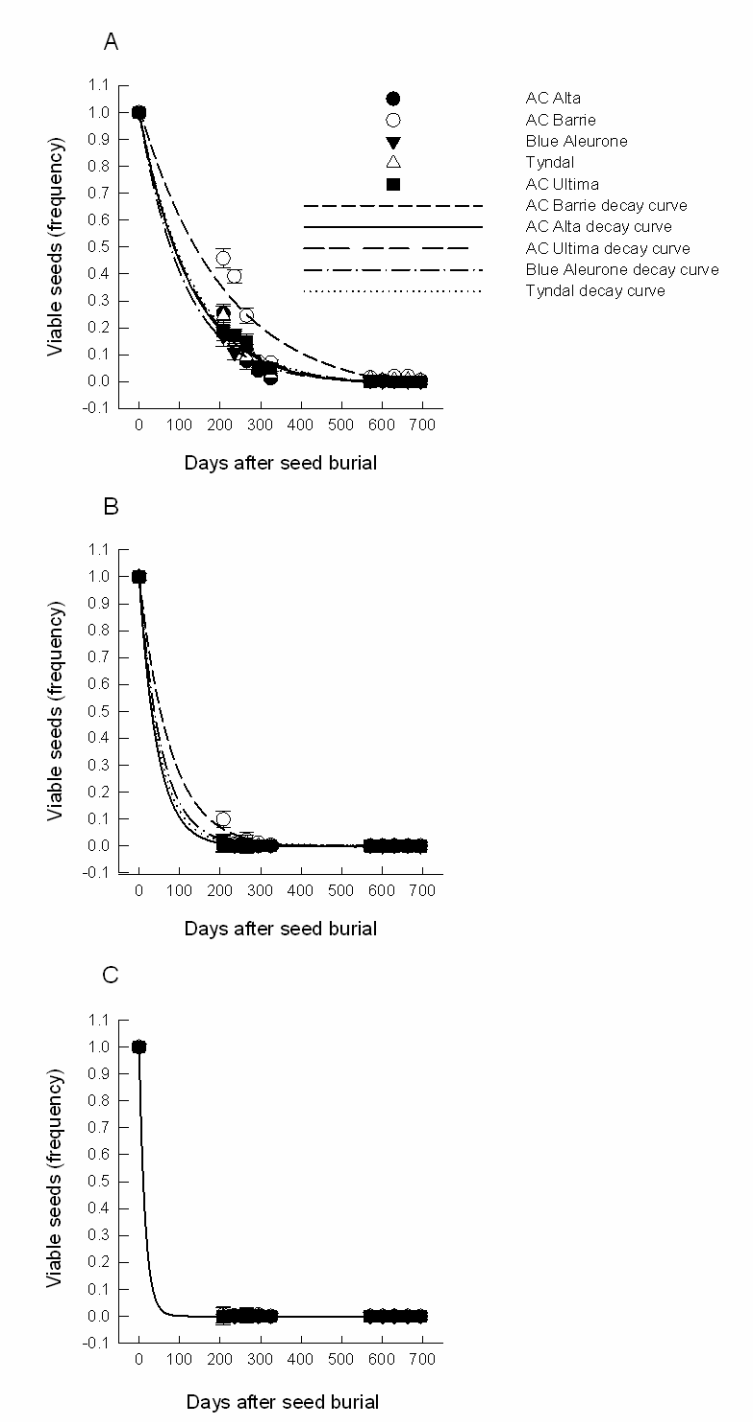

Figure 3.5. Seed survival curves for triticale cultivars AC Alta, Blue Aleurone, Tyndal, and AC Ultima and wheat AC Barrie A) on the soil surface; B) buried to 2 cm; and C) buried to 12 cm. Seed survival is estimated from the exponential decay, $P = ae^{-bd}$ where P is the probablility of viable seeds, a is the intercept set at 1, b is the slope of the curve and the rate of seed viability decline, and d is the number of days after seed burial) . Symbols are the

observed mean frequency of viable seeds ± standard error of the mean. Standard error bars may be obscured by symbols.

3.4 Literature Cited

Alberta Agriculture, Food and Rural Development. 2005. Triticale production and utilization manual 2005: spring and winter triticale for grain, forage and value-added. Alberta Agriculture, Food and Rural Development, Edmonton, Alberta, Canada.

Anderson, R.L., and G. Soper. 2003. Review of volunteer wheat (*Triticum aestivum*) seedling emergence and seed longevity in soil. Weed Tech. 17:620-626.

Baraibar, B., P.R. Westerman, and J. Recasens. 2009. Effects of tillage and irrigation in cereal fields on weed seed removal by seed predators. J. Appl. Ecol. 46:380-387.

Baskin, C.C., and J.M. Baskin. 1998. Seeds: ecology, biogeography, and evolution of dormancy and germination. Academic Press, San Diego, California, USA.

Beckie, H.J. and M.D.K. Owen. 2007. Herbicide-resistant crops as weeds in North America. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources. 44:1-22.

Beckie, H.J., L.M. Hall, M.J. Simard, J.Y. Leeson, and C.J. and Willenborg. 2010. A framework for post-release environmental monitoring of second-generation crops with novel traits. Crop Sci. 50:1587-1604.

Beres, B.L., R.E. Blackshaw, R.J. Graf, E. Bremer, K.N. Harker, and G.W. Clayton. 2010. Weed-competitive ability of spring and winter cereals in the northern Great Plains. Weed Tech. 24:108-116.

Biddulph, T.B., J.A. Plummer, T.L. Setter, and D.J. Mares. 2008. Seasonal conditions influence dormancy and preharvest sprouting tolerance of wheat (*Triticum aestivum* L.) in the field. Field Crops Res. 107:116-128.

Biddulph, T.B., J.A. Plummer, T.L. Setter, and D.J. Mares. 2007. Influence of high temperature and terminal moisture stress on dormancy in wheat (*Triticum aestivum* L.). Field Crops Res. 103:139-153.

Biddulph, T.B., T.L. Setter, J.A. Plummer, and D.J. Mares. 2005. Drought and high temperature increases preharvest sprouting tolerance in a genotype without grain dormancy. Euphytica: Int. J. Plant Breeding 143:277-283.

Brust, G.E., and G.J. House. 1988. Weed seed destruction by arthropods and rodents in low-input soybean agroecosystems. Am. J. Alternative Ag. 3:19-25.

Buraas, T., and H. Skinnes. 1985. Development of seed dormancy in barley, wheat and triticale under controlled conditions. Acta Agric. Scand. 35:233-244.

Burger, J.C., J.M. Holt, and N.C. Ellstrand. 2007. Rapid phenotypic divergence of feral rye from domesticated cereal rye. Weed Sci. 55:204-211.

Burnside, O.C., R.G. Wilson, S. Weisberg, and K.G. Hubbard. 1996. Seed longevity of 41 weed species buried 17 years in eastern and western Nebraska. Weed Sci. 44:74-86.

Canadian Food Inspection Agency. 2004. Assessment criteria for determining environmental safety of plants with novel yraits, Directive Dir94-08. Available at http://www.inspection.gc.ca/english/plaveg/bio/dir/dir9408e.shtml (verified 9 Nov. 2011). CFIA, Plant Bio-Safety Office, Ottawa, Ontario, Canada.

Canadian Triticale Biorefinery Initiative. 2011. Canadian triticale biorefinery intiative. Available at http://www.ctbi.ca (verified 12 Mar. 2012). Edmonton, Alberta, Canada.

Chee-Sanford, J.C., M.M. Williams, A.S. Davis, and G.K. Sims. 2006. Do microorganisms influence seed-bank dynamics? Weed Sci. 54:575-587.

Clarke, J.M. 1981. Effect of delayed harvest on shattering losses in oats, barley and wheat. Can. J. Plant Sci. 61:25-28.

Clarke, J.M. 1985. Harvesting losses of spring wheat in windrower/combine and direct combine harvesting systems. Agron. J. 77:13-17.

Conn, J.S., K.L. Beattie, and A. Blanchard. 2006. Seed viability and dormancy of 17 weed species after 19.7 years of burial in Alaska. Weed Sci. 54:464-470.

Cussans, G.W. 1978. The problem of volunteer crops and some possible means of their control. p. 915-921. *In* Proc. of the 14th British crop protection conference: weeds. Brighton, UK. 20-23 Nov. 1978. British Crop Protection Council. Brighton, UK.

Davis, A.S., K.A. Renner, E.C. Luschei, M.M. Williams II, C.L. Sprague, J.L. Lindquist, F. Forcella, J. Cardina, G. Kegode, and G.A. Johnson. 2005. Environmental factors affecting seed persistence of annual weeds across the U.S. corn belt. Weed Sci. 53:860-868.

De Corby, K.A., R.C. Van Acker, A. Brûlé-Babel, and L.F. Friesen. 2007. Emergence timing and recruitment of volunteer spring wheat. Weed Sci. 55:60-69.

DePauw, R.M., M.R. Fernandez, R.E. Knox, J.G. McLeod, T.N. McCaig, and J.M. Clarke. 1997. Registration of 'AC Barrie' wheat. Crop Sci. 37:289-289.

Dexter, J.E., A.J. Jhala, R.C. Yang, M.J. Hills, R.J. Weselake, and L.M. Hall. 2011. Harvest loss and seed bank longevity of flax (*Linum usitatissimum* L.): Implications for seed-mediated gene flow. Weed Sci. 59:61-67.

Eudes, F. 2006. Canadian triticale biorefinery initiative. p. 85-88. *In* W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G. Oettler and D. Salmon (eds.) Proc. of the 6th Int. Triticale Symp., Stellenbosch, South Africa. 3-7 Sept. 2006. Faculty of AgriSciences, Stellenbosch, South Africa.

FAOSTAT. 2010. Food and agriculture organization statistical database. Available at http://faostat.fao.org/site/567/DesktopDefault.aspx?PageID=567#ancor (verified 12 Mar. 2012). Rome, Italy.

Finkelstein, R., C. Steber, T. Ariizumi, and W. Reeves. 2008. Molecular aspects of seed dormancy. Ann. Rev. Plant Biol. 387-415.

Gilbert, G.S. 2002. Evolutionary ecology of plant diseases in natural ecosystems. Annu. Rev. Phytopathol. 40:13-43.

Government of Alberta Agriculture and Rural Development. 2010. Crop information: Spring triticale. Available at http://www.agric.gov.ab.ca/app95/loadCrop?action=display&id=53 (verified 9 Nov. 2011). Edmonton, Alberta.

Goyal A., B.L. Beres, H.S. Randhawa, A. Navabi, D.F. Salmon, and F. Eudes. 2011. Yield stability analysis of broadly adaptive triticale germplasm in southern and central Alberta, Canada for industrial end-use suitability. Can. J. Plant Sci. 91: 125-135.

Grabe, D.F. (ed.) 1970. Tetrazolium testing handbook for agricultural seeds. Contribution No. 29 to the handbook on seed testing. Association of Official Seed Analysts of North America. Ithaca, New York, USA.

Graziani, F., E. Pannacci, G. Covarelli and F. Tei. 2007. Post-dispersal weed seed predation by vertebrates and invertebrates in organic and conventional cereal fields. p. 196. *In* E. Fløistad (ed.) European Weed Research Society, 14th EWRS Symposium, Hamar, Norway. 17-21 June 2007. EWRS, Doorwerth, Netherlands.

Grundy, A.C., A. Mead, and S. Burston. 2003. Modelling the emergence response of weed seeds to burial depth: interactions with seed density, weight and shape. J. Appl. Ecol. 40:757-770.

Gulden, R. H., A. G. Thomas, and S. J. Shirtliffe. 2004. Secondary dormancy, temperature, and burial depth regulate seedbank dynamics in canola. Weed Sci. 52: 382-388.

Hagemann, M.G., and A.J. Ciha. 1987. Environmental × genotype effects on seed dormancy and after-ripening in wheat. Agron. J. 79:192-196.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y.T. Gan, H.A. Frederick, K.L. Sapsford, R.B. Irvine, and R.C. Van Acker. 2005. Glyphosate-resistant wheat persistence in western Canadian cropping systems. Weed Sci. 53:846-859.

Himi, E., D.J. Mares, A. Yanagisawa, and K. Noda. 2002. Effect of grain colour gene (R) on grain dormancy and sensitivity of the embryo to abscisic acid (ABA) in wheat. J. Exp. Bot. 53:1569-1574.

Hucl, P. 1996. Out-crossing rates for 10 Canadian spring wheat cultivars. Can. J. Plant Sci. 76:423-427.

Kalaitzandonakes, N. 2011. The economic impacts of asynchronous authorizations and low level presence: an overview. International Food and Agricultural Trade Policy Council. Washington, D.C., USA.

Komatsuzaki, M., and O. Endo. 1996. Seed longevity and emergence of volunteer wheat in upland fields. Weed Res. Jap. 41:197-204.

Leeson, J.Y., A.G. Thomas, L.M. Hall, C.A. Brenzil, T. Andrews, K.R. Brown, and R.C. Van Acker. 2005. Prairie weed surveys of cereal, oilseed and pulse crops from the 1970s to the 2000s. Rep. Weed Survey Series Publication 05-1CD. Agriculture and Agri-Food Canada, Saskatoon Research Centre, Saskatoon, Saskatchewan, Canada.

Légère, A., F.C. Stevenson, and D.L. Benoit. 2011. The selective memory of weed seedbanks after 18 years of conservation tillage. Weed Sci. 59:98-106.

Li, S., D.A. Lobb, and M.J. Lindstrom. 2007. Tillage translocation and tillage erosion in cereal-based production in Manitoba, Canada. Soil & Tillage Res. 94:164-182.

McKenzie, R.H., R.F. Dunn, B.L. Beres, P.G. Pfiffner, E. Bremer, and A.B. Middleton. 2007. Efficacy of high seeding rates to increase grain yield of

winter wheat and winter triticale in southern Alberta. Can. J. of Pl. Sci. 87:503-507.

McLeod, J.G., J.M. Clarke, R.M. DePauw. and W.H. Pfeiffer. 2000. Registration of 'AC Ultima' spring triticale. Crop Sci. 41:924-925.

McLeod, J.G., T.F. Townley-Smith, R.M. DePauw. and J.M. Clarke. 1996a. Registration of 'AC Alta' spring triticale. Crop Sci. 36:1415-1415.

McLeod, J.G., T.F. Townley-Smith, R.M. DePauw and J.M. Clarke. 1996b. AC Alta spring triticale. Can. J. of Pl. Sci. 76:139-141.

McLeod, J.G., J.B. Thomas, P.D. Brown, C.L. Vera, W.E. May, D.F. Salmon, and K. Sosulski. 2010. Changes in ethanol production potential due to species, cultivar and location on the Canadian prairie. Can. J. of Pl. Sci. 90:163-171.

McPherson, M.A., R.C. Yang, A.G. Good, R.L. Nielson, and L.M. Hall. 2009. Potential for seed-mediated gene flow in agroecosystems from transgenic safflower (*Carthamus tinctorius* L.) intended for plant molecular farming. Transgenic Res. 18:281-299.

Miller, S.D., C.M. Alford, and W.L. Stump. 2004. Feral rye: a serious threat to high quality wheat. B-1157. Available at http://www.wyomingextension.org/agpubs/pubs/B1157.pdf (verified 12 Mar. 2012) University of Wyoming Cooperative Extension Service, Laramie, Wyoming, USA.

Murdoch, A.J., and R.H. Ellis. 2000. Dormancy, viability and longevity. p. 183-214. *In* M. Fenner (ed.) Seeds: The ecology of regeneration in plant communities. 2nd ed. CABI Publishing, Wallingford, Oxon, UK.

Nielson, R.L., R. Yang, L.M. Hall, K.N. Harker, M.A. McPherson, and J.T. O'Donovan. 2009. Seed-mediated gene flow in wheat: seed bank longevity in western Canada. Weed Sci. 57:124-132.

O'Rourke, M.E., A.H. Heggenstaller, M. Liebman, and M.E. Rice. 2006. Post-dispersal weed seed predation by invertebrates in conventional and low-external-input crop rotation systems. Agr. Ecosyst. Environ. 116:280-288.

Paulsen, G.M., and A.S. Auld. 2004. Preharvest sprouting of cereals. p. 199-219. *In* R.L. Benech-Arnold and R.A. Sanchez (eds.) Handbook of seed physiology: applications to agriculture. 1st ed. The Haworth Press, New York, USA.

Pickett, A.A. 1993. Cereals: seed shedding, dormancy and longevity. Asp. Appl. Biol. 35:17-28.

Porter, R.H., M. Durrell, and H.J. Romm. 1947. The Use of 2, 3, 5 -Triphenyl-Tetrazoliumchloride as a measure of seed germinability. Plant Physiol. 22:149-159.

SAS Institute Inc. 2007. SAS/STAT User's Guide: Statistics. SAS Institute, Cary, North Carolina, U.S.A.

Salmon, D.F., and J.H. Helm. 1985. Preharvest and postharvest dormancy in spring triticale. Agron. J. 77:649-652.

Salmon, D.F., C. Bergen, M. Oro, B. Chapman, S. Albers, J.H. Helm, P.E. Juskiw, and J. Nyachiro. 2007. Registration of 'Tyndal' Triticale. J. of Pl. Registrations 1:100.

Saxton, A.M. 1998. A macro for converting mean separation output to letter groupings in Proc Mixed. p. 1243-1246 *In* Proc. 23rd SAS Users Group Intl., Nashville, TN. 22-25 March 1998. SAS Institute, Cary, NC, USA.

Seerey, N., S.J. Shirtliffe, and P. Hucl. 2011. Seeds from unthreshed wheat (*Triticum aestivum* L.) spikes have reduced field emergence compared with threshed seed regardless of cultivar. Can. J. Plant Sci. 91:583-585.

Skovmand, B., P.N. Fox, and R.L. Villareal. 1984. Triticale in commercial agriculture: progress and promise. Adv. Agron. 37:1-45.

Stump, W.L. and P. Westra. 2000. The seedbank dynamics of feral rye (*Secale cereale*). Weed Tech. 14:7-14.

Torresen, K.S., M.B. Hagemo, H.J. Tandsaether, and R. Skuterud. 2003. Long-term experiments with reduced tillage in spring cereals. I. Effects on weed flora, weed seedbank and grain yield. Crop Protection. 22:185-200.

Van Acker, R. C., W. J. Bullied, and M. J. du Croix Sissons. 2004. Tillage index predicts weed seedling recruitment depth. Can. J. Plant Sci. 84: 319-236.

Van Mourik, T.A., T.J. Stomph, and A.J. Murdoch. 2005. Why high seed densities within buried mesh bags may overestimate depletion rates of soil seed banks. J. Appl. Ecol. 42:299-305.

Warwick, S.I., and C.N. Stewart Jr. 2005. Crops come from wild plants - how domestication, transgenes, and linkage together shape ferality. p. 9-25. *In* J.B. Gressel (ed.) Crop ferality and volunteerism. Taylor & Francis Group, Boca Raton, Florida, USA.

Weidner, S., R. Amarowicz, M. Karamac, and G. Dabrowski. 1999. Phenolic acids in caryopses of two cultivars of wheat, rye and triticale that display

different resistance to pre-harvest sprouting. Eur. Food Res. and Tech. 210:109-113.

White, A.D., D.J. Lyon, C. Mallory-Smith, C.R. Medlin, and J.P. Yenish. 2006. Feral rye (*Secale cereale*) in agricultural production systems. Weed Tech. 20: 815-823.

Willenborg, C.J., and R.C. Van Acker. 2008. The biology and ecology of hexaploid wheat (*Triticum aestivum* L.) and its implications for trait confinement. Can. J. Plant Sci. 88:997-1013.

Zadoks, J.C., T.T. Chang, and C.F. Konzak. 1974. A decimal code for the growth stages of cereals. Weed Res. 14: 415-421.

Chapter 4. Control and Fecundity of Volunteer Triticale (× *Triticosecale* Wittmack)

4.1. Introduction

Cereal crop volunteers become weeds in subsequent crops in western Canada (Leeson et al., 2005). Volunteer cereal densities are variable but often warrant control measures. Within the following rotational crops across the Canadian prairies, volunteer wheat was the 7th, 13th, 5th, and 5th most abundant weed in canola (*Brassica napus* L.), barley (*Hordeum vulgare* L.), field pea (*Pisum sativum* L.), and flax (*Linum usitatissimum* L.) fields, respectively (Leeson et al., 2005). Wild oat (*Avena fatua* L.) and volunteer cereals are the dominant weeds in canola fields on the Canadian prairies (O'Donovan et al., 2005). Volunteer cereals result from natural seed losses such as shatter, crop lodging, animal herbivory, or hail, as well as through losses as a result of mechanical harvest operations (Anderson and Soper, 2003; Clarke, 1985; Willenborg and Van Acker, 2008). Alberta field surveys have shown that volunteer wheat had a high field density of 128 plants m^{-2} (Leeson et al., 2002), although densities have been increasing over decadal periods (1970's to 2000's) (Leeson et al., 2005). Prespray densities of volunteer wheat in Canada range from 1 to 171 plants m^{-2} (Marginet, 2001), although following incrop weed control measures Leeson et al. (2005) showed densities averaged 6 plants m^{-2} in fields where it was found. However, densities can be as high as 60 to 280 plants m^{-2} (Leeson et al. 2005). Volunteer

barley at densities from 3 to 48 plants m^{-2} caused yield loss in the following wheat crop in western Canada and produced up to 7,663 kg ha^{-1} seed in low-density wheat crops (O'Donovan et al., 2007a). In addition to competing with a crop, volunteers that escape or survive control measures produce seed and contribute to adventitious presence (AP), the inadvertent and undesirable inclusion of seeds or other materials within harvested crop seed (Kershen and McHughen, 2005).

The potential introduction of genetically modified (GM) cereal crops necessitates more stringent volunteer control efforts. Since the first GM crops were commercially cultivated in 1996, the GM crop production area has been expanding globally (James, 2010). Concerns over potential market harm has blocked the introduction of GM wheat in North America (Wilson et al., 2008) and to date, only GM cultivars of canola, corn (*Zea mays* L.), and soybean [*Glycine max* (L.) Merr.] are commercially grown on significant areas in Canada (Demeke et al., 2006). GM seed may pose potential economic risks to established cereal markets, but also environmental risks in the event that the introduced transgenes confer a fitness advantage to wild or weedy relatives ([CFIA] Canadian Food Inspection Agency, 2004; Parker and Kareiva, 1996). Prior to the release of GM crops, risk assessments are conducted using case-by-case trait-based evaluations to predict invasiveness, including the potential for pollen- and seed-mediated gene flow (Wolt, 2009). Control of GM volunteers is vital to minimize AP of GM seeds and mitigate environmental risk of weediness.

There is growing interest within Canada to develop triticale (× *Triticosecale* Wittmack), an intergeneric hybrid between wheat and rye

(Kavanagh et al., 2010), as a feedstock for bio-ethanol production, monomer and polymer production, and bio-refining for chemical production (Goyal et al., 2011). These uses are predicated on genetic modification, and are predicted to increase the Canadian triticale production area to 400, 000 ha by 2015 (Canadian Triticale Biorefinery Initiative, 2010). Triticale is currently a minor crop on the Canadian Prairies primarily grown for livestock forage and grazing. In 2010, triticale was grown on over 3.9 million ha worldwide with ~0.6% in Canada. In comparison, wheat was grown on over 216 million ha in 2010 with ~3.8% grown in Canada (FAOSTAT, 2012). Triticale, like wheat, is primarily self-pollinating (Kavanagh et al., in press), therefore gene flow is more likely to occur via seed than pollen (Beckie et al., 2010). Assessing environmental risk includes documenting volunteer frequency and abundance through the entire lifecycle by determining relative plant density, growth, fecundity, germination, dormancy, and seed survival among various other fitness characteristics (Parker and Kareiva, 1996). The density of volunteer triticale depends on the initial crop harvest loss and seed viability and persistence. Triticale and wheat share similar cultural practices for seeding and harvesting; however, triticale can be more competitive than wheat and can have higher yields (Alberta Agriculture, Food and Rural Development, 2005; Beres et al., 2010; McKenzie et al., 2007; Goyal et al., 2011; Harker et al., 2011). Neither triticale harvest losses nor volunteer densities have been documented. However, because harvest practices and seed sizes are similar to wheat, triticale harvest loss may be similar to wheat.

Control of volunteer triticale is not well studied, but herbicides used for grass weed control and other volunteer cereals are expected to be effective. Chambers et al. (1995) showed that a number of aryloxyphenoxypropionate and cyclohexandione herbicides effectively controlled volunteer winter triticale in Australia. Blackshaw et al. (2006) determined that volunteer wheat could be controlled effectively with recommended rates of clethodim and clodinafop-P alone and with broadleaf tank mix partners 2, 4-D ester, bromoxynil, and bromoxynil + MCPA ester. Likewise, Harker et al. (2005) showed that the volunteer GM wheat seed bank could be rapidly depleted if volunteers were prevented from setting seed using tillage and preseed and incrop herbicides. Volunteer cereals represent part of the monocot weed spectrum and are usually controlled with the same herbicides used for wild oat in broadleaf crops, but no selective incrop herbicide options are possible in most cereal crops. O'Donovan et al. (2007b) indicated that wild oat populations decreased over time in barley crops when wild oats were prevented from setting or shedding seeds by early cutting for silage. Because canola is often grown in rotation with cereal crops and most canola grown in western Canada is glyphosate or glufosinate tolerant, farmers will use these systems with their intended herbicides (O'Donovan et al., 2005). Effective management strategies that include herbicide options to control volunteer GM triticale need to be established.

Managing GM volunteers within agro-ecological systems is necessary to limit AP in subsequent crops and is critical to mitigating environmental harm. Triticale volunteers should be eliminated prior to seed set in order to minimize

replenishment of the seed bank or prevent AP during harvest of the following crop. We examined best management practices for controlling volunteer triticale: 1) by evaluating preseeding and incrop herbicide applications in subsequent crops and 2) by assessing the fecundity of three spring triticale cultivars grown in the absence of competition.

4.2. Materials and Methods

4.2.1. Volunteer triticale control

Field experiments were established in Alberta, Canada at two locations each in 2006 and 2007 to assess the control and fecundity of volunteer triticale in following crops. A split-plot design with four replicates was used at all locations where cropping system was the main plot and herbicide application timing was the sub-plot. In 2006, trials were conducted at Calmar (53^{o}17'15" N, 113^{o}52'48" W) and University of Alberta Ellerslie Research Station (53^{o}25'24"N, 113^{o}43'52"W) and in 2007 trials were conducted at University of Alberta Ellerslie Research Station and University of Alberta Edmonton Research Station (53^{o}29'19"N, 113^{o}34'8"W). The Ellerslie, Edmonton, and Calmar sites have Eluviated Black Chernozemic soils.

Soil available nutrients were determined from composite 0 to 15 cm soil samples taken for each location and year (Table 4.1). Approximately 20 soil samples (roughly 50 cm^{3} each) were taken randomly from within each trial area, combined, stirred, and analyzed by Exova. In 2006, experiments at Ellerslie and Calmar were established on Roundup Ready® canola stubble. Volunteer canola

and other broad-leaved weeds were controlled with glyphosate + bromoxynil (glyphosate 540 g/L at 270 g ae/ha and bromoxynil 280 g/L at 330 g ai/ha) on 11 May and a second pre-emergent application of bromoxynil (bromoxynil 280 g/L at 330 g ai/ha) was made at Calmar on 1 June 2006. In 2007, Ellerslie was established on chemical fallow (glyphosate + quizalofop) and glyphosate (glyphosate 540 g/L at 270 g ae/ha) was applied pre-emergence on 14 May. Edmonton trials were placed on barley silage in 2007 and glyphosate applied 2 May to control weeds. A simulated stand of volunteer triticale 'Pronghorn' was direct-seeded at Calmar and Ellerslie on 11 May in 2006 and 1 May at Edmonton and 5 May at Ellerslie in 2007 at 27 to 30 kg ha^{-1} or 80 seeds m^{-2} (targeting a density of 75 plants m^{-2}) similar to volunteer wheat densities established in Blackshaw et al. (2006) and Rainbolt et al. (2004).

Glyphosate-tolerant canola (Roundup Ready® 'DKL 3465' (RR), an open-pollinated cultivar); glufosinate-tolerant canola (Liberty Link® 'Invigor 5030' (LL), a hybrid cultivar), field pea ('DS Admiral'), and imidazolinone-tolerant wheat (Clearfield® 'CDC Imagine') were seeded on 30 May in 2006 and 24 May in 2007 using a double disk seeder[1]. Target seeding rates for each crop were: canola (DKL 3465) 160 to 180 seeds m^{-2}; canola (Invigor 5030) 160 to 170 seeds m^{-2}; pea 100 to 115 seeds m^{-2}; and wheat 290 to 310 seeds m^{-2}. Seeding rates were based on 1000-kernel weights, percent germination, and an assumption of 5% mortality. Nitrogen (46-0-0, urea) at 112 kg ha^{-1} was broadcast and phosphorus (0-45-0, P_2O_5) at 45 kg ha^{-1} was placed with the seed. Each plot was comprised of 6 crop-rows at 20 cm spacing and 7 m length. Because all crops

were seeded at the same time and randomized within the trial area, seeding depth could not be adjusted for each crop, but was kept at 2 cm. Seasonal and long-term average climate data were summarized for each location from the nearest weather station (Figure 4.1).

Preseeding applications were made when triticale was at the 2 leaf stage on 24 May in 2006 and 22 and 26 May in 2007 at Edmonton and Ellerslie, respectively. In 2006, incrop applications were made on 28 June once the wheat had reached the appropriate 4 leaves for application with imazamox + 2, 4-D ester. Volunteer triticale ranged from the 5 leaf, 2 tillers to flag leaf stage. Glyphosate tolerant canola had 5 to 6 leaves, glufosinate tolerant canola had 6 to7 leaves, pea had 6 nodes and wheat had 4 leaves, 2 tillers. In 2007, incrop applications were made on 13 and 15 June at Ellerslie and Edmonton, respectively. Volunteer triticale was at the 5 leaf, 2 tillers stage. Glyphosate tolerant canola had 2 leaves, glufosinate tolerant canola had 2 to 3 leaves, pea had 4 nodes and wheat had 3 to 4 leaves. Recommended herbicide label rates were used for both timings and an untreated control was included for each crop (Table 4.2). In 2006, herbicides were applied with a self-propelled high clearance Spider Trac research sprayer[2] traveling at 5.3 km h^{-1} and equipped with multiple shrouded 2 m booms. Booms contained four Tee Jet® XR 110015 flat fan nozzles[3] calibrated to deliver a volume of 100 L ha^{-1} at 200 to 214 kPa with CO_2. In 2007, Ellerslie preseeding and Edmonton incrop treatments were applied with a 2 m backpack sprayer; however the same nozzles and water volumes were used as in

2006. The Spider Trac sprayer was used in 2007 for the Edmonton preseeding and Ellerslie incrop applications using the same settings as in 2006.

Three 0.25 m^2 quadrats were established in each plot and data were taken from within these quadrats throughout the season. Quadrats were subsamples within each treatment and were averaged for statistical analysis. In both 2006 and 2007, volunteer triticale and crop densities were assessed in each quadrat shortly after the preseeding herbicide application. Crop and volunteer triticale biomass were taken from within the quadrats at time of crop maturity, dried at 30^{o}C for 7 days, and weighed. At biomass harvest, fertile volunteer triticale densities were evaluated.

Seeds were threshed from surviving volunteers within established quadrats and thousand kernel weights (TKW) calculated from total seed weights and total seed numbers per quadrat. Germination was quantified by dividing the total amount of triticale seeds per quadrat into three replicates and placing seeds into 24×16×4 cm acrylic germination boxes[4] on top of absorbent non-toxic 15×23 cm steel blue blotter paper[5] designed to retain moisture, and covered with non-toxic white filter paper 15×23 cm No. 601 Whatman #1 equivalent[5]. A total water volume of 40 mL was added to each germination box which included a 0.2% solution of Helix Xtra® (thiamethoxam + difenoconazole + metalaxyl-M + fludioxonil) to reduce fungal growth. Seeds were maintained in the dark at ambient room temperature for one week. Germination was defined as radicle shoot >1 mm and seeds that germinated were recorded and discarded.

Plots were harvested with a small plot combine[6], seeds dried for 1 week at 30^{o}C and triticale was separated from wheat using a rotating drum-style indent seed separator[7] and hand-sieves were used to manually separate triticale from peas and canola.

4.2.2. Triticale biomass and fecundity

Field experiments were conducted at University of Alberta Edmonton Research Station in 2008, 2009, and 2010 to assess the biomass and fecundity of three triticale cultivars in the absence of competition. A randomized complete block design was used each year with 12 replicates of 'AC Alta', 'Pronghorn', and 'AC Ultima' seeded 1 m apart in every direction. Plots consisted of one plant. Competition from weeds was minimized by hoeing approximately twice weekly and chlorpyrifos (Lorsban® 4E 480 g L^{-1}; Dow AgroSciences Canada, Inc., Calgary, AB, Canada, www.dowagro.com/ca) was applied for cutworm control.

Triticale was seeded by hand on 8 May, 12 May, and 12 May in 2008, 2009, and 2010, respectively. Nitrogen (100 kg ha^{-1} of urea, 46-0-0) was side banded and seed-placed phosphorus at 34 kg ha^{-1} (phosphate, 0-45-0) applied. Plants were thinned to a single plant within 2 weeks of seeding and tomato cages were placed around each plant to provide structural support from the wind. In 2008, plants sustained mammalian predation and hail damage. Data from damaged plants were excluded (4, 6, and 6 replicates remained intact for each of 'AC Alta', 'Pronghorn', and 'AC Ultima', respectively). Below average rainfall was received in 2009 (Figure 4.1); and plants were watered approximately twice weekly in June and July and every second day during a period of daytime highs

over 30^{o}C (approximately 25 mm per plant; approximately 475 mm total supplemented water).

At crop maturity (grain moisture <14%), plants were cut at the soil surface, and the number of tillers with seeds recorded, bagged and dried at 30^{o}C for 7 days and total dry weight biomass was recorded. Harvest occurred on 7 Oct, 23 Sept, and 6 Oct in 2008, 2009, and 2010, respectively. Each plant was threshed, total seed weight and seed numbers per plant recorded, and TKW were calculated as the total seed weight per plant divided by the total number of seeds produced per plant.

4.2.3. Statistical analysis

4.2.3.1. Volunteer triticale control

Because locations varied between years, locations within years were considered as four environments: Calmar 2006, Ellerslie 2006, Ellerslie 2007, and Edmonton 2007. Triticale preharvest density (plants m^{-2}) and triticale fecundity (seeds m^{-2}) were square-root transformed; triticale biomass (g m^{-2}), triticale AP (kg ha^{-1}), and crop biomass (g m^{-2}) were $\log_{10}$ transformed; and triticale germination frequencies were arc-sin transformed to conform to assumptions of normality and homogeneity of variance; crop yields did not require transformation. Differences between environments were assessed with ANOVA using mixed model procedures (SAS Institute Inc., 2007) where environment, crop, and timing were fixed effects and replicate, replicate × environment, replicate × crop, and replicate × environment × crop were random effects (Table 4.3). Environments were separated and ANOVA performed on a reduced model

where crop and timing were fixed effects and replicate and replicate × crop were random effects. Initial volunteer triticale densities were not evaluated for Ellerslie 2006 or Calmar 2006, but were used as a covariate for measures of triticale fecundity and production when significant for Ellerslie 2007 and Edmonton 2007. Least Square Means and standard errors were back-transformed and all possible comparisons of herbicide timing were made within each crop using Bonferroni-adjusted p-values (p<0.0083 for six possible comparisons of four herbicide timings).

4.2.3.2. Triticale fecundity

Analyses of variance using mixed model procedures were initially performed to test significance between years (SAS Institute Inc., 2007). Biomass data were square root transformed and thousand kernel weights were $\log_{10}$ transformed to meet the assumptions of normality and homogeneity of variances. Because year and the interaction between year and cultivar were significant, years were analyzed seperately where cultivar was a fixed effect and replicate was a random effect. For the reduced model, untransformed data for each measure met assumptions of ANOVA.

4.3. Results and Discussion

4.3.1. Volunteer triticale control

The four environments (locations within years) were significantly different for measures of volunteer triticale density and fecundity and crop yield (Table 4.3) and we therefore analyzed them separately. Because crops were a component

of a crop-specific herbicide system, differences between crop systems were not statistically compared.

4.3.1.1. Volunteer triticale density

Volunteer triticale densities were evaluated following crop-specific incrop herbicide applications with LL canola, RR canola, field pea and wheat (Figure 4.2). Volunteer triticale dry weight biomass generally followed the same trends as triticale densities (data not shown).

Preseeding herbicides significantly reduced volunteer triticale densities in 14 of 16 possible crop and environment combinations. Incrop herbicides were not as effective in 2006 as 2007, probably because applications in 2006 were made later and thus to more mature plants. De Corby et al. (2007) observed that volunteer wheat which had emerged early in spring and was at an advanced stage at the time of incrop application was not effectively controlled. Depending upon environmental conditions, some volunteer triticale could emerge the previous fall and then not survive the Canadian winter (Gruber et al., 2008), thereby reducing volunteer densities. In this study, application of both preseed and incrop herbicides was the most effective at minimizing volunteer triticale escapes and was more effective than all other treatment timings in 4 of 16 possible crop and environment combinations.

Preseeding glufosinate was not consistently effective on triticale; it is not registered as a preseeding herbicide. In LL canola, preseeding glufosinate significantly reduced triticale densities relative to the untreated control (where Bonferroni-adjusted p-values, $p<0.0083$ were used to separate means) in Calmar

2006 (p<0.0001) and Ellerslie 2007 (p=0.0035), but not in Ellerslie 2006 (p=0.0340) or Edmonton 2007 (p=0.0615) (Table 4.4; Figure 4.2). Incrop applications of glufosinate were similarly inconsistent, with significant triticale density reductions in 2007, but not 2006 (Table 4.4). The 2006 incrop applications were made when volunteer triticale plants were at an advanced stage, having between 2 tillers to flag leaf initiation, which resulted in reduced herbicide efficacy. The combination of preseed and incrop glufosinate consistently reduced volunteer densities in all environments.

In RR canola, volunteer triticale densities were consistently reduced with preseed glyphosate in all environments (p<0.0001) (Table 4.4; Figure 4.2). Incrop glyphosate and the combination of preseed and incrop glyphosate significantly reduced triticale densities below preseed glyphosate alone (p<0.0001); densities of volunteer triticale within incrop and the combination of preseed and incrop treatments were consistently <1 plant m^{-2} in all environments (Table 4.4).

In field pea, preseeding glyphosate significantly reduced triticale densities in all environments. Incrop imazamox/imazethapyr significantly reduced triticale densities at Ellerslie 2006 (p<0.0001), Ellerslie 2007 (p<0.0001), and Edmonton 2007 (p<0.0001) (Table 4.4). The combination of preseed glyphosate and incrop imazamox/imazethapyr significantly reduced triticale densities below densities found in preseed glyphosate alone in all environments (Table 4.4; Figure 4.2).

In wheat, preseed glyphosate reduced triticale densities in all environments (Table 4.4; Figure 4.2). Incrop imazamox + 2, 4-D ester did not significantly affect triticale in the 2006 environments because many triticale

plants were at advanced stages when incrop applications were made. However, incrop applications were effective in the 2007 environments (Table 4.4). When both preseed glyphosate and incrop imazamox + 2, 4-D ester were applied, triticale densities were significantly lower than that of preseed glyphosate alone at Calmar 2006 (p<0.0001) and Ellerslie 2007 (p<0.0001) (Table 4.4; Figure 4.2).

4.3.1.2. Volunteer triticale fecundity

Seed production was measured for surviving volunteer triticale plants from within individual quadrats (seeds m^{-2}) as a measure of triticale fecundity (Figure 4.3). We also quantified recovered triticale seeds separated from within harvested crops (kg ha^{-1}), which we describe as triticale AP (data not shown). AP is a function of the volunteer seed harvested and difference in seed size between weed and crop which influences seed separation at harvest. Because it varies with harvest conditions, AP is expected to vary between years and growers.

In LL canola, preseed glufosinate significantly reduced triticale fecundity by 59 to 93% in all environments (p<0.0001), relative to untreated controls (where Bonferroni-adjusted p-values, p<0.0083 were used to separate means) (Table 4.5; Figure 4.3), although at Ellerslie 2006 (p=0.0525) (Table 4.6) and Edmonton 2007 (p=0.1863) triticale AP was not significantly lower than untreated controls (data not shown). Incrop glufosinate significantly reduced triticale fecundity by 85 to 98% (Table 4.5; Figure 4.3) and AP by 88 to 96% (data not shown) (Table 4.6). In the 2006 environments, the combination of preseed and incrop glufosinate significantly further reduced volunteer triticale fecundity relative to preseeding or incrop applications alone (Table 4.5; Figure

4.3); however, triticale AP was lowest with both preseed and incrop glufosinate in all environments (data not shown) (Table 4.6).

In RR canola, volunteer triticale fecundity was significantly reduced by 80 to 96% following preseed glyphosate in all environments ($p<0.0001$) (Table 4.5; Figure 4.3). Incrop glyphosate was significantly more effective at reducing triticale fecundity than preseeding glyphosate alone, with fecundity reduced by >99% in all environments (Table 4.5). Preseeding and incrop glyphosate did not significantly contribute further to preventing triticale seed production (Table 4.5; Figure 4.3). Similar trends occurred with triticale AP (Table 4.6).

In field pea, the response of volunteer triticale fecundity was similar to RR canola. Triticale fecundity was reduced by 75 to 89% following preseed glyphosate (Table 4.5; Figure 4.3). Incrop imazamox/imazethapyr significantly reduced triticale fecundity by ≥ 99% ($p<0.0001$); however, the combination of preseed glyphosate and incrop imazamox/imazethapyr did not further significantly reduce triticale fecundity relative to the incrop application alone (Table 4.5; Figure 4.3). Triticale AP followed similar trends, although preseed glyphosate did not significantly affect triticale AP at Calmar 2006 ($p=0.0545$) and Ellerslie 2007 ($p=0.0352$) (data not shown) (Table 4.6).

In wheat, preseed glyphosate significantly reduced triticale fecundity in all environments by 88 to 91% ($p<0.0001$) (Table 4.5; Figure 4.3). Incrop imazamox + 2, 4-D ester significantly reduced volunteer triticale fecundity by 90 to 99% ($p<0.0001$) (Table 4.5). While the combination of preseed glyphosate and incrop imazamox + 2, 4-D ester further reduced triticale fecundity, they were not

significantly different than incrop treatments alone except at Ellerslie 2006 (p=0.0031) (Table 4.5; Figure 4.3). Triticale AP followed similar trends with some exceptions (data not shown) (Table 4.6).

Compared to untreated controls, preseeding herbicides generally reduced triticale fecundity by at least 60% and triticale AP was reduced by at least 48%. While the delayed incrop application showed reduced efficacy on older triticale plants, the survivors still produced less seed than preseed applications alone. Higher seed production in treatments receiving only a preseed herbicide application may be partly attributed to late triticale germination. Incrop applications reduced fecundity by 85 to >99% and AP was similarly reduced. The combination of both preseed and incrop herbicide applications effectively reduced triticale fecundity and AP by >97% and was more effective than incrop applications alone in two of 16 possible crop and environment combinations.

Triticale seed weight (thousand kernel weight or TKW) and percentage germination were evaluated to determine whether seeds produced by survivors were viable (Figure 4.4). Because fecundity was negligible for some treatments that received incrop applications, there were many missing values and no specific comparisons between treatments were made. In all environments, TKW was lower for seeds produced following incrop applications for all crops (data not shown). While fecundity was reduced by all herbicide timings, seed produced by survivors was generally capable of germinating, ranging from 93 to over 99% germination in untreated and preseed treatments (Figure 4.4). Germination was reduced following incrop applications of imidazolinone herbicides in pea and

wheat at Calmar 2006, Ellerslie 2006, and Ellerslie 2007 (40 to 86%). Seeds produced within these treatments were noticeably shriveled and small (data not shown). However, seeds produced by triticale that survived incrop glufosinate or glyphosate were capable of germinating (96 to over 99%).

4.3.1.3. Crop response to volunteer triticale

As expected, crop biomass and yields were reduced by increased volunteer triticale competition (Figure 4.5). In the 2006 environments, biomass was maximized in all crops when a preseed herbicide application was included (Table 4.7; Figure 4.5). Because triticale was at an advanced stage when incrop applications were made in 2006, crop-weed competition occurred over a longer period resulting in reduced crop biomass (data not shown). With some exceptions, all herbicide timings in 2007 improved crop biomass relative to untreated controls.

Yields for all crops were lower in 2006 than in 2007 (Figure 4.5) with the exception of field pea which had pods selectively predated by large mammals (deer feeding suspected although not observed) at Ellerslie 2006, Ellerslie 2007, and Edmonton 2007, although this was not directly measured or observed. The application of preseed herbicides improved yields in 12 of 16 possible crop and environment combinations (Table 4.8; Figure 4.5). Similar to effects on crop biomass, because triticale was at an advanced stage at the time of incrop application in 2006, delayed crop competition resulted in poor yields that were similar to untreated controls for all crops. Early weed removal is essential for maximizing crop yields, crop quality, and ultimately maximizing on-farm profits

(Harker et al., 2008; O'Donovan et al., 2007a; Harker et al., 2001; Martin et al., 2001; Upadhyay et al., 2006; May et al., 2003; Sikkema et al., 2005). The combination of preseed and incrop application significantly improved crop yields relative to untreated controls in 12 of 16 crop and environment combinations (Table 4.8; Figure 4.5). However, the use of both herbicide timings only further improved yields relative to preseed or incrop applications alone in 3 instances. In this study, maximum crop yields were obtained with only one herbicide application and the combination of both preseeding and incrop applications frequently did not further improve crop yields. From a strictly economical perspective of reducing the input costs of herbicide applications and maximizing yields, the recommendation for only one herbicide application could be argued. However, this study did not take into account the later emerging broadleaf and grass weeds that would likely also be present and require management in addition to volunteer triticale. Furthermore, in the event of GM triticale development, the need to minimize AP in the subsequent crop necessitates both a preseed and incrop herbicide application.

4.3.2. Triticale fecundity in the absence of competition

All measures of triticale productivity were significantly different between years and there were significant interactions between year and cultivar. Years were subsequently analyzed separately. As a measure of plant production, dry weight biomass and the number of tillers were assessed per plant. In 2008, there were no significant differences in cultivar dry weight biomass or number of tillers. In 2009 and 2010, ‘AC Alta’ had significantly more biomass per plant than

either ‘Pronghorn’ or ‘AC Ultima’, which resulted from ‘AC Alta’ having more tillers (42 to 47) than either ‘Pronghorn’ or ‘AC Ultima’(30 to 39) (Figure 4.6). Extensive tillering occurs when plants are grown in the absence of competition, however, when grown in a typical crop stand of 280 plants m^{-2}, Beres et al. (2010) showed that spring triticale produced an average of 1.4 tillers per plant, while wheat produced between 1.7 to 2 tillers per plant. As a crop, triticale is as or more competitive with weeds than wheat, barley, or rye (Beres et al., 2010; Harker et al., 2011).

Triticale fecundity was measured as the total number of seeds produced per plant and TKW. There were no significant differences in number of seeds produced per plant between cultivars in 2008, averaging 1723 seeds $plant^{-1}$. ‘AC Alta’ had significantly higher TKW than ‘Pronghorn’. In 2009 and 2010, ‘AC Alta’ had the highest fecundity (3180 to 3401 seeds $plant^{-1}$). In 2009, there were no significant differences in seed size attributed to cultivar where the average TKW was 62 g (Figure 4.6). In 2010, while ‘AC Alta’ had greater seed production, the thousand kernel weight was significantly lower (44 g) than either ‘Pronghorn’ (50 g) or ‘AC Ultima’ (49 g) (Figure 4.6). Beres et al. (2010) reported that spring and winter triticale grown in a typical crop stand produced the largest number of seeds per spike relative to barley, wheat, and rye, averaging 60 to 70 seeds per plant where the TKW averaged 33 to 43 g. In this study, as a volunteer in the following crops, untreated volunteer triticale produced between 76 and 143 seeds $plant^{-1}$. In the absence of herbicide application across all four environments, volunteer triticale seed production per plant was greatest in peas >

wheat > RR canola > LL canola. In a study determining relative competitive ability of spring crops on the Canadian prairies, Harker et al. (2011) found that under cooler, wetter conditions early in the season, hybrid canola cultivars were as competitive as barley with dicot weeds and in some instances with monocot weeds; hybrid cultivars were generally more competitive than open-pollinated canola cultivars. Traditionally, barley is a strong competitor with weeds relative to wheat, canola, flax or field pea (Harker, 2001). However, triticale is also a competitive crop under similar conditions (Harker et al., 2011; Beres et al., 2010). As a volunteer in the following crops, it is expected that triticale would be capable of supplying relatively large amounts of seed into the seed bank if left uncontrolled.

Recropping to spring cereals is not recommended because triticale volunteers cannot be selectively removed with the exception of imidazolinone-tolerant wheat. Canola commonly follows wheat or other cereals and triticale volunteers are manageable within this rotation. Volunteer triticale was consistently controlled in glyphosate-tolerant canola where incrop applications effectively minimized seed production. While preseed herbicides effectively remove early emerging volunteer triticale, late emergence can occur because crop seeding, similar to tillage practices, creates soil disturbance, enhances seed to soil contact, and stimulates weed emergence. Later emerging triticale volunteers avoid preseeding herbicides, necessitating an incrop application. However, relying on incrop applications alone may be risky from a crop yield and quality perspective if weather conditions delay application; herbicides are less effective on larger

more advanced weeds and delayed application also prolongs the period of competition with the crop.

Because minimizing seed production by volunteers is critical for managing GM crops, application of both preseed and crop-specific incrop herbicides will ensure that the least possible amount of seed is being returned to the seed bank or harvested in subsequent crops. Volunteer winter wheat was effectively controlled by preseed and incrop herbicides within herbicide-susceptible and glyphosate-tolerant winter canola (Bushong et al., 2011). Bushong et al. (2011) suggest that canola is a suitable crop to follow winter wheat in Oklahoma. Like triticale, flax is also being considered as a platform for genetic transformation in the form of novel oil creation. Flax has been shown to emerge as volunteers in high numbers following the year of production; however volunteers decrease dramatically following an incrop herbicide application and do not contribute to population growth (Dexter et al., 2010). However, McPherson et al. (2009) showed that safflower (*Carthamus tinctorius* L.) harvest losses ranged from 231 to 1069 seeds m^{-2}, although less than 50% were viable, and the density of volunteers that emerged the following spring ranged from 3 to 11 seedlings m^{-2}. Safflower volunteers that emerged were effectively controlled in chemical fallow fields, but some volunteers successfully produced seed when followed by a cereal crop. Volunteer wheat plants that escaped preseed and incrop management practices in flax can reseed and can cause volunteer populations to persist (De Corby et al., 2007). In a western Canadian weed survey, which assessed weed densities following incrop herbicide applications, volunteer wheat escapes

averaged 6 plants m^{-2} in fields where it was found (Leeson et al., 2005). GM triticale volunteer escapes may contribute to AP and persistence or movement of transgenes.

In this study, volunteer triticale was managed in several typical crop rotations including glyphosate and glufosinate tolerant canola, field pea and imidazolinone tolerant wheat. Crop rotations and the timing of herbicide applications are two integrated management tools for minimizing the effects of weeds. Herbicide timing recommendations for GM triticale are more complex than those based on economics where costs of the herbicide, fuel and time and commodity prices are the main decision drivers. Because GM traits are undesirable within the following harvested crop AP must be kept below thresholds for export markets. The effectiveness of the two herbicide timings was inconsistent between environments although the application of both timings provided the most consistent results. However, other integrated management tools, such as crop seeding rates; rates and placements of soil fertility; competitive crop cultivars; perennial crops; silage; or tillage for minimizing volunteer seed returns to the seed bank, were not explored in this study. In the event that GM triticale is developed, further assessment of other integrated management tools is warranted.

4.4. Sources of Materials

[1]Custom-made research seeders, Fabro Enterprises Ltd., Swift Current, SK, Canada

[2]Spider Trac Sprayer, West Texas Lee, Co., Idalou, TX, USA, www.westtexaslee.com

[3]Tee Jet® XR 110015 flat fan nozzles, Spraying Systems Co., Wheaton, IL, USA. www.teejet.com/english/home.aspx

[4]Acrylic germination boxes, 24×16×4 cm, Hoffman Manufacturing, Inc.

[5]Non-toxic 15×23 cm steel blue blotter paper and non-toxic white filter paper 15×23 cm No. 601 Whatman #1 equivalent, Hoffman Manufacturing, Inc.

[6]Small plot combine, Wintersteiger Nurserymaster Expert combine, Seedmech, Salt Lake City, UT, USA, 1996.

[7]Rotating drum-style indent seed separator, Westrup, Type LA-T/G LAT-0602, Slagelse, Denmark, www.westrup.com

Table 4.1. Soil characteristics for Ellerslie and Calmar in 2006 and Ellerslie and Edmonton in 2007.

					Soil Texture			Soil Fertility			
Location	Year	Soil pH	Soil OM[a] %	Soil EC[b] dS m^{-1}	Sand ----------%	Silt ----------	Clay ----------	N^{c} ---------kg ha^{-1}---------	P^{d}	K^{e}	S^{f}
Ellerslie	2006	5.8	11.5	0.47	27	42	31	64	50	293	40
Calmar	2006	6.5	8.3	0.42	24	45	31	39	25	368	45
Ellerslie	2007	5.8	10.9	0.25	32	41	27	28	41	353	10
Edmonton	2007	5.9	11.5	0.25	26	40	34	35	35	380	9

[a] OM, organic matter
[b] EC, electrical conductivity
[c] N, available nitrate
[d] P, available phosphate
[e] K, potassium
[f] S, sulfur

Table 4.2. Preseeding and incrop herbicide applications within each cropping system.

Cropping system Cultivar	Application Timing	Herbicide[a]	Formulation[b] g L^{-1}	Rate[c]
Roundup Ready® canola	Untreated	none		
DKL 3465	Preseeding	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	glyphosate	540 L	443 g ae ha^{-1}
	Preseed and	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	glyphosate	540 L	443 g ae ha^{-1}
Liberty Link® canola	Untreated	none		
Invigor 5030	Preseeding	glufosinate	150 L	400 g ai ha^{-1}
	Incrop	glufosinate	150 L	400 g ai ha^{-1}
	Preseed and	glufosinate	150 L	400 g ai ha^{-1}
	Incrop	glufosinate	150 L	400 g ai ha^{-1}
Pea	Untreated	none		
AC Admiral	Preseeding	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	imazamox	35% WDG	15 g ai ha^{-1}
		imazethapyr	35% WDG	15 g ai ha^{-1}
		adjuvant		0.5% v v^{-1}
	Preseed and	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	imazamox	35% WDG	15 g ai ha^{-1}
		imazethapyr	35% WDG	15 g ai ha^{-1}
		adjuvant		0.5% v v^{-1}
Clearfield® wheat	Untreated	none		
CDC Imagine	Preseeding	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	imazamox	20 SC	20 g ai ha^{-1}
		2, 4-D ester	560 SC	553 g ae ha^{-1}
		NIS		0.25%v v^{-1}
	Preseed and	glyphosate	540 L	270 g ae ha^{-1}
	Incrop	imazamox	20 SC	20 g ai ha^{-1}
		2, 4-D ester	560 SC	553 g ae ha^{-1}
		NIS		0.25%v v^{-1}

[a]glyphosate, Roundup Weathermax®; glufosinate, Liberty®; imazamox/imazethapyr, Odyssey®; adjuvant, Merge®; imazamox/2, 4-D Ester, Adrenalin SC®; NIS (non-ionic surfactant), AgSurf®.
[b]L, liquid; WDG, water dispersible granular; SC, suspension concentrate.
[c]rates given as grams acid equivalent per hectare (g ae ha^{-1}), grams active ingredient per hectare (g ai ha^{-1}) or percent volume per volume (% v v^{-1}).

Table 4.3. Analysis of variance for expanded model to determine whether the four environments (Ellerslie 2006, Calmar 2006, Ellerslie 2007, Edmonton 2007) can be combined for all measures of volunteer triticale density and fecundity and crop production.

Source	df	Triticale pre-harvest density[a] *p*-value	Triticale biomass[b] *p*-value	Triticale fecundity[a] *p*-value	Triticale AP[b] *p*-value	Triticale germination[c] *p*-value	Triticale TKW *p*-value	Crop biomass[b] *p*-value	Crop yield *p*-value
Environment	3	<0.0001	<0.0001	<0.0001	0.0008	<0.0001	0.0026	<0.0001	<0.0001
Crop	3	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.3845	<0.0001	<0.0001
Env × Crop	9	<0.0001	0.0001	0.0482	0.2616	0.0291	0.0027	<0.0001	<0.0001
Timing	3	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Env × Timing	9	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Crop × Timing	9	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.9934	<0.0001	<0.0001
Env × Crop × Timing	27	0.0005	0.0010	0.1437	0.0003	0.0038	0.0510	0.0325	<0.0001

Factors are considered to be significant at $p<0.05$.
[a]Square root transformed data
[b]Log_{10} transformed data
[c]Arc-sin frequency transformed data

Table 4.4. Analysis of variance for square root transformed volunteer triticale pre-harvest densities (plants m^{-2}) for crops, herbicide timings, and the covariate (if applicable) and significance (p-values) for all possible comparisons of herbicide timings within crop systems at Calmar and Ellerslie, 2006 and Ellerslie and Edmonton, 2007.

		Environment			
	Source	Calmar 2006	Ellerslie 2006	Ellerslie 2007	Edmonton 2007
Volunteer triticale pre-harvest density (plants m^{-2})					
	Crop	<0.0001	0.0002	<0.0001	<0.0001
	Timing	<0.0001	<0.0001	<0.0001	<0.0001
	Crop × Timing	<0.0001	<0.0001	0.0112	<0.0001
Covariate (Volunteer triticale preseed densities)		.	.	0.0003	0.0456
	Contrasts				
LL Canola	Untreated vs. Preseed[†]	<0.0001	0.0340	0.0035	0.0615
	Untreated vs. Incrop	0.2478	0.0497	<0.0001	0.0005
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	<0.0001	0.7390	0.0007	0.0675
	Preseed vs. Preseed + Incrop	0.0004	<0.0001	<0.0001	0.0008
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.1167	0.0815
RR Canola	Untreated vs. Preseed	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	0.6311	0.6602	0.8096	0.5429
Pea	Untreated vs. Preseed	0.0022	<0.0001	<0.0001	0.0037
	Untreated vs. Incrop	0.0126	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.5252	0.3472	<0.0001	0.0002
	Preseed vs. Preseed + Incrop	<0.0001	0.0007	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.9284	0.0785
Wheat	Untreated vs. Preseed	<0.0001	<0.0001	<0.0001	0.0002
	Untreated vs. Incrop	0.2127	0.5667	<0.0001	0.0051
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0006	<0.0001	0.0217	0.2270
	Preseed vs. Preseed + Incrop	0.0188	0.4208	0.0008	0.3773
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.2275	0.0406

[†]Comparisons of all possible herbicide timings within each crop system were made using Bonferroni-adjusted p-values, where $p<0.0083$.

Table 4.5. Analysis of variance for square root transformed volunteer triticale seed amount (seeds m^{-2}) or fecundity for crops, herbicide timings, and the covariate (if applicable) and significance (p-values) for all possible comparisons of herbicide timings within crop systems at Calmar and Ellerslie, 2006 and Ellerslie and Edmonton, 2007.

		Environment			
	Source	Calmar 2006	Ellerslie 2006	Ellerslie 2007	Edmonton 2007
Volunteer triticale seed amount (seeds m^{-2})					
	Crop	0.0067	<0.0001	0.6091	<0.0001
	Timing	<0.0001	<0.0001	<0.0001	<0.0001
	Crop × Timing	<0.0001	0.0015	0.0035	0.0506
Covariate (Volunteer triticale preseed densities)		.	.	0.0104	0.0173
	Contrasts				
LL Canola	Untreated vs. Preseed[†]	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0841	0.0049	<0.0001	<0.0001
	Preseed vs. Preseed + Incrop	0.0006	<0.0001	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	<0.0001	0.0061	0.4897	0.6503
RR Canola	Untreated vs. Preseed	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0022	<0.0001	<0.0001	0.0077
	Preseed vs. Preseed + Incrop	0.0042	<0.0001	<0.0001	0.0317
	Incrop vs. Preseed + Incrop	1.0000	1.0000	0.8689	0.5549
Pea	Untreated vs. Preseed	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	<0.0001	<0.0001	<0.0001	0.0014
	Preseed vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	0.0001
	Incrop vs. Preseed + Incrop	0.6713	0.4324	0.9500	0.3650
Wheat	Untreated vs. Preseed	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0014	0.9757	<0.0001	0.0183
	Preseed vs. Preseed + Incrop	<0.0001	0.0034	<0.0001	0.0015
	Incrop vs. Preseed + Incrop	0.1608	0.0031	0.3680	0.3515

[†]Comparisons of all possible herbicide timings within each crop system were made using Bonferroni-adjusted p-values, where $p<0.0083$.

Table 4.6. Analysis of variance for $\log_{10}$ transformed volunteer triticale yield (kg ha^{-1}) or adventitious presence (AP) for crops, herbicide timings, and the covariate (if applicable) and significance (p-values) for all possible comparisons of herbicide timings within crop systems at Calmar and Ellerslie, 2006 and Ellerslie and Edmonton, 2007.

		Environment			
	Source	Calmar 2006	Ellerslie 2006	Ellerslie 2007	Edmonton 2007
Volunteer triticale yield (kg ha^{-1})					
	Crop	0.0096	0.0020	0.0172	<0.0001
	Timing	<0.0001	<0.0001	<0.0001	<0.0001
	Crop × Timing	0.0025	0.0002	<0.0001	0.0018
Covariate (Volunteer triticale preseed densities)		.	.	ns	ns
	Contrasts				
LL Canola	Untreated vs. Preseed[†]	0.0015	0.0525	0.0041	0.1863
	Untreated vs. Incrop	0.0021	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.9061	0.0002	0.0065	<0.0001
	Preseed vs. Preseed + Incrop	0.0004	<0.0001	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	0.0003	<0.0001	0.0009	0.0462
RR Canola	Untreated vs. Preseed	<0.0001	0.0004	0.0001	0.0017
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0066	<0.0001	<0.0001	<0.0001
	Preseed vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	0.0104	0.2846	0.8427	0.9639
Pea	Untreated vs. Preseed	0.0545	0.0006	0.0352	0.0009
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Incrop vs. Preseed + Incrop	0.1921	0.1393	0.3681	0.5849
Wheat	Untreated vs. Preseed	0.0107	0.0001	<0.0001	0.0008
	Untreated vs. Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	<0.0001	<0.0001	0.0015	0.1787
	Preseed vs. Preseed + Incrop	<0.0001	<0.0001	0.0003	0.0013
	Incrop vs. Preseed + Incrop	0.0232	0.0162	0.5289	0.0456

[†]Comparisons of all possible herbicide timings within each crop system were made using Bonferroni-adjusted p-values, where $p<0.0083$.

Table 4.7. Analysis of variance for log_{10} transformed crop biomass (g m^{-2}) for crops, herbicide timings, and the covariate (if applicable) and significance (p-values) for all possible comparisons of herbicide timings within crop systems at Calmar and Ellerslie, 2006 and Ellerslie and Edmonton, 2007.

		Environment			
	Source	Calmar 2006	Ellerslie 2006	Ellerslie 2007	Edmonton 2007
Crop biomass (g m^{-2})					
	Crop	<0.0001	0.5575	<0.0001	<0.0001
	Timing	<0.0001	<0.0001	<0.0001	<0.0001
	Crop × Timing	0.0008	0.0011	0.7516	0.0059
	Covariate (Crop emergence)	ns	0.0104	0.0020	ns
	Contrasts				
LL Canola	Untreated vs. Preseed[†]	<0.0001	0.0013	0.0152	0.0123
	Untreated vs. Incrop	<0.0001	0.0015	0.0324	0.0013
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	0.0379	<0.0001
	Preseed vs. Incrop	0.0006	0.6640	0.7443	0.4206
	Preseed vs. Preseed + Incrop	0.5379	0.4185	0.7110	0.0193
	Incrop vs. Preseed + Incrop	<0.0001	0.2555	0.9638	0.1135
RR Canola	Untreated vs. Preseed	<0.0001	<0.0001	0.0007	0.0391
	Untreated vs. Incrop	<0.0001	0.0018	0.0011	0.0229
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	0.0012
	Preseed vs. Incrop	0.0044	0.0361	0.6484	0.8153
	Preseed vs. Preseed + Incrop	0.5714	0.8260	0.7489	0.1753
	Incrop vs. Preseed + Incrop	0.0017	0.0557	0.4249	0.2587
Pea	Untreated vs. Preseed	<0.0001	<0.0001	0.0242	<0.0001
	Untreated vs. Incrop	0.0035	0.6779	0.0004	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	0.0016	<0.0001
	Preseed vs. Incrop	0.1769	<0.0001	0.2093	0.0225
	Preseed vs. Preseed + Incrop	0.0016	0.2278	0.3793	0.0459
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.6970	0.7582
Wheat	Untreated vs. Preseed	<0.0001	<0.0001	0.0031	<0.0001
	Untreated vs. Incrop	0.0783	0.9514	0.0051	0.3371
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	0.0210	<0.0001
	Preseed vs. Incrop	<0.0001	<0.0001	0.9622	<0.0001
	Preseed vs. Preseed + Incrop	0.5022	0.7157	0.4507	0.7123
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.4984	<0.0001

[†]Comparisons of all possible herbicide timings within each crop system were made using Bonferroni-adjusted p-values, where $p<0.0083$.

Table 4.8. Analysis of variance for crop yield (kg ha^{-1}) for crops, herbicide timings, and the covariate (if applicable) and significance (p-values) for all possible comparisons of herbicide timings within crop systems at Calmar and Ellerslie, 2006 and Ellerslie and Edmonton, 2007.

		Environment			
	Source	Calmar 2006	Ellerslie 2006	Ellerslie 2007	Edmonton 2007
Crop yield (kg ha^{-1})					
	Crop	<0.0001	<0.0001	<0.0001	<0.0001
	Timing	<0.0001	<0.0001	<0.0001	<0.0001
	Crop × Timing	<0.0001	<0.0001	0.0002	<0.0001
	Covariate (Crop emergence)	ns	ns	ns	ns
	Contrasts				
LL Canola	Untreated vs. Preseed[†]	<0.0001	<0.0001	0.0002	0.0186
	Untreated vs. Incrop	0.4162	0.0220	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0007	0.0003	0.1638	0.0013
	Preseed vs. Preseed + Incrop	0.9192	0.0079	0.1546	<0.0001
	Incrop vs. Preseed + Incrop	0.0005	<0.0001	0.9742	0.0060
RR Canola	Untreated vs. Preseed	0.0004	<0.0001	<0.0001	<0.0001
	Untreated vs. Incrop	0.2704	0.0105	<0.0001	<0.0001
	Untreated vs. Preseed + Incrop	0.0008	<0.0001	<0.0001	<0.0001
	Preseed vs. Incrop	0.0088	<0.0001	0.0795	0.0155
	Preseed vs. Preseed + Incrop	0.9007	0.1145	0.0532	0.0618
	Incrop vs. Preseed + Incrop	0.0118	<0.0001	0.8475	0.5438
Pea	Untreated vs. Preseed	<0.0001	0.9176	0.9940	0.8821
	Untreated vs. Incrop	0.2271	0.9546	0.8768	0.9343
	Untreated vs. Preseed + Incrop	<0.0001	0.9518	0.8892	0.9813
	Preseed vs. Incrop	<0.0001	0.8726	0.8709	0.9476
	Preseed vs. Preseed + Incrop	<0.0001	0.9657	0.8813	0.9006
	Incrop vs. Preseed + Incrop	<0.0001	0.9066	0.9894	0.9529
Wheat	Untreated vs. Preseed	<0.0001	<0.0001	0.0003	<0.0001
	Untreated vs. Incrop	0.0111	0.2125	0.0228	0.0559
	Untreated vs. Preseed + Incrop	<0.0001	<0.0001	0.0759	<0.0001
	Preseed vs. Incrop	<0.0001	<0.0001	0.1102	0.0023
	Preseed vs. Preseed + Incrop	0.0197	0.2895	0.0352	0.8883
	Incrop vs. Preseed + Incrop	<0.0001	<0.0001	0.5855	0.0016

[†]Comparisons of all possible herbicide timings within each crop system were made using Bonferroni-adjusted p-values, where $p<0.0083$.

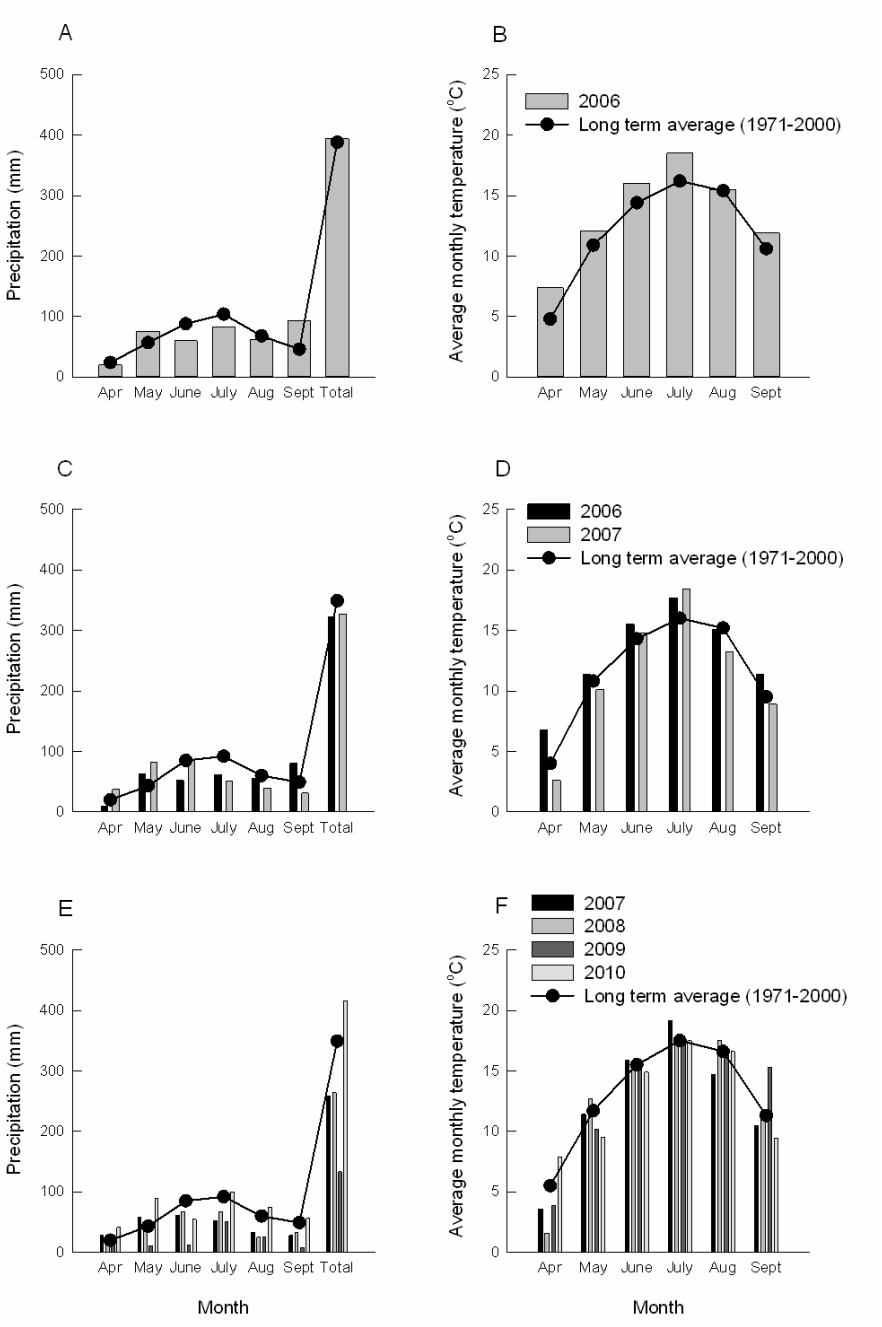

Figure 4.1. Summary of monthly weather data for 2006, 2007, 2008, and 2009 along with the long-term (30 year) average A. total precipitation at Calmar 2006; B. average temperatures at Calmar 2006; C. total precipitation at Ellerslie 2006 and 2007; D. average temperatures at Ellerslie in 2006 and 2007; E. total precipitation at Edmonton in 2007, 2008, 2009, and 2010, and F. average temperature at Edmonton in 2007, 2008, 2009, and 2010. Weather data were compiled from the Environment Canada website for Ellerslie (Edmonton International Airport), Calmar (Calmar weather station), and Edmonton

(University of Alberta Edmonton Research Station, Metabolic Unit available at http://www.climate.weatheroffice.ec.gc.ca/climateData/canada_e.html).

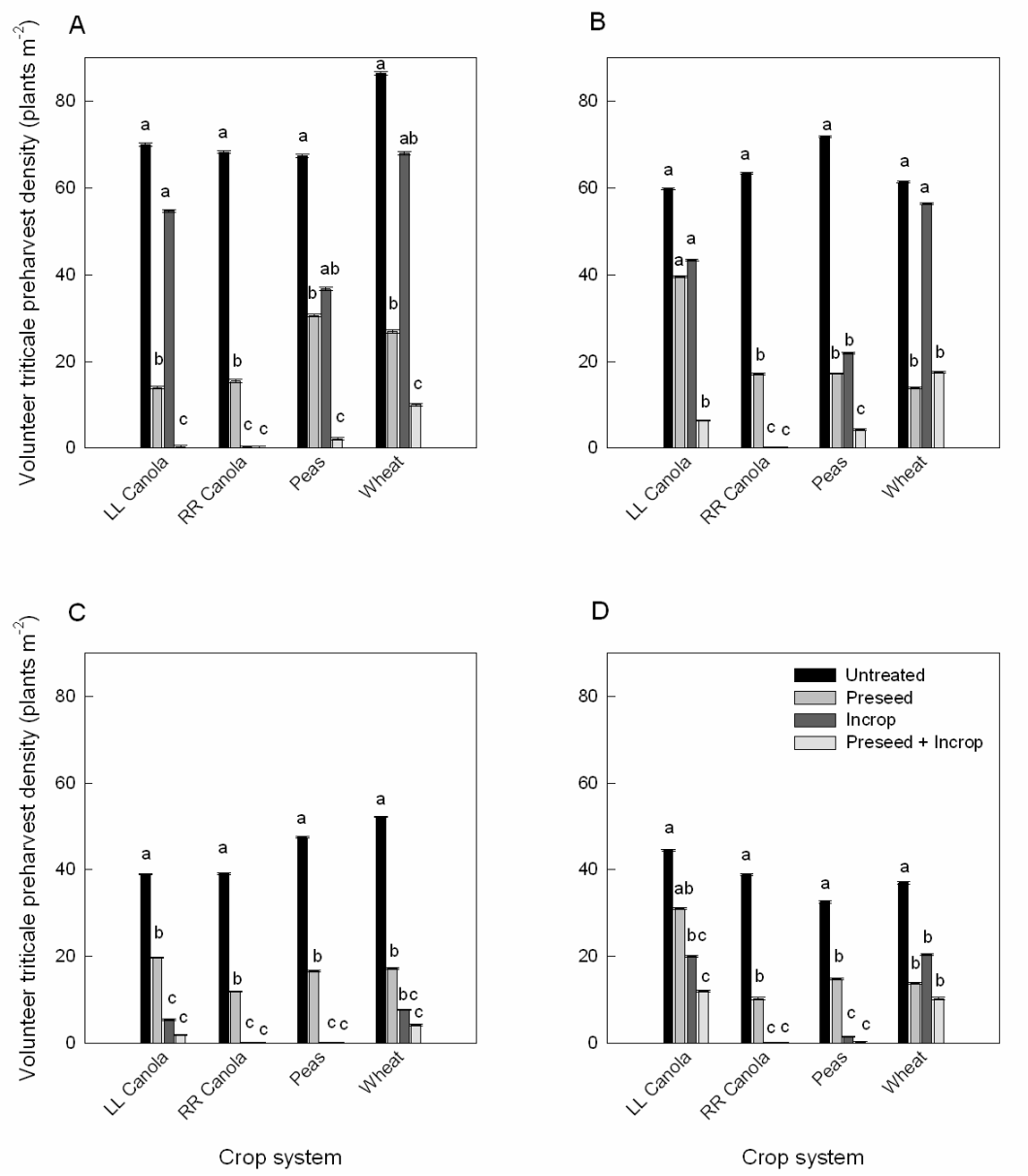

Figure 4.2. Effect of preseed, incrop, and preseed followed by incrop herbicide applications on volunteer triticale preharvest densities (plants m^{-2}) relative to untreated controls for each of four cropping systems: glufosinate-tolerance canola (LL canola), glyphosate-tolerant canola (RR canola), field pea, and imidazolinone-tolerant wheat at A. Calmar 2006, B. Ellerslie 2006, C. Ellerslie 2007, and D. Edmonton 2007. Densities are expressed as back-transformed (the analysis was conducted on square-root transformed data) LSMeans ± standard error bars for each treatment. Treatment timings with the same letter are not significantly different for a given crop and environment (Bonferroni-adjusted p-values, where $p<0.0083$) within each crop system. Crop systems were not statistically compared.

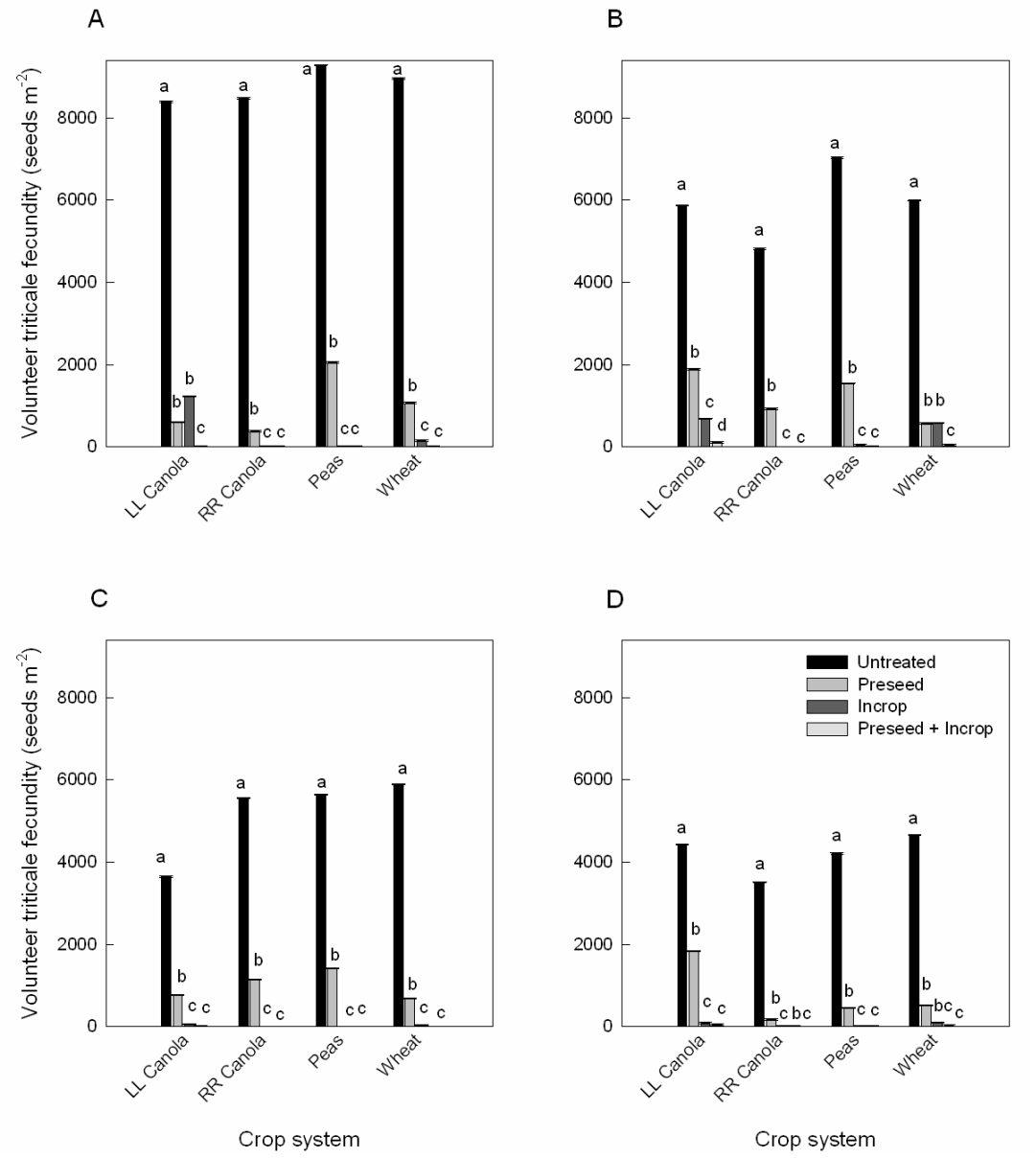

Figure 4.3. Effect of preseed, incrop, and preseed followed by incrop herbicide applications on volunteer triticale fecundity (seeds m^{-2}) relative to untreated controls for each of four cropping systems: glufosinate-tolerance canola (LL canola), glyphosate-tolerant canola (RR canola), field pea, and imidazolinone-tolerant wheat at A. Calmar 2006, B. Ellerslie 2006, C. Ellerslie 2007, and D. Edmonton 2007. Fecundity is expressed as back-transformed (the analysis was conducted on square-root transformed data) LSMeans ± standard error bars for each treatment. Treatment timings with the same letter are not significantly different for a given crop and environment (Bonferroni-adjusted p-values, where $p<0.0083$) within each crop system. Crop systems were not statistically compared.

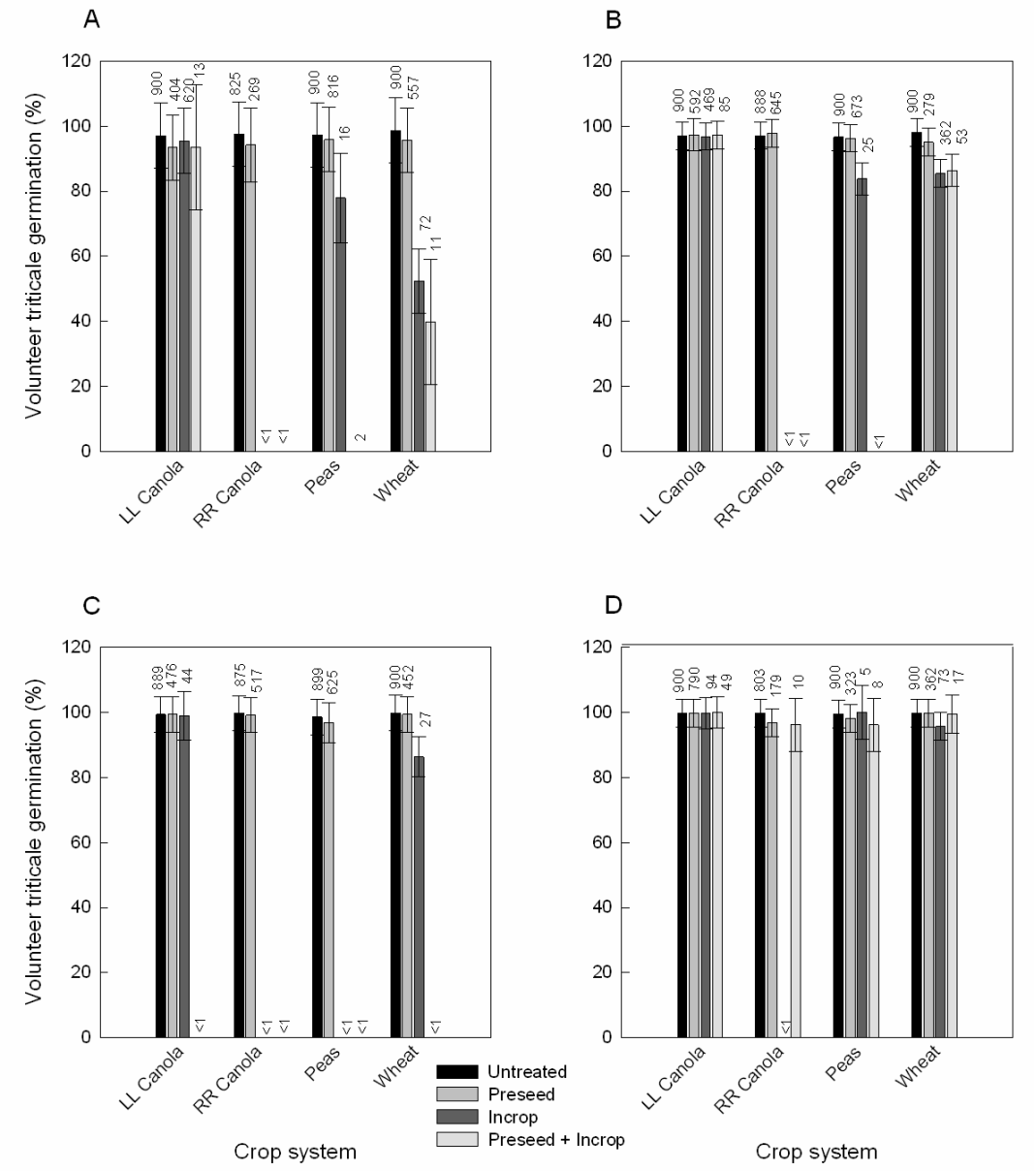

Figure 4.4. Effect of preseed, incrop, and preseed followed by incrop herbicide applications on volunteer triticale germination (%) relative to untreated controls for each of four cropping systems: glufosinate-tolerance canola (LL canola), glyphosate-tolerant canola (RR canola), field pea, and imidazolinone-tolerant wheat at A. Calmar 2006, B. Ellerslie 2006, C. Ellerslie 2007, and D. Edmonton 2007. Germinations are expressed as LSMeans ± standard error bars for each treatment. Numbers above treatment bars denote the average number of seeds tested per replicate. Treatment timings and crop systems were not statistically compared because many treatments or replicates had very few or no seeds for germination testing.

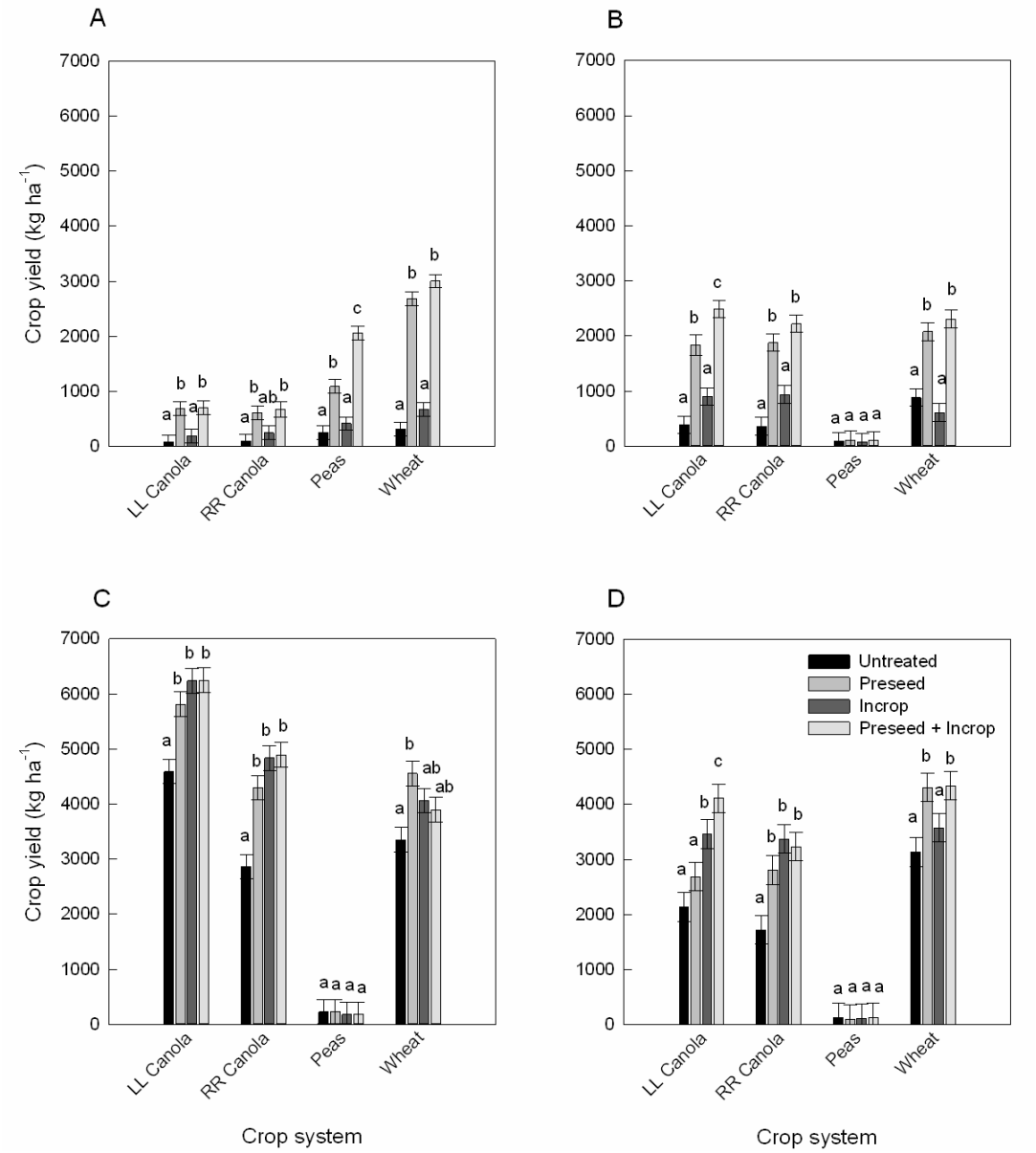

Figure 4.5. Effect of preseed, incrop, and preseed followed by incrop herbicide applications on crop yields (kg ha^{-1}) relative to untreated controls for each of four cropping systems: glufosinate-tolerant canola (LL canola), glyphosate-tolerant canola (RR canola), field pea, and imidazolinone-tolerant wheat at A. Calmar 2006, B. Ellerslie 2006, C. Ellerslie 2007, and D. Edmonton 2007. Yields are expressed as LSMeans ± standard error bars for each treatment. Treatment timings with the same letter are not significantly different, for a given crop and environment (Bonferroni-adjusted p-values, where $p<0.0083$) within each crop system. Crop systems were not statistically compared.

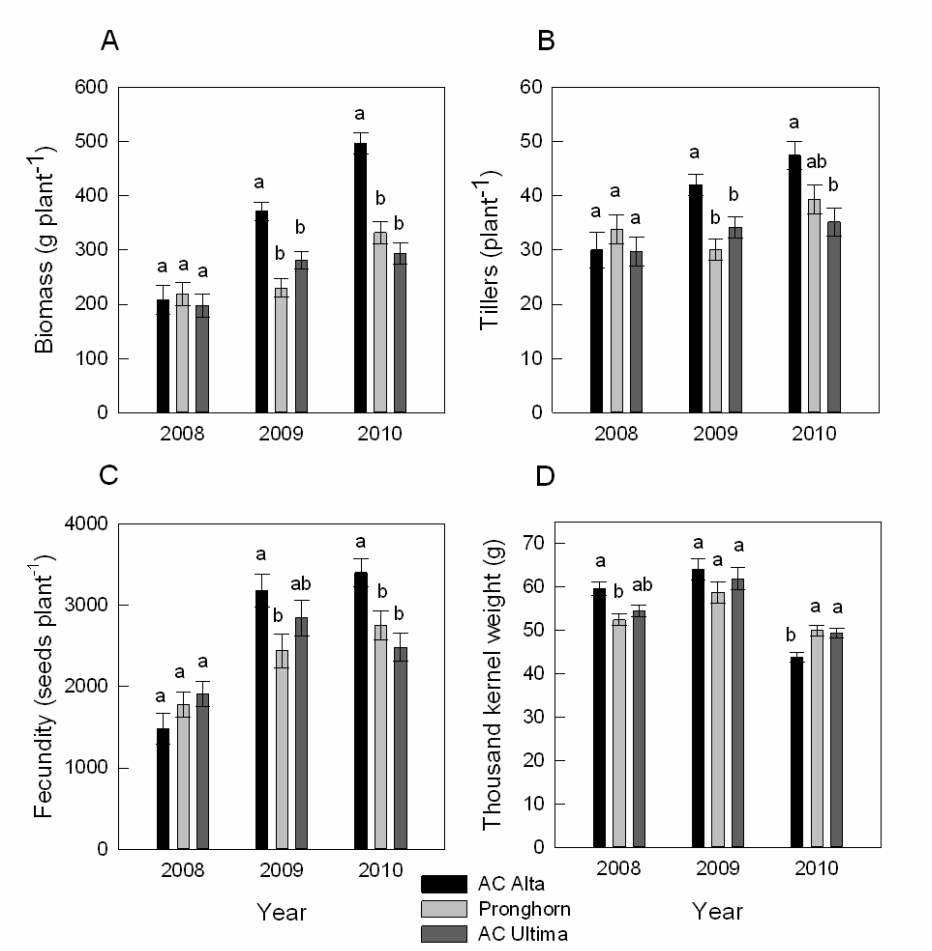

Figure 4.6. Triticale productivity per plant for 'AC Alta', 'Pronghorn' and 'AC Ultima' in 2008, 2009 and 2010 grown in the absence of competition and measured as: A. dry weight biomass (g), B. number of tillers, C. fecundity (seeds plant^{-1}), and D. calculated thousand kernel weights (TKW) (g). Cultivars with the same letter are not significantly different, for a given year.

4.5. Literature Cited

Alberta Agriculture, Food and Rural Development. 2005. Triticale production and utilization manual 2005: spring and winter triticale for grain, forage and value-added. Alberta Agriculture, Food and Rural Development, Edmonton, Alberta, Canada.

Anderson, R.L. and G. Soper. 2003. Review of volunteer wheat (*Triticum aestivum*) seedling emergence and seed longevity in soil. Weed Tech. 17:620-626.

Beckie, H.J., L.M. Hall, M.J. Simard, J.Y. Leeson and C.J. and Willenborg. 2010. A framework for post-release environmental monitoring of second-generation crops with novel traits. Crop Sci. 50:1587-1604.

Beres, B.L., R.E. Blackshaw, R.J. Graf, E. Bremer, K.N. Harker and G.W. Clayton. 2010. Weed-competitive ability of spring and winter cereals in the northern Great Plains. Weed Tech. 24:108–116.

Blackshaw, R.E., L.J. Molnar and H.H. Janzen. 2004. Nitrogen fertilizer timing and application method affects weed growth and competition with spring wheat. Weed Sci. 52:614-622.

Blackshaw, R.E., K.N. Harker, G.W. Clayton and J.T. O'Donovan. 2006. Broadleaf herbicide effects on clethodim and quizalofop-P efficacy on volunteer wheat (*Triticum aestivum*). Weed Tech. 20:221-226.

Blackshaw, R.E., K.N. Harker, J.T. O'Donovan, H.J. Beckie and E.G. Smith. 2008. Ongoing development of integrated weed management systems on the Canadian prairies. Weed Sci. 56:146-150.

Bushong, J., T. Peeper, M. Boyles and A. Stone. 2011. Italian ryegrass (*Lolium perenne*), feral cereal rye (*Secale cereale*), and volunteer wheat (T*riticum aestivum*) control in winter canola. Weed Tech. 25:344-349.

[CFIA] Canadian Food Inspection Agency. 2004. Assessment criteria for determining environmental safety of plants with novel traits. Directive Dir94-08. Available at http://www.inspection.gc.ca/plants/plants-with-novel-traits/applicants/directive-94-08/eng/1304475469806/1304475550733 (verified Mar. 12, 2012). CFIA, Plant Bio-Safety Office. Ottawa, Ontario, Canada.

Canadian Triticale Biorefinery Initiative. 2010. Canadian triticale biorefinery initiative. Available at www.ctbi.ca (verified Nov 10, 2011). CTBI, Edmonton, Alberta, Canada.

Chambers, A., G. Scammell and G. Code. 1995. Annual ryegrass and volunteer cereal control in lupins using selective post-emergent herbicides. Aust. J. Exp. Agric. 35:1141-1149.

Clarke, J.M. 1985. Harvesting losses of spring wheat in windrower/combine and direct combine harvesting systems. Agron. J. 77:13-17.

De Corby, K.A., R.C. Van Acker, A. Brûlé-Babel and L.F. Friesen. 2007. Emergence timing and recruitment of volunteer spring wheat. Weed Sci. 55:60-69.

Demeke, T., D.J. Perry and W.R. Scowcroft. 2006. Adventitious presence of GMOs: scientific overview for Canadian grains. Can. J. Plant Sci. 86:1-23.

Dexter, J.E., A.J. Jhala, R.C. Yang, M.J. Hills and Weselake, R.J. and Hall, L.M. 2011. Harvest loss and seed bank longevity of flax (*Linum usitatissimum* L.): Implications for seed-mediated gene flow. Weed Sci. 59:61-67.

Dexter, J.E., A.J. Jhala, M.J. Hills, R.C. Yang, A.K. Topinka, R.J. Weselake and L.M. Hall. 2010. Emergence and persistence of volunteer flax (*Linum usitatissimum* L.) in western Canadian cropping systems. Agron. J. 102:1321-1328.

FAOSTAT. 2012. Food and agriculture organization statistical database. Available at http://faostat.fao.org/site/567/DesktopDefault.aspx?PageID=567#ancor (verified Mar. 12, 2012). Rome, Italy.

Goyal, A., B.L. Beres, H.S. Randhawa, A. Navabi, D.F. Salmon and F. Eudes. 2011. Yield stability analysis of broadly adaptive triticale germplasm in southern and central Alberta, Canada, for industrial end-use stability. Can. J. Plant Sci. 91:125-135.

Gruber, S., N. Colbach, A. Barbottin and C. Pekrun. 2008. Post-harvest gene escape and approaches for minimizing it. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources 3:17.

Harker, K.N., R.E. Blackshaw and G.W. Clayton. 2001. Timing weed removal in field pea (*Pisum sativum* L.). Weed Tech. 15:277-283.

Harker, K.H. 2001. Survey of yield losses due to weeds in central Alberta. Can. J. Plant Sci. 81:339-342.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y.T. Gan, H.A. Frederick, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2005. Glyphosate-resistant wheat persistence in western Canadian cropping systems. Weed Sci. 53:846-859.

Harker, K.N., J.T. O'Donovan, G.W. Clayton and J. Mayko. 2008. Field-scale time of weed removal in canola. Weed Tech. 22:747-749.

Harker, K.N., J.T. O'Donovan, R.E. Blackshaw, E.N. Johnson, F.A. Holm and G.W. Clayton. 2011. Environmental effects on the relative competitive ability of canola and small-grain cereals in a direct-seeded system. Weed Sci. 59:404-415.

James, C. 2010. Global Status of Commercialized Biotech/GM Crops: 2010. ISAAA Brief No. 42. ISAAA, Ithaca, New York, USA.

Kavanagh, V.B., J.C. Hall and L.M. Hall. 2010. Potential hybridization of genetically engineered triticale with wild and weedy relatives in Canada. Crop Sci. 50:1128-1140.

Kavanagh. V., M. Hills, F. Eudes, K. Topinka, R.-C. Yang, L. Hall. In Press. Pollen-mediated gene flow in triticale (× *Triticosecale* Wittmack). Crop Sci.

Kershen, D.L. and A. McHughen. 2005. Adventitious presence: Inadvertent commingling and coexistence among farming methods. CAST Commentary QTA 2005-1:1-4.

Leeson, J.Y., A.G. Thomas and L.M. Hall. 2002. Alberta weed survey of cereal, oilseed and pulse crops in 2001. Weed Survey Series 02-1, Agriculture and Agri-Food Canada, Saskatoon, Saskatchewan, Canada.

Leeson, J.Y., A.G. Thomas, L.M. Hall, C.A. Brenzil, T. Andrews, K.R. Brown and R.C. Van Acker. 2005. Prairie weed surveys of cereal, oilseed and pulse crops from the 1970s to the 2000s. Rep. Weed Survey Series Publication 05-1CD. Agriculture and Agri-Food Canada, Saskatoon Research Centre, Saskatoon, Saskatchewan, Canada.

Marginet, A.M. 2001. Effect of tillage system and eco-regional field location cluster on the emergence periodicity of wild oat and green foxtail. M.Sc. diss. Univ. of Manitoba, Winnipeg, Manitoba, Canada.

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

May, W., G. Lafond, E. Johnson, T. Hogg, A. Johnston, B. Nybo, N. Harker and G. Clayton. 2003. An assessment of the concept of early time of weed

removal in field pea using natural weed populations. Can. J. Plant Sci. 83:423-431.

McKenzie, R.H., R.F. Dunn, B.L. Beres, P.G. Pfiffner, E. Bremer and A.B. Middleton. 2007. Efficacy of high seeding rates to increase grain yield of winter wheat and winter triticale in southern Alberta. Can. J. Plant Sci. 87:503-507.

McPherson, M.A., R.C. Yang, A.G. Good, R.L. Nielson and L.M. Hall. 2009. Potential for seed-mediated gene flow in agroecosystems from transgenic safflower (*Carthamus tinctorius* L.) intended for plant molecular farming. Transgenic Res. 18:281-299.

O'Donovan, J.T., R.E. Blackshaw, K.N. Harker, G.W. Clayton and D.C. Maurice. 2005. Field evaluation of regression equations to estimate crop yield losses due to weeds. Can. J. Plant Sci. 85:955-962.

O'Donovan, J.T., K.N. Harker, G.W. Clayton, L.M. Hall, J. Cathcart, K.L. Sapsford, F.A. Holm and K. Hacault. 2007a. Volunteer barley interference in spring wheat grown in a zero-tillage system. Weed Sci. 55:70-74.

O'Donovan, J.T., R.E. Blackshaw, K.N. Harker, G.W. Clayton, J.R. Moyer, L.M. Dosdall, D.C. Maurice and T.K. Turkington. 2007b. Integrated approaches to managing weeds in spring-sown crops in western Canada. Crop Protect. 26:390-398.

Parker, I.M. and P. Kareiva. 1996. Assessing the risks of invasion for genetically engineered plants: Acceptable evidence and reasonable doubt. Biol. Conserv. 78:193-203.

Rainbolt, C.R., D.C. Thill and F.L. Young. 2004. Control of volunteer herbicide-resistant wheat and canola. Weed Tech. 18:711-718.

SAS Institute Inc. 2007. SAS/STAT User's Guide: Statistics. SAS Institute, Cary, North Carolina, USA.

Sikkema, P., Deen W. and S. Vyas. 2005. Weed control in pea with reduced rates of imazethapyr applied pre emergence and post emergence. Weed Tech. 19:14-18.

Upadhyay, B.M., E.G. Smith, G.W. Clayton, K.N. Harker and R.E. Blackshaw. 2006. Economics of integrated weed management in herbicide-resistant canola. Weed Sci. 54:138-147.

Willenborg, C.J. and R.C. Van Acker. 2008. The biology and ecology of hexaploid wheat (*Triticum aestivum* L.) and its implications for trait confinement. Can. J. Plant Sci. 88:997-1013.

Wilson, W.W., E.A. DeVuyst, R.D. Taylor, W.W. Koo and B.L. Dahl. 2008. Implications of biotech traits with segregation costs and market segments: the case of Roundup Ready(R) Wheat. Eur. Rev. Agric. Econ. 35:51-73.

Wolt, J.D. 2009. Advancing environmental risk assessment for transgenic biofeedstock crops. Available at www.biotechnologyforbiofuels.com/content/2/1/27 Biotech. for Biofuels 2:27.

Chapter 5. Conclusions

5.1. Background

Transgenes have the potential to move within the environment via pollen-mediated gene flow because genetically modified (GM) crops may cross with crops of the same or closely related species or with wild or weedy relatives. For crops that are primarily self-pollinated or for small seeded species that produce large amounts of seed, gene flow is more likely to occur through seed movement (Beckie and Hall, 2008). The European Union has a labeling threshold of 0.9% for approved GM traits within non-GM products, although there are no thresholds specific to GM seeds (European Union, 2003). In order to maintain trade and export markets for conventional cereals, Canada must meet these standards. Minimizing seed returns to the seed bank the year GM crops are grown and controlling volunteers within the following crop are paramount for minimizing adventitious presence (AP) of GM seed. The research from this thesis contributes to baseline data on triticale biology and provides best management practices following the production of GM triticale.

5.2. Triticale seed persistence

5.2.1. General conclusions

Research conducted for this thesis determined that in central and southern Alberta, spring triticale seeds rapidly exit the seed bank and are no more likely than wheat to persist in the seed bank. The spring triticale cultivars used in this

research exited the seed bank more rapidly than AC Barrie wheat when seeds were placed on the soil surface or those buried shallowly. Triticale seeds are not likely to persist, partly because they do not exhibit prolonged primary dormancy and there is no evidence to suggest that deep seed burial induces secondary dormancy. It can be speculated that triticale may exit the seed bank more rapidly than wheat because triticale seeds are relatively large and elliptical-shaped and therefore may be more prone to mechanical abrasion during the harvest process than wheat. Broken, chipped or abraded seeds may be more susceptible to disease and decay. In the first growing season, the majority of buried seeds exited the seed bank through germination, while those on the soil surface tended to remain quiescent.

Rapid seed bank depletion is desirable for GM crops. Triticale seeds that were buried exited the seed bank rapidly within one year when there was seed-to-soil contact and adequate moisture and temperature for stimulating germination. However, tillage is not recommended as a means of hastening seed bank depletion because maintaining soil structure and standing stubble have been shown to conserve soil moisture (Arshad et al., 1999) and reduce soil erosion. Seeds on the soil surface will remain quiescent when conditions are dry, but will be more exposed to seed predators relative to seeds that are buried. Seeds on the soil surface may eventually become buried through natural means such as rainfall- or wind-moved soil, seed movement into cracks in the soil, or fertilizer is applied or the following crop is seeded.

5.2.2. Summary of results

Crop seed losses at or prior to harvest replenish the seed bank and seeds may remain viable and ungerminated over a period of time or exhibit dormancy which would contribute to seed persistence. Secondary dormancy has been shown to be induced for buried volunteer canola seeds (Gulden et al., 2004). The persistence of spring triticale can be compared to the persistence of its progenitor, wheat (reviewed in Chapter 3). Results from this research indicate:

- Seeds of spring triticale cultivars that remain on the soil surface, buried shallowly, or buried deeply do not persist longer than wheat within the seed bank (Figure 3.5).
- Seeds of spring triticale cultivars on the soil surface may be exposed to arid conditions. Surface-placed triticale seeds in this study persisted for as long as 19 months whereas for those that were shallowly buried, 99% were removed from the seed bank within 8 months. Deep seed burial results in fatal germination and 99% were removed from the seed bank within 6 months (Table 3.5).
- Blue Aleurone and 'AC Alta', one of the parental cultivars for the Blue Aleurone line, persisted for similar amounts of time when on the soil surface or buried deeply (Table 3.5). Blue Aleurone and 'AC Alta' behave similar to each other at the soil surface and when buried deeply, although 'AC Alta' exited the seed bank more rapidly than Blue Aleurone when buried to 2 cm.
- Spring triticale cultivars tested do not exhibit prolonged primary dormancy relative to AC Barrie wheat in central Alberta; however, primary dormancy was generally more pronounced under warm and dry conditions than

under cool and moist conditions at harvest (Table 3.4 and Figure 3.4). ‘AC Ultima’ showed increased primary dormancy prior to harvest relative to other spring triticale cultivars when conditions were conductive to pre-harvest sprouting.

- Spring triticale cultivars do not appear to exhibit induced secondary dormancy as a result of deep seed burial (Table 3.5 and Figure 3.5).

5.2.3. Considerations

Conclusions drawn from artificial seed banks must be made with caution. Seeds placed densely within mesh bags may provide an environment conducive to the spread of pathogenic fungi and may have caused more disease than would occur under natural conditions. Artificial seed banks do not model the fate of seeds within intact heads which may be lost to the seed bank as a result of crop lodging or animal herbivory. Morphological structures such as palea, lemma, and awns may protect or prevent seeds from germinating and may prolong persistence compared to threshed seeds. Wheat seed heads have been shown to persist for a longer period of time than threshed seeds (Seerey et al., 2011), but the persistence of triticale heads has not been investigated. Seed persistence may have been under-estimated in this research as a result of using mesh bags and threshed seeds.

The role of avian, mammalian, and insect seed predators is not well understood and may be underestimated, particularly for cereal crops. In this research, mammalian seed predation (via mice) was observed, although not quantified. Seed predators likely play a role in dispersing, caching, and removing

potential volunteer crop seeds from the seed bank and further study is required to elucidate their effects.

In this research, only spring triticale and wheat cultivars were studied. However, fall triticale seeds in the seed bank may behave differently than spring triticale because of their obligate requirement for a period of vernalization in order to produce reproductive structures. For example, feral rye, a winter annual, has been shown to persist in the seed bank for up to 5 years, and exhibits some secondary dormancy (Stump and Westra, 2000). The persistence of fall triticale should be investigated.

In years or environments that are drier, it is expected that seeds on the soil surface and those buried shallowly would persist for a longer period of time. Seeds would remain quiescent until moisture conditions were conducive for germination. Environments with periods of aridity may not allow for sufficient moisture or time for seeds to completely imbibe water; in these instances, seeds would remain ungerminated. However, excess moisture would likely cause seeds to enter a hypoxic or anoxic state and accelerate degradation. When conditions or environments are moist to wet, seeds would be expected to germinate readily when temperatures are conducive and seeds would exit the seed bank more rapidly than in drier environments.

The intention for GM triticale cultivars and the specific traits required is still unclear, therefore careful selection of lines may contribute to best management practices. Currently, breeders select for some pre-harvest sprouting tolerance or primary dormancy in wheat and triticale; however, prolonged

primary dormancy is undesirable and would impose limitations on future uses of the crop seed. Therefore, it would be expected that some lines would have more primary dormancy than others, but lines with longer lived primary dormancy would not likely be selected for crop production. Selection of lines or cultivars with requirements for longer or shorter seasons will determine the environments most suited for production. Central Alberta typically has shorter, wetter growing seasons than those of southern Alberta and cultivars with requirements for long growing seasons may be unable to reach maturity if seed production and quality are required. However, if biomass production is the goal, moisture may limit the production environment more than season length. The length of the growing season may influence seed persistence only when the season is too short for cultivars to reach maturity. Immature seeds are difficult to combine harvest and seeds could be subject to detrimental early frosts that may halt seed development, reducing quality and yield. In these instances, seeds may have reduced viability.

5.3. Volunteer triticale control and fecundity

5.3.1. General conclusions

Volunteer cereals typically emerge early in spring and can be controlled with pre-seeding or pre-emergent herbicide applications (Blackshaw et al., 2006; De Corby et al., 2007; Harker et al., 2005; Rainbolt et al., 2004). The results from this research indicate that volunteer triticale may escape or survive control measures or may emerge following application. Additionally, volunteer triticale at an advanced stage when in-crop applications are made may recover

and produce seed; this has also been shown to occur with volunteer wheat (De Corby et al., 2007). In the event of GM triticale production, the application of both pre-seed and in-crop herbicides would be most effective for prevention of survivors and reduction of seed returns to the seed bank. Glyphosate tolerant canola or field pea appeared to be the best crops to follow triticale for minimizing volunteer seed returns. Volunteer seed production was higher in LL canola because glufosinate provided less effective control of triticale. Conventional wheat is not recommended following triticale because the seeds are similar in appearance and size so AP is difficult to identify and no selective herbicides exist for removing volunteer cereals; thus AP may have been under-estimated within the wheat crop in this study. However, development of a visual marker, for example triticale with a blue aleurone like that of the Blue Aleurone research line, is visually identifiable from wheat or conventional triticale. A visual marker could assist in the identification of GM triticale and could assist with facilitating coexistence with conventional cereals, particularly if the identification of AP can be performed with a mechanized or automated test.

Minimizing seed inputs from volunteer GM crops is vital in following crops in order to limit AP and not exceed the EU threshold of 0.9%. Triticale seeds are generally retained prior to harvest, but could be lost during the combining process through sieves or air used to clean the seed as it is threshed. In this study, following triticale with glyphosate tolerant canola or field pea and applying both a pre-seed and in-crop herbicide was most effective at

minimizing seed production. However, should winter triticale be transformed, further research will be required to determine the most effective cropping rotations and herbicides, as well as the effects of overwintering seeds that are returned to the seed bank. Volunteer control has been assessed for wheat (De Corby et al., 2007; Anderson and Soper, 2003; Blackshaw et al., 2006; Beckie and Owen, 2007; Rainbolt et al., 2004; Lyon et al., 2002; Nielson, 2007; Harker et al., 2005); the research within this thesis contributes to the control assessment of volunteer spring triticale on the Canadian prairies.

5.3.2. Summary of results

Cereal crop seeds lost at harvest succumb to disease, mortality, or predation, but most germinate readily and form volunteer populations in following crops (Anderson and Soper, 2003; Brust and House, 1988). Best management practices for the control and fecundity of volunteer spring triticale within four rotational crops was reviewed in Chapter 4. Results from this research indicate:

- In central Alberta, control of volunteer triticale with pre-seeding herbicides (glyphosate or glufosinate) provides inconsistent and variable control (Figure 4.2). Volunteer triticale densities were reduced by 30 to 80% with pre-seeding herbicides alone, resulting in triticale fecundity from 179 to 2050 seed m^{-2} (Figure 4.3). When conditions are conducive to later emergence, control is less effective.
- The use of in-crop herbicides in LL canola, field pea, and imidazolinone tolerant wheat reduced volunteer triticale densities by 8 to >99% (Figure 4.2),

where triticale fecundity ranged from 1 to 1230 seeds m^{-2} (Figure 4.3). Inclement weather can delay herbicide application so that volunteer triticale is at an advanced stage resulting in plants that survive and recover from herbicide symptoms with overall reduced efficacy. However, in-crop glyphosate in RR canola provided consistent control and reduced densities by >99% with seed returns of no more than 2 seeds m^{-2} despite advanced stages of volunteer triticale.

- Application of pre-seed and in-crop herbicides provides the most consistent control of volunteer triticale in following crops, reducing densities by 72 to >99% and fecundity ranged from 0 to 109 seeds m^{-2}.
- When both pre-seed and in-crop herbicide applications are made, volunteer triticale fecundity is least for RR canola and field pea followed by imidazolinone tolerant wheat and LL canola.
- Adventitious presence (AP) of volunteer triticale within the harvested portion of all following crops was reduced by over 97% when both a pre-seed and in-crop herbicide application were made, although AP was reduced by 89 to >99% when only an in-crop application was made.
- While few seeds were produced by triticale survivors following pre-seed and in-crop herbicides, only the imidazolinone herbicides used on field pea and imidazolinone tolerant wheat caused injury to seeds and reduced germination in three of four environments (Figure 4.4). Seeds produced by survivors in RR and LL canola had high levels of germination.

- Triticale is a competitive plant capable of producing 30 to 47 tillers per plant and yielding 1478 to 3400 seeds per plant in the absence of competition. However, within following crops in this study, volunteer triticale produced only between 76 and 142 seeds per plant when left untreated.

5.3.3. Considerations

The results from this experiment highlight how herbicide timings can influence volunteer triticale survival and resulting fecundity. Volunteers that emerge following or survive pre-seeding applications will contribute to adventitious presence. However, leaving volunteers uncontrolled until in-crop herbicides are applied will result in larger, more mature plants which may be more difficult to control. In the event of poor weather conditions that delay the in-crop herbicide application (e.g. prolonged rainy period), the volunteer plants may be too mature for effective control and plants may survive and produce seed. When only an in-crop herbicide was applied within all following crops, the crop yields were reduced because volunteers remained for a greater time to compete directly with the crop. As much as possible, other weeds within these experiments were removed so that only the effects of volunteer triticale were being evaluated. On the Canadian prairies, a number of broadleaved (e.g. wild buckwheat) and grass weeds (e.g. wild oat) would also be present that would require herbicides in order to maximize yields and may require appropriate registered tank mixes. All herbicides used in this experiment effectively control of a number of grass and broadleaved weeds to provide a more realistic scenario.

Most of the pre-seeding applications in this experiment involved the use glyphosate by itself, however because of recent concerns around selecting glyphosate resistant weed populations, appropriate pre-seeding tank mixes such as CleanStart® (glyphosate + carfentrazone) or PrePass® (glyphosate + florasulam) have more than one mode of action to reduce the selection pressure. In the event of herbicide resistant GM spring triticale, the selection of following crops and appropriate tank mixes will become more critical. Herbicide tolerance can sometimes be used as a linked genetic marker for inserted traits of interest to help identify future volunteers or plants that are the result of out-crossing. Herbicides other than those used as the marker would need to be applied to avoid volunteer triticale survival. Additionally, the insertion of traits that may provide abiotic stress tolerance, for example: waxier leaves to increase drought tolerance, may reduce the efficacy of herbicides or have unpredictable effects on volunteer control and fecundity and would require re-evaluation in various environments.

5.4. Management recommendations

Tillage is not recommended for hastening seed bank depletion of volunteer triticale seeds. While deep seed burial exhibited the most rapid reduction in seed viability, it is accomplished by using implements such as a mold board plow. Deep plowing is a management tool that is rarely recommended or used because it is destructive to the soil structure and can increase the rate of soil drying and soil erosion. Shallow tillage is still practiced to stimulate weed seed germination, to

remove early emerged seedlings, and prepare a seed bed. However, on the Canadian prairies, minimum- or zero-tillage practices have been widely adopted to conserve soil moisture and reduce soil erosion. Seeds that fall to the soil surface will either remain there for a period of time before eventually being covered by soil and surface chaff. Shallow burial will promote germination once soil moisture is adequate and these plants will then form a volunteer population that may be controlled with the use of herbicides or by winter kill if they germinate in fall on the Canadian prairies. The act of seeding a following crop will also act to bury seeds and stimulate them to germinate. Spring triticale seedlings are the recommended stage for applying control measures.

Pre-seeding herbicides will control broadleaf and grass weeds, including volunteer spring triticale, that emerge early in spring while the in-crop application will control later emerging volunteers and those that are stimulated to germinate by the process of seeding the next crop. Spring triticale is best followed by a broadleaf crop such as canola or field pea. A number of herbicides can be used to selectively remove grass weeds from within broadleaf crops, in addition to using herbicide tolerant crops such as the Roundup Ready® glyphosate tolerant canola or Liberty Link® glufosinate tolerant canola systems that were used in this study. Following triticale with a cereal crop is not recommended because there are no selective herbicides to remove volunteer triticale from wheat, barley, oat, or rye in-crop with the exception of Clearfield® imidazolinone-tolerant wheat that was used in this study. Additionally, using label or recommended herbicide rates are the best option to minimize survival of GM volunteer triticale. Choosing to use

reduced rates may be economical and efficacious under some conditions, however, higher volunteer densities, poor environmental conditions at the time of application, and advanced weed stages may result in poorer efficacy. Because minimizing AP in following crops is crucial following a GM triticale seed crop, it is recommended to rotate to competitive following crops that use different herbicide modes of action, use appropriate pre-seed and in-crop herbicides and label recommended herbicide rates within the following crop.

5.5. Recommendations for future research

Future research should include assessing the role of seed mammalian, avian, and/or insect seed predators on depletion of crop seed banks. Within agricultural environments, the majority of seed bank depletion research has involved the study of weed species. However, within this thesis research, we observed (although were unable to quantify) seed predation of surface placed and shallowly buried triticale and wheat cultivars. Ideally, crop seed predation should be assessed from within a field setting where seeds have been spread uniformly and are not patchy or limiting so that seeds within the research experiment are not simply 'baiting' predators. Seed predation may also take place prior to seed release by small birds. It is expected that on the Canadian prairies, migratory birds such as Canada geese or snow geese, as well as mice and other small mammals may use crop seed in recently harvested fields as a seasonal food source. Factors such as the proximity to water, time of harvest, surrounding vegetation, crop density, the patchiness/distribution of harvest losses, among other factors would

likely influence the rate and extent of seed depletion. Crop seed predation also has implications for GM crops because predators also play a role in seed-mediated gene flow by moving and caching seeds and also requires assessment. Similarly, the role of endemic soil pathogens on crop seed bank depletion should be assessed.

Intact seed heads may enter the seed bank as a result of hail, crop lodging, mammalian or insect herbivory, or when combine settings inadequately thresh seeds from heads. Seerey et al. (2011) showed that intact wheat heads persisted for a significantly longer period of time than threshed wheat seeds. The persistence of whole spring triticale seed heads should be evaluated and compared with those of threshed seeds. Persistence of whole seed heads should also be assessed for transformed spring or winter triticale and compared to non-transformed lines. Morphological structures around seeds have been speculated to have inhibitory effects on seed germination and this should be evaluated.

Winter triticale is typically seeded in fall on the Canadian prairies and overwinters when the plants have entered the tillering stage. Winter types have an obligate requirement for vernalization in order to enter the reproductive stage. In the event that winter types become the target of genetic modification for bio-product development, the persistence, control and fecundity will need to evaluated using cultivars common to the Canadian prairies and compared with the data produced in this research.

Upon genetically transforming spring or winter triticale lines, the persistence and control and fecundity of the transformed lines needs to be

evaluated and compared with untransformed lines. Depending upon what traits are introduced, the persistence may be altered particularly if there is introduction of traits that directly alter the seeds. A change to lignin content within the stems, for example, may alter efficacy of some herbicides. However, future research using conventional volunteer spring triticale is not required at this time because the most common cropping rotations were evaluated.

5.6. In summary

Developing triticale as a platform crop for plant-based bio-energy or bio-industrial uses on the Canadian prairies may require genetic modification to enhance environmental tolerances or energy conversion (Goyal et al., 2011; Eudes, 2006; Canadian Triticale Biorefinery Initiative, 2011). The Canadian Food Inspection Agency (CFIA) compiles biological information for the purposes of regulating a number of potential GM crops and to assess their behaviour in the environment relative to their untransformed comparators ([CFIA] Canadian Food Inspection Agency, 2011). CFIA collects information about the propensity for a transformed crop to become weedy or invasive and baseline data can be assessed prior to genetic transformation. Seed persistence has been assessed for conventional wheat (Nielson et al., 2009; De Corby et al., 2007; Anderson and Soper, 2003), glyphosate tolerant GM wheat (Lyon et al., 2002; Nielson, 2007); Harker et al., 2005), and feral rye (Stump and Westra, 2000). This research will contribute to the compilation of a biology document for spring triticale. The research from this thesis contributes baseline biological information about triticale

seed persistence and fecundity of triticale that survives management practices. Additionally, this research makes recommendations for volunteer triticale control in order to minimize seed production in following crops. This baseline data was conducted prior to genetic modification; however, the findings have implications for continuing the development of GM triticale lines. In the event of that GM triticale lines are developed, further research would be required comparing seed persistence and volunteer control of transformed lines with conventional lines such as those used in this research. Additionally, in the event of GM triticale development, researchers in cooperation with CFIA and industrial stakeholders must develop stewardship or co-existence plans to minimize AP and protect conventional cereal markets and the interests of Canadian farmers.

5.7. Literature Cited

Anderson, R.L. and G. Soper. 2003. Review of volunteer wheat (*Triticum aestivum*) seedling emergence and seed longevity in soil. Weed Tech. 17:620-626.

Arshad, M.A., A.J. Franzleubbers and R.H. Azooz. 1999. Components of surface soil structure under conventional and no-tillage in northwestern Canada. Soil Tillage Res. 53:41-47.

Beckie, H.J. and L.M. Hall. 2008. Simple to complex: Modelling crop pollen-mediated gene flow. Plant Sci. 175:615-628.

Beckie, H.J. and M.D.K. Owen. 2007. Herbicide-resistant crops as weeds in North America. CAB Reviews: Perspectives in agriculture, veterinary science, nutrition and natural resources 044:1-22.

Blackshaw, R.E., K.N. Harker, G.W. Clayton and J.T. O'Donovan. 2006. Broadleaf herbicide effects on clethodim and quizalofop-P efficacy on volunteer wheat (*Triticum aestivum*). Weed Tech. 20:221-226.

Brust, G.E. and G.J. House. 1988. Weed seed destruction by arthropods and rodents in low-input soybean agroecosystems. Am. J. Alternative Agric. 3:19-25.

Canadian Triticale Biorefinery Initiative. 2011. The Canadian Triticale Biorefinery Initiative. Available at http://www.ctbi.ca (verified Mar. 12, 2012). CTBI, Edmonton, Alberta, Canada.

[CFIA] Canadian Food Inspection Agency. 2011. Companion documents for Directive 94-08, assessment criteria for determining environmental safety of plants with novel traits. Available at http://www.inspection.gc.ca/plants/plants-with-novel-traits/applicants/directive-94-08/eng/1304475469806/1304475550733 (verified November 15, 2011). Plant Biosafety Office, Government of Canada, Ottawa, Ontario, Canada.

De Corby, K.A., R.C. Van Acker, A. Brûlé-Babel and L.F. Friesen. 2007. Emergence timing and recruitment of volunteer spring wheat. Weed Sci. 55:60-69.

Eudes, F. 2006. Canadian triticale biorefinery initiative. p. 85-88. *In* W.C. Botes, D. Boros, N. Darvey, P. Gustafson, R. Jessop, G.F. Marais, G. Oettler and D. Salmon (eds.) Proc. 6th International Triticale Symp., Stellenbosch,

South Africa. 3-7 September 2006 Faculty of AgriSciences, Stellenbosch, South Africa.

European Union. 2003. Regulation (EC) No 1830/2003 of the European parliament and of the council. Available at http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2003:268:0024:0028:EN:PDF (verified Mar. 12, 2012). Official Journal of the European Union 1830/2003:L 268/24-L 268/28. Brussels, Belgium.

Goyal, A., B.L. Beres, H.S. Randhawa, A. Navabi, D.F. Salmon and F. Eudes. 2011. Yield stability analysis of broadly adaptive triticale germplasm in southern and central Alberta, Canada, for industrial end-use stability. Can. J. Plant Sci. 91:125-135.

Gulden, R.H., A.G. Thomas and S.J. Shirtliffe. 2004. Secondary dormancy, temperature, and burial depth regulate seedbank dynamics in canola. Weed Sci. 52:382-388.

Harker, K.N., G.W. Clayton, R.E. Blackshaw, J.T. O'Donovan, E.N. Johnson, Y.T. Gan, H.A. Frederick, K.L. Sapsford, R.B. Irvine and R.C. Van Acker. 2005. Glyphosate-resistant wheat persistence in western Canadian cropping systems. Weed Sci. 53:846-859.

Lyon, D.J., A.J. Bussan, J.O. Evans, C.A. Mallory-Smith and T.F. Peeper. 2002. Pest management implications of glyphosate-resistant wheat (*Triticum aestivum*) in the western United States. Weed Tech. 16:680-690.

Nielson, R.L., R. Yang, L.M. Hall, K.N. Harker, M.A. McPherson and J.T. O'Donovan. 2009. Seed-mediated gene flow in wheat: Seed bank longevity in western Canada. Weed Sci. 57:124-132.

Nielson, R. 2007. Volunteer wheat biological parameters to support the development of a mechanistic model. M.Sc. diss. Univ. of Alberta, Edmonton, Alberta, Canada.

Rainbolt, C.R., D.C. Thill and F.L. Young. 2004. Control of volunteer herbicide-resistant wheat and canola. Weed Tech. 18:711-718.

Seerey, N., S.J. Shirtliffe and P. Hucl. 2011. Seeds from unthreshed wheat (*Triticum aestivum* L.) spikes have reduced field emergence compared with threshed seed regardless of cultivar. Can. J. Plant Sci. 91:583-585.

Stump, W.L. and P. Westra. 2000. The seedbank dynamics of feral rye (*Secale cereale*). Weed Tech. 14:7-14.