Evaluating disease reaction of western Canadian spring wheat cultivars (*Triticum* spp.) to natural and artificial infection with *Claviceps purpurea* (Fr.) Tul.

A Thesis Submitted to

The College of Graduate Studies and Research

In Partial Fulfillment of the Requirements

For the Degree of Master of Science

In the Department of Plant Sciences

University of Saskatchewan

Saskatoon, Saskatchewan

By

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ABSTRACT

Ergot, caused by the fungal pathogen *Claviceps purpurea* (Fr.) Tul., attacks the floral organs of many grassy species resulting in sclerotia production rather than grain. Infection causes reduced yields, downgrading, and poisoning if consumed by humans or animals. Few recent studies have been conducted on ergot in wheat (*Triticum* spp.), and prevention is the only means of control. The objectives of this study were to determine if western Canadian spring wheat differed in reaction to infection with *C. purpurea* and if levels of inoculum would affect disease intensity in a field setting. Three variables were measured for the field experiments to determine disease reaction, including percent sclerotia by weight, number of sclerotia per spike, and weight per sclerotium. In the first experiment, nine wheat cultivars were tested using three inoculum levels. No significant differences were detected among inoculum levels. In the second and third experiments, ninety-two cultivars were studied in field and controlled conditions. Honeydew production, sclerotial size, and the percent of florets aborted were added as variables in the growth chamber experiment. Pearson correlations were calculated using cultivar means for the field and controlled environments. Results indicate that there are differences in disease reaction among cultivars and market classes, but these differences varied depending on the evaluation method used. In the field, CWAD wheat had the smallest sclerotia, but had more per spike compared to the CWRS and CWES market classes. There were no significant differences among these market classes for percent sclerotia by weight. In the growth chamber, CWAD wheat generally had the lowest ergot infection levels. When comparing the market classes within *T. aestivum* (CWRS, CPS, and CWES), there were no significant differences except for honeydew production. The correlation between environments was not significant for any of the variables, suggesting alternate resistance mechanism expression. In the field, reduced infection may be due to an escape mechanism, while artificial inoculation in a controlled environment may detect a physiological resistance mechanism. However, a group of cultivars with Grandin parentage showed promising results in both environments, and might confer resistance that could be integrated into disease resistance breeding programs.

ACKNOWLEDGEMENTS

I would like to thank my supervisor Dr. Pierre Hucl for his continued guidance, assistance, and valuable knowledge during the course of this project, as well as my advisory committee members Dr. Sabine Banniza, Dr. Randy Kutcher, and Dr. Bruce Coulman for their help in my thesis. I would also like to acknowledge the technical assistance of Mike Grieman, Glen Trowell and the bread wheat field research group; Tim Dament and the flax/cereal pathology group; and Dr. Jim Menzies (CRC-AAFC). I appreciate the financial support provided by the Robert P. Knowles Scholarship (College of Graduate Studies and Research) and the Syngenta Sustainable Agriculture Scholarship (College of Agriculture and Bioresources), as well as the research funding provided by NSERC and The Saskatchewan Ministry of Agriculture through the Agriculture Development Fund (ADF). I would also like to include a special thanks to my family for their support throughout my schooling, and for all of their love and encouragement that has gotten me to where I am today.

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LIST OF ABBREVIATIONS

C: Control(s) (Sandro triticale and Gazelle rye)

CPSR: Canada Prairie Spring Red

CPSW: Canada Prairie Spring White

CWAD: Canadian Western Amber Durum

CWES: Canada Western Extra Strong

CWGP: Canada Western General Purpose

CWHWS: Canada Western Hard White Spring

CWRS: Canadian Western Red Spring

CWSWS: Canada Western Soft White Spring

HD: Honeydew production

O: Overall (all cultivars of wheat and controls)

OC: Outcrossing

PDA: Potato dextrose agar

Percent (%) sclerotia (scl): Percent sclerotia by weight

Percent (%) aborted (ab): Percent of florets aborted of the total inoculated

PSB: Potato sucrose broth

Scl: Sclerotia

Sclerotia / Spike: Average number of sclerotia per spike

SE: Standard Error

T.a.: *Triticum aestivum*

T.t.d.: *Triticum turgidum* ssp. *durum*

T.s.: *Triticum spelta*

W: Wheat cultivars

Weight (Wt) / Sclerotium: Average sclerotium weight (mg)

1.0 INTRODUCTION

Canada is one of the world's leading producers of wheat (*Triticum* spp.), with an average of over twenty-five million tonnes produced annually in the last twenty years (FAO, 2013). Half of the wheat produced annually in Canada is exported to other countries as an important food and feed commodity. Approximately twenty percent of the total calories and protein consumed by the world's population are from bread (*Triticum aestivum*) and durum (*Triticum turgidum* ssp. *durum*) wheat (Breiman & Graur, 1995; Gupta et al., 2008). Because of the great demand for Canadian wheat it is important to maintain quality and efficient production of this commodity. High yields in the past have been accomplished through breeding efforts that enhance agronomic characteristics of the wheat, including, but not limited to, increased harvest index, straw strength, kernel quality, and disease resistance.

Ergot is a disease that has the potential to become a large threat to wheat production. In Saskatchewan, Agriculture and Agri-Food Canada (2012) report ergot disease as having a "widespread yearly occurrence with low to moderate pest pressure OR widespread sporadic occurrence with moderate pressure OR sporadic localized occurrence with moderate pressure". In Alberta, Manitoba, Ontario, and Quebec, the disease has a "localized yearly occurrence with low to moderate pest pressure OR widespread sporadic occurrence with low pressure OR localized sporadic occurrence with low to moderate pest pressure".

Ergot is caused by the *Clavicipitaceae* fungus, *Claviceps purpurea* (Fr.) Tul. (White et al., 2003). This biotrophic fungal pathogen is able to infect a wide range of grassy plants (family *Poaceae*, formerly *Gramineae*), which includes many of the important cereal crops grown on the plains of North America (Bove, 1970). When *C. purpurea* infects its host, it attacks the ovary and produces a sclerotium (pl. sclerotia) rather than a kernel of grain (Knox & McLeod, 2003). When the grain is harvested, the sclerotia may enter into the food or feed chain. The sclerotia can cause poisoning if consumed by humans or animals due to the toxic alkaloids they contain (White et al., 2003). Economic losses may also arise due to ergot infections. The first economic loss

occurs through yield losses when the sclerotia replace the kernels and the second loss occurs due to downgrading at the elevator, which requires minimal amounts of sclerotia contamination. Only 0.01% sclerotia by weight of the total sample weight is acceptable for No.1 wheat, ranging to only 0.1% by weight for feed wheat (Canadian Grain Commission, 2011a).

There are a number of biotic and abiotic factors that influence the pathogen's opportunity to infect the host. Biotic factors include the host's floret morphology and flowering habit; abiotic factors include temperature, moisture, sources and amounts of inoculum, and micronutrient deficiencies. These factors affect the host's susceptibility to infection and the pathogen's ability to germinate and colonize the host.

Floret morphology and timing of flowering has been suggested to be one of the most important factors of disease occurrence, as this is the site of entry for the pathogen (Mantle et al., 1977; Knox and McLeod, 2003). When open pollination occurs, either naturally or because of environmental stresses, the period of susceptibility increases. This is why rye and triticale are most susceptible, followed by wheat, barley, and oat (Platford & Bernier, 1976).

Environmental conditions must be favourable for the ergot sclerotia to germinate and cause the initial infection. This was the case in Saskatchewan in 1999 and 2008, where major ergot outbreaks occurred (Saskatchewan Ministry of Agriculture, 2009). The final crop report released by the Saskatchewan Ministry of Agriculture (2011 and 2012) also reported losses and downgrading due to ergot infection in the 2011 and 2012 growing seasons. Cool and wet conditions lead to poor plant emergence and uneven flowering, and rain splash acted as a means of spreading the pathogen, thus increasing disease spread. Ergot epidemics leave sclerotia in the field to cause infection in succeeding years when conditions are favourable for germination. Environment Canada (2011) reported record levels of precipitation on the prairies in 2010 that had not occurred since 1948, with a departure of 30% from normal. These conditions were conducive for germination of the sclerotia that had been deposited in the previous year(s). The spring of 2012 also experienced some extreme weather, including cool temperatures and wet conditions that are favoured by the ergot fungus. In fact, 2012 was ranked as the third

wettest year for spring precipitation since 1948, with a departure of 52% from normal (Environment Canada, 2012).

There are many factors that contribute to the occurrence and severity of ergot infections, but currently there is little knowledge on specificity of resistance or controls for the disease in wheat. Currently there are no cultivars that are resistant to *C. purpurea* and there are very few management practices available for growers. Prevention and/or avoidance are the only means of control. However, in 2004 Menzies published an article stating that Canada Western Amber Durum (CWAD) and Canada Prairie Spring (CPS) wheat suffer fewer symptoms compared to Canada Western Extra Strong (CWES) and Canada Western Red Spring (CWRS) wheat. Fernandez et al. (2000) also reported low levels of infection in CPS wheat, but found both CWRS and CWAD to be among the most susceptible market classes.

Information on the genetic control of ergot resistance is lacking, but research is currently being conducted to map two populations of durum wheat, Avonlea/9260B-173A and Avonlea*2/9260B-173A, to eventually develop molecular markers to aid in future breeding efforts (Menzies et al., unpublished). In these lines, there are two additive genes that appear to confer resistance to honeydew production, but genetic control of sclerotia number and size has not been determined. However, one study did suggest that the spring and durum wheat cultivars Kenya Farmer and Carleton, respectively, carried resistance genes in their B genomes, which resulted in reduced honeydew production, as well as decreased sclerotial size and frequency (Platford et al., 1977). Kenya Farmer, which contained the genes on chromosome 6B, appeared more resistant compared to Carleton, which contained the genes on chromosome 1B and 3B. Both cultivars were considered to be moderately resistant.

The objective of this study was to determine if there is a method to measure ergot severity among western Canadian spring wheat cultivars. Currently, ergot disease screening is not required for variety registration, and therefore breeders do not make assessments for it on potentially new lines. Field protocols were established to evaluate differences among wheat cultivars. Disease reaction, or resistance traits, were determined by a number of variables that measured disease incidence and severity, which included honeydew production; sclerotial size, number, and weight; and kernel development in

relation to sclerotia formation. Western Canadian spring wheat cultivars were assessed for ergot disease reaction in both field and controlled (growth chamber) conditions. Based on the limited existing knowledge, it was hypothesized that (1) greater levels of inoculum would result in the production of more sclerotia as a percentage of the total harvested (grain + sclerotia) biomass, (2) there would be significant differences in disease reaction among wheat cultivars and among market classes, and (3) wheat cultivars would differ in their capacity to produce secondary inoculum. Results from the two environments were tested for correlations to determine if there was a genetic component conferring disease resistance. The results of this study may provide valuable information on lines to use for future disease resistance breeding efforts.

2.0 LITERATURE REVIEW

2.1 Canadian Wheat Production: A Look at the Host

The *Triticum* species (tribe *Triticeae*, family *Poaceae*) were first domesticated in approximately 7500-6500 BC (Feldman, 1976) in the Fertile Crescent, an area now composed of Turkey, Iraq, Iran, and Syria, where some species still naturally occur. The domestication of wheat allowed the subsequent domestication of cultures, where people could grow large quantities of food for an increasing population. The major production areas today include the central plains of North America, north-west Europe, the Mediterranean basin, southern Russia and the Ukraine, India, north-central China, south-western Australia, and Argentina (Feldman et al., 1995).

Triticum aestivum L. is generally known as common or bread wheat. It is a hexaploid spring wheat with three sub-genomes, each containing seven chromosomes (Gupta et al., 2008). Each sub-genome came from a diploid progenitor to make up the full AABBDD genome, with 2n=6x=42. *Triticum turgidum* ssp. *durum* is a tetraploid spring wheat with the genome AABB, with 2n=4x=28.

A number of hybridization/crossing events of early wheat led to the development of modern day wheat. *T. monococcum* (wild einkorn) and *T. searsii*, contributed the A and B genomes, respectively, to first create *Triticum turgidum* (wild emmer) (Peleg et al., 2011). This allotetraploid was the first successful wheat crop to be cultivated (Salamini et al., 2002). It later gave rise to both *T. turgidum* ssp. *durum* and *T. aestivum*. The D genome for *T. aestivum* was donated by *Aegilops tauschii* (Gupta et al., 2008).

Certain traits were selected in the process of domesticating wheat, many of which were important for increasing yields and preventing yield losses at harvest (Peleg et al., 2011). Some of these traits included glume reduction for easier threshing, larger kernel size, and non-shattering plants (Gupta et al., 2008; Peleg et al., 2011). Other traits selected were loss of seed dormancy, increased carbohydrate content, lower protein and mineral concentrations, and photoperiod insensitivity. Wild emmer wheat had large seeds that were easily shattered at maturity, domesticated emmer wheat contained seeds

that were hulled, and these gave rise to the free-threshing wheat seen today (Salamini et al., 2002).

The two major species of wheat grown in Canada are *T. aestivum* and *T. turgidum* ssp. *durum*. *Triticum aestivum* was first recorded to have been grown in western Canada, with little success, in the winter of 1812 and spring of 1813 (Buller, 1919). The Selkirk settlers arrived from Scotland and settled in the area of the Red River. It was not until the 1820s, with new seed wheat from the United States, that this crop became more of a success. In 1870, the cultivar Red Fife was introduced to western Canada and the number of acres grown increased considerably over the next several years (Dickenson, 1980). Red Fife was the first well-known cultivar in Canada and it gave rise to many of the cultivars that are grown today. Marquis was the next major cultivar to be bred, which was released in 1911 and replaced Red Fife due to its superior qualities in terms of yield, maturity, stature, and shattering resistance (Morrison, 1960).

As of 2010, there were over nine million hectares of wheat grown in Canada (Agriculture and Agri-Food Canada, 2012). There are both spring and winter wheat types, but the majority of production is spring wheat. Spring wheat is predominantly grown in the western Prairies, with Alberta, Saskatchewan, and Manitoba producing 32.4, 51.7, and 14.3 % of the Canadian total, respectively (Statistics Canada, 2011). There are more than 175 different varieties of spring wheat, which are placed into twelve categories based on certain properties and therefore end use markets (Agriculture and Agri-Food Canada, 2012). Durum wheat is produced only in Alberta and Saskatchewan and makes up approximately sixteen percent of spring wheat production, whereas non-durum wheat can be found in all production regions, and makes up the other eighty-four percent (Statistics Canada, 2011). The market classes of spring wheat commonly grown in western Canada are determined by their major characteristics and end uses (Table 2.1).

Triticum spelta L. is another type of wheat grown in Canada, but is considered a specialty crop and is not covered under the Canada Grain Act (Canadian Grain Commission, 2009). Its grading requirements are listed under “other cereal grain” but the end use of spelt is similar to *T. aestivum* in that it is ground into flour and used in a number of baked goods. *T. spelta* was derived from free-threshing tetraploid wheat through hybridization with hulled emmer (Dvorak et al., 2012). It has the genome

AABBDD, just as in *T. aestivum*. The D sub-genome has been used extensively to study the differences between the two species, as the A and B sub-genomes are quite similar. Spelt, unlike bread wheat, has its seeds tightly enclosed within the glumes, and requires mechanical force to separate them.

Table 2.1. Select western Canadian wheat milling class characteristics and end uses (adapted from Canadian Grain Commission, 2012)

	Colour	**Size**	**Shape**	**End Uses**
Canadian Western Red Spring (CWRS)	Translucent red	Small to midsize	Oval to ovate	High volume pan bread; hearth, steamed, or flat breads; noodles
Canada Western Extra Strong (CWES)	Dark to medium red	Large	Ovate, s-shaped base	Specialty products when high gluten strength needed; ideal for blending
Canada Prairie Spring Red (CPSR)	Opaque red to orange	Midsize to large	Ovate to elliptical, incurved base	Hearth, steamed, or flat breads; noodles
Canada Prairie Spring White (CPSW)	White	Midsize to large	Ovate to elliptical, incurved base	Noodles, chapatis, flat breads
Canadian Western Amber Durum (CWAD)	Amber	Large to midsize	Elliptical	Semolina (for pasta), couscous
Canada Western Soft White Spring (CWSWS)	White	Small to midsize	Ovate to oval	Flat breads, steamed breads, noodles, chapatis, cookies, cakes, pastry
Canada Western Hard White Spring (CWHWS)	White	Small to midsize	Oval to ovate	Bread and noodles
Canada Western General Purpose (CWGP)	No specifications			Ethanol products, animal feed

2.2 Ergot Disease

2.2.1 Historical Significance

Claviceps purpurea (Fr.) Tul. is a fungal pathogen that has been around for centuries, but was first recorded to cause disease, later known as ergotism, in Germany in 857 (White et al., 2003). In the following years, the occurrence of ergotism became more common throughout the major grain producing regions of Europe. Rye was the major food source for most people, and it is the most susceptible cereal to infection by the fungus. Consumption of ergot contaminated grain products resulted in ergotism, which was categorized into two forms (De Costa, 2002). These could occur simultaneously or separately; one form is termed acute or convulsive ergotism, and the other is chronic or gangrenous ergotism.

Gangrenous ergotism was named so because of the vasoconstriction of veins that often resulted in gangrene of hands, feet, or whole limbs (De Costa, 2002). In France circa 1100 A.D. followers of St. Anthony created refuge centers for people who suffered from ergotism, and because of the burning sensation associated with this form of the disease, it became known as "St. Anthony's Fire". In the most severe cases, ergotism even resulted in death. This was the case in Sologne, France in 1778, where an ergot epidemic lead to the death of more than eight thousand people.

The major symptoms of acute (convulsive) ergotism are hallucinations and dementia, which are caused by the lysergic acid diethylamide (LSD) properties in ergot alkaloids acting on the hormone serotonin (De Costa, 2002). This type of ergotism may have led to the instances where people were accused of being witches and werewolves in the Middle Ages (White et al., 2003). There have been other legends associated with ergotism that involved consuming ergot sclerotia on purpose. In some Greek initiation ceremonies, ergot was consumed in the form of water soluble alkaloids, which caused sweating, trembling, vertigo, and fear. In religious ceremonies in Israel and New England, it was proposed that ergot consumption caused people to hallucinate thereby allowing them to experience the presence of God.

In the sixteenth century awareness of ergot increased and it became of use in the medical world (Moir, 1955). Ergot sclerotia were given to pregnant women to increase

uterine contractions and quicken the process of child birth. However, fatalities, including abortion, often occurred due to lack of knowledge on the proper dosage.

In the seventeenth century, ergotism was finally associated with the consumption of contaminated grain (De Costa, 2002). The first people to recognized ergot sclerotia on plants were farmers who observed infected cereal crops and forage grasses (Bove, 1970). Rye (*Secale cereale* L.) was the major crop infected, but ergot was also seen in wheat (*Triticum* spp.), barley (*Hordeum vulgare* L.), and oat (*Avena sativa* L.) crops.

Throughout the eighteenth century, the origin and control of ergot disease were discovered, which led to a decrease in cases of ergotism in the nineteenth century (De Costa, 2002). It became known that there were a number of different species of *Claviceps*, and that they produced a wide range of alkaloids. The nineteenth century also saw an introduction of ergot usage in pharmacology (Moir, 1955). In 1820 ergot entered into the United States Pharmacopoeia and the British Pharmacopoeia in 1836 (Taber, 1985). Alkaloids were extracted from the sclerotia and purified chemical substances were used in making medicinal products (Moir, 1955). Work continues to be done today on the biosynthesis of the ergot alkaloids and their biochemical pathways, including research on the genes and enzymes involved in creating these biologically active substances for use in the pharmaceutical industry.

2.2.2 Current Significance

Claviceps purpurea is considered as the most common or most widely known species in the *Clavicipitaceae* family (White et al., 2003).This species can infect a number of the cereal crops grown in North America and it produces a number of alkaloids that are poisonous to both humans and animals when consumed. Depending on the species of the original host and the newly infected host, the alkaloids can vary in their amounts and level of toxicity (Knox & McLeod, 2003). Rye, for example, produces some of the highest levels of alkaloids compared to many other cereals (Shelby, 1999).

Due to the extreme toxicity of the ergot alkaloids, very little is tolerated in grain samples (Table 2.2), and only 0.01% in a sample of most Canadian wheat will be allowed before downgrading occurs (Canadian Grain Commission, 2011a). The Department of Justice has also set out regulations in the Canadian Seeds Act (2012) that indicates the

allowable number of sclerotia, or ergot bodies, in one kilogram of grain. There are limits to the amounts of sclerotia in seed of common (*T. aestivum*) and durum (T. *turgidum* subsp. *durum*) wheat, barley and oats, as well as for rye and triticale (Table 2.3).

Table 2.2. Ergot sclerotia as a percentage of net weight of a seed sample for western Canadian wheat classes and their resulting grades. (Adapted from the Official Grain Grading Guide; Canadian Grain Commission, 2011a).

Wheat Class	Grade				
	No. 1	**No. 2**	**No. 3**	**No. 4**	**Feed**
Canadian Western Red Spring (CWRS) Canada Western Hard White Spring (CWHWS) Canadian Western Amber Durum (CWAD)	0.01%	0.02%	0.04%	0.04%	0.1%
Canada Western Red Winter Canada Western Soft White Spring (CWSWS)	0.01%	0.02%	0.04%	NA*	0.1%
Canada Western Extra Strong (CWES) Canada Prairie Spring White (CPSW) Canada Prairie Spring Red (CPSR)	0.03%	0.06%	NA	NA	0.1%
Canada Western General Purpose (CWGP)	0.1%	0.1%	NA	NA	NA

*NA – Not Applicable

Table 2.3. Maximum number of sclerotia per kilogram seed allowed in various cereals, adapted from the Canadian Seeds Act (2012).

	Maximum number of sclerotia/kg		
Grade	**Wheat**	**Barley & oats**	**Triticale and rye**
Canada Foundation No.1	1	1	2
Canada Foundation No.2	8	8	10
Canada Registered No.1	1	1	2
Canada Registered No.2	8	8	10
Canada Certified No.1	2	2	4
Canada Certified No.2	8	8	15
Common No.1	2	2	4
Common No.2	8	8	15

Ergot alkaloids remain quite stable during end-use processing (Fajardo et al., 2003). Even when ground into flour and cooked in various products, the toxins can remain fully active (Knox & McLeod, 2003). Therefore, it is imperative that sclerotia are

properly removed before processing to minimize risk to consumers. There are six predominant alkaloids in hard red spring wheat that are retained, which include ergonovine, ergosine, ergocornine, α-ergokryptine, ergotamine, and ergocristine, the latter two being the most common. In the early 1990s a number of Canadian foods produced from cereal grains were tested for the presence of ergot alkaloids (Scott et al., 1992). Over fifty percent of the products tested positive, with the highest levels in products made from rye and triticale. Although the amounts detected were usually inadequate to cause ergotism symptoms, the potential for harm exists if the diet is high enough in contaminated grain products. Rye produces even greater amounts of alkaloids, with the two major ones being ergocristine and ergotamine (Scott et al., 1992). Three others are produced in lesser amounts including ergosine, ergocornine, and ergonovine. Alkaloids produced in the ergot of triticale are similar to that of wheat.

Ergot-based alkaloids are considered one of five major mycotoxins in North America that impairs growth and reproductive efficiency in the animal industry (Diekman & Green, 1992). Depending on the species and age of the animal, many symptoms can arise at varying levels of ergot consumption (Knox & McLeod, 2003). In several livestock species, problems that can occur include reduced weight gain, lowered reproductive efficiency, and agalactia (Diekman & Green, 1992). There are a number of signs of ergotism that range in severity. Animals may have convulsions, become paralyzed in their posterior, and lose blood flow to extremities leading to gangrene and possible loss of limbs, ears, and tail. If alkaloid ingestion is great enough, the animal may die (Knox & McLeod, 2003). Less severe symptoms include diarrhea, heat stress, and salivation.

Individual species of *Claviceps* are largely unknown and therefore the type and potency of alkaloids they produce may cause varying reactions. The potential for poisoning exists regardless of the source, and therefore there is a general rule that there should never be more than 0.1% ergot by weight in any feed ration. Young and pregnant animals are especially susceptible to poisoning, but often minor symptoms will subside if clean grain is provided within a certain period of time.

Before 1999, ergot in cereal crops [in Saskatchewan] had not been observed since the 1980s (Saskatchewan Ministry of Agriculture, 2009). Recent outbreaks are likely due

environmental conditions. In other areas, increased incidence has also been attributed to the use of male-sterile plants for hybrid breeding, which are especially susceptible (Tudzynski et al., 1995). The infrequency of ergot outbreaks makes it difficult to isolate the cause of the problem.

Environmental conditions are frequently the most influential factor for epidemics, but host characteristics are also important. Since environmental conditions cannot be predicted from year to year, it may become increasingly important to discover cereal cultivars that are resistant to ergot infection to prevent the negative consequences of ergot consumption. To date, there are no known wheat cultivars that are resistant to infection by *C. purpurea,* and significant economic losses, including grade penalties and decreased yield, can occur in years with ergot outbreaks (Menzies, 2004). Yield losses occur because kernels are replaced with sclerotia, and seed set can be impaired beyond the florets that have been infected (Rapilly, 1968).

2.3 *Claviceps purpurea* (Fr.) Tul. Disease Epidemiology

2.3.1 Biology

Claviceps purpurea is an important floral pathogen that infects cereal and grassy species in temperate growing regions (Mantle & Swan, 1995). It is a tissue specific, biotrophic fungus that infects the ovaries of the host plant and replaces them with an ergot body, or sclerotium (White et al., 2003). *Claviceps purpurea* is one of the most widely known species in the *Clavicipitaceae* family, from the order *Hypocreales,* and has the ability to infect a wide range of hosts. Of the six hundred species in the *Poaceae* family, *C. purpurea* has been found to parasitize over four hundred (Bove, 1970). These include many forage grasses and cereal crops such as wheat, barley, and rye. In wheat, races, or *forma speciales,* are not known (Campbell, 1957), but there are strains that vary in the degree of pathogenicity (Darlington et al., 1977). Pažoutová et al. (2000) examined the sclerotia of a number of grassy species from Europe, North America, Australia, and South Africa and identified two groups that were based largely on habitat. Tools used to separate the isolates included random amplified polymorphic DNA (RAPD), *Eco*RI

restriction site polymorphism in 5.8S ribosomal DNA, alkaloid analyses, conidia size, and sclerotia densities determined by floatation. Group G1 isolates are found in open fields and meadows as well as along road sides where conditions are sunny and often dry (Pažoutová et al., 2000). Group G2 isolates are found in areas that are shady or wet, such as river banks, ponds, ditches, and forests. Some species of grasses can be infected by both G1 and G2 isolates. However, the incidence of finding members from different groups in the same location is quite rare.

Rye (*Secale cereale*) and wheat (*Triticum* spp.) are host species grown in North America in areas favourable for the G1 strains of *C. purpurea*. An additional group, G3, was identified by Pažoutová et al. (2002) that inhabits salt marshes. The majority of *C. purpurea* strains belong to either G1 or G2. These three groups have been described by Douhan et al. (2008) as ecotypes, and work has been done to try to distinguish speciation among them. Using phylogenetic and population genetic analyses they found that G2 and G3 are chemoraces. They are more closely related compared to G1, which has significantly diverged from either of these groups. Between these ecotypes there is little to no gene flow, likely driven by the adaptations they have for their different ecological habitats.

Claviceps purpurea is distinguished from other *Clavicipitaceae* fungi based on a few major characteristics. The stroma develops from an exposed sclerotium and within it are multiple capitate perithecia (Bischoff & White, 2003). The stromata range from 5-25 mm in length and there may be as many as 60 or as few as 1 per sclerotium. The perithecia produce many cylindrical asci (pl.), and each ascus (sing.) contains 8 ascospores. The ascospores are the sexual spores, which range in size from 50-76 x 0.6-0.7 μm. The ascospores are long and threadlike and extend the entire length of the ascus (Alexopoulos et al., 1996). They are forcibly discharged from the asci upon maturity. The asexual spores, called conidia, are much smaller, and range in size from 4-6 x 2-3 μm. Sexual spores are only formed when the fungus develops in a host plant, whereas the asexual spores can be produced in axenic culture (Esser & Tudzynski, 1978).

Sclerotia (pl.) are the resting stage of *C. purpurea*, and are commonly found to be black in colour and resemble the kernels they replace (White et al., 2003). The surface of a sclerotium is composed of pseudoparenchymatous cells and the inside is usually white-

grey (Bischoff & White, 2003). The length can range from 2-30 mm and be straight, slightly curved or spur-like. The size and shape is often dependent on how much space is available within the floral cavity of the host (Pažoutová, 2003). In wheat, for example, the sclerotia are commonly more rounded than in rye.

Sclerotial size has been found to influence the rate of germination, with larger sclerotia having higher rates (Cooke & Mitchell, 1966). It is suggested that the larger sclerotia have more energy available to use in the germination process. There may also be a direct linear relationship between the size of sclerotia and the number of stromata produced. Furthermore, the stromata may be taller than those produced from smaller sclerotia. Under natural conditions, when the sclerotia may be buried below the soil surface, larger sclerotia would have an advantage due to the taller stromata they produce. If the stromata are able to get to the surface, spores will have a better chance of being discharged and be able to cause infection.

2.3.2 Life Cycle

There are four major stages in the life cycle of *Claviceps purpurea* (Bove, 1970), which are shown in Figure 2.1. Germination is followed by the conidial stage, the ascigerous stage, and finally the resting or sclerotial stage.

In the spring, the sclerotia that were left on or in the soil germinate under favourable conditions (Figure 2.1 a). For germination to occur, the sclerotia must undergo a cold treatment requiring temperatures between 0 and 10°C (Alderman, 2003). In nature these requirements for the sclerotia are fulfilled during the cold or freezing periods of winter (Mitchell & Cooke, 1968). When the temperature is near the higher end of the range, the time required to activate germination will be lengthened. However, severe cold periods may reduce the viability of sclerotia. After the incubation period, the greatest germination occurs under moist conditions with temperatures of approximately 10°C, though sclerotia will germinate up to 20°C (Rapilly, 1968). Beyond 25°C, germination rates begin to decrease. If temperatures fall below 10°C, the development of stromata may be inhibited.

The pedicel of the stromata first grows down into the soil and then upwards towards the light (Rapilly, 1968). This is due to the positive phototropism of the head

region. If the pedicel were to be cut, sclerotia have the potential to grow new ones. The pedicel holds up the head that contains the perithecia, within which are the asci, and they contain the ascospores. The stromata can grow to be almost 1 cm, and can be seen with the naked eye (Alexopoulos et al., 1996). Within the head region, plasmogamy takes place starting with the migration of the male nuclei into the female ascogonium. The perithecia are produced through the growth of hyphae around the sexual apparatus. Eventually the mature perithecia will contain the asci that contain eight mature ascospores each.

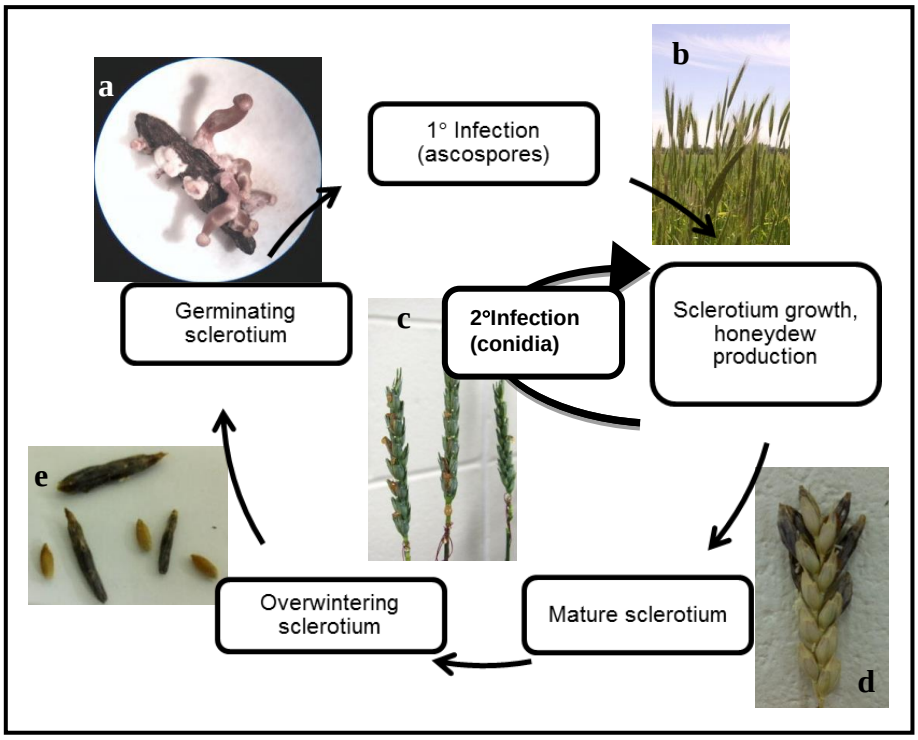

Figure 2.1. The life cycle stages of *Claviceps purpurea*: a) germination; b) primary infection; c) honeydew/conidial stage; d) ascigerous stage; e) resting/sclerotial stage

For an infection to occur there must be air-borne ascospores present to infect the florets (Cooke & Mitchell, 1966). The ascospores require adequate humidity to be released (Alderman, 2003). Each sclerotium is capable of producing up to one million ascospores (Bove, 1970). Once they are released, the ascospores are carried by wind to the florets where they can infect the ovary, stigma, or style, which occurs within 24 hours (Figure 2.1 b). Rain splash and insects can also disperse the ascospores. This stage most

commonly occurs in early flowering plants, such as wild grasses, fall-sown cereals, and early-sown spring crops.

The second stage of infection is known by several names, which include honeydew, conidial, or sphacelial stage (Bove, 1970). In this stage, the ascospores must germinate to begin infection, which requires moisture at the base of the ovary. Since *Claviceps* is a homothallic fungus, only a single ascospore is required for infection and ultimately the formation of a sclerotium (Esser & Tudzynski, 1978).

It has been suggested that spores germinate in a manner similar to the pollen tube in plants (Tudzynski et al., 1995). First the hyphae penetrate directly into the pistil without the help of any specialized infection structures, but rather with disintegrative enzymes (Bove, 1970). The hyphae then grow intercellularly in the epidermal walls of the ovary before branching occurs at sites appropriate for entry (Tudzynski et al., 1995). The hyphae will grow to the tip of the rachilla. The fungus is thought to get its nutrition during its intercellular growth through the use of pectolytic enzymes, which degrade the cell wall constituents of the ovary. The intercellular hyphae have characteristics of haustoria common to other biotrophic fungi.

Within six to eight days, the mycelia grow and cover the entire pistil, with the exception of the stigma (Bove, 1970). The intercellular hyphae become connected to the upper part of the vascular bundle at the tip of the rachilla (Tudzynski et al., 1995). Once the connection is made, the photoassimilates will nourish the fungus rather than the seed that would have otherwise grown. The connection is also very important for the exudation of honeydew. The digested tissue of the pistil will be replaced with a white mass called the sphacelial stroma. During the time of infection, the enzyme catalase is also secreted by the fungus, which helps to suppress the host's defensive responses (Agrios, 2005). Throughout this time, the ovary will continue to grow, but a sclerotium will start to form rather than a kernel, beginning at the basal end of the ovary (Bove, 1970). At the upper end, sporodochia are formed and will begin to produce small sphacelia type conidia (Agrios, 2005). The conidia are covered in a sweet, yellow, mucilaginous substance known as honeydew (Bove, 1970).

The honeydew facilitates secondary infections (Bove, 1970). This sticky sweet substance exudes from the infected florets (Figure 2.1 c). It is highly concentrated in

sugar (Rapilly, 1968), which attracts insects such as flies and weevils that can transfer the honeydew, and consequently the conidia, to healthy florets (Bove, 1970). The honeydew may be exuded in large drops that are heavy enough to fall to lower spikelets or cause the spike to bend and physically come into contact with other spikes to cause infections. For the conidia to produce germ tubes and cause infection, the honeydew must first be diluted from rain water or dew (Schwartingl & Hiners, 1945). It takes as few as 1 to 10 conidia to infect an ovary, but the maximum level of infection occurs when there are upwards of 2000 conidia per ovary (Puranik & Mathre, 1971). The honeydew stage lasts for approximately 72 hours (Rapilly, 1968), but can persist for longer periods of time when conditions are cloudy and humid (Schwartingl & Hiners, 1945). Secondary spread can occur between cereals of the same species, as in a cultivated crop, or from other species, as would occur from the surrounding grasses or weeds to the crops within a field (Knox & McLeod, 2003).

The third stage of the ergot disease cycle is called the ascigerous stage, which is when the production of the sclerotium is completed (Bove, 1970). In this stage, the production of honeydew ceases and the conidial stroma withers (Tudzynski et al., 1995). At the lower end of the ovary, the hyphae will continue to penetrate its walls (Bove, 1970). The mass of mycelia will become thicker and interwoven as it penetrates upwards into the ovary. The large mass of mycelia eventually encompasses the entire ovary, and the pressure it creates causes formation of a dense tissue known as pseudoparenchyma. This cortex then becomes black in colour due to the deposition of pigments. A full grown sclerotium will replace the entire kernel (Figure 2.1 d). The average size of sclerotia may decrease as the frequency of sclerotia per spike increases (Rapilly, 1968). It takes approximately twelve to sixteen days from the time of honeydew production to the formation of sclerotia (Wood & Coley-Smith, 1982). In some cases, honeydew can be produced until plant maturity. The ovaries that are not infected by the fungus can grow normally, undisturbed by the activities occurring in the neighboring florets (Alexopoulos et al., 1996). It has also been found that even when there are no sclerotia, shriveled ovaries or undeveloped kernels form rather than fully developed structures (Platford & Bernier 1970).

The final stage of the ergot life cycle is the resting stage, in which the sclerotium reaches maturity, occurring approximately five weeks after inoculation (Tudzynski et al., 1995). Sclerotia are able to over-winter and are therefore critically important for the disease cycle to continue and maintain inoculum levels from year to year (Menzies, 2004). When crops mature and are harvested, the sclerotia either fall from the glumes to the ground (Figure 2.1 e) or become mixed with the harvested grain (Knox & McLeod, 2003). The sclerotia that were left on or in the ground, or are seeded with a new crop, are then available as inoculum in the following year if conditions are favourable for germination. Sclerotia usually remain viable for approximately one year but are known to survive for two springs after formation (Mitchell & Cooke, 1968). After three years in the soil, the sclerotia are no longer viable under field conditions, but can remain viable in a laboratory setting (Rapilly, 1968).

2.3.3 Symptoms in Cereals

The first signs of an ergot infection are noticeable at flowering when the honeydew exudes from infected florets (Knox & McLeod, 2003). Although it may be difficult to see the actual honeydew, it is easy to recognize when there is dust or pollen stuck to the honeydew on the spike, making the infected plants stand out amongst the healthy ones. Depending on the species of cereal, the time from inoculation to the first signs of honeydew varies, with barley showing signs first at 6.9 days, followed by 8.5 days for rye, and 10.3 days for wheat (Campbell, 1957). Fewer sclerotia may be produced when there is less honeydew (Platford & Bernier, 1976).

The presence of sclerotia is the most notable indication of an ergot infection. They are easily seen as large black or purple objects protruding from the spikes in a standing cereal crop (Knox & McLeod, 2003). Rye often exhibits the largest sclerotia compared to either wheat or barley. In many cases, the sclerotia are the same size as the kernel, but can be much larger. The sclerotia from cereal grains are usually much bigger than the small slender sclerotia found in forage grasses. Once the crop is harvested, the sclerotia can still be seen mixed amongst the grain, but require a microscope for identification once crushed up for flour or feed. There are also physiological and chemical tests that can be used to determine if a sample contains ergot.

2.3.4 Sources of Inoculum

Inoculum in headland grasses is almost always available, but in the field, infection fluctuates from year to year (Campbell, 1957). In years when conditions are favourable for ergot disease, the grasses surrounding a field may become an important source of inoculum for the crop. Weedy grasses within the field can also be a significant source of inoculum (Mantle et al., 1977).

Sclerotia in the headland areas often remain at the soil surface undisturbed (Mantle et al., 1977). Increased amounts of sclerotia at or near the surface results in an increase in the amount of ascospores released (Cooke & Mitchell, 1966). Germination of sclerotia bodies often occurs during flowering in grasses (Alderman, 2003). The earlier the grasses flower, the more likely it is that the grass will become infected and in turn be able to infect the cereal crop (Mantle et al., 1977). There are also some cereals that flower early enough for anthesis to coincide with ergot germination, in which case the flowering date of the grass would not impact the infection of the crop. However, the cross-infection from grass to cereal host has been considered to be the major cause of infection (Bretag & Merriman, 1981). The grasses become infected and then after one to two weeks, begin to exude honeydew that contains conidia. The honeydew can be passed to the cereal spikes by rain splash or by insects. For infection to occur in the cereal host, the honeydew must be produced at anthesis, and the ergot fungus must be pathogenic not only to the grass, but to the cereal as well. Infection of cereals in this manner means that they do not have to be flowering when ascospores are released (Bove, 1970).

Campbell (1957) conducted a study where he collected ergot from thirty-eight different host species and cultured the resulting 421 isolates of *C. purpurea* on artificial media. The conidia from these cultures were then used to inoculate rye, wheat and barley in greenhouse conditions, in which all but one were successful in causing infection. The honeydew produced from the infected rye was then used to inoculate forty-six gramineous species in both greenhouse and field conditions, and was successful in both environments. Campbell's study illustrates the potential for indigenous and forage grasses to be a reservoir for ergot infection of cultivated cereals.

Bretag and Merriman (1981) examined wheat in the state of Victoria in Australia to determine the main source of inoculum for infection. Both primary and secondary

inoculum was considered. The treatments included different combinations of wheat and ryegrass with sclerotia from either wheat or ryegrass. The sclerotia were buried just below the soil surface and left to germinate and produce ascospores to cause the primary infections. The mixed wheat/ryegrass plots always had a higher infection rate compared to plots with wheat alone. Ryegrass flowering occurs for a longer period of time and peaks at the time of ascospore discharge. The wheat was mainly infected due to the spread of conidia from the ryegrass in secondary infections.

The infected cereal can also be a source of inoculum (Puranik & Mathre, 1971). Under field conditions, tillers are produced over a considerable amount of time. This leads to spikes that flower much later and allows for the infected spike on the main stem to develop honeydew to cause infection of tillers.

2.4 Influence of Environmental Conditions on Infection

The disease triangle states that for a disease to establish itself there must be a susceptible host, the presence of the pathogen, and a suitable environment. Environmental conditions have been considered by many to be the most important factor contributing to ergot epidemics, specifically temperature and moisture (Campbell, 1957; Watkins & Littlefield, 1976; Menzies, 2004).

2.4.1 Temperature

Cool temperatures, in conjunction with moist soil or humidity, encourage the germination of sclerotia and the release of ascospores early in the growing season (Knox & McLeod, 2003). Germination usually occurs when the temperature is warm and stable during early to late June (Mitchell & Cooke, 1968). Temperatures between 10 and 20°C are most conducive. At 25°C germination has been shown to drop below 50% and continues to decline as temperatures increase. Warmer temperatures (between 20 and 30°C) are optimum for mycelial growth once the infection has been initiated.

Cool conditions during flowering can reduce pollination and create periods of uneven flowering and out-crossing (Mantle et al., 1977). The duration of flowering may also be extended. Flowering can occur as much as one to two weeks earlier than normal if

weather conditions are warm and sunny in spring and the early part of summer. Under unfavourable flowering conditions, the infection period increases, which is mentioned in greater detail in the following section.

Cool and wet soils in the spring can lead to a reduction in the root growth of new plants (Government of Saskatchewan, 2006). This may cause problems with the uptake of micronutrients, thus causing deficiencies in the plant that may predispose it to infection by certain pathogens such as *C. purpurea* (Graham, 1983).

2.4.2 Moisture

Disease presence has been known to increase with an increase in soil surface moisture in the spring and early summer (Menzies, 2004). This is due to the need for moisture for the overwintering ergot bodies to germinate. Moisture is also required for the dispersal of ascospores, which can be present in soil or as rainfall (Alderman, 2003). The greatest release of ascospores occurs at 100% relative humidity (RH), and declines significantly below 30% RH (Rapilly, 1968). Once the ascospores reach the floret, moisture at the base of the ovary is required for the ascospores to penetrate the plant tissue and begin development (Bove, 1970). Moisture may be present in greater amounts from precipitation or from as little as morning dew. Irrigation has also been shown to result in increased infection since it provides a source of moisture for the fungus to flourish (Puranik & Mathre, 1971). Irrigation in the spring would give the sclerotia the moisture they need to germinate. Water splash from irrigation can also cause transfer of the honeydew, resulting in secondary spread of the disease. Rain splash under natural conditions would likewise spread the honeydew.

Insects can be a major factor in the transportation of secondary inoculum (Mantle et al., 1977), and in wet conditions populations of insects often escalate (Saskatchewan Ministry of Agriculture, 2009). Some of the most common vectors include aphids, thrips, midge, and leaf hoppers (Saskatchewan Ministry of Agriculture, 2009), as well as flies, weevils, and ants (Bove, 1970).

2.5 Influence of Flowering on Infection

There are two proposed mechanisms of resistance to ergot infection under natural conditions (Platford & Bernier, 1976). The first is due to the physiology of the host, and the second is an escape mechanism based on flowering habit and floret morphology. The following sections discuss the latter, as little is known on the physiological resistance mechanism within the host.

2.5.1 Floret Morphology and Male-sterility

A number of studies have illustrated the importance of floret morphology in regards to susceptibility to ergot (Campbell, 1957; Puranik & Mathre, 1971; Platford & Bernier, 1976; Mantle et al., 1977). Access to the floret for infection may be made easier for the fungus depending on the composition of the floret. Grasses with a protogynous flowering mechanism can be infected more easily than self-fertilizing plants (Mantle et al., 1977). A longer period of susceptibility results when the stigma protrudes from the floret before the anthers release pollen.

However, a recent study (Bayles et al., 2009) examined a number of flowering characteristics (anther extrusion, anther size, blind florets, and ear density) and found results contrary to those published previously. There were some cultivars that had characteristics that would likely predispose them to infection, what were called a "greater tendency to open-ness", that did not result in more severe infection. The same was true for less open flowers, which exhibited a relatively high level of infection. The study suggests that the difference in susceptibility to infection between cultivars is primarily due to tissue resistance.

Hybrid production in wheat and barley has the potential to increase yields as much as thirty percent above conventional varieties (Done, 1973). The F1 hybrids have been shown to have greater kernel size and number, number of tillers, and total yield. There may also be potential for an increase in disease resistance and drought tolerance, and a decrease in vulnerability to soil conditions such as acidity (Rajaram, 2001). To obtain these hybrids in wheat, male sterility must first be obtained (Done, 1973). This is accomplished through either cytoplasmic or genetic male sterility, which prevents self-

pollination. The problem with male sterility in wheat is that the flowers must open for the plant to receive pollen for fertilization. When the flowers are open, the floral organs are exposed to diseases that attack the ovaries. Male sterile florets have been found to contain approximately three times more sclerotia compared to fertile florets (Rapilly, 1968). One study found a significant correlation between incidence of ergot disease and impaired seed set in male sterile wheat (Mantle & Swan, 1995). The mean incidence of ergot sclerotia in the fertile wheat was less than 1% but was over 20% in the male-sterile line, which set less seed due to cytoplasmic male sterility and limited pollination from pollen donors. These results illustrate the risk of F1 hybrid seed production, as florets not pollinated are an easy target for *C. purpurea*.

2.5.2 Flowering Habit

Escape or avoidance of infection is often the mechanism associated with ergot resistance under natural field conditions (Platford & Bernier, 1976). In some cereals, in particular barley, the florets reach anthesis before the spike fully emerges from the boot. Others undergo self-fertilization after the spike has emerged, but because the glumes remain closed, the plant is still less susceptible to infection (Campbell, 1957).

Rye is reported to be the most susceptible cereal to ergot, followed closely by triticale, then wheat, barley and oat (Menzies, 2004). Rye is an outcrossing plant and therefore must open its flowers for pollination to occur. This gives the fungus a considerably longer time to attack compared to in wheat, barley, and oat, which are largely self-pollinating plants. In wheat, the glumes may open, but very briefly and irregularly (Bretag & Merriman, 1981). In fact, out-crossing in wheat is often less than one percent (Lawrie et al., 2006). After anthesis occurs and the florets are fertilized, susceptibility has been shown to decrease (Rapilly, 1968).

A number of studies have indicated the importance of the mechanics and duration of flowering in regards to escaping infection. Platford & Bernier (1976) used oat and barley, which usually reach anthesis while still in the boot, to inoculate artificially and to be inoculated naturally in a field setting. They reported that when these two cereals were inoculated artificially, they had a much higher rate of infection compared to infection under field conditions. Campbell (1957) reported that if inoculation of barley was

delayed until after flower emergence from the leaf sheath and the anthers appeared, no infection would occur. Wood and Coley-Smith (1982) used germinating sclerotia as sources of inoculum in field trials using male sterile cereals. They observed in the two years of the study that many of the main spikes escaped infection due to timing of flowering. The successive flushes of tillers, however, saw a greatly increased infection rate, with 84% of the total diseased spikes in one year and 99% in the other year.

There are other factors that may cause a delay in the time of flowering or extend the flowering period. Improper herbicide application can injure a plant and cause delayed maturity and floret sterility (Knox & McLeod, 2003). Excess nitrogen has also been shown to delay heading (Rapilly, 1968). When flowering is delayed, the time of anthesis in the cereal crop is more likely to coincide with the production of secondary inoculum. When flowering periods are extended, the florets remain unfertilized and open longer and increase the length of time that the plant is susceptible to infection.

2.5.3 Effect of Fertilization

The pollination of an ovary and subsequent fertilization has been found to decrease the chance of infection by *C. purpurea* (Puranik & Mathre, 1971; Darlington & Mathre, 1976; Watkins & Littlefield, 1976). In barley, the decline in susceptibility following fertilization has been attributed to changes in morphology and biochemistry in the ovary that restricts access to the fungus and stops its penetration and/or development within the ovary (Cunfer et al., 1975). Many studies have used a combination of inoculation dates relative to anthesis to determine when and if resistance would decrease with fertilization in wheat.

Watkins and Littlefield (1976) used the hard red spring wheat, Waldron, a cultivar they found to be most susceptible to ergot, to test the effects of fertilization. A spore suspension was injected into the florets beginning five days before anthesis and continuing for seven days after anthesis. They observed significantly more sclerotia per spike in the plants inoculated before anthesis compared to plants inoculated after anthesis. As the date of infection moved further from anthesis, the incidence of ergot decreased, and on the seventh day after anthesis, no infection occurred.

Darlington and Mathre (1976) used cytoplasmically male sterile spring wheat cultivars to determine if fertilization and host genotype differed in their susceptibility to ergot. They hand pollinated the wheat and then inoculated it on the day of pollination and each day thereafter for seven days. Results were similar to those reported by Watkins and Littlefield in that the incidence of ergot decreased as time after pollination increased. When inoculations were performed more than seven days after pollination, essentially all lines tested were able to escape infection. The male and female parental lines of these cultivars were also inoculated. The infection of the progeny was mostly like that of the maternal genotype, likely because the first few days of the host-pathogen interaction is between the fungus and maternal tissue (stigma, style, or ovary).

Puranik and Mathre (1971) reported similar results in field and greenhouse conditions. Using male sterile wheat and barley, they observed differences in susceptibility between fertilized and unfertilized ovaries. Immediately after fertilization, the ovaries were susceptible for approximately four days, but then susceptibility started to decline until they were no longer susceptible after nine days. When the ovaries were not fertilized, the decline in susceptibility did not occur until ten days after anthesis and it took fifteen days until the spikes were no longer susceptible.

2.6 Ergot and Soil Micronutrient Deficiencies

Fertile soil is important for optimal plant growth. Trace elements are known to be particularly important in terms of certain host-pathogen interactions (Graham, 1983). There are two ways that a nutrient can help to sustain a healthy plant against disease pressure. The first is through greater tolerance to disease, and the second is through the resistance mechanisms within the host. When there are elements missing from the soil profile, or if there is not enough of a particular element, the host plant may be more susceptible to the disease. However, greater amounts of the nutrient do not increase the resistance and too much can be phytotoxic.

Certain micronutrient deficiencies have been shown to increase the susceptibility of a plant to fungal-borne pathogens (Graham, 1983). In particular, copper and boron deficiencies have been linked to an increased incidence of infection by *C. purpurea* as well as an increase in severity of symptoms. Copper and boron can affect the synthesis of

lignin and simple phenols, interfering with cell wall integrity. The cell wall is a common target for many pathogens and reduced lignin makes penetrating the cell wall much easier.

When plants are copper deficient, the anthers are smaller and the pollen is often sterile (Franzen et al., 2008). A lack of copper can interfere with microsporogenesis, which is an important stage of meiosis that occurs during the early boot stage (Graham, 1976). When the plant is sterile the florets are forced to open for cross-pollination, exposing them to the fungus (Pageau & Lajeunesse, 2006). Flower maturation may also be delayed, which would lengthen the time of susceptibility for infection (Knox & McLeod, 2003). Similar effects are seen with the florets of boron deficient plants (Graham, 1983). However, fungal pathogens do not require boron to survive and therefore the fungus can thrive even in a deficient environment. Boron deficiencies are common in soils that have little organic matter, especially in areas with high rainfall that would cause boron to be easily leached.

Soils can be replenished with micronutrients to alleviate problems with plant pathogens (Graham, 1983). However, there is a fine line between deficiency and toxicity, which must be considered when applying fertilizers. Copper and boron are best applied with a fertilizer containing nitrogen, phosphorous, and potassium (NPK fertilizer). When the limiting nutrient is supplied, the host has increased vigour and disease resistance. However, it has been shown that if copper is applied instead of boron in a boron deficient soil, the opposite is true, and ergot incidence will increase.

There are some soils that are more predisposed to micronutrient deficiencies than others. Properties of these soils include low organic matter, sandy texture and/or little clay content, and high pH (Government of Saskatchewan, 2006). Coarse soils that are low in copper as well as organic carbon have a greater tendency for copper deficiency in cereals compared to other soil types (Franzen et al., 2008). In Saskatchewan, copper and boron are particularly deficient in the northeastern part of the province in the Gray and sometimes in the Black soil zones (Government of Saskatchewan, 2006). The deficiencies often occur in peaty and sandy patches of land and on eroded knolls. In sandy soils, copper deficiency can be widespread. The edges of fields and grassy sloughs are often less deficient in copper compared to the rest of the field.

2.7 Analyses of Ergot Infection in Cereals

2.7.1 Inoculating Technique

For inoculating flowers experimentally, there are two major categories of practices used: natural and artificial (Peach & Loveless, 1975). Natural, as the name implies, are inoculation procedures that mimic the way infection might take place in nature. Methods include spraying the head with inoculum, spraying inoculum into florets while holding apart the glumes, and dipping the heads into a spore suspension. Artificial, or unnatural, procedures are those in which there are no comparisons in nature. These methods often include invasive procedures where the florets may be damaged. Induced infections will cause more sclerotia per head to be formed compared to a natural infection (Schwartingl & Hiners, 1945).

Campbell (1957) considered inoculation technique to be very important when studying the susceptibility of cereals and grasses to ergot infection. The best methods he found were to either cut the tips of the glumes off to blow the spores onto the floret with an atomizer or to inject the spore suspension directly into the floret. The methods he found to be less successful included dipping the spikes of the host into a spore suspension and spraying inoculum on the spikes without removing any parts.

Peach and Loveless (1975) tested two methods of inoculation. The first was a natural method in which an atomizer was used to spray a spore suspension when the glumes were open and the second an artificial method, where a hypodermic syringe injected the spore suspension into each floret. In both cases, ten heads of wheat (*T. aestivum*) were inoculated with spore suspensions of 3000 spores/ml from isolates of a number of different host species. Spraying occurred during flowering and injections were done just after the head emerged from the leaf sheath. When the wheat was inoculated by either method with *C. purpurea* spores of an isolate from *T. aestivum*, there were more sclerotia per head compared to any other isolate. However, injection resulted in more sclerotia per head compared to the spraying method. These results indicate that there may be some type of natural barrier preventing infection from certain isolates unless there is direct penetration into the floret as is the case with the injection method. However, in

nature this is not the case, and thus the spraying method might actually resemble more closely what would occur under natural conditions.

Puranik and Mathre (1971) also observed lower infection rates when the plants were sprayed with a conidial spore suspension. When the glumes were cut back 2-4 mm the infection rate increased to 100% when using a concentration of 10^6 conidia/ml. However, higher levels of infection resulted from the atomizer compared to dropping the inoculum into the floret with a capillary tube. This is suggested to be from the force of the discharge of the conidia into the floret, resulting in more conidia reaching the ovary.

When comparing similar inoculation procedures, Bayles et al. (2009) suggests that field conditions are more conducive to successful ergot infection compared to the greenhouse. This could be due to overheating in sunny conditions, and a lack of humidity and low light intensity. The ability to control the environment was shown to be extremely important when studying the host-pathogen relationship of ergot.

2.7.2 Differentiating Market Classes and Cultivars

There have been few studies performed to determine if there are differences among wheat market classes and among cultivars within classes. Some studies have observed that there are genotypic or cultivar differences that can impact the severity of ergot disease (Menzies, 2004; Pageau & Lajeunesse, 2006) and others have observed differences only at the market class level (Rapilly, 1968; Coley-Smith & Watkinson, 1987).

Pageau and Lajeunesse (2006) tested seventeen cultivars of spring wheat under natural field conditions for susceptibility to ergot. The study was conducted over three seasons (2003-2005) in the Lac-Saint-Jean area of Québec, with twelve to seventeen cultivars tested each year. The most resistant cultivars, AC Napier and Nass, had ergot contents below 0.1 % in all three years. The least resistant cultivars included AC Barrie, AC Gabriel, Saku, SS Fundy, and Torka. A concurrent study was performed using barley, which yielded similar results for ergot content.

In 1999, twenty-two locations in Saskatchewan were investigated for the presence of ergot contaminated grain (Fernandez et al., 2000). Multiple entries were collected from three wheat market classes common to the prairies: CPS, CWRS, and CWAD. At each

site, multiple samples were collected and a bulk sample of 150 g was analyzed for ergot sclerotia. At 55% of the locations the harvested grain was contaminated with sclerotia. All sites contained at least one entry that had enough ergot for the grain to be downgraded from number one to number two wheat, which is 0.1 and 0.2%, respectively (Canadian Grain Commission, 2011a). The location found to be most infected was Girvin, which is located in central Saskatchewan. The CWRS and CWAD market classes had similar infection levels, with 0.32 and 0.25% respectively, and the CPS class was slightly lower, with 0.15%. Over all the entries and locations, the average percent ergot in grain ranged from 0.01 to 0.10%, which is within the range of allowable levels for number one to feed grade wheat.

In a greenhouse study, Platford and Bernier (1976) used two isolates of *C. purpurea*, one from wheat and the other from triticale. They inoculated a number of cultivated cereals with spore suspensions at seven different concentrations, which were injected into the florets two days before anthesis. By inoculating before anthesis, any genetic resistance the host contained could be expressed before fertilization. This type of experiment also eliminates the effect flowering habit or morphology could have on infection. The differences in disease reaction were measured using sclerotial size, percentage of inoculated florets with sclerotia, and honeydew production. Inoculum concentration was found to affect only the frequency of sclerotia, not sclerotial size or honeydew production, in the species and cultivars tested. In spring wheat, they found that differences between the cultivars were best expressed at an inoculum concentration of 10^4 conidia/ml of either isolate. Barley, oat, rye, and triticale did not show significant differences in disease reaction.

A similar experiment was conducted by Menzies (2004) on currently registered Canadian wheat cultivars and experimental lines in which the reaction to artificial inoculation with *C. purpurea* was tested. In his study, a mixture of six isolates of *C. purpurea* was used to inoculate a variety of cultivars from five wheat market classes [Canadian Western Red Spring (CWRS), Canadian Prairie Spring (CPS), Canadian Western Extra Strong (CWES) Canadian Western Soft White Spring (CWSWS), and Canadian Western Amber Durum (CWAD)]. Inoculations were carried out just after the spikes emerged from the boot by injecting a spore suspension at a concentration 10^4

conidia/ml into the floral cavity of the wheat, which is similar to the protocol of Platford and Bernier (1976). Disease reaction was determined by counting the number of sclerotia per spike, and rating the size of the sclerotia and the amount of honeydew produced. The fewest number of sclerotia per spike were observed in the CWAD and CWSWS wheat and the CWES and CWRS wheat produced the greatest number of sclerotia per spike. There were no differences among market classes in regards to honeydew production. Within many of the classes, there were differences for all three measures of disease reaction among individual genotypes. One CWAD genotype (9260B-173A) had the fewest sclerotia, the lowest ratings for sclerotial size and the least amount of honeydew produced. The CWAD cultivars Kyle and Morse, however, had smaller sclerotia. Of the CPS genotypes, AC Vista and AC Taber were among the most susceptible. However, there were no differences observed in the CWES or CWSWS cultivars and only differences in honeydew production in the CWRS cultivars.

Coley-Smith and Watkinson (1987) reported ergot to be more predominant in durum than in bread wheat. They inoculated a number of cultivars from each market class with a spore suspension of 10^6 conidia/ml using an atomizer. The inoculum originated from the conidia and honeydew produced on male sterile wheat plants in the previous year. The inoculum was sprayed three times during various stages of anthesis. Once the plants matured, the spikes were removed and the numbers of sclerotia were counted. Infection was based on the percentage of infected spikes for each cultivar, which ranged from 50-97% for the durum cultivars and only 14-35% for the bread wheat cultivars over the two years of the study.

Bayles et al. (2009) tested a number of UK and European wheat cultivars and observed significant differences in resistance to ergot infection. Comparable to previous studies, resistance was measured based on the frequency of sclerotia produced (including whole and partial sclerotia), weight of sclerotia per spike, and mean weight of an individual sclerotium. Three different concentrations of inoculum were used (10^4, 10^5, and 10^6 spores/ml) for hypodermic inoculations in field conditions. With a higher inoculum rate (10^6 spores/ml), it was found that the number of sclerotia produced was not a good measure of disease reaction among cultivars, as all were infected. However, resistance was recognized based on the lower weight per sclerotium and the resulting

total weight per spike. At the lowest spore concentration (10^4 spores/ml), only very low levels of infection occurred.

2.8 Control Options

There are no control options to date that have been proven to be effective against ergot in wheat (Knox & McLeod, 2003). However, there have been some studies in other crops that have suggested that fungicide treatments may prevent ergot. These crops include Kentucky bluegrass, which is also infected by *C. purpurea* (Schultz et al., 1993; Alderman & Barker. 2003), and grain sorghum, which is infected by another *Claviceps* species, *C. africana* (McLaren, 1994; Ryley et al., 2003). Fungicides may also work in barley, but timing is critical as the fungicides do not work as eradicants (Wood & Coley-Smith, 1980). In rye, fungicidal seed treatments have been suggested to delay or suppress sclerotia germination and ascocarp formation (Dabkevičius & Mikaliūnaite, 2006).

There are some fungicides that have been suggested for use in wheat, but none that have been economical or that worked well enough to warrant their application (Bretag, 1981; Gladders et al., 2001). Gladders et al. (2001) reported mixed results for lab and greenhouse studies for use of fungicide to control ergot in rye and wheat, with some products providing sufficient control, and others not being effective at all. In the field, the fungicides did not provide acceptable control of ergot, and application of some products resulted in an increase in the number and size of the sclerotia. Since there are no reliable fungicides or resistant cultivars, management practices have been employed as the most efficient and effective way to reduce severity of ergot disease (Gladders et al., 2001).

Uniformity of the crop stand has been shown to be an important control measure (Knox & McLeod, 2003). Seeds with a high germination rate should be seeded into an even seedbed to ensure that all plants emerge at the same time. Appropriate levels of fertilizers should also be applied to ensure there are no limiting nutrients that would cause stress to the plant. These factors encourage plant development to occur uniformly, which is important for reduction of flowering time and therefore length of susceptibility. Flowering can also be affected by herbicide applications, in which poor timing or improper rates may cause sterility or damage that would set back the flowering date, once again increasing susceptibility (Knox & McLeod, 2003). Manipulating the seeding date

of the crop can also impact infection by altering the timing of flowering to allow better synchronization of early or late-maturing cultivars with pollen release (Hucl, 1996).

Using clean seed prevents the introduction of ergot into a clean field, and therefore only seed that is free from contamination should be used (Puranik & Mathre, 1971). If the seed lot contains any sclerotia, it can often be cleaned using a gravity table, which separates the grain and sclerotia based on differences in their densities (Canadian Grain Commission, 2011b). This is the most commonly used method, but new technology is emerging, with colour sorters becoming another means to separate the sclerotia and grain kernels using differences in colour. The Carter dockage tester, which uses riddles and sieves for separation, does not sufficiently remove the sclerotia from the grain (Fajardo et al., 1995).

Infection can be avoided through control of the primary and secondary inoculum (Knox & McLeod, 2003). The grasses surrounding a field are especially important in the production of secondary inoculum (honeydew), which can be transferred to neighboring plants and the cultivated crop by rain splash or insect vectors. If the ditch grasses are cut before they flower, they cannot be infected by the germinating sclerotia, and therefore will not produce the secondary inoculum that would otherwise infect the crop. This would also result in the production of fewer sclerotia and therefore a reduction in the number that would over-winter. Secondary inoculum can also be produced from weeds within the crop, which can be controlled through the application of herbicides (Mantle et al., 1977).

Controls at harvest time can be implemented to reduce the contamination of grain with ergot (Knox & McLeod, 2003). Since surrounding grasses are sources of inoculum, the headland rounds (around the outside of the field) are most susceptible to infection by these grasses. The headland round(s) that are severely infected should be harvested separately from the rest of the field. This will ease the separation process, and if the grain is too heavily contaminated, it can be discarded. The sclerotia can be destroyed by burning or burial. At a depth of at least five centimeters below the soil surface, the germination rate of sclerotia is greatly reduced (Bretag, 1981).

Sclerotia viability is reduced after one year under natural conditions, making crop rotation an important means of preventing ergot infection (Knox & McLeod, 2003). By

planting non-cereal crops in consecutive years, the risk of inoculum build-up is decreased (Rapilly, 1968). Non-host crops, such as broadleaf species, that are not susceptible to ergot are therefore a good option in a rotation (Bretag, 1987). Sclerotia viability can also be decreased by burning the cereal stubble (Bretag, 1985; Johnston et al., 1996). Bretag (1985) studied the effect of stubble burning in a field setting and found that leaving the sclerotia on the soil surface or burying them just under the trash can reduce their germination to virtually zero percent. Germination viability decreased less as the sclerotia were buried further from the soil surface, but eventually burial was deep enough to inhibit germination completely. Johnston et al. (1996) did a similar study using sclerotia collected from Kentucky bluegrass burned at various temperatures in a kiln as well as under field conditions. In the kiln it took longer for germination to cease at lower temperatures, with requirements of 15 seconds at 400ºC, 116 seconds at 200ºC, until germination was only slightly affected at 100 and 50ºC. In the field, various temperatures were also reached in open conditions and using machine burners, which showed similar results to the kiln experiment. The open field did not reach temperatures as high or burn as consistently as the machine burners. However, similar to Bretag (1985), they found that temperatures at 1 and 3 cm below the soil surface did not significantly impact the capacity to germinate.

A number of studies have examined the use of biological agents as a control measure against *Claviceps* species (Cunfer, 1975; Mower et al., 1975; Singh & Navi, 2000; Bhuiyan et al., 2003). There are commercial products registered for use in other fungal disease, such as Contans ® WG (PROPHYTA Biologischer Pflanzenschutz GmbH, Malchow/Poel, Germany), which is used to control *Sclerotinia* species. This product could potentially be tested for efficacy against *Claviceps purpurea*. Contans ® WG utilizes the fungus *Coniothyrium minitans* as a parasite to degrade sclerotia in the soil. The product can be used before planting or after harvest, and has been used in oilseed rape (*Brassica napus*), soya bean (*Glycine max*), and sunflower (*Helianthus annuus*), among others.

Another potential biological agent that has been tested on *C. purpurea* is *Fusarium heterosporum* (Cunfer, 1975). This fungal organism does not prevent infection by the pathogen, but feeds on its honeydew and prevents sclerotial formation. *Fusarium*

heterosporum proved to be successful at sclerotial inhibition starting three days prior to ergot infection until three days after honeydew appeared. *Fusarium roseum* 'Sambucinum' acted in a different way; it broke down ergotamine to inert substances that did not harm rabbits or rats after feeding them the digested substance (Mower et al., 1975).

Among other *Claviceps* species, experiments have been conducted with biological controls under both field and greenhouse conditions. In sorghum (*Sorghum bicolor*), mixed results were obtained, with two *Trichoderma* species and two isolates of *Penicillium citrinum* being effective against *Claviceps africana* macroconidial germination *in vitro*, while others failed to inhibit infection (Bhuiyan et al., 2000). In another study, *Claviceps sorghi*, another species to infect sorghum, was inhibited through the use of crude garlic extract (Singh & Navi, 2000). The garlic extract was 98-100% affective under greenhouse conditions, and approximately 90% in the field, as long as there was not an abundance of rain after application.

3.0 MATERIALS AND METHODS

3.1 Introduction

Two field experiments and one Growth Chamber Experiment were conducted to measure the disease response (incidence and severity) of wheat cultivars and market classes to ergot (*Claviceps purpurea*) infection. One of the field experiments was conducted to determine if inoculum level would affect the incidence and severity of the disease. The other field trial and growth chamber experiment were conducted to determine disease reaction among species, market classes, and cultivars.

3.2 Site Information for Field Experiments

The field experiments were carried out over two seasons (2011 and 2012) at the North Seed Farm research location at the Crop Development Center at the University of Saskatchewan in Saskatoon, Saskatchewan (NW 36 36 5 W3) on a loam soil in the moist Dark Brown soil zone. This location allowed for irrigation, in combination with a lighter soil, to lengthen the flowering period of the crop to allow for maximization of disease pressure. The previous crops in rotation at this site was barley green manure in 2010, fallow in 2009, barley in 2008, and fall rye in 2007, which was seeded in the fall of 2006. The two experiments were positioned next to each other with rye in between (Figure 3.1). In 2012 the experiments were seeded west of the 2011 site. In the summer of 2011 (July 29) perennial rye, cultivar ACE-1 (Acharya et al., 2004), was seeded in a strip next to the location of the 2012 experiments. The purpose of the rye around the experimental plots was to provide susceptible plants that flowered early and could be infected by the primary ascospores when the sclerotia germinated. Seeding on two different dates increased the risk of infection due to staggered flowering dates. Once the rye became infected it could act as a source of inoculum for the wheat plots through the spread of honeydew, allowing infection by both primary and secondary spores.

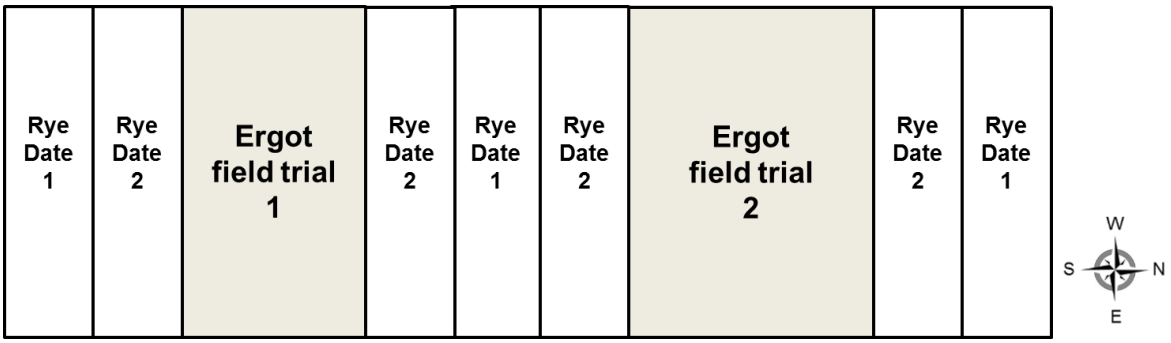

Figure 3.1. Organization of the two field experiments (same setup for 2011 and 2012), with rye spreader plots seeded at two different dates on both sides of the wheat to enhance disease pressure.

The inoculum used for both field experiments in 2011 was composed of a mixture of sclerotia from the 2009 and 2010 crop seasons. The mixture included sclerotia from a pedigreed wheat grown in east-central Saskatchewan and Gazelle rye from a breeder plot grown at Saskatoon. In 2012, the inoculum included sclerotia from 2010 and 2011, as well as a small portion of the inoculum used in 2011 (approximately 10% of the mixture). The 2011 sclerotia were obtained from the University of Saskatchewan Kernen Research Farm spring wheat and barley breeder plots (approximately 8%) as well as a small portion from intergeneric plots (*Triticum aestivum* x *Lophopyrum ponticum* F1 hybrids) grown at the Seed Farm (approximately 1%). In addition, sclerotia were obtained from the rye spreader plots from the 2011 field experiment (approximately 15%), and from rye pedigreed seed grown in north-eastern Saskatchewan (approximately 67%).

All of the sclerotia were stored at a temperature of -18°C and the different sources were thoroughly blended together prior to being spread on the field experimental plots. The mixture was divided into individual packets with the appropriate amounts weighed out for each plot.

In both years, the experimental plots were maintained using the appropriate herbicides. To control any weeds present, they were sprayed with a tank mix of Horizon ® 240EC (Syngenta Canada Inc., Guelph, ON) and Buctril M® (Bayer CropScience Inc., Calgary, AB) at the label rates, which required the inclusion of Score® Adjuvant (Syngenta Canada Inc., Guelph, ON). The herbicide was applied when the rye was at approximately Zadok stage 15-16, 22 (Zadok et al., 1974) and the wheat had not yet emerged (May 31, 2011 and May 29, 2012). On June 27, 2011, the wheat was sprayed at

the label rate with Velocity® M3 (Bayer CropScience Inc., Calgary, AB) for further weed control. Following harvest from the 2011 site, soil samples were collected on the 2011 site and 2012 site (October 6). The soil cores (30 cm deep) were taken at random throughout the experiment plot area and bulked to determine the soil characteristics (ALS Laboratory Group Analytical Chemistry and Testing Services, Saskatoon, SK). Table 3.1 summarizes the levels of micronutrients from each year and Figure 3.2 indicates the relative amounts of each micronutrient. Application of a micronutrient fertilizer was deemed unnecessary, in particular copper and boron, which as previously stated, are considered important in regards to ergot infection.

Table 3.1. Summary of micronutrient levels (kg/ha) for 2011 and 2012 ergot field experiments located at the North Seed Farm at the University of Saskatchewan.

	Micronutrient (kg/ha)*									
	NO_3^-N	P	K	SO_4^-S	Cu	Mn	Zn	B	Fe	Cl
2011	20	73	1211	27	3.6	28.0	3.3	3.3	119	18
2012	59	>135	>1345	29	2.4	24.5	4.6	4.3	104	15

* NO_3^-N = nitrate- nitrogen; P = phosphorus; K = potassium; SO_4^-S = sulfate-sulfur; Cu = copper; Mn = manganese; Zn = zinc; B = boron; Fe = iron; Cl = chloride

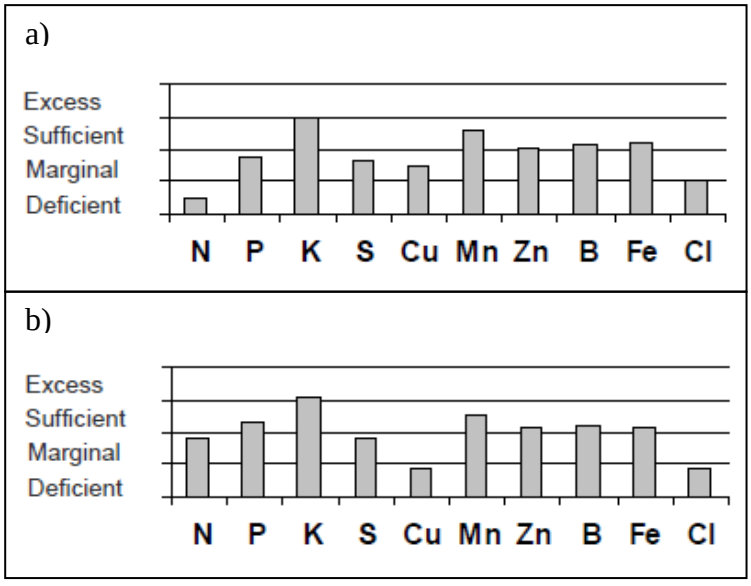

Figure 3.2. Bar graph showing adequacy of micronutrient levels, with amounts ranging from deficient to excess for the 2011 (a) and 2012 (b) field experimental sites.

3.3 Field Experiment #1 – Reaction of Cultivars to Varying Levels of Inoculum

3.3.1 Plant Material

Nine cultivars of spring wheat were used to determine if the level of inoculum in a field setting would affect infection intensity and whether or not differences in reaction would vary among cultivars. Of the nine cultivars, eight were hexaploid spring wheat, including five from the market class Canada Western Red Spring (CWRS), one from Canada Prairie Spring (CPS), one from Canada Western Hard White Spring (CWHWS), and one from Canada Western Soft White Spring (CWSWS). The ninth wheat cultivar was a tetraploid cultivar, from the market class Canada Western Amber Durum (CWAD). These market classifications and their cultivars are listed in Table 3.2. Positive controls for the experiment included 'Gazelle' spring rye (Sosulski & Curran, 1975) and 'Sandro' triticale.

Table 3.2. Wheat market classes and respective cultivars for ergot Field Experiment #1.

Canadian Wheat Market Classification	Cultivar(s)
Canada Western Red Spring (CWRS)	AC Barrie, Superb, CDC Teal, 5602HR, Harvest
Canada Prairie Spring (CPS)	5702PR
Canada Western Hard White Spring (CWHWS)	Snowstar
Canada Western Soft White Spring (CWSWS)	AC Andrew
Canada Western Amber Durum (CWAD)	AC Avonlea

3.3.2 Experimental Design

Field Experiment #1 was conducted to evaluate the effect of inoculum level on western Canadian spring growth habit wheat cultivars. In 2009 and 2010, preliminary experiments were conducted at the University of Saskatchewan Research Farm in a split-plot design where ergot treatments and controls were the main plot and the wheat cultivars were the subplots. In 2009, sclerotia added in-furrow increased the amount of sclerotia in the harvested sample by approximately 50% by weight compared to the non-inoculated control, which was statistically significant. In 2010, the sclerotia were broadcast but there were no statistical differences between the inoculum rates (0, 4 and 8

grams per plot) in terms of the amount of sclerotia in the harvested sample. However, the cultivars did show a range in sclerotia formation.

The current experiment was conducted as a split-plot design with four complete blocks. The main plots were the varying levels of inoculum and control and the sub-plots were the wheat cultivars. Each cultivar was sown using a 4-row disc seeder in a single 1.3 m long row at a rate of approximately 100 seeds per plot (approximately 77 seeds/m). Row spacing was 30.5 cm and seed depth was approximately 2.5 cm. A granular fertilizer (11-51-0) was applied at a rate of 56 kg per hectare with the seed. The plots were seeded on May 26, 2011 and on May 21, 2012.

The rates of inoculum used were based on volume of ergot equivalent to seed volume planted in each plot, which included the control (0 g), 1X (8 g) and 2X (16 g) the weight, which was equivalent to 0 g/m^2, and 20.5 g/m^2 and 41.0 g/m^2 respectively. Sclerotia were applied evenly on the soil surface on each of the rows, for the entire length of the row at the appropriate rates. The application was done by hand broadcasting the sclerotia from the pre-weighed envelopes. Inoculation of the wheat occurred when it reached Zadok stage 13-14, 21 (Zadok et al., 1974). The plots were bordered by spring rye, which was sown at early (April 28, 2011 and April 24, 2012) and late seeding dates, the latter of which coincided with plot seeding. The second seeding of rye occurred directly adjacent to the wheat plots. For each date, one seeder pass of rye was sown at a rate of 224 seeds/m^2. The rye was inoculated by hand broadcasting 80 grams of sclerotia per 3.65 meter length of the seeder pass, or 23.9 g/m^2, and acted as a spreader plot for infection. The rye was inoculated at Zadok stage 14-15, 21-22 (Zadok et al., 1974). In 2012, the perennial rye was also inoculated, which occurred at the same time as the first seeded rye. At this time the perennial rye was at approximately Zadok growth stage 37-40 (Zadok et al., 1974).

3.3.3 Data Collection

The wheat remained standing until maturity in the field, at which time all the spikes from each plot were harvested using scissors and put into brown paper bags to be dried for one to two days. The temperature in the drier averaged 35°C and the humidity

approximately 40%. After drying, the numbers of spikes per plot were counted and bulk threshed using a de-awner. The belts on the de-awner were loosened slightly so that the sclerotia were not damaged. The chaff was then separated from the grain and sclerotia using a Carter-Day dockage tester. Since the dockage tester did not separate all of the sclerotia from the grain, the sclerotia were further separated manually. After separation, each component was weighed individually and the numbers of sclerotia were counted. The sclerotia content as a percentage of the total harvested biomass (grain + sclerotia) was determined (Formula 1) as well as the mean sclerotia per spike (Formula 2) and mean weight of individual sclerotium in milligrams (Formula 3). The following three formulas show how the variables were calculated:

1) Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)
2) Sclerotia per spike = number of sclerotia / number of spikes
3) Sclerotium weight (mg) = 1000 * (weight of the sclerotia (g) / number of sclerotia)

3.3.4 Data Analyses

The field experiments were carried out in two seasons (2011 and 2012) in which all plots had data collected on an individual basis. Once all data was collected it was entered into SAS Version 9.3 (SAS Institute, 2010) and the mixed procedure was used for analysis. If any significant differences were found ($P < 0.05$), the Tukey-Kramer test was used to separate the means. Mean separation output was converted to letter groupings using a macro for SAS (Saxton, 1998).

3.4 Field Experiment #2 – Disease Reaction Differences among Wheat Species, Market Classes, and Cultivars

3.4.1 Plant Material

Ninety-two wheat cultivars were assessed for differences in reactions to *C. purpurea* (Fr.) Tul. The cultivars were chosen from a number of wheat market classes (Table 3.3). The hexaploid spring wheat (*Triticum aestivum* L.) included 55 cultivars of Canada Western Red Spring (CWRS), 9 Canada Prairie Spring (CPS), 6 Canada Western Extra Strong (CWES), 2 Canada Western Hard White Spring (CWHWS), 3 Canada Western Soft White Spring (CWSWS), and 3 Canada Western General Purpose (CWGP). Durum wheat (*Triticum turgidum* ssp. d*urum*) included 12 cultivars of Canada Western Amber Durum (CWAD). Two cultivars of spelt (*Triticum spelta*) were also tested. The positive control for this experiment was Sandro triticale, which showed infection rates great than or equal to the rye in the 2010 preliminary study.

3.4.2 Experimental Design

Field Experiment #2 was located at the North Seed Farm. The experiment was conducted as a randomized complete block design with four replications.

Each cultivar was sown in a single 1.3 m row. The seeding rate was 100 seeds per plot (approximately 77 seeds/m). The same seeder and settings were used for both ergot field experiments. The experiment was bordered by spring rye, which was seeded on two different dates, and the rye was inoculated in the same way as in Field Experiment #1. In the wheat, sclerotia were applied at a single rate of 16 grams per row, or 41.0 g/m^2. The sclerotia were hand broadcast evenly onto the soil surface within each of the rows, for the entire length of the row.

Table 3.3. Wheat market classes and respective cultivars used for Field Experiment #2 and for Growth Chamber Experiment.

*CWRS	CPS	CWES	CWHWS	CWSWS	CWGP	CWAD	Spelt
AC Barrie	AC Crystal	Glenlea	Snowbird	AC Andrew	Minnedosa	AC Morse	CDC Zorba
CDC Bounty	AC Foremost	CDC Rama	Snowstar	Bhishaj	NRG003	Kyle	CDC Origin
AC Cadillac	AC Taber	CDC Walrus		Sadash	NRG010	AC Avonlea	
AC Elsa	5700PR	Burnside				Napoleon	
Harvest	5701PR	GlencrossVB				AC Navigator	
CDC Imagine	AC Vista	CDN Bison				Strongfield	
AC Intrepid	5702PR					Commander	
Journey	AC ConquerVB					CDC Verona	
AC Abbey	SY985					Brigade	
Eatonia						Eurostar	
Lillian						Enterprise	
Lovitt						Transcend	
McKenzie							
Prodigy							
AC Splendor							
AC Superb							
CDC Teal							
5500HR							
5600HR							
5601HR							
Roblin							
Katepwa							
Marquis							
Red Fife							
CDC Merlin							
CDC Osler							
CDC Go							
CDC Alsask							
PT 559							
Infinity							
CDC Abound							
Alvena							
Goodeve VB							
Helios							
KANE							
Somerset							
Unity VB							
Waskada							
5602HR							
Fieldstar VB							
5603HR							
859CL							
Stettler							
Shaw							
Glenn							
Carberry							
Muchmore							
5604HR CL							
CDC Stanley							
CDC Kernen							
CDC Utmost							
CDC Thrive							
Vesper							
CDC VR Morris							
CDC Plentiful							

*Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES), Canada Western Hard White Spring (CWHWS), Canada Western Soft White Spring (CWSWS), Canada Western General Purpose (CWGP), Canadian Western Amber Durum (CWAD)

3.4.3 Data Collection

The data for Field Experiment #2 were collected and processed in the same way as in Field Experiment #1. See section 3.2.3 for details.

3.4.4 Data Analyses

The field experiments were carried out in two seasons (2011 and 2012). Each year there were four replications of each plot that had data collected on an individual basis. Once all data was collected it was entered into SAS Version 9.3 (SAS Institute, 2010). After preliminary analyses using the mixed procedure, the year was deemed to have a significant effect on results ($P < 0.05$), and it was decided that each of the years be analyzed separately. The *r*-value determined from the Pearson Correlation test also suggested that there was not enough correlation between years to analyze them together.

The data for all variables were tested for basic assumptions including homogeneity of variance, and normality of residual and random (replicate) effects. The distributions of the residual and replicate effects were checked for normality with the univariate procedure in SAS, where the Shapiro-Wilk statistic (W) was utilized. These effects were considered to be normally distributed when the *P*-value of W was greater than 0.05. Square root, natural logarithmic, log to base 10, and arcsin transformations were all considered.

Homogeneity of variance was tested with the Levene's test option using the GLM procedure in SAS. To accommodate the heterogeneous variances of any given variable, the data were modeled using a number of variance-covariance structures. The model with the fewest parameters and goodness-of-fit statistics closest to zero was chosen for that particular variable.

After the assumptions were tested, the MIXED procedure was used to determine significant differences among cultivar means for each variable. The Tukey-Kramer test was used to separate the means when significant. Mean separation output was converted to letter groupings using a macro for SAS (Saxton, 1998). Contrasts determined differences among selected wheat market classes, as well as among the *Triticum* species

(Table 3.4). Pearson's correlation coefficients were also calculated to determine the strength of the relationships among variables.

Table 3.4. List of contrasts for select market classes and wheat species for Field Experiment # 2.

Contrast	C*	CWRS+	CWAD	CPS	CWES	CWSWS	CWGP	CWHWS	Spelt
control vs wheat	-8	1	1	1	1	1	1	1	1
T. aestivum vs T.*turgidum* ssp. *durum*	0	1	-6	1	1	1	1	1	0
T. spelta vs T. aestivum	0	1	0	1	1	1	1	1	-6
CWRS vs CWAD	0	1	-1	0	0	0	0	0	0
CPS vs CWAD	0	0	1	-1	0	0	0	0	0
CWRS vs CPS	0	1	0	-1	0	0	0	0	0
CPS vs CWES	0	0	0	1	-1	0	0	0	0
CWRS vs CWES	0	1	0	0	-1	0	0	0	0

* C = control = triticale

+ Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES), Canada Western Hard White Spring (CWHWS), Canada Western Soft White Spring (CWSWS), Canada Western General Purpose (CWGP), Canadian Western Amber Durum (CWAD)

3.5 Growth Chamber Experiment – Disease Reaction Differences among Wheat Species, Market Classes, and Cultivars

3.5.1 Plant Material

The same ninety-two wheat cultivars examined in Field Experiment #2 were used in the Growth Chamber Experiment (see Table 3.3). The seeds were pre-germinated in the dark at room temperature (22 ± 1°C) for two days. Fifty seeds per cultivar were placed on Whatman No.1 filter paper in a 9 cm Petri dish with 2.5 ml of distilled water. The seeds were then transplanted into 15 cm diameter pots containing Sunshine #3 potting mix (Sun Gro Horticulture Canada Ltd., Seba Beach, AB). Eight seeds were sown in each pot at a depth of 2 cm and were thinned to six seedlings per pot at Zadok stage 13 (Zadok et al., 1974). The rye cultivar was sown ten days after the wheat to accommodate its earlier flowering time. The plants were placed in a growth chamber (Conviron Model GR48, Conviron, Winnipeg, MB) in a completely randomized fashion and were watered every two days (more often at flag leaf to grain filling due to higher water requirements). The temperature regime in the chamber was 21°C day/17°C night with an 18:6 hour light/dark period. A split application of Type 100 Nutricote controlled-release granular fertilizer (14-14-14) (Plant Products Co. Ltd. Brampton, ON) was applied at the 1-2 leaf and 5-6 leaf stages at a rate of approximately 0.8 kg/m^2. At the 4-leaf stage, Micromax Micronutrients granular micronutrient fertilizer (The Scotts Company, Marysville, OH) was applied at a rate of 100 g/m^2.

The experiment was replicated three times. Within each replication, there were ninety-four genotypes (ninety-two wheat cultivars plus one rye and one triticale as controls). The seeding dates were staggered to spread the work load of inoculating and rating. Genotypes were separated into two groups, the first group included forty-six wheat cultivars and the triticale, and the second group, which was seeded ten days later, included the other forty-six wheat cultivars and the rye. The seeding date of the subsequent replications occurred approximately two months after the preceding one.

3.5.2 Inoculum Preparation

(Adapted from Menzies, 2004)

Ergot inoculum for the Growth Chamber Experiment was prepared from the same source of sclerotia as that used for field inoculations. The sclerotia were all collected from similar locations, the dry and open field regions favourable to G1 populations, as recommended by Pažoutová et al. (2002) for testing differences among cultivars. The sclerotia were cultured separately and then combined for inoculation purposes.

Before plating on potato dextrose agar (PDA), the sclerotia were surface sterilized for three minutes in a buffered bleach solution. The bleach solution was prepared by diluting 20 ml of 10% KH_2PO_4 (monopotassium phosphate) and 20 ml of NaClO (sodium hypochlorite) and two drops of Tween 20 in distilled water to a total volume of 100 ml. It was recommended to boil potatoes to make fresh PDA media rather than to use commercially prepared PDA, but both were used and were equally successful. When making fresh PDA media, the potatoes should be white, as red-skinned potatoes contain toxins that may impact fungal growth. The sclerotia were cut aseptically into two to four pieces (depending on size) and placed onto the PDA plates (Figure 3.3 a). The plates were kept at room temperature (22 ± 1°C) under a light regime of 16:8 hours light:dark to allow mycelial growth. After sufficient growth (approximately 3.5-5 cm in diameter) a plug from each of the cultures was re-isolated onto new PDA (Figure 3.3 b). Cultures were selected based on the rate of growth and purity of white of the mycelium, with the fastest growing and whitest cultures being chosen. Any plates with poor growth or contamination were discarded.

A potato-sucrose broth (PSB) was prepared as a liquid medium to further increase the isolates. First, 400 g of white-skinned potatoes, cultivar Russet Norkotah (Canadian Food Inspection Agency, 2009), were autoclaved in 600 ml of water followed by filtration of the mixture through cheesecloth. The amount of liquid produced from the autoclaved water and potatoes was approximately 500 ml, to which 20 g of sucrose was added and water to 1 L. This was now considered 2% PSB, and 125 ml was added to six Erlenmeyer flasks (500 ml) to be autoclaved. Six *C. purpurea* isolates were chosen from a number of plates that originated from a range in size of sclerotia. One plug from each plate was added to each of the six flasks and allowed to incubate for two weeks at room

temperature on a rotary shaker at 125 rpm (Figure 3.3 c). The plates and the liquid cultures were grown under the same light regime and temperature as the PDA plates (16:8 hours light:dark at 22 ± 1°C).

The liquid cultures were incubated on the rotary shaker for two weeks. Cheesecloth was used to filter each individual culture into a 500 ml Erlenmeyer flask (Figure 3.3 d). The cheesecloth was placed into a small funnel in the opening of the Erlenmeyer flask, covered with foil, autoclaved for 20 minutes and cooled before using. After filtration, the liquid containing the fungal spores was divided into two, 50 ml centrifuge tubes. The tubes were centrifuged at 2500 g for 15 minutes. The supernatant was decanted from the centrifuge tubes and sterile distilled water was added to one of the tubes of each isolate. A vortex mixer was used to re-suspend the pellet before transferring the liquid to the second tube of the same isolate. This was done twice and resulted in one tube per isolate. Each tube was mixed thoroughly on the vortex mixer before a second centrifuge cycle was performed using the same conditions as in the first cycle. The supernatant was decanted again and the final pellet was suspended in 20 ml of sterile 60% sucrose solution and stored at 5°C. The sucrose solution was prepared by dissolving 170 g of sucrose in 200 ml of warm distilled water, followed by vacuum sterilization through a 0.2 µm vacuum filtration system. The spore suspension was usable for approximately two months.

A hemocytometer was used to count the number of spores per ml, which was done using a dilution of 10 µl of the spore suspension in 90 µl of distilled water. Ten counts were completed under 400x magnification and then averaged (two times five squares of the hemocytometer). After the average was determined, the number was multiplied by twenty-five for the total number of squares of the hemocytometer, then by ten for the dilution, and by 10,000 to get the number of spores per ml. Some isolates had extremely high spore counts, so a 1:10 dilution was carried out and this solution was used in the spore mixture for inoculating. The final spore suspension used for inoculations consisted of equal parts of each isolate for a total 10,000 spores/ml.

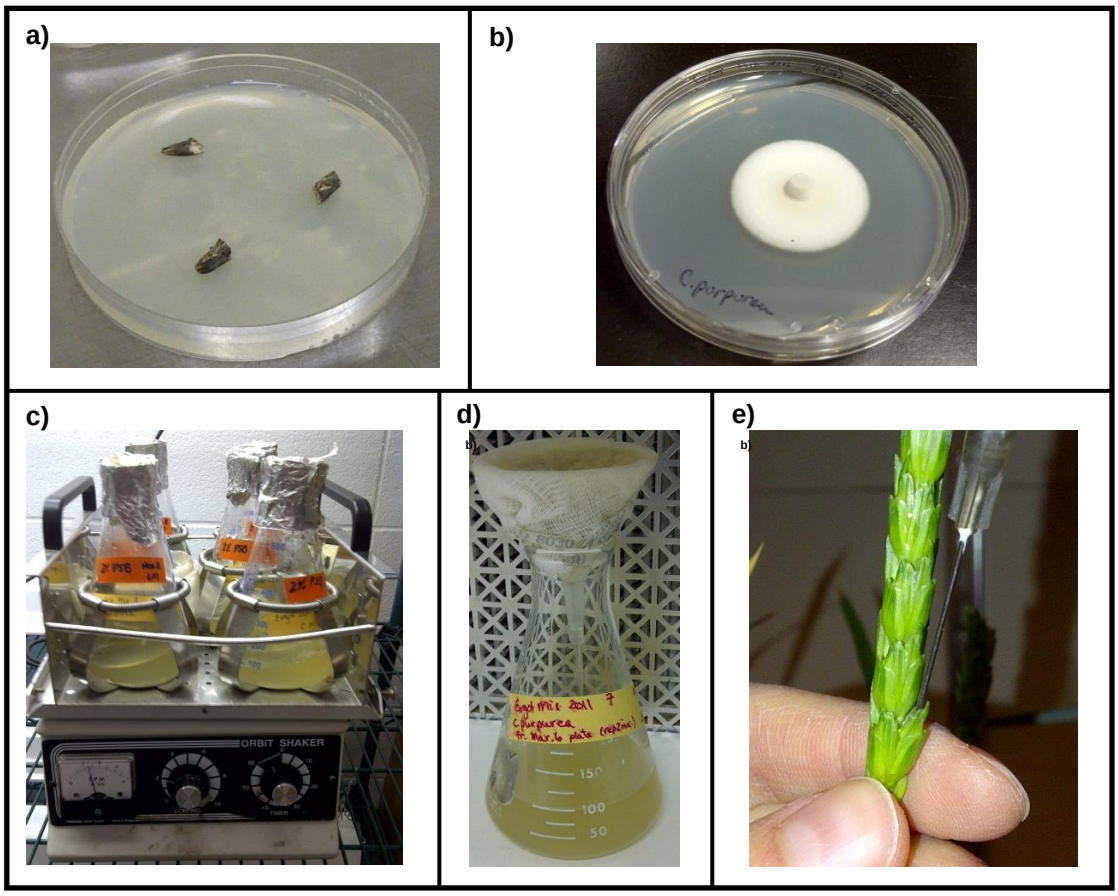

Figure 3.3. Steps for inoculation procedure; a) sclerotia in pieces on potato dextrose agar, b) *C. purpurea* mycelium on PDA, c) *C. purpurea* in liquid potato sucrose broth on rotary shaker d) separation of spores from PSB, e) injection of spore suspension into wheat floret between lemma and palea.

Since the inoculum was viable for only two months, and the inoculations spanned over approximately six months, new cultures were prepared for each replication. Fresh isolates of *C. purpurea* were grown on the PDA and in 2% PSB and subsequent spore suspensions were prepared when needed. The PDA plates containing the mycelia were stored in the fridge and warmed to room temperature to continue isolations, which could be used for further growth in the liquid medium.

3.5.3 Inoculation

Currently there are no known races of *C. purpurea*, but isolates vary in their degree of pathogenicity (Darlington et al., 1977). Because of this, it is recommended that inoculations be made with a mixture of isolates. In this study, six isolates were selected, but only five were used due to poor sporulation of one isolate. This occurred in the first replication, so in the following replications the same five isolates were used. The isolates were combined in equal concentrations in distilled water to establish a final concentration of 10^4 conidia per ml. Since 250 ml of water were used, 500,000 spores from each isolate were needed ($2.5x10^6$ spores total divided by five isolates). Over the period of inoculations, a fresh mixture of conidia was prepared every four days to ensure an adequate number of active spores.

Inoculations were performed when the spikes completely emerged from the boot, but before they reached anthesis (Menzies, 2004). Twenty florets per spike were inoculated, in which five healthy spikelets were selected from each side of the spike. Awned cultivars had their awns clipped prior to inoculation, and all cultivars had the basal and apical spikelets removed from the spikes. Any additional spikelets were removed so that only twenty florets developed on each spike. Furthermore, the middle floret(s) was removed from each spikelet, leaving the two outermost florets to be inoculated with the spore suspension. The suspension was injected between the lemma and palea using a syringe and hypodermic needle (Figure 3.3 e).

3.5.4 Data Collection

Collection of data from the Growth Chamber Experiment was slightly different from that in the field experiments. The first assessment of ergot disease reaction took place twelve to thirteen days after inoculation. The production of honeydew was rated on a scale of 1 to 5 (Figure 3.4), where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike (modified from Menzies, 2004). The remaining data collection occurred once the plants reached maturity. Each individual spike was harvested, dried for two to three days, and hand threshed using a bicycle inner tube.

Sclerotial size was rated relative to kernel size, which was based on a 1 to 3 scale, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel, extending beyond the lemma and palea (Figure 3.5).

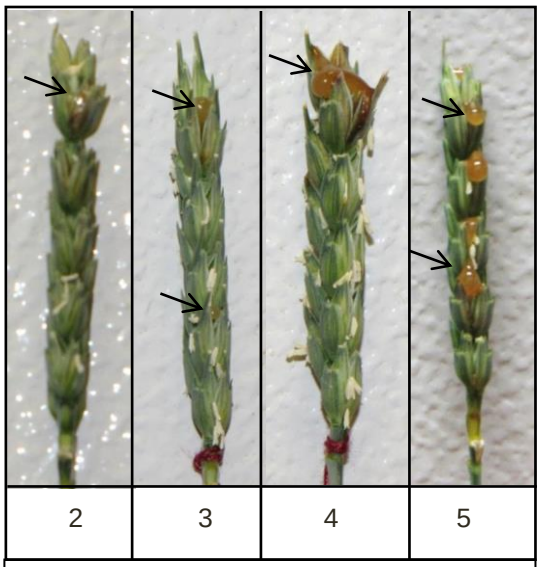

Figure 3.4. Honeydew production rating scale, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike.

Figure 3.5. Sclerotial size rating scale, where 1 = smaller than kernel, 2 = same size as kernel, and 3 = larger than kernel.

The number of sclerotia from each individual spike was counted and the sclerotia were bulked (one sample per pot) and weighed. The number of kernels per pot was also counted and weighed. The percent sclerotia by weight and average sclerotium weight

(mg) were calculated in the same manner as the field experiments (Formulas 1 and 3, respectively).

The number of florets aborted (those not completely formed into kernels or sclerotia) were determined using the total counts (kernels and sclerotia) subtracted from the total number of inoculated florets (120 per pot), and was expressed as a percent of the total. Some pots had fewer than 120 inoculated florets, which was accounted for in the calculations.

3.5.5 Data Analyses

In the Growth Chamber Experiment, there was one pot with six spikes for each replication, each of which had individual data collected and then averaged (one pot of six plants = one experimental unit). After all three replications were completed the data were pooled for statistical analysis in SAS Version 9.3 (SAS Institute, 2010).

The data for all variables were tested for basic assumptions including homogeneity of variance and normality of residual and random (replicate) effects. The distributions of the residual and replicate effects were checked for normality with the univariate procedure in SAS, where the Shapiro-Wilk statistic (W) was utilized. These effects were considered to be normally distributed when the *P*-value of W was greater than 0.05.

Homogeneity of variance was tested in SAS with the Levene's test option using the GLM procedure. To accommodate for variances that were not homogenous, the data were modeled using a number of variance-covariance structures. The model in which the fewest parameters and goodness-of-fit statistics closest to zero was chosen to be used for that particular variable.

After the assumptions were tested, the MIXED procedure was used to determine if there were any significant differences among cultivars. When significant differences were detected ($P < 0.05$), the Tukey-Kramer test was used to separate the means. Mean separation output was converted to letter groupings using a macro for SAS (Saxton, 1998). Contrasts were performed to determine differences among selected wheat market classes, as well as among the different *Triticum* species (Tables 3.5). Pearson's

correlation coefficients were also calculated to determine if any of the variables were related.

Table 3.5. List of contrasts for select market classes and wheat species for Growth Chamber Experiment.

Comparison	C1*	C2	CWRS+	CWAD	CPS	CWES	CWSWS	CWGP	CWHWS	Spelt
control vs wheat	-4	-4	1	1	1	1	1	1	1	1
T. aestivum vs *T. durum*	0	0	1	-6	1	1	1	1	1	0
T. spelta vs *T. aestivum*	0	0	1	0	1	1	1	1	1	-6
CWRS vs CWAD	0	0	1	-1	0	0	0	0	0	0
CPS vs CWAD	0	0	0	1	-1	0	0	0	0	0
CWAD vs CWES	0	0	0	1	0	-1	0	0	0	0
CWRS vs CPS	0	0	1	0	-1	0	0	0	0	0
CPS vs CWES	0	0	0	0	1	-1	0	0	0	0
CWRS vs CWES	0	0	1	0	0	-1	0	0	0	0

*C1 and C2 = controls = rye and triticale
+ Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES), Canada Western Hard White Spring (CWHWS), Canada Western Soft White Spring (CWSWS), Canada Western General Purpose (CWGP), Canadian Western Amber Durum (CWAD)

3.6 Additional Data Analyses

3.6.1 Pearson Correlations Between Experiments

The means of the common variables from the Growth Chamber Experiment and Field Experiment #2 were calculated, which included percent of sclerotia by weight, number of sclerotia per spike, and average sclerotium weight. The rye and triticale were not included in the means, as the comparisons were made solely among wheat cultivars. Pearson correlations were calculated using cultivar means among environments for the three aforementioned variables, which were considered significant if the *P*-value was less than 0.05.

4.0 RESULTS

4.1 Field Disease Reaction Evaluation

4.1.1 Reaction of Cultivars to Varying Levels of Inoculum

The effect of inoculum level in a field setting was tested over two seasons using nine wheat cultivars. The data was analyzed combined over years and as separate years. The mean of each inoculum level is presented in Table 4.1. The results from the type 3 tests of fixed effects of the mixed procedure are presented in the Table 4.2. At the 5% significance level the rates were not significantly different for any of the variables measured ($P > 0.05$) with the exception of the sclerotium weight ($P < 0.05$). However, upon comparison using the Tukey-Kramer test there was no separation among the three inoculum levels for the sclerotium weight.

Since there were no differences among inoculum levels, the means and ranges of each of the three variables are presented for the entire data set, regardless of inoculum level, in Table 4.3. The mean for percent sclerotia for both years of the wheat was 1.30, and ranged from 0.10 to 10.17. For the sclerotia per spike, the mean was less than one, but ranged from 0.03 to 2.56. Sclerotium weight also varied greatly, with a mean of 26.99, and a range of 5.71 to 61.62 mg. The values of each variable were higher for the control, with a mean of 13.49% sclerotia by weight, 3.53 sclerotia per spike, and 38.79 mg per sclerotium. The ranges were also greater for the control than the wheat, with 3.06 to 31.76%, 1.3 to 9.71, and 12.84 to 78.82 mg for percent sclerotia by weight, number of sclerotia per spike, and sclerotium weight, respectively.

The means were higher in 2011 compared to 2012 (Table 4.3), which could be due to the timing of precipitation and/or temperature patterns. Regardless of differences between years, there were differences among cultivars in both seasons ($P < 0.05$) without considering the inoculum level (Table 4.2). These results were not surprising as the variables had large ranges in values. However, this was not the objective of Field Experiment #1, and a wider range of cultivars were examined in the Field Experiment #2.

When the interaction between wheat cultivar and inoculum rate was examined, there was no significant effect ($P > 0.05$), indicating that there was no preferential reaction from one cultivar to another to inoculum rate and the resulting disease reaction.

Table 4.1. Disease reaction mean scores and standard error (SE) by rate for % sclerotia, sclerotia/spike, and wt/sclerotium for all wheat cultivars and controls in 2011 and 2012 individually and combined (grown at the North Seed Farm, Saskatoon, SK).

			Triticum spp.		Controls (Rye + Triticale)	
	Rate	Variable	Mean(±SE)	Range	Mean (±SE)	Range
2011	0 g	% Scl [a]	1.77 (± 0.294)	0.41 - 10.17	13.58 (± 1.815)	6.19 - 19.94
		Sclerotia/spike [b]	0.47 (± 0.080)	0.08 - 2.56	3.58 (± 0.533)	1.50 - 5.52
		Wt/sclerotium [c]	34.27 (± 1.221)	18.14 - 46.05	50.06 (± 6.680)	26.68 - 74.33
	8 g	% Scl	1.88 (± 0.320)	0.39 - 8.72	14.07 (± 2.075)	7.97 - 22.75
		Sclerotia/spike	0.47 (± 0.081)	0.09 - 2.46	4.05 (± 0.705)	2.06 - 7.94
		Wt/sclerotium	34.74 (± 1.484)	17.25 - 53.03	46.40 (± 6.526)	26.42 - 70.44
	16 g	% Scl	1.69 (± 0.188)	0.40 - 5.40	15.23 (± 2.692)	6.46 - 24.30
		Sclerotia/spike	0.40 (± 0.053)	0.10 - 1.44	3.98 (± 0.783)	1.57 - 7.01
		Wt/sclerotium	38.28 (± 1.676)	23.13 - 61.62	49.62 (± 6.538)	29.87 - 78.82
2012	0 g	% Scl	0.94 (± 0.236)	0.10 - 7.69	13.76 (± 3.251)	4.64 - 27.99
		Sclerotia/spike	0.26 (± 0.046)	0.05 - 1.54	3.32 (± 0.543)	2.01 - 6.40
		Wt/sclerotium	17.99 (± 0.960)	5.71 - 29.50	29.31 (± 4.405)	12.84 - 48.74
	8 g	% Scl	0.78 (± 0.181)	0.14 - 6.20	12.68 (± 3.500)	3.70 - 31.76
		Sclerotia/spike	0.23 (± 0.037)	0.03 - 1.21	3.54 (± 0.989)	1.47 - 9.71
		Wt/sclerotium	18.22 (± 1.037)	7.89 - 33.38	27.93 (± 3.950)	15.21 - 43.58
	16 g	% Scl	0.74 (± 0.125)	0.10 - 3.31	11.60 (± 2.870)	3.06 - 24.79
		Sclerotia/spike	0.21 (± 0.027)	0.03 - 0.74	2.72 (± 0.432)	1.30 - 4.68
		Wt/sclerotium	18.46 (± 1.012)	8.15 - 33.97	29.46 (± 4.990)	14.16 - 46.44
Overall	0 g	% Scl	1.35 (± 0.194)	0.10 - 10.17	13.67 (± 1.799)	4.64 - 27.99
		Sclerotia/spike	0.36 (± 0.048)	0.05 - 2.56	3.45 (± 0.369)	1.50 - 6.40
		Wt/sclerotium	26.13 (± 1.236)	5.71 - 46.05	39.69 (± 4.702)	12.84 - 74.33
	8 g	% Scl	1.33 (± 0.194)	0.14 - 8.72	13.37 (± 1.974)	3.70 - 31.76
		Sclerotia/spike	0.35 (± 0.047)	0.03 - 2.46	3.79 (± 0.590)	1.47 - 9.71
		Wt/sclerotium	26.48 (± 1.330)	7.89 - 53.03	37.16 (± 4.389)	15.21 - 70.44
	16 g	% Scl	1.22 (± 0.126)	0.10 - 5.40	13.42 (± 1.957)	3.06 - 24.79
		Sclerotia/spike	0.30 (± 0.032)	0.03 - 1.44	3.35 (± 0.461)	1.30 - 7.01
		Wt/sclerotium	28.37 (± 1.526)	8.15 - 61.62	39.54 (± 4.750)	14.16 - 78.82

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)

[b] Sclerotia/spike = number of sclerotia / number of spikes

[c] Wt/sclerotium (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

Table 4.2. Type 3 tests of fixed effects for % sclerotia, sclerotia/spike, and wt/sclerotium for all wheat cultivars and controls in 2011 and 2012 individually and combined (grown at the North Seed Farm, Saskatoon, SK) with asterisks indicated significance level.

		2011	2012	Both years
	Effect	**F-Value**	**F-Value**	**F-Value**
Percent sclerotia by weight	rate	0.47	0.81	0.61
	cultivar	37.08***	21.28***	51.93***
	rate*cultivar	1.15	0.62	1.01
Number of sclerotia per spike	rate	0.97	1.48	1.48
	cultivar	31.38***	19.84***	38.86***
	rate*cultivar	0.55	0.87	0.61
Weight per sclerotium	rate	3.89*	0.11	3.07*
	cultivar	9.86***	13.97***	17.65***
	rate*cultivar	1.20	1.45	1.27

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

Table 4.3. Disease reaction mean scores and standard error (SE) for % sclerotia, sclerotia/spike, and wt/sclerotium for all wheat cultivars and controls in 2011 and 2012 individually and combined (grown at the North Seed Farm, Saskatoon, SK).

		***Triticum* spp.**		**Controls (Rye + Triticale)**	
		Mean (± SE)	**Range**	**Mean** (± SE)	**Range**
Overall	Percent Sclerotia [a]	1.30 (±0.100)	0.1-10.17	13.49 (± 1.080)	3.06-31.76
	Sclerotia / Spike [b]	0.34 (±0.024)	0.03-2.56	3.53 (± 0.274)	1.3-9.71
	Weight / Sclerotium [c]	26.99 (±0.790)	5.71-61.62	38.79 (± 2.613)	12.84-78.82
2011	Percent Sclerotia	1.78 (± 0.156)	0.39-10.17	14.29 (± 1.236)	6.19-24.30
	Sclerotia / Spike	0.45 (± 0.042)	0.08-2.56	3.87 (± 0.378)	1.50-7.94
	Weight / Sclerotium	35.76 (± 0.860)	17.25-61.62	48.69 (± 3.647)	26.42-78.82
2012	Percent Sclerotia	0.82 (± 0.107)	0.10-7.69	12.68 (± 1.784)	3.06-31.76
	Sclerotia / Spike	0.23 (± 0.022)	0.03-1.54	3.19 (± 0.391)	1.30-9.71
	Weight / Sclerotium	18.22 (± 0.574)	5.71-33.97	28.90 (± 2.469)	12.84-48.74

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)

[b] Sclerotia/spike = number of sclerotia / number of spikes

[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

4.1.2 Disease Reaction of Wheat Species and Market Classes

The market classes were compared using non-modeled data. Select comparisons were carried out in SAS using orthogonal contrasts within the mixed procedure for percent sclerotia by weight, number of sclerotia per spike, and average sclerotium weight. Table 4.4 shows the means for each of the market classes tested and Table 4.5 shows the

results of the contrasts for each year, with asterisks indicating the significance level of the comparison.

Table 4.4. Disease reaction mean scores for % sclerotia, sclerotia/spike, and wt/sclerotium for all wheat market classes and spelt in 2011 and 2012 (grown at the North Seed Farm, Saskatoon, SK).

		Percent Sclerotia [a]		Sclerotia / Spike [b]		Weight / Sclerotium [c]	
Market Class[+]	n	2011	2012	2011	2012	2011	2012
CWES	6	1.90	1.22	0.56	0.38	38.31	27.37
CPS	9	1.37	0.70	0.43	0.24	31.82	20.78
CWAD	12	1.05	0.51	0.49	0.29	25.36	13.43
CWRS	55	0.77	0.49	0.18	0.13	32.17	19.49
CWHWS	2	0.52	0.51	0.13	0.15	30.79	18.31
CWGP	3	0.50	0.40	0.18	0.15	33.34	22.81
CWSWS	3	0.39	0.27	0.34	0.20	15.75	14.71
Spelt	2	0.36	0.13	0.08	0.06	41.61	16.85

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)

[b] Sclerotia/spike = number of sclerotia / number of spikes

[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

+ Canada Western Extra Strong (CWES), Canada Prairie Spring (CPS), Canadian Western Amber Durum (CWAD), Canadian Western Red Spring (CWRS), Canada Western Hard White Spring (CWHWS), Canada Western General Purpose (CWGP), Canada Western Soft White Spring (CWSWS)

Table 4.5. Summary of contrast results for % sclerotia, sclerotia/spike, and wt/sclerotium for species and selected market classes in 2011 and 2012 (grown at the North Seed Farm, Saskatoon, SK).

	Percent Sclerotia [a]		Sclerotia / Spike [b]		Weight / Sclerotium [c]	
Comparison[+]	2011	2012	2011	2012	2011	2012
Triticale vs *Triticum* spp.	***	***	***	***	***	***
T. aestivum vs *T. t.* ssp. *durum*	NS	NS	***	NS	**	***
T. aestivum vs *T. spelta*	NS	NS	*	NS	***	NS
CWRS vs CWAD	NS	NS	***	***	***	***
CPS vs CWAD	NS	NS	NS	NS	***	***
CWES vs CWRS	***	***	***	***	**	***
CWES vs CPS	*	*	*	NS	**	**
CPS vs CWRS	***	NS	***	*	NS	NS

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; NS = not significant = $P > 0.05$

+ Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES)

In both years, for all three variables, the control (triticale) had significantly greater values than the combined average of all wheat cultivars ($P < 0.001$). This was to be expected as triticale has been shown in previous studies to be highly susceptible to infection by *C. purpurea*.

The comparison between species did not show any differences for percent sclerotia by weight, but there were significant differences within species (*T. aestivum*). Results varied for the number of sclerotia per spike, and the weight per sclerotium showed significant differences in almost all cases.

Weight per sclerotium is the only variable in both years with differences between wheat species, *T. aestivum* and *T. turgidum* ssp. d*urum (*$P < 0.01$ and < 0.001 in 2011 and 2012, respectively). In both of these years, *T. aestivum* had sclerotia with greater weight than *T. turgidum* ssp. d*urum*. In 2011 there were also differences in the number of sclerotia per spike ($P < 0.001$); there were more in *T. turgidum* ssp. d*urum* than *T. aestivum*. Neither year showed significant differences between species for the percent sclerotia by weight ($P > 0.05$). When comparing species, *T. aestivum* and *T. spelta*, there were no significant differences in 2012 for percent sclerotia by weight ($P > 0.05$). In 2011 there were significantly more sclerotia per spike for *T. aestivum* ($P < 0.05$) but a greater weight per sclerotium for *T. spelta* ($P < 0.001$).

When further dividing *T. aestivum* into market classes to compare with the *T. turgidum* ssp. *durum* market class (CWAD), there were subtle differences found compared to results for the collective average. CWAD was greater than CWRS in both years for the number of sclerotia per spike ($P < 0.001$), but CWRS was greater for sclerotium weight ($P < 0.001$). The sclerotium weight was greater in both years for CPS compared to CWAD ($P < 0.001$), but there were no differences between the numbers of sclerotia per spike ($P > 0.05$). Similar to the collective average of *T. aestivum*, neither CWRS nor CPS had percentage values different from the CWAD market class ($P > 0.05$).

The three major market classes within *T. aestivum* were also compared. The largest difference was between CWES and CWRS, where the values for all three variables were greater for CWES ($P < 0.001$ for all variables in both years, except 2011 sclerotium weight, in which $P < 0.01$). CWES was also greater than CPS for these variables, but the differences were less than between CWES and CWRS ($P < 0.05$). The

number of sclerotia per spike in 2012 was not different between CWES and CPS ($P > 0.05$). Sclerotium weight in both years also showed CWES to be significantly greater for this comparison ($P < 0.01$). When comparing CPS to CWRS however, there were no differences in either year for sclerotium weight ($P > 0.05$). Percent sclerotia by weight was only greater for CPS over CWRS in 2011 ($P < 0.001$), but both years showed CPS to have more sclerotia per spike compared to CWRS ($P < 0.001$ and < 0.05 for 2011 and 2012, respectively).

4.1.3 Disease Reaction of Wheat Cultivars

When inoculated with *Claviceps purpurea*, spring wheat cultivars exhibited a range of reactions. After testing each variable for residual and replicate normality, it was determined that there was no benefit to transforming the data and therefore the analyses were on non-transformed data. Using the mixed procedure in SAS, the mean disease reaction scores for each of the three variables, which included the percent sclerotia by weight, the average number of sclerotia per spike, and the average sclerotium weight, were calculated for the wheat cultivars grown at the North Seed Farm. These results are presented for the ninety-two cultivars in each year in Appendix A. However, since the means of the cultivars had heterogeneous variances, the data were modeled to be compared appropriately. These results are presented in Appendix C, which contains the new estimates and their standard errors. Differences among means are indicated by letters; means followed by the same letter are not different. The F-values for each variable are presented in Table 4.6, which are based on the modeled data. For all three variables, there were significant differences among cultivars ($P < 0.001$). The cultivar means before modeling (Table 4.7) indicate a range of values and therefore likely significant differences among cultivars. The means were generally higher in 2011 than 2012, but mean values of the control remained very similar between years.

Table 4.6. F-values from Type 3 tests of fixed effects using modeled data for cultivar differences for % sclerotia, sclerotia/spike, and wt/sclerotium (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012).

	F-Value	
	2011	**2012**
Percent Sclerotia [a]	320.35***	3.29***
Sclerotia / Spike [b]	7.38***	3.04***
Weight / Sclerotium [c]	1.99***	2.35***

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
*** $P < 0.001$

Table 4.7. Disease reaction mean scores for % sclerotia, sclerotia/spike, and wt/sclerotium for all 92 wheat genotypes and control for 2011 and 2012 individually and combined, including standard error (SE) and range (North Seed Farm, Saskatoon, SK).

		Percent Sclerotia [a]		**Sclerotia / Spike** [b]		**Weight / Sclerotium** [c]	
	Year	Mean (± SE)	Range	Mean (± SE)	Range	Mean (± SE)	Range
O[x]	2011	1.08(±0.098)	0.01 - 24.61	0.31(±0.026)	0.00 - 6.55	31.82(±0.555)	8.62 - 96.52
	2012	0.71(±0.097)	0.00 - 25.22	0.23(±0.030)	0.00 - 8.33	19.46(±0.437)	3.33 - 64.62
W[y]	2011	0.90(±0.045)	0.01 - 6.70	0.27(±0.013)	0.00 - 1.43	31.33(±0.495)	8.62 - 71.82
	2012	0.54(±0.037)	0.00 - 7.19	0.18(±0.011)	0.00 - 1.95	19.26(±0.429)	3.33 - 64.62
C[z]	2011	16.55(±2.691)	13.54 - 24.61	4.25(±0.781)	3.21 - 6.55	77.22(±9.331)	60.07 - 96.52
	2012	16.42(±3.047)	11.20 - 25.22	4.92(±1.161)	3.26 - 8.33	38.20(±3.238)	34.14 - 47.86

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[x] O = overall (all cultivars of wheat and control)
[y] W = all wheat cultivars averaged
[z] C = control, Sandro triticale

Once the data were modeled, the Tukey-Kramer test was used to rank the ninety-two cultivars from highest to lowest for each variable, and a macro assigned letters to indicate significant differences (Saxton, 1998). In many cases, there was no difference among cultivars, indicated by common letters. Tables 4.8 through 4.13 contain the highest and lowest ten cultivars for the 2011 and 2012 field experiments.

In 2011, the CWES cultivar Glenlea ranked as the cultivar with the highest percent sclerotia by weight (Table 4.8). Similar results were observed in 2012 for Glenlea, but Marquis was also ranked as one of the cultivars with the highest percent sclerotia by weight. There was a greater separation among cultivars in 2011 compared to 2012 for percent sclerotia. The distinction within the first ten cultivars was clear, with SY985, 5702PR, CDN Bison, and Burnside having the highest means. These cultivars are

all within the CWES or CPS class and ranged from approximately 2.3-2.6 % sclerotia by weight. The remaining cultivars were from the CWRS and CWAD classes. In 2012, the cultivars ranked from 2-20 were all statistically the same, and the remaining 72 cultivars had no significant differences among them. These results indicate that there are very few differences for percent sclerotia by weight. The ten cultivars with the lowest mean percent sclerotia in 2011 were also very similar, all with less than 0.5%sclerotia by weight (Table 4.9).

When both years were examined together, there were some further commonalities. The cultivars 5702PR, Red Fife, AC Navigator, and 5601HR had the highest percent sclerotia by weight while CDC Utmost, Vesper, Stettler, AC Splendor, Prodigy, and Waskada had the least.

Table 4.8. Ten cultivars with the highest mean percent sclerotia by weight (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 0.104)		Market Class	Cultivar	Actual	Estimate (± 0.331)	
CWES	Glenlea	3.30	3.32	A [z]	CWES	Glenlea	3.57	3.93	A
CPS	SY985	2.55	2.60	B	CWRS	Marquis	2.40	2.72	AB
CPS	5702PR	3.22	2.52	BC	CWRS	5601HR	2.03	1.86	BC
CWES	CDN Bison	2.28	2.41	BCD	CWAD	Brigade	1.20	1.50	BC
CWES	Burnside	2.70	2.30	CDE	CWRS	CDC Imagine	1.36	1.48	BC
CWRS	Red Fife	2.47	2.29	CDE	CWRS	Red Fife	1.76	1.46	BC
CWAD	AC Navigator	2.25	2.28	DE	CWRS	CDC Stanley	1.25	1.45	BC
CWRS	CDC Merlin	2.18	2.15	EF	CPS	5702PR	1.97	1.44	BC
CWRS	5601HR	2.23	2.02	F	CWRS	Somerset	1.39	1.27	BC
CWAD	Enterprise	2.09	1.95	FG	CWAD	AC Navigator	1.31	1.24	BC

[z] Means with different letters are considered to be significantly different from each other

+ Canada Western Extra Strong (CWES), Canada Prairie Spring (CPS), Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD)

Table 4.9. Ten cultivars with the lowest mean percent sclerotia by weight (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 0.104)		Market Class	Cultivar	Actual	Estimate (± 0.331)	
CWRS	CDC Plentiful	0.15	0.35	(2)IJKLMNO [z]	CWRS	Stettler	0.08	0.10	C
CWAD	Commander	0.36	0.33	(2)IJKLMNO	Spelt	CDC Zorba	0.08	0.10	C
CWRS	5604HRCL	0.14	0.33	(2)JKLMNO	CWRS	CDC Utmost	0.05	0.09	C
CWRS	Waskada	0.25	0.31	(2)JKLMNO	CWRS	Fieldstar VB	0.08	0.09	C
CPS	AC Crystal	0.54	0.30	(2)JKLMNO	CWRS	AC Splendor	0.05	0.06	C
CWRS	Prodigy	0.19	0.30	(2)KLMNO	CWRS	Waskada	0.08	0.05	C
CWRS	AC Splendor	0.15	0.29	(2)LMNO	CWRS	Prodigy	0.04	0.04	C
CWRS	Stettler	0.25	0.27	(2)MNO	CWSWS	Bhishaj	0.02	0.04	C
CWRS	Vesper	0.05	0.23	(2)NO	CWRS	AC Abbey	0.11	0.04	C
CWRS	CDC Utmost	0.10	0.21	(2)O	CWRS	Vesper	0.02	0.03	C

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD), Canada Prairie Spring (CPS), Canada Western Soft White Spring (CWSWS)

Table 4.10. Ten cultivars with the highest mean number sclerotia per spike (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 0.078)		Market Class	Cultivar	Actual	Estimate (± 0.089)	
CWES	Glenlea	0.95	0.90	A [z]	CWES	Glenlea	1.00	0.79	A
CWAD	Enterprise	0.89	0.83	AB	CWES	Burnside	0.63	0.65	AB
CPS	5702PR	0.88	0.78	ABC	CWAD	Brigade	0.66	0.60	ABC
CPS	SY985	0.69	0.78	ABC	CWRS	Red Fife	0.62	0.56	ABCD
CWES	Burnside	0.81	0.77	ABC	CWRS	Marquis	0.63	0.55	ABCDE
CWRS	Red Fife	0.73	0.75	ABCD	CWAD	AC Navigator	0.53	0.54	ABCDEF
CWAD	AC Navigator	0.75	0.75	ABCD	CWSWS	Sadash	0.40	0.45	ABCDEF
CWES	CDN Bison	0.62	0.61	ABCDE	CPS	5702PR	0.57	0.41	ABCDEF
CWAD	CDC Verona	0.52	0.60	ABCDEF	CPS	AC Taber	0.38	0.39	ABCDEF
CWAD	Napoleon	0.57	0.56	ABCDEFG	CWRS	5601HR	0.45	0.35	ABCDEF

[z] Means with different letters are considered to be significantly different from each other
+ Canada Western Extra Strong (CWES), Canadian Western Amber Durum (CWAD), Canada Prairie Spring (CPS), Canadian Western Red Spring (CWRS), Canada Western Soft White Spring (CWSWS)

In 2011 and 2012, Glenlea was among the cultivars with the highest mean sclerotia per spike (Table 4.10), but there was less separation between it and the other cultivars compared to percent sclerotia. The cultivars 5702PR, Burnside, Red Fife, and AC Navigator had the most sclerotia per spike in both years. Vesper, CDC Utmost, AC

Splendor, Alvena, Waskada, and Prodigy were among the cultivars with the fewest number of sclerotia per spike in both years (Table 4.11).

Table 4.11. Ten cultivars with the lowest mean number sclerotia per spike (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 0.078)		Market Class	Cultivar	Actual	Estimate (± 0.089)	
CWRS	Prodigy	0.08	0.09	HI [z]	CWRS	Fieldstar VB	0.04	0.06	DEF
CWRS	KANE	0.08	0.09	HI	CWRS	CDC Utmost	0.03	0.05	DEF
CWRS	CDC Go	0.09	0.09	HI	CWRS	Alvena	0.04	0.04	DEF
CWRS	Waskada	0.08	0.09	I	CWRS	Stettler	0.03	0.04	DEF
CWRS	CDC Plentiful	0.06	0.09	I	CWRS	AC Abbey	0.03	0.04	EF
CWRS	Alvena	0.05	0.08	I	CWSWS	Bhishaj	0.02	0.04	EF
CWRS	5604HRCL	0.06	0.08	I	CWRS	Waskada	0.04	0.04	EF
CWRS	AC Splendor	0.04	0.06	I	CWRS	Prodigy	0.02	0.03	EF
CWRS	CDC Utmost	0.04	0.06	I	CWRS	AC Splendor	0.02	0.03	EF
CWRS	Vesper	0.02	0.04	I	CWRS	Vesper	0.01	0.02	F

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Western Soft White Spring (CWSWS)

As was observed with the number of sclerotia per spike, there was little separation among cultivars for mean sclerotium weight (Tables 4.12 and 4.13 for highest and lowest, respectively). In 2011, the cultivar Marquis had sclerotia that were significantly greater in weight than Bhishaj, Commander, and Sadash. In 2012, Marquis was not among the cultivars with the highest mean, but CDC Imagine was the cultivar with the highest sclerotium weight, and it was one of few cultivars that showed any significant difference for this variable. The CWAD cultivars Napoleon and AC Avonlea were among the cultivars with the lowest mean sclerotium weight, with the only estimates below 10 mg.

Table 4.12. Ten cultivars with the highest mean weight per sclerotium (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 5.094)		Market Class	Cultivar	Actual	Estimate (± 3.664)	
CWRS	Marquis	47.34	47.85	A [z]	CWRS	CDC Imagine	31.45	35.14	A
CWRS	KANE	45.14	46.56	AB	CWRS	AC Superb	27.80	32.29	AB
Spelt	CDC Zorba	47.78	46.22	AB	CWRS	5603HR	28.17	30.43	ABC
CWES	CDN Bison	43.35	44.25	AB	CWES	Glenlea	28.79	29.16	ABCD
CWES	Glenlea	41.94	42.26	ABC	CWES	Glencross VB	28.10	28.92	ABCD
CWES	CDC Walrus	42.38	41.86	ABC	CWRS	Unity VB	33.40	27.81	ABCD
CWRS	Unity VB	40.05	41.48	ABC	CWES	CDC Walrus	29.39	27.33	ABCD
CPS	SY985	40.68	40.91	ABC	CPS	AC Conquer VB	24.98	26.56	ABCD
CWRS	5500HR	40.17	40.60	ABC	CWRS	AC Intrepid	22.45	25.64	ABCD
CWRS	CDC Bounty	40.46	40.50	ABC	CWRS	CDC Bounty	25.35	25.63	ABCD

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Western Extra Strong (CWES), Canada Prairie Spring (CPS),

Table 4.13. Ten cultivars with the lowest mean weight per sclerotium (grown at the North Seed Farm, Saskatoon, SK, 2011 and 2012). Estimate values are from modeled data.

2011					2012				
Market Class[+]	Cultivar	Actual	Estimate (± 5.094)		Market Class	Cultivar	Actual	Estimate (± 3.664)	
CWAD	Kyle	22.04	21.52	ABC [z]	CWRS	Glenn	14.24	12.40	BCD
CWRS	CDC Plentiful	19.47	19.85	ABC	CWAD	Strongfield	11.44	12.37	BCD
CWRS	CDC Utmost	20.03	19.84	ABC	CWRS	Vesper	14.58	11.49	BCD
CPS	AC Crystal	19.50	19.49	ABC	CWAD	Kyle	10.71	11.22	BCD
CWAD	Strongfield	19.06	19.24	ABC	CWSWS	Bhishaj	9.88	10.85	BCD
CWAD	Transcend	19.09	19.15	ABC	CWSWS	AC Andrew	19.83	10.71	BCD
CWSWS	AC Andrew	17.94	18.13	ABC	CWAD	Transcend	10.40	10.7	BCD
CWSWS	Bhishaj	16.08	16.59	BC	CWAD	AC Navigator	11.19	10.15	BCD
CWAD	Commander	15.97	16.27	BC	CWAD	AC Avonlea	8.76	8.69	CD
CWSWS	Sadash	13.23	13.14	C	CWAD	Napoleon	8.08	7.43	D

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Amber Durum (CWAD), Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western Soft White Spring (CWSWS)

4.2 Growth Chamber Disease Reaction Evaluation

The Growth Chamber Experiment yielded results similar to those found in Field Experiment #2. There were significant differences among cultivars and within market classes of western Canadian spring wheat. However, the highest ranked cultivars were

rarely the same in both environments. Also, the means of the variables measured were much greater in the growth chamber than in the field. This was likely due to the way in which the plants were inoculated. In the field, natural infection was simulated through the spreading of sclerotia on the soil surface, whereas a more invasive technique was used to inoculate in the growth chamber. By injecting a spore suspension into the wheat floret, the fungus had direct access to the ovary and did not have to overcome physical barriers. Table 4.14 presents the non-modeled means of the six variables collected from the growth chamber.

Table 4.14. Disease reaction mean scores, standard error (SE), and range for % sclerotia, sclerotia/spike, wt/sclerotium, honeydew production, sclerotial size, and % florets aborted in the Growth Chamber Experiment.

		O[x]	W[y]	C[z]
Percent sclerotia [a]	Mean (± SE)	51.41 (±1.028)	51.50 (±1.039)	47.49 (±7.561)
	Range	2.28 - 91.89	2.28 - 91.89	17.05 - 63.68
Sclerotia / spike [b]	Mean (± SE)	9.18 (±0.194)	9.27 (±0.192)	4.88 (±1.449)
	Range	1.25 - 16.50	1.33 - 16.50	1.25 - 9.33
Weight / sclerotium (mg) [c]	Mean (± SE)	48.61 (±1.092)	47.13 (±0.898)	116.89 (±12.488)
	Range	10.00 - 160.71	10.00 - 108.24	82.50 - 160.71
Honeydew rating [d]	Mean (± SE)	3.6 (±0.05)	3.6 (±0.05)	4.9 (±0.04)
	Range	1.2 - 5.0	1.2 - 5.0	4.8 - 5.0
Sclerotial size rating [e]	Mean (± SE)	2.6 (±0.02)	2.6 (±0.02)	2.9 (±0.11)
	Range	0.9 - 3.0	0.9 - 3.0	2.3 - 3.0
Percent florets aborted [f]	Mean (± SE)	8.95 (±0.470)	8.77 (±0.458)	17.49 (±6.184)
	Range	0.00 - 42.50	0.00 - 42.50	5.00 - 41.00

[a]Percent sclerotia = Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b]Sclerotia / Spike = Sclerotia/spike = number of sclerotia / number of spikes
[c]Weight / Sclerotium (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[d]Honeydew rating = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)
[e]Sclerotial size rating (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)
[f] Percent florets aborted = 100 * (florets aborted / total inoculated florets)
[x] O = overall (all cultivars of wheat and controls)
[y] W = all wheat cultivars averaged
[z] C = control, Sandro triticale and Gazelle rye

4.2.1 Disease Reaction of Wheat Species and Market Classes

The market classes were compared using non-modeled data for percent sclerotia by weight, number of sclerotia per spike, sclerotium weight, honeydew production, sclerotial size, and percent florets aborted. Select comparisons were carried out in SAS

using orthogonal contrasts within the mixed procedure. Additionally, the controls were compared to the wheat species and wheat species were compared amongst each other. The means for each of the market classes tested and the results of the contrasts, with asterisks indicating the significance level of the comparison, are presented in Tables 4.15 and 4.16, respectively. The mean values for the species (*T. aestivum, T. turgidum* ssp. *durum,* and *T. spelta*) can be found in Appendix C.

Table 4.15. Disease reaction mean scores for % sclerotia, sclerotia/spike, wt/sclerotium, honeydew production, sclerotial size, and % florets aborted for all the wheat market classes in the Growth Chamber Experiment.

Market Class[+]	% Scl[a]	Scl/ Spike[b]	Wt/Scl[c]	HD[d]	Scl Size[e]	% Ab[f]
CWGP	64.28	10.70	67.38	3.1	2.9	9.35
CPS	57.36	10.56	50.69	4.1	2.6	5.69
CWSWS	54.33	9.93	55.91	4.3	2.7	7.80
CWRS	53.21	9.43	46.81	3.7	2.7	7.00
CWES	52.61	10.62	48.14	3.2	2.7	4.26
CWHWS	41.67	7.67	36.00	3.6	2.4	7.92
CWAD	39.22	7.18	42.37	3.2	2.0	18.46
Spelt	34.64	6.03	32.93	1.8	1.8	28.06

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia / (weight grain + sclerotia)
[b] Scl / Spike = Sclerotia/spike = number of sclerotia / number of spikes
[c] Wt / Scl = Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[d] HD = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)
[e] Scl Size = sclerotial size (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)
[f] % Ab = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)
+ Canada Western General Purpose (CWGP), Canada Prairie Spring (CPS), Canada Western Soft White Spring (CWSWS), Canadian Western Red Spring (CWRS), Canada Western Extra Strong (CWES), Canada Western Hard White Spring (CWHWS), Canadian Western Amber Durum (CWAD)

There was no difference between the control and the *Triticum* species for the percent sclerotia by weight ($P > 0.05$), but there were differences for the remaining six variables (Table 4.16). The control had fewer sclerotia per spike ($P < 0.001$) but the sclerotia present were heavier and larger ($P < 0.001$ and <0.01, respectively). The control also produced more honeydew ($P < 0.001$), and had more florets aborted than the *Triticum* species ($P < 0.05$).

When *T. aestivum* was compared with *T. turgidum* ssp. *durum,* values for all variables differed significantly. *T. aestivum* had a higher percent sclerotia by weight, a greater number of sclerotia, and higher sclerotium weight ($P < 0.001$, 0.001, and 0.01,

respectively). The ratings for honeydew production and sclerotial size were also greater for *T. aestivum* than for *T. turgidum* ssp. d*urum* ($P < 0.01$ and 0.001, respectively). Finally, *T. aestivum* had fewer florets aborted ($P > 0.001$).

Similar results were found when *T. aestivum* was compared to *T. spelta*. *Triticum aestivum* had higher means except for the percent of florets aborted, which was greater for *T. spelta* ($P < 0.01$ for all but HD and sclerotial size, where $P < 0.001$).

Table 4.16. Results of specific contrasts for % sclerotia, sclerotia/spike, wt/sclerotium, honeydew production, sclerotial size, and % florets aborted for species and selected market classes in the Growth Chamber Experiment.

Comparison	% Scl [a]	Scl / Spike [b]	Wt / Scl [c]	HD [d]	Scl Size [e]	% Ab [f]
C[w] vs *Triticum*	NS	***	***	***	**	*
T.a [x]. vs *T.t.d.* [y]	***	***	**	**	***	***
T.a. vs *T.s.* [z]	**	**	**	***	***	***
CWRS vs CWAD	***	***	NS	***	***	***
CPS vs CWAD	***	***	*	***	***	***
CWES vs CWAD	**	***	NS	NS	***	***
CPS vs CWRS	NS	NS	NS	*	NS	NS
CPS vs CWES	NS	NS	NS	***	NS	NS
CWRS vs CWES	NS	NS	NS	**	NS	NS

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; NS = not significant = $P > 0.05$

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia / (weight grain + sclerotia)

[b] Scl / Spike = Sclerotia/spike = number of sclerotia / number of spikes

[c] Wt / Scl = Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

[d] HD = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)

[e] Scl Size = sclerotial size (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)

[f] % Ab = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)

[w] C = control = rye + triticale

[x] *T.a. = T. aestivum*

[y] *T.t.d. = T. turgidum* ssp. *durum*

[z] *T.s. = T. spelta*

+ Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES)

The three major market classes of *T. aestivum* (CWRS, CPS, and CWES) were compared on an individual basis to the *T. turgidum* ssp. d*urum* market class CWAD. The results (Table 4.16) were the same as for *T. aestivum* as a whole when examining all variables except sclerotium weight and honeydew rating, in which there were some non-significant differences. The CPS market class had higher sclerotium weights than CWAD ($P < 0.05$). There was no difference in the weight of sclerotia between CWRS and CWAD ($P > 0.05$). The CWES market class was not different from CWAD for

sclerotium weight or for honeydew production ($P > 0.05$). The number of sclerotia per spike was greater for CWES compared to CWAD ($P < 0.01$).

When comparing market classes within *T. aestivum*, there were no differences for any of the variables ($P > 0.05$) except for honeydew production. The CPS market class produced more honeydew than CWRS ($P < 0.05$) and CWES ($P < 0.001$), and CWRS produced more honeydew than CWES ($P < 0.01$).

When the table of contrasts is examined as a whole, a few patterns emerge. There are differences in honeydew production for all comparisons, with the exception of CWES versus CWAD. Also, it is interesting to note that there are differences in values for all variables between species, but not within hexaploid wheat. These differences are quite unlike those from the field experiment, where more differences were seen within the species, and fewer differences between the species.

4.2.2 Disease Reaction of Wheat Cultivars

The mean scores were calculated for each of the variables from the Growth Chamber Experiment for all the cultivars; these results can be found in Appendix B. The variables measured and/or calculated included percent sclerotia by weight, number of sclerotia per spike, sclerotium weight, honeydew production, sclerotial size, and percent of florets aborted.

After testing each variable for residual and replicate normality, only three parameters (sclerotium weight, sclerotial size, and percent florets aborted) were tested with other transformations as the rest had normal distributions with the non-transformed data. The only benefit gained was with the square root transformation of the weight per sclerotium and percent of florets aborted. The other variables were left untransformed for the remainder of the analyses.

Due to heterogeneity of variances, the data were modeled to standardize the comparisons, and the new estimates and their standard errors can be found in Appendix D. The estimates calculated for each cultivar were used to assign letters to indicate differences. All six variables were significant ($P < 0.001$). The F-values for each are

presented in Table 4.17. Tables 4.18 through 4.29 show the highest and lowest cultivars for the variables used to evaluate disease reaction in the growth chamber.

Table 4.17. F-values from Type 3 tests of fixed effects using modeled data for cultivar differences for % sclerotia, sclerotia/spike, wt/sclerotium, honeydew production, sclerotial size, and % florets aborted for the Growth Chamber Experiment.

	F-Value
Percent sclerotia [a]	6.8***
Sclerotia / spike [b]	11.46***
Weight / sclerotium (mg) [c]	445.75***
Honeydew rating [d]	28.69***
Sclerotial size rating [e]	7.67***
Percent aborted [f]	4.07***

[a] Percent sclerotia = Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Sclerotia / Spike = Sclerotia/spike = number of sclerotia / number of spikes
[c] Weight / Sclerotium (mg) = 1000 * (weight of the sclerotia / number of sclerotia) – square root transformation
[d] Honeydew rating = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)
[e] Sclerotial size rating (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)
[f] Percent aborted = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)
*** $P < 0.001$

The only significant differences among cultivars for both percent sclerotia by weight or for number of sclerotia per spike were those that were the very highest or the very lowest. The ten cultivars with the highest percent sclerotia by weight and most sclerotia per spike were all *Triticum aestivum*, with the majority from the CWRS market class (Tables 4.18 and 4.20). The ten cultivars with the lowest percent sclerotia by weight and least number of sclerotia per spike were also very similar, with four from the market class CWAD (Tables 4.19 and 4.21). The cultivar Eurostar should be noted as being the lowest for both variables; it had only 14.31% sclerotia and less than three sclerotia per spike. The remaining nine cultivars listed were all essentially the same, with values almost all below 30% and contained five or fewer sclerotia per spike.

Table 4.18. Ten cultivars with the highest mean percent sclerotia by weight (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 5.598)	
CWRS	AC Abbey	83.50	86.23	A [z]
CWRS	CDC Kernen	79.33	78.32	AB
CPS	AC Conquer VB	79.45	77.84	ABC
CWGP	NRG010	68.68	77.12	ABCD
CWRS	CDC Osler	70.22	74.29	ABCDE
CWRS	Roblin	69.13	73.40	ABCDEF
CWRS	859CL	73.64	73.10	ABCDEFG
CWRS	CDC Merlin	67.41	71.01	ABCDEFG
CWRS	AC Barrie	68.98	69.48	ABCDEFGH
CWRS	CDC Utmost	67.01	68.97	ABCDEFGH

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western Extra Strong (CWES), Canada Western General Purpose (CWGP)

Table 4.19. Ten cultivars with the lowest mean percent sclerotia by weight (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 5.598)	
CWRS	Carberry	31.20	30.51	MNOPQRSTUV
CWRS	Waskada	33.29	29.30	NOPQRSTUV
CWRS	AC Superb	33.45	29.12	OPQRSTUV
CWAD	AC Avonlea	32.02	28.40	PQRSTUV
CWRS	Muchmore	26.85	25.83	QRSTUV
CWRS	Journey	31.28	24.41	RSTUV
Spelt	CDC Zorba	23.11	22.92	STUV
CWAD	Transcend	26.07	20.90	TUV
CWAD	Strongfield	18.87	19.10	UV
CWAD	Eurostar	11.86	14.31	V

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD)

Table 4.20. Ten cultivars with the highest mean number sclerotia per spike (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.896)	
CWRS	AC Abbey	15.06	15.16	A [z]
CWRS	CDC Kernen	14.50	14.09	AB
CPS	SY985	14.17	13.94	ABC
CPS	AC Conquer VB	14.00	13.74	ABCD
CWRS	859CL	13.50	13.68	ABCDE
CWRS	AC Barrie	13.39	13.41	ABCDEF
CWRS	Roblin	13.05	13.15	ABCDEFG
CWGP	NRG010	11.67	13.11	ABCDEFG
CWRS	CDC Osler	13.11	12.92	ABCDEFGH
CPS	5702PR	12.50	12.68	ABCDEFGHI

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Prairie Spring (CPS), Canada Western General Purpose (CWGP)

Table 4.21. Ten cultivars with the fewest mean number sclerotia per spike (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.896)	
CWAD	AC Avonlea	5.61	5.33	TUVWXYZ
CWAD	Strongfield	5.17	5.07	UVWXYZ
CWRS	Journey	5.44	4.75	VWXYZ
Spelt	CDC Zorba	4.67	4.64	VWXYZ
CWRS	AC Superb	4.89	4.42	WXYZ
CWAD	Transcend	5.00	4.33	XYZ
CWRS	Carberry	4.02	4.22	YZ
CWRS	Glenn	4.23	4.18	YZ
CWRS	Muchmore	4.33	4.16	YZ
CWAD	Eurostar	2.66	2.77	Z

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Amber Durum (CWAD), Canadian Western Red Spring (CWRS)

The cultivars with the highest and lowest sclerotium weight were nearly all different compared to the previous two variables. The cultivars AC Conquer VB and NRG010 were similarly high for all three variables measured, while Eurostar, CDC Zorba, and Strongfield were among the lowest (Tables 4.22 and 4.23). Unlike percent sclerotia and number of sclerotia per spike, a larger number of cultivars could be distinguished from each other in terms of sclerotium weight. Within the top ten cultivars for highest sclerotium weight were two of the three CWGP cultivars, and the remainder

an assortment of other market classes. A number of CWAD and CWRS cultivars, as well as one of the two spelt cultivars, were the lowest for weight per sclerotium.

The cultivars with the highest weight per sclerotium (> 70 mg) included NRG003, Carberry, Brigade, AC Superb, and Glenn. The cultivar with the lowest weight per sclerotium was Eurostar, followed by CDC Zorba, Strongfield, Goodeve VB, Snowbird, and 5601HR, all of which had weights at or below 31.5 mg. Carberry, AC Superb, and Glenn also had low percent sclerotia by weight and number of sclerotia per spike.

Table 4.22. Ten cultivars with the highest mean weight per sclerotium (grown in the growth chamber). Estimate values are from modeled data; square root transformation.

Market Class[+]	Cultivar	Actual	Square Root	Estimate (± 0.157)	
CWGP	NRG003	73.57	8.57	8.50	A [z]
CWRS	Carberry	80.02	8.84	8.45	A
CWAD	Brigade	71.72	8.43	8.36	A
CWRS	AC Superb	74.49	8.55	8.32	A
CWRS	Glenn	70.58	8.34	8.10	B
CWSWS	Sadash	64.98	7.99	7.88	C
CPS	AC Crystal	59.51	7.68	7.76	CD
CWRS	Marquis	60.89	7.80	7.75	CD
CPS	AC Conquer VB	60.58	7.77	7.75	CD
CWGP	NRG010	66.50	8.07	7.72	CD

[z] Means with different letters are considered to be significantly different from each other
+ Canada Western General Purpose (CWGP), Canadian Western Red Spring (CWRS), Canadian Western Amber Durum (CWAD), Canada Western Soft White Spring (CWSWS), Canada Prairie Spring (CPS)

Table 4.23. Ten cultivars with the lowest mean weight per sclerotium (grown in the growth chamber). Estimate values are from modeled data; square root transformation.

Market Class[+]	Cultivar	Actual	Square Root	Estimate (± 0.157)	
CWAD	Kyle	35.59	5.95	5.89	(2)LM
CWRS	5600HR	34.30	5.84	5.81	(2)LMN
CWRS	Somerset	30.74	5.52	5.79	(2)LMNO
CWRS	CDC VR Morris	31.33	5.59	5.77	(2)MNOP
CWRS	5601HR	29.10	5.38	5.62	(2)NOPQ
CWHWS	Snowbird	30.68	5.54	5.60	(2)OPQ
CWRS	Goodeve VB	31.49	5.60	5.58	(2)PQ
CWAD	Strongfield	29.29	5.39	5.56	(2)PQ
Spelt	CDC Zorba	31.58	5.60	5.53	(2)Q
CWAD	Eurostar	21.92	4.57	4.80	(2)R

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Amber Durum (CWAD), Canadian Western Red Spring (CWRS), Canada Western Hard White Spring (CWHWS)

The florets aborted were those that did not form complete kernels of grain or sclerotia and therefore did not contribute to disease, but also were unable to contribute to the final yield. Of the ten cultivars with the highest percent florets aborted, six were CWAD and two were spelt (Table 4.24). Only two of the cultivars, Prodigy and Red Fife (both CWRS), were from *T. aestivum*. The cultivars with the lowest percent florets aborted were mainly from the CWRS and CWES classes, and included Helios, 5602HR, AC Intrepid, and 5701PR (Table 4.25). Only eight cultivars, which had the greatest percent florets aborted, were different from the lowest two cultivars, indicating very little difference among cultivars.

Table 4.24. Ten cultivars with the highest mean percent of florets aborted of total inoculated (grown in the growth chamber). Estimate values are from modeled data; square root transformation.

Market Class[+]	Cultivar	Actual	Square Root	Estimate (± 0.476)	
CWAD	Eurostar	38.98	6.24	6.25	A[z]
CWAD	Napoleon	30.09	5.49	5.57	AB
Spelt	CDC Origin	28.06	5.30	5.33	ABC
Spelt	CDC Zorba	28.06	5.30	5.26	ABCD
CWAD	Kyle	20.28	4.50	4.55	ABCDE
CWAD	AC Navigator	19.36	4.40	4.50	ABCDEF
CWRS	Prodigy	22.78	4.77	4.46	ABCDEF
CWAD	Commander	18.89	4.35	4.43	ABCDEF
CWRS	Red Fife	15.55	3.94	4.00	ABCDEFG
CWAD	CDC Verona	16.04	4.01	3.96	ABCDEFG

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Amber Durum (CWAD), Canadian Western Red Spring (CWRS)

Table 4.25. Ten cultivars with the lowest mean percent of florets aborted of total inoculated florets (grown in the growth chamber). Estimate values are from modeled data; square root transformation.

Market Class[+]	Cultivar	Actual	Square Root	Estimate (± 0.476)	
CWRS	5500HR	3.33	1.83	1.80	EFGHI
CWRS	AC Splendor	4.17	2.04	1.79	EFGHI
CWES	Glencross VB	4.72	2.17	1.75	EFGHI
CWES	CDC Walrus	4.44	2.11	1.74	EFGHI
CWRS	Roblin	2.78	1.67	1.70	FGHHI
CWES	Burnside	3.06	1.75	1.57	FGHI
CPS	5701PR	2.03	1.42	1.45	GHI
CWRS	AC Intrepid	1.95	1.40	1.30	GHI
CWRS	5602HR	2.22	1.49	1.00	HI
CWRS	Helios	2.22	1.49	0.99	H

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Western Extra Strong (CWES), Canada Prairie Spring (CPS)

The cultivars with the highest ratings for honeydew production and sclerotial size were very similar, with most belonging to the CPS, CWRS, and CWGP market classes (Tables 4.26 and 4.28). Among the ten cultivars with the highest ratings for both were AC Conquer VB and 5603HR. The cultivars with the lowest ratings for both included Eurostar, CDC Zorba, CDC Origin, Commander, and AC Morse (Tables 4.27 and 4.29). These all belong to either the CWAD market class or spelt. There were also a few CWRS cultivars in the lower ranked for honeydew production, but all the remaining cultivars for smaller sized sclerotia were CWAD cultivars. Strongfield was also among the lowest for sclerotial size, but was not included in the lowest for honeydew rating. Prodigy, a CWRS cultivar, also showed a different pattern, with less honeydew production but larger sclerotia.

There were fewer differences among cultivars for sclerotial size compared to honeydew production. Sclerotial size was very similar for the first eighty cultivars. Differences were only observed among the few cultivars that produced the very largest and the very smallest sclerotia.

Table 4.26. Ten cultivars with the highest mean honeydew production rating (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.15)	
CPS	AC Conquer VB	4.9	4.9	A[z]
CWRS	Unity VB	4.8	4.9	AB
CWRS	KANE	4.8	4.9	AB
CPS	AC Taber	4.6	4.7	ABC
CPS	AC Crystal	4.7	4.7	ABC
CWRS	Infinity	4.6	4.6	ABCD
CWRS	5603HR	4.5	4.6	ABCDE
CWRS	McKenzie	4.5	4.6	ABCDE
CPS	AC Foremost	4.5	4.6	ABCDEF
CWSWS	Sadash	4.5	4.6	ABCDEF

[z] Means with different letters are considered to be significantly different from each other
+ Canada Prairie Spring (CPS), Canadian Western Red Spring (CWRS), Canada Western Soft White Spring (CWSWS)

Table 4.27. Ten cultivars with the lowest mean honeydew production rating (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.15)	
CWRS	Journey	2.8	2.9	WXYZ(2)A
CWES	CDC Rama	3.0	2.9	WXYZ(2)A
CWRS	Vesper	2.8	2.8	XYZ(2)AB
CWRS	Glenn	2.8	2.7	YZ(2)AB
CWRS	Prodigy	2.6	2.6	Z(2)AB
CWAD	AC Morse	2.4	2.4	(2)ABC
CWAD	Commander	2.4	2.4	(2)ABC
Spelt	CDC Origin	2.0	2.0	(2)BCD
Spelt	CDC Zorba	1.6	1.6	(2)CD
CWAD	Eurostar	1.3	1.3	(2)D

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Western Extra Strong (CWES), Canadian Western Amber Durum (CWAD)

Table 4.28. Ten cultivars with the highest mean sclerotial size rating (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.12)	
CWRS	Marquis	2.9	3.0	A
CWRS	Carberry	2.9	2.9	A
CWGP	NRG003	2.9	2.9	A
CWRS	CDC Utmost	2.9	2.9	A
CWSWS	Bhishaj	2.9	2.9	A
CWGP	Minnedosa	2.9	2.9	A
CWRS	CDC Plentiful	2.8	2.9	A
CPS	AC Conquer VB	2.9	2.9	A
CWRS	5603HR	2.9	2.9	A
CWRS	Prodigy	2.9	2.9	A

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Red Spring (CWRS), Canada Western General Purpose (CWGP), Canada Western Soft White Spring (CWSWS), Canada Prairie Spring (CPS)

Table 4.29. Ten cultivars with the lowest mean sclerotial size rating (grown in the growth chamber). Estimate values are from modeled data.

Market Class[+]	Cultivar	Actual	Estimate (± 0.12)	
CWAD	AC Morse	2.1	2.0	DEFGHIJK
CWAD	AC Avonlea	2.1	2.0	EFGHIJK
Spelt	CDC Origin	2.0	1.9	FGHIJK
CWAD	Napoleon	2.1	1.9	FGHIJK
CWAD	Kyle	1.9	1.9	GHIJK
CWAD	Transcend	1.9	1.9	GHIJK
CWAD	Commander	1.7	1.7	HIJK
Spelt	CDC Zorba	1.7	1.6	IJK
CWAD	Eurostar	1.3	1.5	JK
CWAD	Strongfield	1.5	1.4	K

[z] Means with different letters are considered to be significantly different from each other
+ Canadian Western Amber Durum (CWAD)

4.3 Correlation Between and Within Environments

Pearson correlation coefficients were calculated for each of the three variables within each environment, as well as between environments.

The Pearson's correlation coefficients (*r*) were calculated for 2011 and 2012 (Table 4.30) for each of the variables in Field Experiment #2. A significant correlation was found between the percent sclerotia by weight and number of sclerotia per spike, as well as between the percent sclerotia by weight and weight per sclerotium ($P < 0.001$ for both). There was no correlation between the number of sclerotia per spike and the sclerotium weight ($P > 0.05$). Results were similar in both years. When examining the correlation coefficient values, the only correlation that might be considered substantial was that between the percent sclerotia and the number of sclerotia per spike, with an *r* value close to 1, indicating a strong positive correlation.

Table 4.30. Pearson's correlation coefficients for disease reaction means for % sclerotia by weight, sclerotia/spike, and weight/sclerotium for 92 cultivars in 2011 and 2012 (grown at the North Seed Farm, Saskatoon, SK).

		Percent Sclerotia [a]	Sclerotia / Spike
Sclerotia / Spike [b]	2011	0.865***	
	2012	0.907***	
Weight / Sclerotium [c]	2011	0.352***	0.055
	2012	0.217***	0.032

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

[a] Percent sclerotia by weight = 100 * weight sclerotia / (weight grain + sclerotia)

[b] Sclerotia/spike = number of sclerotia / number of spikes

[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

Table 4.31 contains the Pearson's correlation coefficients (*r*) for each of the variables in the growth chamber experiment. There were significant correlations between all variables; however, the significance level varied for many of the correlations. Also, many of the correlation coefficients were not very strong. The most notable correlation was that between the percent sclerotia by weight and number of sclerotia per spike, with an *r* value of 0.904 ($P < 0.001$).

Table 4.31. Correlation coefficients between the disease reaction means for % sclerotia, sclerotia/spike, wt/sclerotium, honeydew production, sclerotial size, and % florets aborted for the wheat cultivars in the growth chamber experiment.

	% Scl [a]	Scl/Spike [b]	Wt/Scl [c]	HD [d]	Scl Size [e]
Scl / Spike [b]	0.904***				
Wt / Scl [c]	0.178**	-0.140*			
HD [d]	0.402***	0.317***	0.214***		
Scl Size [e]	0.461***	0.286***	0.589***	0.357***	
% Ab [f]	-0.209***	-0.319***	-0.149*	-0.394***	-0.506***

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)

[b] Scl / Spike = Sclerotia/spike = number of sclerotia / number of spikes

[c] Wt / Scl = Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

[d] HD = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)

[e] Scl Size = sclerotial size (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)

[f] % Ab = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)

Using the means from the three common variables from the growth chamber and Field Experiment #2 (percent sclerotia by weight, number of sclerotia per spike, and average sclerotium weight), Pearson's correlation coefficients (*r*) were calculated to determine if there were any commonalities between environments. The growth chamber data was correlated with the field data on a yearly basis as well as with both years combined. In all cases, no significant correlations were detected at the 5% confidence level between the growth chamber, in which artificial inoculation methods were used, and the field, in which a more natural inoculation method was used. Table 4.32 contains the values of the test's correlation coefficients for each comparison.

Table 4.32. Pearson correlation coefficients (*r*) between each field year of data (combined and individually for 2011 and 2012) and the growth chamber data for % sclerotia, sclerotia/spike, and wt/sclerotium.

	Percent Sclerotia[a]			Sclerotia / Spike[b]			Weight / Sclerotium[c]		
	--- Field ---								
	2011	2012	Both	2011	2012	Both	2011	2012	Both
Growth Chamber	0.1811	0.1979	0.1988	0.1582	0.1304	0.1544	-0.0041	0.1174	0.0550

[a] Percent sclerotia by weight = 100 * weight sclerotia / (weight grain + sclerotia)

[b] Sclerotia/spike = number of sclerotia / number of spikes

[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

5.0 DISCUSSION

5.1 Field Disease Reaction Evaluation

There have been few recent studies examining ergot infection in wheat under artificial conditions (greenhouse or growth chamber) and even fewer under field conditions. There are a number of published studies that examine ergot infection in other crops such as Kentucky blue grass, sorghum, and rye. This study is the first to quantify ergot reactions of a large set of western Canadian wheat cultivars in a field setting. Disease severity was estimated from percent sclerotia by weight, number of sclerotia per spike, and sclerotium weight. By inoculating the wheat with *C. purpurea* in a more natural manner, in this case by spreading sclerotia on the soil surface, reactions could be examined. The only other study that has looked at wheat cultivars on a field level was conducted in the United Kingdom, which consequently means that different cultivars were evaluated (Bayles et al., 2009). In both cases, precipitation and temperature during the studies had an impact on the final results, although not directly correlated in this study. The overall means for each of the three variables in 2011 were greater than in 2012, which was likely due to environmental conditions (see Appendix E for daily and cumulative precipitation and Appendix F for temperatures). The development of *C.* purpurea can be greatly affected depending on the timing of optimal growth conditions (i.e. temperature and moisture). Environmental conditions are also important for crop development and may influence the period of susceptibility to the pathogen. In both years, ample precipitation was available for germination in the spring, but timing of conditions later in the season likely impacted final disease levels. In early- to mid-July, when the wheat would be going through anthesis, temperatures were greater in 2012 compared to 2011, and this could have reduced fungal growth in the hotter year.

In many cases, the samples collected from each plot did not appear to be infected with ergot, as there were no sclerotia protruding from the glumes. However, upon harvest and subsequent threshing of the samples, it was evident that the ergot fungus was present, but the sclerotia were contained within the glumes.

5.1.1 Reaction of Cultivars to Varying Levels of Inoculum

In Field Experiment #1 there were no differences detected among the rates of inoculum (0, 8, or 16 grams per plot), nor were there differential reactions to the inoculum levels among the cultivars (no interaction). In this experiment, it is likely that the rye spreader plots acted as the most important source of inoculum compared to the sclerotia that was spread on the soil surface. The rye flowered considerably earlier than the wheat, with some seeded before and some at the same time as the wheat plots, and therefore would have been infected by the ascospores which were released after germination of the sclerotia in late spring/early summer. Once infected, the fungus would mature and eventually produce honeydew that contained conidia, and these spores could then be spread to the wheat once they were at the susceptible stage.

These results agree with those reported by Bretag and Merriman (1981), who stated that the wheat in their experiment was mainly infected by secondary infections where conidia were spread via honeydew from the ryegrass. Puranik and Mathre (1971) also found that tillers, which are produced later and often over an extended period of time, can be easily infected by honeydew. One experiment determined that as much as 98% of the total number of infected spikes were actually tillers, rather than the main stem spikes (Wood and Coley-Smith, 1982).

Although the honeydew stage often lasts for only 72 hours (Rapilly, 1968), it is obvious from this experiment, and others, that under conducive conditions, secondary infections may be the most important cause of an epidemic. This might be especially true in years where growing conditions are less than optimal for the crop. Uneven flowering, caused by environmental and other stresses, allow the fungus a longer period of time to infect the crop. When conditions are humid and cloudy, the period of honeydew production may be increased (Schwartingl & Hiners, 1945), exacerbating the problem.

In future studies, testing inoculum rates in a field setting may prove to be more successful with some sort of barrier for isolation among plots. Rye spreader rows may also need to be removed, as they are obviously an important source of inoculum. Preventing spread of honeydew among plots would permit only the sclerotia present to cause infection, allowing a true detection, if there is one, of the effect of inoculum rate on infection intensity and severity.

5.1.2 Disease Reaction of Wheat Species, Market Classes, and Cultivars

In Canada, wheat cultivars are divided into market classes based on certain characteristics, and so those within similar classes would come from similar genetic backgrounds. In this experiment, seven different market classes, plus spelt, were tested to determine if differential disease reactions to ergot could be associated with certain market classes or genetic backgrounds. Some market classes contained very few cultivars relative to others, and so orthogonal contrasts were used for comparisons. The most commonly grown market classes in western Canada were compared in this study, including CWRS (55 cultivars), CPS (9 cultivars), CWES (6 cultivars), and CWAD (12 cultivars). The market classes were further categorized by species: *Triticum aestivum*, *T. turgidum* ssp. *durum*, and *T. spelta*, and were also compared using contrasts. Each of the cultivars were compared on an individual basis, and subsequently evaluated based on the combinations of three variables.

The contrasts were performed for all three sclerotia variables, but for most of these comparisons, the results obtained were not consistently highest or lowest for all three. Differences between *T. aestivum* and *T. turgidum* ssp. d*urum* or *T. spelta* were not detected for the percent sclerotia by weight. The species *T. turgidum* ssp. *durum* had more sclerotia per spike, but they were smaller than for *T. aestivum*, whereas *T. spelta* had fewer, but larger sclerotia compared to *T. aestivum*.

Some market classes within *T. aestivum* were more similar in disease reaction to durum wheat than other market classes. The two most commonly grown market classes, CPS and CWRS (McCallum & DePauw, 2008), were each compared to durum on an individual basis. The CPS class was not different from durum wheat for the number of sclerotia per spike, whereas the CWRS class had fewer sclerotia per spike compared to the durum wheat in both years.

Weight per sclerotium was the only variable for which consistent differences (both years) were detected between the species; however, when examining cultivars within *T. aestivum*, greater differences were detected within this species compared to among species. The CWES and CWRS market classes were different for all three variables in both years, as were the CWES and CPS market classes with the exception of the number of sclerotia in 2012. The highest values were for the CWES market class for

all three variables, followed by CPS, and then CWRS. Fernandez et al. (2000) conducted a survey for the presence of ergot contamination in a number of samples of grain from different locations in Saskatchewan. Based on percent sclerotia by weight, they found that the CWRS and CWAD market classes had similar infection levels and that CPS was slightly lower than the other market classes (0.32, 0.25, and 0.15% respectively). In this study, CPS wheat was actually higher than both CWRS and CWAD wheat for percent sclerotia, ranging from 0.70-1.37% over the two field seasons. The CWAD wheat ranged from 0.51-1.05% and CWRS wheat from 0.49-0.77%, and these two market classes were considered to be statistically the same in the contrast analysis for this variable.

In this study, there were more differences detected within the species *T. aestivum* than among species. One might assume that the results would be opposite, with greater divergence between the durum and bread wheat. The high and low data points may have averaged out when *T. aestivum* was compared to *T. turgidum* ssp. *durum*, masking any major differences, or there may just be greater differences within the gene pools due to many years of breeding and selecting for certain traits. There are differences among cultivars in maturity and flowering time, and potentially differences in flower morphology itself. Some wheat may also be less susceptible to stresses such as copper deficiency and excessive moisture or cool temperatures, resulting in flowering patterns that avoid infection. Many studies have illustrated the importance of floret morphology and its effect on ease of access and therefore susceptibility to the ergot fungus (Campbell, 1957; Puranik & Mathre, 1971; Platford & Bernier, 1976; Mantle et al., 1977).

Floret morphology of the host is important in regards to the final size of sclerotia, as size is often attributed to the size of the spikelets (Pažoutová, 2003). When there is more space within the spikelet, then the fungus has more room to grow. Spelt wheat typically has very tight glumes, with harvested grain often tightly enclosed in the hulls. This would make it more difficult for the fungus to access the ovary. However, once the fungus has access to the ovary and is able to infect, there would be more room within the spikelets compared to most bread wheat cultivars. Durum wheat was the opposite of spelt, producing many, but smaller sclerotia than produced on bread wheat. There was less room within the spikelets for the fungus to grow, but easier access for infection due to looser glumes. The similar percentages of sclerotia by weight between the species or

market classes can be attributed to the size and number, with a greater number of smaller sclerotia for the durum and larger but fewer sclerotia for the bread and spelt wheat. These results agree with those of Rapilly (1968), who reported that the average size of the sclerotia decreased as the frequency increased.

The size of sclerotia produced may influence infection levels in succeeding years. Some studies have recognized the importance of sclerotial size in the process of germination. Cooke & Mitchell (1966) reported that larger sclerotia may have more energy to use in the germination process and therefore be more successful compared to smaller sclerotia. This may be important in the event that the sclerotia get buried below the soil surface, as the larger sclerotia would produce a greater number of taller stromata to reach the surface and discharge the ascospores. The smaller sclerotia would be expected to produce less primary inoculum compared to the larger sized sclerotia.

The CWES market class had some of the highest values for all three variables, whereas the other market classes were more inconstant. The CWES class could therefore be considered the worst of the market classes, as it had many large sclerotia, resulting in a greater percentage by weight. The CPS market class is also poor in terms of susceptibility to ergot. The CWAD market class, which is especially important in Southern Alberta and Saskatchewan, had moderate to high percent sclerotia by weight and sclerotia per spike, but a low average sclerotium weight. The lower weight may not be advantageous as sclerotia of similar size to the kernels may prove to be difficult to separate, thereby affecting the final grade of a sample. The Grain Grading Guide (Canadian Grain Commission, 2011a) states that only 0.01% ergot sclerotia by weight are allowed in a sample of No.1 CWAD, and only 0.1% for feed grade wheat. These regulations are the same for CWRS, which is the most common market class in western Canada. The CWRS and CWAD market classes had similar percentages by weight; however, CWRS had fewer sclerotia per spike and larger sclerotia compared to CWAD. If the weight of the sclerotia is greater, while still maintaining a low percentage and few sclerotia per spike, a higher grade may be more easily obtained. Larger sclerotia may be more easily and more cheaply removed via gravity table versus colour sorter, which may need to be used for samples containing sclerotia the size of wheat kernels. However, as mentioned

previously, smaller sclerotia may actually be preferable in terms of reduced germination success and therefore reduced production of the primary inoculum.

Individual cultivars in this study generally behaved in a similar manner as their respective market classes. However, with a further division of test entries, there was some variation between years as to where the cultivars ranked. With the species and market classes, the same general pattern was observed in both years. When there were similarities between years for the cultivars, it may be an indication of the physiological component conferring resistance, rather than simply escape.

Glenlea (CWES) was ranked as the most infected cultivar in both field seasons, as was 5702PR (CPS). Other susceptible cultivars included SY985 (CPS), which is a recently registered cultivar, and Burnside and CDN Bison (both CWES). Two very old lines, Marquis and Red Fife (both CWRS), had among the highest percent sclerotia. These cultivars were also among those with the most sclerotia per spike. Among the lowest ranked cultivars for both variables were CDC Utmost, Vesper, Stettler, AC Splendor, and Prodigy, which are all CWRS. These five cultivars also had moderate to low sclerotium weights, although only CDC Utmost was in the bottom ten for 2011 and Vesper in 2012.

The heaviest sclerotia were produced on cultivars largely from the CWES and CWRS market classes and the lightest sclerotia from the CWSWS and CWAD market classes. Some of the cultivars on which heavy sclerotia were produced in 2011 were not the same cultivars in 2012; however, some were notable for having heavy sclerotium weights in both years. These cultivars included Glenlea, CDC Walrus, Unity VB, CDC Bounty, CDC Imagine, CDN Bison, and SY985. The cultivars on which the lightest sclerotia were produced included Sadash, AC Andrew, and Bhishaj in the CWSWS market class and Napoleon, AC Avonlea, Strongfield and Transcend from the CWAD market class.

Glenlea was one of the few cultivars found to be among the highest for all three variables. It was first registered in 1972 as Utility wheat, now in the CWES market class (Evans et al., 1972), and was grown as a popular CWES cultivar until 2007. It has been used as a parent in crosses yielding Burnside, CDC Rama, and CDC Walrus (McCallum & DePauw, 2008). Glenlea has been noted for its outcrossing under both greenhouse and

field conditions (Hucl, 1996; Lawrie et al., 2006). Wheat, however, is normally a self-pollinating plant that is assumed to have out-crossing rates that are less than one percent (Lawrie et al., 2006). There have been cases where wheat plants exhibit rates of greater than 1% under certain conditions (Hucl, 1996).

With self-fertilization, the florets reach anthesis and are pollinated before the fungus has access to the ovary. Platford and Bernier (1976) reported that flowering habit and the ability to escape infection is perhaps the main means of disease avoidance under natural infection in the field. Once the ovary is fertilized, the risk of infection decreases with each day that passes (Puranik & Mathre, 1971; Darlington & Mathre, 1976; Watkins & Littlefield, 1976). In the field, then, it may be critical for plants to maintain fertility and self-pollinate. In circumstances where this cannot occur, the less time the florets are open, the shorter the period of susceptibility to the fungus.

There are a number of factors that can impact a plant's flowering habit and pollination. Cool temperatures during anthesis, improper herbicide application, excess nitrogen, and copper and boron deficiencies are all factors that can lead to uneven or extended flowering and/or floret sterility (Rapilly, 1968; Mantle et al., 1977; Mantle & Swan, 1995; Franzen et al., 2008). Delayed flowering may also increase infection rates because of increased risk that anthesis will coincide with the release of the secondary spores (Rapilly, 1968). In male-sterile wheat used for hybrid production, it is also important that there is adequate pollen from pollen donors to limit the time the florets need to be open for reception (Mantle & Swan, 1995). Some environmental factors that can impact pollen dispersal and outcrossing include precipitation, humidity, wind speed and direction, and temperature (de Vries, 1971). These are risk factors that are also considered important for ergot disease.

In a two year field study Hucl (1996) found that Glenlea was among the highest of ten cultivars for outcrossing (OC), with a rate of 0.95%. Other cultivars examined by Hucl (1996) that were also included in this study, were Roblin, with a high OC rate (1.43%) and Katepwa, which had one of the lowest OC rates (0.38%). The range for the ten cultivars in the Hucl study was between 0.3 and 6.05% over the two years. The study did not detect correlations between OC rate and any of the measured spike characteristics. However, Oslo, Wildcat, and Glenlea, three of the cultivars with the highest OC rate, had

spikes that tapered at the extremities; Biggar and Glenlea had a relatively high degree of spikelet opening at anthesis; and Oslo, Columbus, Glenlea and Roblin had among the lowest pollen viability and Katepwa among the highest. In general, the CPS cultivars were found to have the highest OC rates, followed by CWES, and then CWRS. The results for Katepwa and Roblin from the present study are similar to the results in the study by Hucl (1996). Roblin was found to have relatively high percent sclerotia by weight and Katepwa the least. The same was true for the number of sclerotia.

In a more recent greenhouse study, thirty-five cultivars were examined, and some had similar OC rates regardless of environment, and others were more inconstant (Lawrie et al., 2006). Eighteen of the cultivars had OC rates of more than 1% in at least one year of the study. Glenlea, however, was one of three cultivars that had high OC rates (10.6 and 8.6%) in both years. Most other cultivars had rates below 2.8%. It might be interesting to test if there is a correlation between the rate of OC and the disease reaction to ergot to see if there is any outstanding pattern among cultivars.

Burnside had a similar disease reaction to Glenlea, which would not be unexpected with Glenlea as part of its pedigree. The two CWES cultivars were both among the ten highest for percent sclerotia by weight and for number per spike, and for all remaining variables they were within the twenty highest. Somerset (CWRS) also has some parentage in common with Burnside, with Pasqua as part of their pedigree, and it had some of the highest values for each variable. The other two cultivars with Glenlea in their background were CDC Rama and CDC Walrus, but they did not appear to be as affected by the disease. Their values were in the mid-range for percent sclerotia by weight and number of sclerotia per spike. Both of these cultivars were selected on the basis of reduced shattering at harvest (Hucl, personal communication). The sclerotium weight was the only variable in the higher range, which was very similar to Glenlea. The higher sclerotium weight appears to be the variable most often associated with CWES cultivars, but sclerotia percent and number are more inconstant.

Bayles et al. (2009) reported that flowering characteristics may not actually influence infection rate. There were some cultivars they expected would be more prone to infection that were less affected by the disease than some that they expected would exhibit relatively high tolerance. This may have been due to the fact that there was some

type of physiological resistance within the host that allowed the plant to defend itself, even with unfavourable flowering characteristics.

As mentioned previously, there are few studies of wheat in a field environment that examine specific cultivars and their reaction to ergot. However, one study by Pageau and Lajeunesse (2006) tested seventeen cultivars of spring wheat under natural field conditions in Eastern Canada. The levels of ergot as estimated by percent sclerotia by weight were much lower compared to what was found in the current study (0.01-0.10% compared to 0 to 7.19%). One cultivar in common between the two studies was AC Barrie. Pageau and Lajeunesse (2006) reported that it was one of the least resistant cultivars, but in the current study, AC Barrie did not stand out as either highly susceptible or resistant, but was in the middle range for all three variables. The same was true of the cultivars Superb and McKenzie, with percentages similar to Barrie, within the mid-range of all cultivars compared. In the growth chamber, however, AC Barrie was among the least resistant, but it is difficult to make this comparison based on the method of inoculation used. Environmental conditions from eastern to western Canada could also have an impact on infection levels and susceptibility of certain cultivars.

Although not specifically measured in this study, a few traits were examined for currently registered cultivars to determine if they might coincide with their susceptibility to ergot. The CWAD and CWSWS cultivars require a longer growing season due to their later maturity. One would assume this could affect disease from a flowering standpoint. The later maturity may allow the plants to not only escape primary infections, but also late enough to avoid secondary infections depending on production of honeydew in surrounding areas. The CWAD and CWSWS market classes had moderate to low means over the two field seasons, and so maturity may be a factor to examine for association with ergot disease.

Whether the cultivar was awned or not did not appear to make any difference in infection in the field experiment. All the CPS cultivars were awned, but so were the CWSWS and CWAD cultivars, and the CWRS cultivars were a mixture of both. Since pollen reception can be hindered by awns (de Vries, 1971), and flowering habit and pollination may play a critical role in ergot infection under natural field conditions, it

might be interesting to examine the extent awns and pollen dispersal could have on infection levels as well.

These results all illustrate the importance of measurement method. The species comparisons did not show any significant difference for percent sclerotia by weight, but differences were found for sclerotia per spike and weight per sclerotium. When comparing market classes or cultivars, differences were detected with all three variables. Those with the highest or lowest means may therefore be useful for future tests in ergot reaction, but the traits considered to be most important will need to be determined.

5.2 Growth Chamber Disease Reaction Evaluation

Most studies to date have taken place in a controlled environment. Similar steps were taken in this experiment as done previously, specifically, injecting a spore suspension in the floral cavity of the wheat and collecting data on sclerotial size, frequency, and percentage, as well as honeydew production. In addition, the percent of florets aborted were calculated based on the number of total inoculated florets and the production of fully formed kernels and sclerotia. Similar comparisons were made for the growth chamber data as in the field experiment, including contrasts for the wheat species and select market classes, as well as comparisons among individual cultivars. Evaluation of disease reaction as a whole was based on the combination of the variables.

5.2.1 Disease Reaction of Wheat Species, Market Classes, and Cultivars

Triticum aestivum was contrasted with both *T. turgidum* ssp. *durum* and *T. spelta* and the variables were found to be significantly different in all cases. *T. aestivum* had a higher mean than both of the other *Triticum* species for all variables, with the exception of the percent florets aborted. *T. spelta* had the greatest percent, followed by *T. turgidum* ssp. *durum* and *T. aestivum* with the lowest. The florets aborted may not harbor sclerotia and contribute to disease in such a way that is visible, but the fungus may still be infecting the florets, thus inhibiting kernel formation and decreasing yield. Even though *T. aestivum* had fewer florets aborted, it had the most sclerotia per spike, with close to ten, whereas *T. turgidum* ssp. *durum* and *T. spelta* had only seven and six per spike,

respectively. Sclerotium weight for *T. aestivum* was close to 50 mg, and with its greater number of sclerotia per spike, the percentage of sclerotia by weight was over 50%. The other two species had weights ranging from 30-40 mg and consequently percentages below 40. The results of this portion of the study agree with that found by Platford and Bernier (1976). The percent of inoculated florets with sclerotia was greater for *T. aestivum* than *T. turgidum* ssp. *durum*, however, that test included only one cultivar per species.

As with the field experiment, the size and weight of the sclerotia may be attributed to the size of the floral cavity, which appeared to be the case with the larger florets of *T. aestivum* (2.7 rating and 48 mg per sclerotium) compared to those of *T. turgidum* ssp. *durum* (2.0 rating and 42 mg per sclerotium). Spelt, however, did not follow the same pattern. It had a very low size rating (1.8) and a low sclerotium weight, opposite of that found in the field. It also had the lowest honeydew rating, with a score indicating that the honeydew was often confined within the glumes and not usually visible. It also had the only average below a score of 3. Spelt may not be as receptive to artificial inoculation due to its tight glumes making access difficult, or it may have better resistance at the physiological level. It is difficult to come to any solid conclusions as only two cultivars of spelt were used in this experiment.

The honeydew ratings for *T. aestivum* and *T. turgidum* ssp. *durum* were similar, with 3.7 and 3.2 respectively, indicating that the honeydew was abundant enough to drip in small or large drops from the glumes. With the results at the species level, it would appear that more honeydew led to greater infection levels in terms of all the sclerotia variables (percent, number, weight, size), but the percent of florets aborted could have implications even though visible disease symptoms would have gone unnoticed.

Triticum aestivum can be divided into a number of different market classes, and in the contrast analysis for this study the most widely grown classes (CWRS, CPS, and CWES) were all compared to the *T. turgidum* ssp. *durum* market class (CWAD).

The results were very similar to those found for the species comparison with a few exceptions. The CWRS and CWAD market classes were not significantly different for sclerotium weight, and the same was true for the sclerotium weight comparison of CWES to CWAD. The CWES and CWAD market classes also had similar honeydew

ratings. The CPS and CWAD market classes were the least alike, with differences in all variables. The CWAD market class was lower than the other bread wheat market classes, with the exception of the percent florets aborted. Similarly, Menzies (2004) reported that the fewest number of sclerotia per spike were found in the CWAD wheat market class, and that CWES and CWRS market classes produced the greatest number. In contrast, the CPS market class had relatively few sclerotia in the experiment by Menzies (2004), but had among the most in this experiment. The results were very similar for the sclerotial size rating, with the exception of CPS, which had the largest size in this experiment and smallest sizes according to Menzies (2004). Platford and Bernier (1976) also observed more sclerotia in CWRS than CWAD wheat. Coley-Smith and Watkinson (1987) reported that ergot, based on percentage of infected spikes, was more predominant in durum wheat versus bread wheat, which disagrees with the finding from this study.

When the market classes within *T. aestivum* were compared, there were no differences for any of the variables except honeydew ratings. This was one of the biggest differences when comparing results to those of Menzies (2004), where honeydew production was not different among any of the market classes. Honeydew ratings could be considered somewhat subjective, which could have an impact on the final results.

In contrast to the field results, the comparisons among species and their respective market classes showed much greater variation in the growth chamber. The comparisons within *T. aestivum* were virtually the same for all those in the growth chamber, but quite different among the *T. aestivum* market classes in the field. In general, CWES had highest levels for all variables in the field, but similar to all the other *T. aestivum* market classes in the growth chamber. CWAD was usually the lowest for all variables in the growth chamber, but in the field it was similar to CWRS in percent and weight, and was among the highest for number of sclerotia.

In the growth chamber experiment, just as in the field experiment, there were some variables that showed very little difference in values among cultivars within the highest or the lowest rankings, i.e. many of the cultivars reacted similarly to the ergot pathogen. This was especially true in terms of percent sclerotia and number of sclerotia per spike. The highest fifty cultivars for percent were not different from each other, but AC Abbey, the cultivar with the highest percent, was different from the remaining forty-

two cultivars. CDC Kernen was the second highest and had higher percent sclerotia than the lowest twenty-three cultivars. AC Conquer VB, NRG010, Osler, and Roblin were also among the cultivars for highest percent sclerotia by weight. The lowest ranked cultivars for percent sclerotia were mainly from the CWAD and spelt market classes, including CDC Zorba (spelt), Transcend, Strongfield, and Eurostar. Essentially the same scenario was seen with the number of sclerotia per spike. AC Abbey, CDC Kernen, and AC Conquer VB were among the highest, along with SY985, 859CL, and AC Barrie. Eurostar and Transcend were again found to be the lowest, along with Muchmore, Glenn, Carberry, and AC Superb, which are CWAD and CWRS cultivars. None of the cultivars with the highest scores for percent and number of sclerotia were from the CWAD market class, but were composed of the classes from the *T. aestivum* species. The CWRS market class however, was observed throughout the highest and lowest, but this could be due to the fact that there was the greatest number of cultivars from this class.

There was more distinction among cultivars for the sclerotium weight, with NRG003, Carberry, Brigade, AC Superb, and Glenn containing the heaviest. Brigade was the only CWAD cultivar with a high score for any variable except for florets aborted, which is interesting since it, like many other CWAD cultivars in the study, have Kyle in its pedigree. The lowest sclerotium weights were again mainly CWAD and spelt cultivars, with Eurostar, CDC Zorba, and Strongfield being among the lowest. Also notable for low sclerotium weight was Goodeve VB, a CWRS cultivar, and Snowbird, which is a CWHWS cultivar.

Carberry, AC Superb, and Glenn were cultivars with low percent sclerotia by weight and number of sclerotia, but had large sclerotia. These qualities may prove to be beneficial, as was mentioned with the field data, since larger sclerotia may be more easily removed. Also, if there is a low percentage or few per spike, there would be less to remove.

In contrast to the variables already discussed, the highest percent of florets aborted were seen with CWAD and spelt cultivars and the lowest percent florets aborted were mainly obtained for the CWRS cultivars. A pattern of little differentiation among cultivars is seen again with this variable, as the highest eight (CWAD: Eurostar, Napoleon, Kyle, AV Navigator, and Commander; spelt: CDC Origin and CDC Zorba;

CWRS: Prodigy) are the only cultivars different from the lowest four (Helios, 5602HR, AC Intrepid, 5701PR). It is not uncommon to see undeveloped kernels or shriveled ovaries even when sclerotia are not present, but the reason for this inhibition is still speculation (Platford & Bernier, 1970; Alexopoulos et al., 1996).The cultivars with the lowest percent florets aborted were the ones with the highest percentage and number of sclerotia. This would most likely be due to the fact that there would be more fully formed sclerotia and there would be fewer florets available for abortion, and others may have still formed kernels. The cultivars with the most florets aborted may have had few sclerotia, but they also did not form a large portion of kernels. In a field situation, this would mean a decrease in yield even though disease symptoms may not have been present (as sclerotia). This variable may help to determine which cultivars, although diseased, may be able to better withstand yield losses.

The honeydew ratings were equally highest for the first twenty-three cultivars. Among the highest thirty-one cultivars, only AC Conquer VB, Unity VB, and KANE were significantly different. Large groupings of similar cultivars were seen throughout the results, but there were some CWAD and spelt cultivars that again stood out for having lower scores. AC Morse, Commander, Eurostar (CWAD), and the two spelt cultivars, CDC Origin and CDC Zorba all had ratings below 2.5, which was half on the scale for honeydew production. The spelt wheat and Eurostar were particularly low, with a score of 2 or less, indicating very little honeydew was produced, and what was produced, remained contained within the glumes. Similar cultivars had the lowest sclerotial size rating as honeydew production, with the addition of a number of other CWAD cultivars (Strongfield, Transcend, Kyle, Napoleon, and AC Avonlea). The cultivars within the highest ratings were all statistically the same. In fact, no difference was evident among the first eighty cultivars, and only the top sixteen had significantly greater honeydew production than the lowest twelve, with the split essentially between those with a rating either above or below 2.

There was a wide range of cultivars that had high values for each variable, but the CPS cultivar AC Conquer VB and the CWGP cultivar NRG010 were included among the highest for percent sclerotia by weight, number of sclerotia per spike, as well as for the sclerotium weight, and AC Conquer VB was also highest for honeydew production and

sclerotial size. AC Conquer VB is a newer cultivar that is resistant to the Orange Wheat Blossom Midge (*Sitodiplosis mosellana*), as are Unity VB and CDC Utmost, which have high ratings for honeydew and sclerotial size and among the highest scores for sclerotia percent and number. However, there are other midge resistant cultivars (Saskatchewan Ministry of Agriculture, 2013), such as Fieldstar VB, Glencross VB, and Shaw VB that are neither high nor low for the variables. It would be interesting to look into such a trait as midge resistance in a field setting and see if there is any correlation to ergot disease reaction, especially since insect vectors are a major concern with secondary infections.

All of the cultivars in this study were compared regardless of market class, whereas Menzies (2004) compared the cultivars within each of the classes separately. In both cases, differences were found for all measures of disease reaction. Kyle and Morse had some of the smallest sclerotia for the durum. For the CWRS wheat class, honeydew was the only variable that showed significant differences among cultivars, however, the cultivars in common between the two studies did not show any difference for honeydew ratings. For the CWES (Glenlea among them) and CWSWS cultivars, there were no differences at all. The two cultivars AC Vista and Taber for the CPS class were found to be the most susceptible, which generally agree with the results from this study, but with AC Vista having a slightly lower honeydew rating.

With little deviation among cultivars for most variables, it is difficult to actually rank the cultivars from 1 to 92. However, most variables at least show separation between the very highest and lowest, even if only significant for a few cultivars. Depending on the extent of differentiation needed, some variables may be more valuable than others.

5.3 Correlation of Variables

5.3.1 Correlation Within Environments

In studies where the wheat is infected artificially, usually by means of a spore suspension being injected into the floral cavity before anthesis, levels of infection are much greater than under conditions of natural infection (Campbell, 1957; Platford & Bernier, 1976; Bayles et al., 2009). This was the case in the present experiment, where

percent by weight, number per spike and weight of the sclerotia were much greater in the growth chamber compared to in the field. By infecting the plants before anthesis, self-pollination cannot occur, and the fungus, rather than the pollen, can access the ovary.

Pearson correlation tests were carried out for each variable within the two environments, as well as between each of the three variables in common between the environments. The variables within the environments showed that the highest correlation were percent sclerotia by weight and number of sclerotia per spike. In both years of the field experiment, as well as in the growth chamber, the correlation coefficients were around 0.9 and were highly significant ($P < 0.001$). This indicates a strong positive relationship between these variables. The percent sclerotia by weight was also found to be correlated with the weight per sclerotium in both environments ($P < 0.001$ in the field and < 0.01 in the growth chamber), but the correlation coefficient was only 0.3. In the field, the number of sclerotia per spike and the weight of a sclerotium were not correlated ($P > 0.05$) and in the growth chamber, although significant ($P < 0.05$), had a very low correlation coefficient. These results suggest that the number of sclerotia per spike does not influence the weight of the sclerotia. The weight, although it has a significant correlation with percent, is not high, indicating that the weight of the sclerotia does not have a large impact on the final percentage. It is the number of sclerotia per spike that could be considered to be most significant in terms of the percent sclerotia by weight, and this is the variable that is taken into consideration when the wheat is being graded.

In future studies, it may be possible to measure one variable over another (percent by weight or number per spike), saving time and effort for determining disease reactions. In a study using Kentucky bluegrass, Alderman and Barker (2003) suggest that the precision of severity data (percentage and number of sclerotia per spike) may not be any better than incidence data (number of spikes with sclerotia, which was not used in this study). In future studies with wheat, it may be beneficial to test whether incidence data may be sufficient for detecting differences among cultivars. Increased efficiency may outweigh any sort of precision that may be gained otherwise, and make selection of ergot resistant cultivars more manageable.

Further correlations were performed for the growth chamber with the remaining variables and they were all found to be significant ($P < 0.05$). However, as with the

sclerotium weight, the correlation values were too low to be considered important. Some studies have found that when less honeydew was produced, there was also fewer sclerotia produced (Platford & Bernier, 1976). The correlation coefficient in this experiment between sclerotia number and honeydew was only 0.317, but was significant ($P < 0.001$). More studies would need to be conducted to determine if a meaningful relationship exists. The size of sclerotia has been found to decrease when the number of sclerotia per spike increases (Rapilly, 1968). If the number of sclerotia per spike is correlated with the weight per sclerotium, then a slightly negative relationship may exist (r-value was -0.140, with $P < 0.05$). However, the number of sclerotia per spike was actually found to have a positive relationship with the size rating of the sclerotia, but again, the correlation coefficient was low (0.286) even though the P-value was significant ($P <0.001$).

5.3.2 Correlation Between Environments

Platford and Bernier (1976) suggested that there were two possible means of resistance under natural conditions, the first due to host physiology, and the second due to an escape mechanism by flowering habit and/or floret morphology. With a lack of correlation between the natural field and artificial growth environments, it may be suggested from this study that the resistance mechanism is due to an escape or avoidance mechanism. With a physiological resistance mechanism, one might expect that disease reaction would be similar in both environments. However, many of the cultivars that reacted favourably in the field were quite severely infected in the growth chamber, and vice versa.

When comparing market classes between environments, it was found that there were no CWAD cultivars with the highest values among any of the variables in the growth chamber, with the exception of Brigade for sclerotium weight. In the field, there were CWAD cultivars for the highest percent and number of sclerotia, but not for sclerotium weight. In general, there were more CWAD cultivars ranked among the lowest in the growth chamber compared to the field. The only commonality was for sclerotium weight, where both field and growth chamber had CWAD cultivars among the lowest.

For specific cultivars, CDC Utmost and AC Abbey, both CWRS cultivars, had low values in the field for the percent sclerotia, and AC Abbey had very few sclerotia per

spike. However, in the growth chamber, these cultivars performed the opposite, and had very high percent sclerotia by weight and number of sclerotia. For the sclerotium weight, Glenn (CWRS), Sadash (CWSWS), and AC Crystal (CPS) were among the highest in the growth chamber, but lowest in the field.

The cultivars in the field that were less affected by disease may have gone through anthesis before or after the timing of sclerotia germination, thereby avoiding primary infection by ascospores. Avoidance could have also occurred if anthesis did not coincide with the spread of the secondary inoculum. The number and timing of flowering of tillers may also have an impact on the degree of secondary infections, as the honeydew would be present later in the growing season.

In the growth chamber experiment, the plants were inoculated directly in the floral cavity, which is where the fungus needs to be to attack the ovary. The cultivars that had the least disease, in terms of any combination of the variables, may actually contain some type of physiological resistance, where the plant is able to defend itself from the pathogen.

Of all the cultivars tested, there were very few commonalities between environments for consistently high or low values for all variables. However, when considering only percent sclerotia and number of sclerotia per spike, there were some cultivars that did appear to have a more uniform reaction. The CWAD cultivar Eurostar and the CWRS cultivars Journey, AC Superb, Waskada, Muchmore and Carberry had some of the lowest means for the measured variables in both environments. Many other cultivars in the field differed from year to year, but these were relatively consistent in their ranking, indicating the possibility of physiological resistance. There were also cultivars that were consistently high for percent and number of sclerotia, and these included the CPS cultivars 5702PR and SY985, and the CWRS cultivars Roblin and CDC Merlin.

The cultivars that were highest for percent and number of sclerotia in both environments do not come from a similar pedigree. However, 5702PR and SY985 are both from the Syngenta breeding program in the USA, and so they may have germplasm in common.

The cultivars that were most resistant in both environments and in both years all had a similar background. AC Superb was the result of a backcross to Grandin from AC Domain (Townley-Smith et al., 2010). Carberry and Muchmore are both from a cross between Superb and Alsen (DePauw et al. 2011a and 2011b). Waskada is also from a cross involving Superb, but with BW278 (Fox et al., 2009). Journey, however, derives from the cross CDC Teal//Grandin/PT819 (Graf et al., 2003). It still contains Grandin, which is in common with the cross from which Superb was derived, so the results for Journey are not unexpected. Grandin was the result of the cross Len//Butte * 2//ND507/ND593 (Liu & Kolmer, 1997). It is a hard red spring wheat that was released in 1989 after its development at North Dakota State University Agricultural Experiment Station.

Additional cultivars with Grandin or Superb in their parentage were examined for their disease reaction mean scores. Vesper (Thomas et al., 2013), Stettler (DePauw et al., 2009), CDC Go (Hucl, personal communication), and CDC Abound (Canadian Food Inspection Agency, 2013) were also found to have among the lowest percent sclerotia and number of sclerotia per spike in both the field environment as well as in the growth chamber.

Since Superb has both Grandin and AC Domain in its pedigree, cultivars with AC Domain in their pedigree were also examined. Harvest, KANE, Snowbird, and 5604HR CL had mixed results compared to the previous cultivars. Although some had low means for the variables in either one or both environments, the evidence for resistance was not as strong as with the Grandin cultivars.

With these results, it is quite possible that the germplasm from North Dakota contains some type of resistance to *Claviceps purpurea*, with Grandin the main contributor. Alsen was developed at NDSU, but was released more than a decade later in the year 2000 (Frohberg et al., 2006). However, there is also Grandin parentage in Alsen, as well as some of the other North Dakota lines used in cultivars within the other crosses. Table 5.1 contains the lines that potentially confer resistance and shows the crosses used to make them. In all cases, Grandin, or some variation of this cultivar, was used.

Table 5.1. Potentially resistant lines showing parentage containing Grandin or some variation.

Cultivar	Parentage
AC Superb	Grandin*2/AC Domain
Waskada	BW278/2*Superb
Muchmore	Alsen/Superb
Carberry	Alsen/Superb
Vesper	Augusta/Hard White Alpha//3*AC Barrie x BW150*2//Tp/Tm/ 3/2*Superb/4/94B35-R5C/5/Superb
Stettler	Prodigy/Superb
CDC Go	Grandin/SD3055
CDC Abound	Superb*2/BW755
Journey	CDC Teal//Grandin/PT819

6.0 CONCLUSIONS

The results of this study suggest that there are different reactions among market classes and their respective cultivars when infected by the fungal pathogen *Claviceps purpurea*. The distinction among cultivars or market classes, however, may vary depending on the variable(s) used to determine disease reaction. Some may be more useful than others in determining which cultivars are most resistant to or tolerant of the disease. It is also important to note that using varying levels of inoculum did not affect disease reaction, and that secondary inoculum may have a greater impact on final infection levels than does the primary inoculum. If future studies are to be completed to test inoculum levels in a field setting, proper barriers and the removal of spreader plots would be recommended. Consideration of testing environment, however, does appear to be important when determining which variables are best suited to testing disease reaction to ergot. Unlike inoculum levels, environment had a major influence on which cultivars and market classes reacted more favourably to infection.

There was no correlation observed between the two environments, and this may have implication for future studies. In the growth chamber, artificial inoculation leads to greater levels of infection, and the only means of resistance would have to be through some type of genetic mechanism. In the field, where natural infection can occur, resistance may be accomplished if there is a genetic component, but the floret morphology or flowering habit may have a larger impact. If the plant is able to escape or avoid infection, whether or not it contains some sort of genetic or physiological resistance may not be of importance.

There are currently no requirements for ergot disease screening of new lines that are up for variety registration. The information gained through this study on cultivars that may confer resistance, particularly those within the Grandin lineage, could lead to their introduction into breeding programs and therefore encourage breeders to make assessments for ergot. Possibly the best traits to select would be percent sclerotia by weight and number of sclerotia per spike. However, these types of measurements for severity are quite time consuming and labor intensive, and further field testing should be

conducted to determine if simple incidence ratings may be sufficient. It may likewise be important for breeders to make note of flowering characteristics that may predispose the wheat to ergot infection to make superior selections.

In growth chamber studies, it has been suggested that honeydew is an important indicator of disease reaction, but in this experiment, correlations between the honeydew and other variables were not very robust. Further testing should be conducted to determine if there is a relationship, as assessment of honeydew production can be done a considerable time before maturity, long before all other measurements can be taken.

Experiments that are conducted in growth chambers or other controlled environments may not elucidate what might confer resistance in a field setting. However, there were some instances where cultivars had low disease values in either one or both environments. The best performing cultivars found through this research may prove to be important sources of physiological resistance, or may have favourable flowering characteristic that allow escape of infection, for future studies in ergot disease in wheat.

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8.0 APPENDICES

Appendix A. Mean disease reaction scores for Field Experiment #2 (non-modeled, non-transformed data) in 2011 and 2012 grown at the North Seed Farm, Saskatoon, SK.

Cultivars

	Percent Sclerotia [a]		Sclerotia / Spike [b]		Weight / Sclerotium [c]	
Cultivar	**2011**	**2012**	**2011**	**2012**	**2011**	**2012**
5500HR	1.55	0.51	0.28	0.11	40.17	22.05
5600HR	1.67	0.69	0.35	0.16	36.40	19.74
5601HR	2.23	2.03	0.48	0.45	30.69	19.50
5602HR	0.62	0.18	0.17	0.06	31.20	23.22
5603HR	1.10	0.27	0.27	0.07	35.71	28.17
5604HRCL	0.14	0.12	0.06	0.06	27.64	13.87
5700PR	1.77	0.40	0.50	0.18	36.24	15.50
5701PR	2.11	0.81	0.58	0.21	35.45	22.23
5702PR	3.22	1.97	0.88	0.57	37.48	24.94
859CL	0.72	0.41	0.16	0.11	34.08	19.05
AC Abbey	0.39	0.11	0.10	0.03	26.08	16.85
AC Andrew	0.46	0.25	0.34	0.19	17.94	19.83
AC Avonlea	0.89	0.41	0.55	0.28	22.59	8.76
AC Barrie	0.52	0.35	0.15	0.10	24.17	16.32
AC Cadillac	0.39	0.17	0.09	0.06	35.47	18.00
AC Crystal	0.54	0.25	0.32	0.17	19.50	13.42
AC Elsa	0.73	0.35	0.23	0.09	25.76	18.23
AC Foremost	0.51	0.38	0.20	0.15	29.87	21.03
AC Intrepid	0.93	0.42	0.23	0.10	30.90	22.45
AC Morse	1.66	0.26	0.47	0.12	36.52	17.19
AC Splendor	0.15	0.05	0.04	0.02	35.21	17.08
AC Superb	0.37	0.21	0.12	0.05	25.97	27.80
AC Taber	0.31	1.05	0.22	0.38	25.13	25.19
AC Vista	0.25	0.29	0.08	0.11	35.18	20.70
Alvena	0.15	0.12	0.05	0.04	25.46	18.11
Bhishaj	0.25	0.02	0.21	0.02	16.08	9.88
Brigade	1.02	1.20	0.52	0.66	31.18	16.34
Burnside	2.70	1.73	0.81	0.63	37.08	26.89
CDC VR Morris	1.55	0.71	0.36	0.18	30.20	25.11
Carberry	0.23	0.18	0.06	0.06	31.64	15.30
CDC Abound	0.72	0.31	0.18	0.08	36.72	20.39
CDC Alsask	0.74	0.30	0.16	0.08	33.99	21.53
CDC Bounty	1.31	0.56	0.21	0.13	40.46	25.35

CDC Go	0.37	0.32	0.09	0.11	38.35	19.79
CDC Imagine	1.38	1.36	0.31	0.26	33.64	31.45
CDC Kernen	0.60	0.42	0.16	0.14	35.26	19.13
CDC Merlin	2.18	1.13	0.43	0.21	35.30	18.08
CDC Origin	0.47	0.17	0.09	0.07	35.44	18.77
CDC Osler	0.61	0.28	0.16	0.07	26.04	19.51
CDC Rama	0.44	0.19	0.15	0.07	36.17	26.32
CDC Stanley	0.92	1.25	0.25	0.38	28.56	18.31
CDC Teal	0.94	0.57	0.23	0.15	28.94	22.06
CDC Thrive	0.87	0.60	0.22	0.17	34.99	21.22
CDC Utmost	0.10	0.05	0.04	0.03	20.03	15.25
CDC Verona	0.98	0.44	0.52	0.28	23.41	13.52
CDC Walrus	1.88	0.56	0.56	0.18	42.38	29.39
CDC Zorba	0.26	0.08	0.07	0.05	47.78	14.92
CDN Bison	2.28	0.81	0.62	0.25	43.35	24.72
Commander	0.36	0.71	0.27	0.27	15.97	15.70
Eatonia	0.52	0.43	0.11	0.10	32.05	22.11
Enterprise	2.09	0.47	0.89	0.27	30.87	25.41
Eurostar	0.37	0.09	0.18	0.09	27.17	12.45
Fieldstar VB	0.35	0.08	0.08	0.04	37.96	13.53
Glencross VB	0.78	0.46	0.26	0.16	28.95	28.10
Glenlea	3.30	3.57	0.95	1.00	41.94	28.79
Glenn	0.49	0.13	0.12	0.05	28.81	14.24
Goodeve VB	0.26	0.28	0.07	0.11	31.81	15.87
Harvest	0.72	0.44	0.18	0.13	30.71	20.13
Helios	0.57	0.23	0.12	0.06	33.26	19.52
AC Conquer VB	1.01	0.38	0.38	0.14	26.86	24.98
SY985	2.55	0.79	0.69	0.26	40.68	19.01
Infinity	1.38	0.81	0.34	0.21	35.52	19.47
Journey	0.46	0.23	0.10	0.09	33.51	16.75
KANE	0.50	0.24	0.08	0.07	45.14	20.84
Katepwa	0.59	0.31	0.14	0.06	32.09	27.81
Kyle	0.73	0.45	0.46	0.32	22.04	10.71
Lillian	0.42	0.36	0.16	0.08	26.30	17.42
Lovitt	1.17	1.14	0.28	0.26	29.86	20.92
Marquis	2.16	2.40	0.30	0.63	47.34	13.63
McKenzie	0.70	0.35	0.14	0.09	35.89	20.91
Minnedosa	0.73	0.46	0.20	0.12	41.89	25.19
Muchmore	0.51	0.25	0.13	0.10	32.10	15.32
Napoleon	1.29	0.10	0.57	0.11	29.28	8.08
AC Navigator	2.25	1.31	0.75	0.53	27.20	11.19
NRG003	0.54	0.44	0.18	0.17	35.41	26.15
NRG010	0.25	0.29	0.15	0.17	22.73	17.11
Prodigy	0.19	0.04	0.08	0.02	23.40	15.98
PT559	0.34	0.66	0.08	0.22	36.61	15.56
CDC Plentiful	0.15	0.31	0.06	0.10	19.47	16.66
Red Fife	2.47	1.76	0.73	0.62	36.27	22.04

Roblin	0.87	0.44	0.23	0.22	27.36	13.19
Sadash	0.45	0.55	0.48	0.40	13.23	14.44
Sandro	16.55	16.42	4.25	4.92	77.22	38.20
Shaw	0.27	0.14	0.08	0.07	35.07	17.20
Snowbird	0.64	0.64	0.17	0.21	31.13	16.29
Snowstar	0.40	0.38	0.09	0.09	30.45	20.34
Somerset	2.13	1.39	0.42	0.23	34.39	25.34
Stettler	0.25	0.08	0.10	0.03	25.36	14.61
Strongfield	0.31	0.32	0.20	0.23	19.06	11.44
Transcend	0.71	0.43	0.45	0.33	19.09	10.40
Unity VB	0.61	0.31	0.13	0.08	40.05	33.40
Vesper	0.05	0.02	0.02	0.01	32.25	14.58
Waskada	0.25	0.07	0.08	0.04	27.96	16.40
Mean	**0.90**	**0.54**	**0.27**	**0.18**	**31.33**	**19.26**

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)

Species

		Percent Sclerotia [a]		Sclerotia/Spike [b]		Weight/Sclerotium [c]	
		Mean (± SE)	**Range**	**Mean (± SE)**	**Range**	**Mean (± SE)**	**Range**
2011	*T.a.*[x]	0.90(±0.050)	0.01 - 6.70	0.24(±0.013)	0.00 - 1.43	31.98(±0.525)	8.62 - 71.82
	T.t.d.[y]	1.05(±0.103)	0.14 - 3.00	0.49(±0.035)	0.07 - 1.15	25.36(±0.964)	14.25 - 40.38
	T.s.[z]	0.36(±0.092)	0.07 - 0.91	0.08(±0.010)	0.04 - 0.12	41.61(±5.306)	15.00 - 62.22
2012	*T.a*	0.56(±0.042)	0.00 - 7.19	0.17(±0.012)	0.00 - 1.95	20.22(±0.449)	3.33 - 64.62
	T.t.d.	0.51(±0.073)	0.04 - 2.47	0.29(±0.033)	0.03 - 1.12	13.43(±1.160)	5.00 - 60.00
	T.s.	0.13(±0.022)	0.04 - 0.22	0.06(±0.005)	0.04 - 0.09	16.85(±2.211)	8.33 - 25.56
O[w]	*T.a.*[x]	0.73(±0.034)	0.00 - 7.19	0.20(±0.009)	0.00 - 1.95	26.11(±0.418)	3.33 - 71.82
	T.t.d.[y]	0.78(±0.069)	0.04 - 3.00	0.39(±0.026)	0.03 - 1.15	19.40(±0.968)	5.00 - 60.00
	T.s.[z]	0.24(±0.055)	0.04 - 0.91	0.07(±0.006)	0.04 - 0.12	29.23(±4.234)	8.33 - 62.22

[a] Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[w] O = overall (all cultivars of wheat and control)
[x] *T.a. = T. aestivum*
[y] *T.t.d. = T. turgidum* ssp. *durum*
[z] *T.s. = T. spelta*

Appendix B. Mean disease reaction scores for Growth Chamber Experiment (non-modeled, non-transformed data)

Cultivars

Cultivar	% Scl[a]	Scl / Spike[b]	Wt / Scl[c]	HD[d]	Scl Size[e]	% Ab[f]
5500HR	69.01	12.28	48.93	4.3	2.7	3.33
5600HR	61.96	11.56	34.30	3.8	2.5	6.67
5601HR	49.78	10.28	29.10	3.3	2.1	7.50
5602HR	53.16	10.44	44.00	3.2	2.6	2.22
5603HR	53.02	8.55	53.89	4.5	2.9	5.00
5604HRCL	47.45	6.83	51.69	3.2	2.8	10.83
5700PR	25.67	5.72	38.84	3.7	2.2	6.39
5701PR	52.57	10.61	39.60	3.9	2.5	2.03
5702PR	67.22	12.50	49.77	3.6	2.7	7.50
859CL	73.64	13.50	43.71	3.8	2.7	5.55
AC Abbey	83.50	15.06	41.16	4.4	2.7	8.09
AC Andrew	61.21	12.72	39.65	4.3	2.4	5.56
AC Avonlea	32.02	5.61	53.00	3.8	2.1	12.22
AC Barrie	68.98	13.39	36.39	3.3	2.6	5.55
AC Cadillac	66.77	12.33	47.62	3.9	2.8	4.17
AC Crystal	58.52	10.00	59.51	4.7	2.8	8.61
AC Elsa	46.35	7.89	54.47	3.4	2.9	4.72
AC Foremost	37.65	6.51	57.52	4.5	2.6	5.31
AC Intrepid	45.57	9.39	39.98	4.1	2.6	1.95
AC Morse	48.13	9.33	35.50	2.4	2.1	13.43
AC Splendor	50.42	9.17	47.07	3.5	2.8	4.17
AC Superb	33.45	4.89	74.49	3.7	2.9	4.76
AC Taber	59.44	10.11	47.40	4.6	2.8	7.50
AC Vista	63.01	11.39	57.22	3.1	2.8	5.28
Alvena	47.94	9.78	38.53	2.9	2.5	7.50
Bhishaj	49.95	8.39	63.10	4.0	2.9	7.50
Brigade	65.97	9.67	71.72	4.5	2.8	13.04
Burnside	54.72	11.28	44.27	3.0	2.8	3.06
CDC VR Morris	51.51	10.83	31.33	4.1	2.4	4.17
Carberry	31.20	4.02	80.02	3.2	2.9	5.11
CDC Abound	42.75	7.83	46.15	3.1	2.6	7.78
CDC Alsask	46.00	8.84	37.89	3.5	2.5	5.28
CDC Bounty	42.83	8.50	41.75	3.5	2.7	5.28
CDC Go	37.76	6.36	53.85	3.3	2.7	9.83
CDC Imagine	52.71	9.22	43.57	4.1	2.7	6.39
CDC Kernen	79.33	14.50	44.27	3.5	2.6	7.78
CDC Merlin	67.41	10.95	61.45	4.4	2.9	6.94
CDC Origin	46.17	7.39	34.28	2.0	2.0	28.06
CDC Osler	70.22	13.11	38.30	4.3	2.7	6.67
CDC Rama	57.29	11.89	43.75	3.0	2.6	5.00
CDC Stanley	59.39	11.17	37.85	3.9	2.6	4.72
CDC Teal	42.68	8.67	38.11	3.3	2.6	6.67

CDC Thrive	54.16	10.33	41.90	3.9	2.7	3.89
CDC Utmost	67.01	12.00	49.50	3.9	2.9	5.56
CDC Verona	53.58	8.44	48.71	3.3	2.2	16.04
CDC Walrus	46.15	9.66	47.04	3.2	2.6	4.44
CDC Zorba	23.11	4.67	31.58	1.6	1.7	28.06
CDN Bison	51.63	10.45	45.14	3.4	2.8	3.61
Commander	42.22	8.95	35.73	2.4	1.7	18.89
Eatonia	63.45	10.72	46.46	3.3	2.9	9.79
Enterprise	38.26	7.44	54.98	3.5	2.4	6.94
Eurostar	11.86	2.66	21.92	1.3	1.3	38.98
Fieldstar VB	54.19	8.78	41.13	4.4	2.6	10.00
Gazelle	34.50	1.88	126.15	4.9	2.8	28.08
Glencross VB	55.11	10.72	48.16	3.6	2.7	4.72
Glenlea	50.78	9.72	60.45	3.2	2.8	4.72
Glenn	32.79	4.23	70.58	2.8	2.9	5.11
Goodeve VB	46.19	9.83	31.49	3.7	2.6	6.94
Harvest	37.60	6.17	58.46	4.4	2.7	5.28
Helios	37.91	7.67	38.51	3.4	2.5	2.22
AC Conquer VB	79.45	14.00	60.58	4.9	2.9	4.45
SY985	72.71	14.17	45.73	3.4	2.6	4.17
Infinity	63.53	11.00	44.52	4.6	2.8	7.50
Journey	31.28	5.44	39.21	2.8	2.6	10.83
KANE	56.29	9.06	55.19	4.8	2.6	6.39
Katepwa	65.68	11.72	45.23	4.2	2.8	6.11
Kyle	37.88	8.00	35.59	3.2	1.9	20.28
Lillian	64.15	12.44	34.46	4.3	2.5	7.78
Lovitt	51.71	9.91	42.61	3.1	2.8	6.67
Marquis	54.98	8.17	60.89	3.8	2.9	9.72
McKenzie	50.66	7.11	55.34	4.5	2.8	5.83
Minnedosa	56.78	10.06	62.06	3.0	2.9	5.28
Muchmore	26.85	4.33	53.40	4.1	2.8	7.22
Napoleon	39.01	6.45	39.62	3.6	2.1	30.09
AC Navigator	56.75	9.39	42.47	4.1	2.0	19.36
NRG003	67.37	10.39	73.57	3.2	2.9	12.78
NRG010	68.68	11.67	66.50	3.0	2.9	10.00
Prodigy	55.24	7.17	62.08	2.6	2.9	22.78
PT559	50.31	8.56	47.20	3.4	2.6	11.94
CDC Plentiful	57.32	9.72	50.16	3.7	2.8	9.17
Red Fife	61.54	10.22	45.14	3.0	2.7	15.55
Roblin	69.13	13.05	40.64	4.0	2.7	2.78
Sadash	51.84	8.67	64.98	4.5	2.7	10.33
Sandro	60.48	7.87	107.62	5.0	3.0	6.89
Shaw	48.68	8.78	49.81	4.2	2.7	3.89
Snowbird	52.94	10.22	30.68	3.6	2.5	9.72
Snowstar	30.41	5.11	41.32	3.6	2.4	6.11
Somerset	65.29	13.00	30.74	4.5	2.5	4.17
Stettler	36.57	6.44	49.29	3.2	2.8	8.33
Strongfield	18.87	5.17	29.29	3.3	1.5	15.61
Transcend	26.07	5.00	39.85	3.2	1.9	16.67
Unity VB	64.23	8.33	57.20	4.8	2.8	14.42

Vesper	51.81	9.06	48.10	2.8	2.8	10.00
Waskada	33.29	6.00	45.32	3.6	2.6	6.45
Mean	**51.5**	**9.27**	**47.13**	**3.6**	**2.6**	**8.95**

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia/ (weight grain + sclerotia)
[b] Scl / Spike = Sclerotia/spike = number of sclerotia / number of spikes
[c] Wt / Scl = Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[d] HD = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)
[e] Scl Size = sclerotial size (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)
[f] % Ab = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)

Species

		Mean (±SE)	Range
% Scl[a]	*T. aestivum*	53.82(± 1.051)	12.13 - 91.89
	T. turgidum ssp. *durum*	39.22(± 3.133)	2.28 - 86.38
	T. spelta	34.64(± 5.578)	20.64 - 50.64
Scl/Spike[b]	*T. aestivum*	9.67(± 0.203)	2.67 - 16.50
	T. turgidum ssp. *durum*	7.18 (± 0.456)	1.33 - 12.50
	T. spelta	6.03(± 0.707)	4.00 - 8.33
Wt/Scl[c]	*T. aestivum*	48.22(± 0.950)	24.55 - 108.24
	T. turgidum ssp. *durum*	42.37(± 2.752)	10.00 - 88.80
	T. spelta	32.93(± 1.706)	25.36 - 37.50
HD[d]	*T. aestivum*	3.7(± 0.04)	2.3 - 5.0
	T. turgidum ssp. *durum*	3.2(± 0.16)	1.2 - 4.8
	T. spelta	1.8(± 0.11)	1.5 - 2.2
Scl Size[e]	*T. aestivum*	2.7(± 0.02)	1.8 - 3.0
	T. turgidum ssp. *durum*	2.0(± 0.08)	0.9 - 3.0
	T. spelta	1.8(± 0.07)	1.6 - 2.0
% Ab[f]	*T. aestivum*	6.78(± 0.318)	0.00 - 36.67
	T. turgidum ssp. *durum*	18.46(± 1.700)	4.17 - 42.50
	T. spelta	28.06(± 2.516)	21.67 - 36.67

[a] % Scl = Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)
[b] Scl / Spike = Sclerotia/spike = number of sclerotia / number of spikes
[c] Wt / Scl = Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[d] HD = production of honeydew (scale of 1 to 5, where 1 = none, 2 = confined within the glumes, 3 = exuding from florets in small drops, and 4 = exuding from florets in large drops, and 5 = large drops running down the spike)
[e] Scl Size = sclerotial size (scale of 1 to 3, where 1 = smaller than normal kernel, 2 = same size as kernel, and 3 = larger than kernel)
[f] % Ab = Percent of florets aborted = 100 * (florets aborted / total inoculated florets)

Appendix C. Cultivar estimates and letter separation after data modeling for Field Experiment #2 in 2011 and 2012 grown at the North Seed Farm, Saskatoon, SK.

2011 Field Experiment

Percent Sclerotia [a]				Sclerotia / Spike [b]				Weight / Sclerotium [c]			
	Cultivar	Estimate [w]			Cultivar	Estimate [x]			Cultivar	Estimate [y]	
1	Glenlea	3.32	A [z]	1	Glenlea	0.90	A	1	Marquis	47.85	A
2	SY985	2.60	B	2	Enterprise	0.83	AB	2	KANE	46.56	AB
3	5702PR	2.52	BC	3	5702PR	0.78	ABC	3	CDC Zorba	46.22	AB
4	CDN Bison	2.41	BCD	4	SY985	0.78	ABC	4	CDN Bison	44.25	AB
5	Burnside	2.30	CDE	5	Burnside	0.77	ABC	5	Glenlea	42.26	ABC
6	Red Fife	2.29	CDE	6	Red Fife	0.75	ABCD	6	CDC Walrus	41.86	ABC
7	AC Navigator	2.28	DE	7	AC Navigator	0.75	ABCD	7	Unity VB	41.48	ABC
8	CDC Merlin	2.15	EF	8	CDN Bison	0.61	ABCDE	8	SY985	40.91	ABC
9	5601HR	2.02	F	9	CDC Verona	0.60	ABCDEF	9	5500HR	40.60	ABC
10	Enterprise	1.95	FG	10	Napoleon	0.56	ABCDEFG	10	CDC Bounty	40.50	ABC
11	Somerset	1.80	GH	11	AC Avonlea	0.54	ABCDEFGH	11	5702PR	38.70	ABC
12	5701PR	1.75	GH	12	5701PR	0.49	ABCDEFGHI	12	Minnedosa	38.47	ABC
13	5500HR	1.60	HI	13	AC Morse	0.47	ABCDEFGHI	13	CDC Go	38.44	ABC
14	AC Morse	1.49	IJ	14	Sadash	0.45	ABCDEFGHI	14	Burnside	38.10	ABC
15	Napoleon	1.46	IJ	15	CDC Merlin	0.44	BCDEFGHI	15	Fieldstar VB	38.06	ABC
16	5600HR	1.41	IJK	16	Transcend	0.43	BCDEFGHI	16	PT559	36.63	ABC
17	5700PR	1.38	IJK	17	5700PR	0.43	BCDEFGHI	17	AC Cadillac	36.55	ABC
18	Glencross VB	1.38	JK	18	Somerset	0.42	BCDEFGHI	18	AC Morse	36.53	ABC
19	CDC Bounty	1.29	JKL	19	Kyle	0.41	BCDEFGHI	19	CDC Rama	36.46	ABC
20	Marquis	1.23	KLM	20	Brigade	0.41	BCDEFGHI	20	5600HR	36.43	ABC
21	CDC Walrus	1.22	KLMN	21	5601HR	0.41	BCDEFGHI	21	5603HR	36.21	ABC
22	Roblin	1.22	KLMN	22	CDC Walrus	0.38	CDEFGHI	22	CDC Abound	35.99	ABC
23	Unity VB	1.15	LMNO	23	AC Conquer VB	0.36	CDEFGHI	23	5700PR	35.99	ABC
24	AC Elsa	1.13	LMNO	24	CDC VR Morris	0.35	CDEFGHI	24	Red Fife	35.98	ABC

25	CDC Imagine	1.11	LMNO	25	Glencross VB	0.35	CDEFGHI	25	Shaw	35.95	ABC
26	Infinity	1.09	LMNO	26	CDC Imagine	0.34	CDEFGHI	26	McKenzie	35.88	ABC
27	CDC VR Morris	1.07	LMNOP	27	AC Andrew	0.33	CDEFGHI	27	5701PR	35.86	ABC
28	5603HR	1.03	MNOPQ	28	5600HR	0.30	DEFGHI	28	NRG003	35.32	ABC
29	AC Intrepid	1.01	MNOPQR	29	Infinity	0.29	EFGHI	29	CDC Merlin	35.21	ABC
30	CDC Verona	1.00	NOPQRS	30	5500HR	0.29	EFGHI	30	CDC Thrive	34.98	ABC
31	Snowbird	0.98	OPQRS	31	5603HR	0.29	EFGHI	31	CDC Kernen	34.89	ABC
32	McKenzie	0.86	PQRST	32	Roblin	0.26	EFGHI	32	CDC Alsask	34.56	ABC
33	AC Avonlea	0.83	QRSTU	33	CDC Stanley	0.25	EFGHI	33	859CL	34.25	ABC
34	AC Conquer VB	0.81	QRSTUV	34	Bhishaj	0.25	EFGHI	34	Infinity	34.13	ABC
35	CDC Stanley	0.81	RSTUVW	35	AC Elsa	0.25	EFGHI	35	AC Splendor	34.09	ABC
36	Lovitt	0.80	RSTUVW	36	Strongfield	0.24	EFGHI	36	Somerset	34.09	ABC
37	CDC Teal	0.77	STUVWX	37	AC Intrepid	0.24	EFGHI	37	AC Vista	33.92	ABC
38	CDC Osler	0.74	TUVWXY	38	CDC Bounty	0.23	EFGHI	38	Katepwa	33.88	ABC
39	Eatonia	0.73	TUVWXYZ	39	Snowbird	0.23	EFGHI	39	CDC Origin	33.55	ABC
40	859CL	0.72	TUVWXYZ	40	CDC Teal	0.23	EFGHI	40	CDC Imagine	33.01	ABC
41	Carberry	0.72	TUVWXYZ(2)A	41	Commander	0.22	EFGHI	41	Journey	32.95	ABC
42	Harvest	0.67	TUVWXYZ(2)AB	42	AC Crystal	0.22	EFGHI	42	Carberry	32.88	ABC
43	Sadash	0.67	TUVWXYZ(2)ABC	43	AC Taber	0.21	EFGHI	43	Eatonia	32.56	ABC
44	Transcend	0.65	TUVWXYZ(2)ABCD	44	Minnedosa	0.21	EFGHI	44	Goodeve VB	32.15	ABC
45	AC Cadillac	0.65	TUVWXYZ(2)ABCD	45	Marquis	0.21	EFGHI	45	Vesper	31.98	ABC
46	Helios	0.65	TUVWXYZ(2)ABCD	46	Lovitt	0.20	EFGHI	46	Brigade	31.98	ABC
47	Lillian	0.64	UVWXYZ(2)ABCDE	47	Eurostar	0.19	EFGHI	47	AC Intrepid	31.58	ABC
48	Minnedosa	0.63	UVWXYZ(2)ABCDEF	48	CDC Thrive	0.19	EFGHI	48	5601HR	31.54	ABC
49	5602HR	0.62	UVWXYZ(2)ABCDEF	49	CDC Kernen	0.19	EFGHI	49	Harvest	31.53	ABC
50	Fieldstar VB	0.62	UVWXYZ(2)ABCDEFG	50	CDC Osler	0.18	EFGHI	50	Helios	31.51	ABC
51	Katepwa	0.60	VWXYZ(2)ABCDEFG	51	Harvest	0.17	EFGHI	51	Snowbird	31.30	ABC
52	CDC Kernen	0.58	WXYZ(2)ABCDEFGH	52	Lillian	0.17	EFGHI	52	5602HR	31.29	ABC
53	Kyle	0.58	WXYZ(2)ABCDEFGH	53	Unity VB	0.17	EFGHI	53	Enterprise	31.14	ABC
54	Goodeve VB	0.56	XYZ(2)ABCDEFGHI	54	AC Foremost	0.17	EFGHI	54	Snowstar	31.07	ABC
55	Snowstar	0.55	XYZ(2)ABCDEFGHI	55	5602HR	0.16	EFGHI	55	Muchmore	30.98	ABC
56	AC Foremost	0.55	XYZ(2)ABCDEFGHI	56	NRG003	0.16	EFGHI	56	AC Foremost	30.58	ABC
57	Strongfield	0.53	YZ(2)ABCDEFGHIJ	57	CDC Rama	0.16	EFGHI	57	Lovitt	30.22	ABC

58	KANE	0.52	YZ(2)ABCDEFGHIJ	58	859CL	0.15	EFGHI	58	CDC VR Morris	29.49	ABC
59	AC Andrew	0.52	YZ(2)ABCDEFGHIJK	59	NRG010	0.15	FGHI	59	Napoleon	29.43	ABC
60	Eurostar	0.51	YZ(2)ABCDEFGHIJK	60	McKenzie	0.15	FGHI	60	Glenn	29.10	ABC
61	CDC Origin	0.51	Z(2)ABCDEFGHIJKL	61	CDC Alsask	0.15	FGHI	61	CDC Teal	29.08	ABC
62	Muchmore	0.51	Z(2)ABCDEFGHIJKL	62	Helios	0.15	FGHI	62	Glencross VB	28.75	ABC
63	CDC Abound	0.50	Z(2)ABCDEFGHIJKL	63	Muchmore	0.15	FGHI	63	CDC Stanley	28.19	ABC
64	Brigade	0.50	Z(2)ABCDEFGHIJKL	64	AC Barrie	0.14	FGHI	64	Waskada	28.18	ABC
65	Shaw	0.50	(2)ABCDEFGHIJKLM	65	CDC Abound	0.14	GHI	65	Lillian	27.41	ABC
66	CDC Alsask	0.49	(2)ABCDEFGHIJKLM	66	CDC Origin	0.13	GHI	66	Eurostar	27.40	ABC
67	CDC Go	0.47	(2)BCDEFGHIJKLM	67	Eatonia	0.13	GHI	67	Roblin	27.31	ABC
68	NRG003	0.47	(2)BCDEFGHIJKLM	68	AC Superb	0.13	GHI	68	AC Navigator	27.27	ABC
69	Bhishaj	0.47	(2)BCDEFGHIJKLM	69	AC Cadillac	0.12	GHI	69	AC Conquer VB	26.76	ABC
70	CDC Thrive	0.45	(2)BCDEFGHIJKLM	70	Katepwa	0.12	GHI	70	AC Elsa	26.13	ABC
71	AC Taber	0.44	(2)CDEFGHIJKLMN	71	Glenn	0.12	GHI	71	AC Taber	25.55	ABC
72	AC Barrie	0.44	(2)CDEFGHIJKLMN	72	AC Abbey	0.12	GHI	72	AC Superb	25.31	ABC
73	CDC Zorba	0.44	(2)DEFGHIJKLMN	73	Carberry	0.12	GHI	73	CDC Osler	25.22	ABC
74	NRG010	0.42	(2)EFGHIJKLMNO	74	Goodeve VB	0.11	GHI	74	5604HR CL	25.07	ABC
75	PT559	0.42	(2)EFGHIJKLMNO	75	AC Vista	0.11	GHI	75	AC Abbey	25.07	ABC
76	CDC Rama	0.41	(2)EFGHIJKLMNO	76	Fieldstar VB	0.11	GHI	76	Alvena	24.41	ABC
77	Journey	0.41	(2)EFGHIJKLMNO	77	Snowstar	0.10	HI	77	Stettler	24.37	ABC
78	AC Vista	0.41	(2)FGHIJKLMNO	78	PT559	0.10	HI	78	AC Barrie	24.18	ABC
79	AC Abbey	0.39	(2)GHIJKLMNO	79	Shaw	0.10	HI	79	Prodigy	24.04	ABC
80	Glenn	0.39	(2)GHIJKLMNO	80	Journey	0.10	HI	80	NRG010	23.29	ABC
81	AC Superb	0.37	(2)HIJKLMNO	81	CDC Zorba	0.10	HI	81	CDC Verona	23.21	ABC
82	Alvena	0.36	(2)IJKLMNO	82	Stettler	0.09	HI	82	AC Avonlea	22.22	ABC
83	CDC Plentiful	0.35	(2)IJKLMNO	83	Prodigy	0.09	HI	83	Kyle	21.52	ABC
84	Commander	0.33	(2)IJKLMNO	84	KANE	0.09	HI	84	CDC Plentiful	19.85	ABC
85	5604HR CL	0.33	(2)JKLMNO	85	CDC Go	0.09	HI	85	CDC Utmost	19.84	ABC
86	Waskada	0.31	(2)JKLMNO	86	Waskada	0.09	I	86	AC Crystal	19.49	ABC
87	AC Crystal	0.30	(2)JKLMNO	87	CDC Plentiful	0.09	I	87	Strongfield	19.24	ABC
88	Prodigy	0.30	(2)KLMNO	88	Alvena	0.08	I	88	Transcend	19.15	ABC
89	AC Splendor	0.29	(2)LMNO	89	5604HR CL	0.08	I	89	AC Andrew	18.13	ABC
90	Stettler	0.27	(2)MNO	90	AC Splendor	0.06	I	90	Bhishaj	16.59	BC

91	Vesper	0.23	(2)NO	91	CDC Utmost	0.06	I	91	Commander	16.27	BC
92	CDC Utmost	0.21	(2)O	92	Vesper	0.04	I	92	Sadash	13.14	C

[a] Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[w] SE = 0.104, [x] SE = 0.078, [y] SE = 5.094
[z] Means with different letters are considered to be significantly different from each other

2012 Field Experiment

Percent Sclerotia [a]				Sclerotia / Spike [b]				Weight / Sclerotium [c]			
	Cultivar	Estimate [w]			Cultivar	Estimate [x]			Cultivar	Estimate [y]	
1	Glenlea	3.93	A [z]	1	Glenlea	0.79	A	1	CDC Imagine	35.14	A
2	Marquis	2.72	AB	2	Burnside	0.65	AB	2	AC Superb	32.29	AB
3	5601HR	1.86	BC	3	Brigade	0.60	ABC	3	5603HR	30.43	ABC
4	Brigade	1.50	BC	4	Red Fife	0.56	ABCD	4	Glenlea	29.16	ABCD
5	CDC Imagine	1.48	BC	5	Marquis	0.55	ABCDE	5	Glencross VB	28.92	ABCD
6	Red Fife	1.46	BC	6	AC Navigator	0.54	ABCDEF	6	Unity VB	27.81	ABCD
7	CDC Stanley	1.45	BC	7	Sadash	0.45	ABCDEF	7	CDC Walrus	27.33	ABCD
8	5702PR	1.44	BC	8	5702PR	0.41	ABCDEF	8	AC Conquer VB	26.56	ABCD
9	Somerset	1.27	BC	9	AC Taber	0.39	ABCDEF	9	AC Intrepid	25.64	ABCD
10	AC Navigator	1.24	BC	10	5601HR	0.35	ABCDEF	10	CDC Bounty	25.63	ABCD
11	Infinity	1.03	BC	11	CDC Stanley	0.34	ABCDEF	11	5602HR	25.46	ABCD
12	Burnside	1.02	BC	12	Transcend	0.33	ABCDEF	12	CDC Rama	25.43	ABCD
13	CDC Merlin	1.00	BC	13	Enterprise	0.32	ABCDEF	13	AC Taber	25.21	ABCD
14	Snowbird	0.95	BC	14	Commander	0.31	ABCDEF	14	Katepwa	24.88	ABCD
15	Commander	0.89	BC	15	Kyle	0.28	ABCDEF	15	CDN Bison	24.85	ABCD
16	Lovitt	0.87	BC	16	SY985	0.28	ABCDEF	16	Burnside	24.62	ABCD
17	Sadash	0.87	BC	17	CDC Imagine	0.27	ABCDEF	17	McKenzie	24.49	ABCD
18	PT559	0.85	BC	18	PT559	0.26	BCDEF	18	CDC Abound	24.16	ABCD
19	CDN Bison	0.83	BC	19	AC Avonlea	0.25	BCDEF	19	AC Cadillac	23.72	ABCD
20	5600HR	0.81	BC	20	Snowbird	0.24	BCDEF	20	Eatonia	23.40	ABCD
21	AC Taber	0.67	C	21	AC Andrew	0.21	BCDEF	21	5702PR	23.38	ABCD

22	CDC Teal	0.67	C	22	5701PR	0.20	BCDEF	22	Somerset	23.34	ABCD
23	SY985	0.67	C	23	Infinity	0.20	BCDEF	23	NRG003	23.27	ABCD
24	CDC Bounty	0.63	C	24	NRG010	0.20	BCDEF	24	Lovitt	23.03	ABCD
25	5701PR	0.58	C	25	Roblin	0.20	BCDEF	25	AC Vista	22.93	ABCD
26	NRG003	0.55	C	26	Somerset	0.20	BCDEF	26	5500HR	22.47	ABCD
27	AC Barrie	0.55	C	27	CDC VR Morris	0.20	BCDEF	27	CDC Go	22.46	ABCD
28	Kyle	0.55	C	28	5700PR	0.20	BCDEF	28	CDC VR Morris	22.44	ABCD
29	AC Intrepid	0.52	C	29	Strongfield	0.19	BCDEF	29	Minnedosa	22.29	ABCD
30	CDC VR Morris	0.52	C	30	CDN Bison	0.19	BCDEF	30	CDC Teal	22.18	ABCD
31	Roblin	0.48	C	31	AC Crystal	0.18	BCDEF	31	Red Fife	22.02	ABCD
32	Transcend	0.48	C	32	NRG003	0.18	BCDEF	32	859CL	21.28	ABCD
33	5700PR	0.46	C	33	5600HR	0.17	BCDEF	33	CDC Alsask	21.11	ABCD
34	McKenzie	0.46	C	34	Lovitt	0.17	BCDEF	34	Infinity	21.01	ABCD
35	Enterprise	0.45	C	35	CDC Merlin	0.17	BCDEF	35	5600HR	20.88	ABCD
36	CDC Walrus	0.44	C	36	CDC Walrus	0.17	BCDEF	36	Helios	20.82	ABCD
37	CDC Verona	0.44	C	37	CDC Verona	0.16	BCDEF	37	Alvena	19.96	ABCD
38	CDC Thrive	0.44	C	38	Glencross VB	0.16	BCDEF	38	Enterprise	19.55	ABCD
39	Lillian	0.44	C	39	CDC Kernen	0.16	BCDEF	39	SY985	19.41	ABCD
40	AC Avonlea	0.43	C	40	CDC Teal	0.16	BCDEF	40	5601HR	19.31	ABCD
41	5500HR	0.42	C	41	CDC Thrive	0.14	BCDEF	41	Shaw	19.27	ABCD
42	Eatonia	0.41	C	42	Harvest	0.13	BCDEF	42	Snowstar	19.17	ABCD
43	Unity VB	0.40	C	43	AC Conquer VB	0.13	CDEF	43	Harvest	19.17	ABCD
44	AC Conquer VB	0.40	C	44	Napoleon	0.13	CDEF	44	CDC Osler	19.10	ABCD
45	Minnedosa	0.39	C	45	5500HR	0.13	CDEF	45	5700PR	19.01	ABCD
46	Snowstar	0.38	C	46	Minnedosa	0.12	CDEF	46	AC Barrie	18.98	ABCD
47	Glencross VB	0.38	C	47	CDC Bounty	0.12	CDEF	47	CDC Thrive	18.46	ABCD
48	Harvest	0.37	C	48	AC Foremost	0.12	CDEF	48	Lillian	18.14	ABCD
49	Goodeve VB	0.37	C	49	Eurostar	0.12	CDEF	49	Snowbird	18.02	ABCD
50	AC Vista	0.37	C	50	AC Vista	0.11	CDEF	50	AC Splendor	17.99	ABCD
51	5603HR	0.37	C	51	Journey	0.11	CDEF	51	KANE	17.75	ABCD
52	859CL	0.35	C	52	Unity VB	0.11	CDEF	52	CDC Kernen	17.70	ABCD

53	CDC Plentiful	0.35	C	53	AC Intrepid	0.11	CDEF	53	CDC Stanley	17.64	ABCD
54	CDC Osler	0.32	C	54	CDC Go	0.11	CDEF	54	5701PR	17.33	ABCD
55	AC Cadillac	0.31	C	55	859CL	0.11	CDEF	55	Goodeve VB	17.29	ABCD
56	CDC Go	0.31	C	56	AC Morse	0.10	CDEF	56	Journey	17.23	ABCD
57	CDC Alsask	0.30	C	57	AC Barrie	0.10	CDEF	57	CDC Origin	17.18	ABCD
58	Katepwa	0.30	C	58	Snowstar	0.10	CDEF	58	CDC Merlin	17.09	ABCD
59	CDC Kernen	0.29	C	59	Goodeve VB	0.10	CDEF	59	Brigade	16.49	ABCD
60	AC Foremost	0.29	C	60	Muchmore	0.10	CDEF	60	CDC Plentiful	16.48	ABCD
61	Helios	0.29	C	61	CDC Plentiful	0.10	CDEF	61	Muchmore	16.29	ABCD
62	AC Crystal	0.28	C	62	5603HR	0.09	CDEF	62	Stettler	15.99	ABCD
63	Strongfield	0.28	C	63	Lillian	0.09	CDEF	63	Waskada	15.94	ABCD
64	Muchmore	0.27	C	64	Shaw	0.09	CDEF	64	PT559	15.76	ABCD
65	Journey	0.27	C	65	CDC Osler	0.09	CDEF	65	Sadash	15.70	ABCD
66	CDC Abound	0.27	C	66	CDC Alsask	0.09	CDEF	66	NRG010	15.51	ABCD
67	NRG010	0.25	C	67	McKenzie	0.09	CDEF	67	CDC Utmost	15.45	ABCD
68	AC Morse	0.25	C	68	CDC Origin	0.09	CDEF	68	Roblin	15.41	ABCD
69	AC Superb	0.24	C	69	AC Cadillac	0.09	CDEF	69	AC Morse	15.21	ABCD
70	AC Elsa	0.23	C	70	CDC Abound	0.08	CDEF	70	CDC Zorba	14.74	ABCD
71	KANE	0.22	C	71	CDC Rama	0.08	CDEF	71	5604HR CL	14.16	ABCD
72	AC Andrew	0.21	C	72	AC Elsa	0.08	CDEF	72	AC Foremost	13.89	ABCD
73	5602HR	0.19	C	73	KANE	0.08	CDEF	73	AC Crystal	13.88	ABCD
74	Shaw	0.16	C	74	Katepwa	0.08	CDEF	74	Commander	13.75	ABCD
75	CDC Origin	0.15	C	75	Eatonia	0.08	CDEF	75	AC Abbey	13.34	ABCD
76	Alvena	0.14	C	76	5602HR	0.07	CDEF	76	Prodigy	13.33	ABCD
77	Carberry	0.14	C	77	5604HR CL	0.07	DEF	77	Carberry	13.31	ABCD
78	5604HR CL	0.14	C	78	Helios	0.07	DEF	78	Marquis	13.17	ABCD
79	Glenn	0.13	C	79	CDC Zorba	0.07	DEF	79	AC Elsa	13.08	ABCD
80	Napoleon	0.13	C	80	Carberry	0.07	DEF	80	Fieldstar VB	12.76	BCD
81	CDC Rama	0.12	C	81	AC Superb	0.06	DEF	81	CDC Verona	12.63	BCD
82	Eurostar	0.11	C	82	Glenn	0.06	DEF	82	Eurostar	12.58	BCD
83	Stettler	0.10	C	83	Fieldstar VB	0.06	DEF	83	Glenn	12.40	BCD
84	CDC Zorba	0.10	C	84	CDC Utmost	0.05	DEF	84	Strongfield	12.37	BCD
85	CDC Utmost	0.09	C	85	Alvena	0.04	DEF	85	Vesper	11.49	BCD

86	Fieldstar VB	0.09	C	86	Stettler	0.04	DEF	86	Kyle	11.22	BCD
87	AC Splendor	0.06	C	87	AC Abbey	0.04	EF	87	Bhishaj	10.85	BCD
88	Waskada	0.05	C	88	Bhishaj	0.04	EF	88	AC Andrew	10.71	BCD
89	Prodigy	0.04	C	89	Waskada	0.04	EF	89	Transcend	10.70	BCD
90	Bhishaj	0.04	C	90	Prodigy	0.03	EF	90	AC Navigator	10.15	BCD
91	AC Abbey	0.04	C	91	AC Splendor	0.03	EF	91	AC Avonlea	8.69	CD
92	Vesper	0.03	C	92	Vesper	0.02	F	92	Napoleon	7.43	D

[a] Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[w] SE = 0.331, [x] SE = 0.089, [y] SE = 3.664
[z] Means with different letters are considered to be significantly different from each other

Appendix D. Cultivar estimates and letter separation after data modeling for the growth chamber experiment.

Sclerotia Variables

Percent Sclerotia [a]				Sclerotia / Spike [b]				Weight / Sclerotium [c]			
	Cultivar	Estimate [w]			Cultivar	Estimate [x]			Cultivar	Estimate^2 [y]	
1	AC Abbey	86.23	A [z]	1	AC Abbey	15.16	A	1	NRG003	72.25	A
2	CDC Kernen	78.32	AB	2	CDC Kernen	14.09	AB	2	Carberry	71.40	A
3	AC Conquer VB	77.84	ABC	3	SY985	13.94	ABC	3	Brigade	69.89	A
4	NRG010	77.12	ABCD	4	AC Conquer VB	13.74	ABCD	4	AC Superb	69.22	A
5	CDC Osler	74.29	ABCDE	5	859CL	13.68	ABCDE	5	Glenn	65.61	B
6	Roblin	73.40	ABCDEF	6	AC Barrie	13.41	ABCDEF	6	Sadash	62.09	C
7	859CL	73.10	ABCDEFG	7	Roblin	13.15	ABCDEFG	7	AC Crystal	60.22	CD
8	CDC Merlin	71.01	ABCDEFG	8	NRG010	13.11	ABCDEFG	8	Marquis	60.06	CD
9	AC Barrie	69.48	ABCDEFGH	9	CDC Osler	12.92	ABCDEFGH	9	AC Conquer VB	60.06	CD
10	CDC Utmost	68.97	ABCDEFGH	10	5702PR	12.68	ABCDEFGHI	10	NRG010	59.60	CD
11	5500HR	68.42	ABCDEFGHI	11	CDC Utmost	12.55	ABCDEFGHIJ	11	Bhishaj	59.60	CD
12	SY985	67.73	ABCDEFGHIJ	12	Somerset	12.48	ABCDEFGHIJ	12	Glenlea	58.98	CD
13	AC Crystal	66.64	ABCDEFGHIJ	13	AC Andrew	12.46	ABCDEFGHIJ	13	CDC Merlin	58.98	CD
14	5702PR	66.44	ABCDEFGHIJK	14	5500HR	12.43	ABCDEFGHIJ	14	Minnedosa	58.22	DE
15	AC Taber	65.90	ABCDEFGHIJKL	15	Lillian	12.38	ABCDEFGHIJ	15	McKenzie	57.76	DEF
16	Infinity	64.00	ABCDEFGHIJKLM	16	CDC Rama	11.83	ABCDEFGHIJK	16	Unity VB	57.61	DEF
17	AC Vista	63.96	ABCDEFGHIJKLM	17	AC Vista	11.79	ABCDEFGHIJK	17	Harvest	57.46	DEFG
18	NRG003	63.69	ABCDEFGHIJKLMN	18	CDC Merlin	11.60	ABCDEFGHIJKL	18	Prodigy	57.15	DEFG
19	KANE	63.50	ABCDEFGHIJKLMNO	19	AC Cadillac	11.53	ABCDEFGHIJKLM	19	AC Foremost	55.50	EFGH
20	Unity VB	63.41	ABCDEFGHIJKLMNO	20	Infinity	11.44	ABCDEFGHIJKLMN	20	KANE	54.76	FGHI
21	Eatonia	63.36	ABCDEFGHIJKLMNO	21	Katepwa	11.20	ABCDEFGHIJKLMNO	21	Muchmore	54.32	GHI
22	Katepwa	63.30	ABCDEFGHIJKLMNO	22	CDC Stanley	11.18	ABCDEFGHIJKLMNOP	22	AC Vista	54.32	GHI
23	Somerset	62.00	ABCDEFGHIJKLMNOP	23	5600HR	11.12	ABCDEFGHIJKLMNOP	23	5603HR	53.58	HIJ
24	Lillian	61.65	ABCDEFGHIJKLMNOP	24	AC Crystal	10.78	ABCDEFGHIJKLMNOPQ	24	5604HR CL	52.27	IJK
25	AC Cadillac	60.18	ABCDEFGHIJKLMNOPQ	25	Glencross VB	10.75	ABCDEFGHIJKLMNOPQ	25	AC Elsa	50.84	JKL
26	Shaw	60.10	ABCDEFGHIJKLMNOPQ	26	AC Taber	10.65	ABCDEFGHIJKLMNOPQ	26	Enterprise	50.55	JKL
27	Brigade	59.64	ABCDEFGHIJKLMNOPQ	27	5701PR	10.59	ABCDEFGHIJKLMNOPQ	27	5500HR	50.41	KL

28	CDC Stanley	59.52	ABCDEFGHIJKLMNOPQ	28	CDN Bison	10.55	ABCDEFGHIJKLMNOPQ	28	CDC Plentiful	50.41	KL
29	AC Andrew	59.33	ABCDEFGHIJKLMNOPQ	29	Snowbird	10.49	ABCDEFGHIJKLMNOPQR	29	CDC Go	50.13	KLM
30	Marquis	58.84	ABCDEFGHIJKLMNOPQ	30	Burnside	10.47	ABCDEFGHIJKLMNOPQR	30	CDC Utmost	49.98	KLM
31	AC Splendor	58.65	ABCDEFGHIJKLMNOPQR	31	NRG003	10.36	ABCDEFGHIJKLMNOPQR	31	AC Taber	49.84	KLM
32	CDC Rama	57.10	ABCDEFGHIJKLMNOPQRS	32	Minnedosa	10.30	ABCDEFGHIJKLMNOPQRS	32	5702PR	49.42	KLMN
33	5701PR	56.84	ABCDEFGHIJKLMNOPQRS	33	Eatonia	10.26	BCDEFGHIJKLMNOPQRS	33	Shaw	49.42	KLMN
34	CDC Verona	55.72	ABCDEFGHIJKLMNOPQRS	34	AC Intrepid	10.18	BCDEFGHIJKLMNOPQRST	34	AC Avonlea	49.00	LMN
35	5600HR	55.65	ABCDEFGHIJKLMNOPQRS	35	5602HR	10.18	BCDEFGHIJKLMNOPQRST	35	Vesper	48.44	LMNO
36	Minnedosa	55.51	ABCDEFGHIJKLMNOPQRS	36	Glenlea	10.10	BCDEFGHIJKLMNOPQRST	36	Eatonia	48.44	LMNOP
37	Vesper	55.36	ABCDEFGHIJKLMNOPQRS	37	Lovitt	10.07	BCDEFGHIJKLMNOPQRST	37	AC Splendor	48.30	LMNOP
38	CDC Plentiful	55.28	ABCDEFGHIJKLMNOPQRST	38	Shaw	10.00	BCDEFGHIJKLMNOPQRST	38	CDC Walrus	47.33	MNOPQ
39	Glencross VB	55.11	ABCDEFGHIJKLMNOPQRST	39	KANE	9.98	BCDEFGHIJKLMNOPQRST	39	Stettler	47.20	MNOPQ
40	Red Fife	54.68	ABCDEFGHIJKLMNOPQRST	40	CDC VR Morris	9.94	BCDEFGHIJKLMNOPQRSTU	40	Red Fife	46.65	NOPQR
41	AC Morse	54.22	ABCDEFGHIJKLMNOPQRST	41	Goodeve VB	9.89	BCDEFGHIJKLMNOPQRSTU	41	CDC Verona	46.65	NOPQRS
42	AC Navigator	53.97	ABCDEFGHIJKLMNOPQRST	42	5601HR	9.79	BCDEFGHIJKLMNOPQRSTU	42	PT559	46.65	NOPQRS
43	Sadash	53.72	ABCDEFGHIJKLMNOPQRST	43	AC Splendor	9.76	BCDEFGHIJKLMNOPQRSTU	43	Glencross VB	45.97	OPQRST
44	Glenlea	53.61	ABCDEFGHIJKLMNOPQRST	44	AC Morse	9.72	BCDEFGHIJKLMNOPQRSTU	44	Katepwa	45.70	OPQRST
45	5603HR	53.50	ABCDEFGHIJKLMNOPQRST	45	CDC Thrive	9.71	BCDEFGHIJKLMNOPQRSTU	45	CDC Abound	45.70	OPQRSTU
46	CDN Bison	53.33	ABCDEFGHIJKLMNOPQRSTU	46	Alvena	9.51	BCDEFGHIJKLMNOPQRSTUV	46	Burnside	45.56	PQRSTUV
47	PT559	53.08	ABCDEFGHIJKLMNOPQRSTU	47	Vesper	9.51	BCDEFGHIJKLMNOPQRSTUV	47	AC Navigator	45.16	QRSTUVW
48	AC Intrepid	52.92	ABCDEFGHIJKLMNOPQRSTU	48	CDC Walrus	9.45	BCDEFGHIJKLMNOPQRSTUV	48	AC Cadillac	45.02	QRSTUVWX
49	Prodigy	52.16	ABCDEFGHIJKLMNOPQRSTU	49	CDC Plentiful	9.44	BCDEFGHIJKLMNOPQRSTUV	49	CDN Bison	44.49	QRSTUVWXY
50	Lovitt	51.88	ABCDEFGHIJKLMNOPQRSTU	50	Red Fife	9.31	BCDEFGHIJKLMNOPQRSTUV	50	CDC Kernen	44.09	RSTUVWXYZ
51	5602HR	51.48	BCDEFGHIJKLMNOPQRSTU	51	Sadash	9.25	BCDEFGHIJKLMNOPQRSTUVW	51	Fieldstar VB	43.69	STUVWXYZ
52	5601HR	51.27	BCDEFGHIJKLMNOPQRSTU	52	Commander	9.14	CDEFGHIJKLMNOPQRSTUVWX	52	CDC Rama	43.69	STUVWXYZ
53	Snowbird	50.92	BCDEFGHIJKLMNOPQRSTU	53	AC Navigator	8.95	DEFGHIJKLMNOPQRSTUVWXY	53	Infinity	43.43	TUVWXYZ(2)A
54	McKenzie	49.59	BCDEFGHIJKLMNOPQRSTU	54	CDC Imagine	8.86	DEFGHIJKLMNOPQRSTUVWXY	54	Lovitt	42.90	UVWXYZ(2)AB
55	Burnside	48.88	BCDEFGHIJKLMNOPQRSTU	55	5603HR	8.84	EFGHIJKLMNOPQRSTUVWXY	55	Waskada	42.77	VWXYZ(2)AB
56	Fieldstar VB	48.51	BCDEFGHIJKLMNOPQRSTUV	56	Brigade	8.82	EFGHIJKLMNOPQRSTUVWXY	56	Transcend	42.77	VWXYZ(2)AB
57	CDC Thrive	48.50	BCDEFGHIJKLMNOPQRSTUV	57	Kyle	8.79	FGHIJKLMNOPQRSTUVWXY	57	CDC Bounty	42.64	WXYZ(2)ABC
58	Goodeve VB	48.21	BCDEFGHIJKLMNOPQRSTUV	58	Marquis	8.76	FGHIJKLMNOPQRSTUVWXY	58	SY985	42.51	WXYZ(2)ABC
59	CDC Abound	47.67	BCDEFGHIJKLMNOPQRSTUV	59	PT559	8.76	FGHIJKLMNOPQRSTUVWXY	59	859CL	42.25	XYZ(2)ABCD
60	CDC Imagine	47.25	BCDEFGHIJKLMNOPQRSTUV	60	CDC Verona	8.58	FGHIJKLMNOPQRSTUVWXY	60	CDC Thrive	41.73	YZ(2)ABCDE
61	CDC Origin	47.17	BCDEFGHIJKLMNOPQRSTUV	61	CDC Teal	8.53	FGHIJKLMNOPQRSTUVWXY	61	CDC Imagine	41.60	Z(2)ABCDEF

62	CDC VR Morris	47.08	BCDEFGHIJKLMNOPQRSTUV	62	Unity VB	8.30	GHIJKLMNOPQRSTUVWXY	62	Roblin	41.47	Z(2)ABCDEF
63	Bhishaj	46.80	BCDEFGHIJKLMNOPQRSTUV	63	Bhishaj	8.30	GHIJKLMNOPQRSTUVWXY	63	Snowstar	41.34	Z(2)ABCDEF
64	Alvena	46.48	BCDEFGHIJKLMNOPQRSTUV	64	CDC Alsask	8.29	GHIJKLMNOPQRSTUVWXY	64	AC Abbey	40.96	(2)ABCDEFG
65	Harvest	45.31	BCDEFGHIJKLMNOPQRSTUV	65	CDC Bounty	8.20	HIJKLMNOPQRSTUVWXY	65	Journey	40.70	(2)BCDEFGH
66	CDC Walrus	45.22	BCDEFGHIJKLMNOPQRSTUV	66	CDC Abound	8.15	HIJKLMNOPQRSTUVWXY	66	5602HR	40.58	(2)BCDEFGH
67	Kyle	44.13	BCDEFGHIJKLMNOPQRSTUV	67	Helios	8.12	HIJKLMNOPQRSTUVWXY	67	AC Intrepid	40.45	(2)BCDEFGHI
68	Napoleon	44.05	BCDEFGHIJKLMNOPQRSTUV	68	Fieldstar VB	7.91	IJKLMNOPQRSTUVWXY	68	Napoleon	40.20	(2)BCDEFGHI
69	5604HR CL	43.74	CDEFGHIJKLMNOPQRSTUV	69	Enterprise	7.87	IJKLMNOPQRSTUVWXY	69	5701PR	39.94	(2)CDEFGHI
70	CDC Bounty	43.38	DEFGHIJKLMNOPQRSTUV	70	AC Elsa	7.74	JKLMNOPQRSTUVWXY	70	CDC Osler	39.56	(2)DEFGHI
71	AC Elsa	42.64	EFGHIJKLMNOPQRSTUV	71	Prodigy	7.30	KLMNOPQRSTUVWXYZ	71	CDC Alsask	39.31	(2)EFGHI
72	Commander	42.26	EFGHIJKLMNOPQRSTUV	72	CDC Origin	7.13	KLMNOPQRSTUVWXYZ	72	AC Andrew	39.19	(2)EFGHI
73	CDC Alsask	41.37	EFGHIJKLMNOPQRSTUV	73	McKenzie	7.06	KLMNOPQRSTUVWXYZ	73	Alvena	38.94	(2)FGHI
74	Helios	41.07	EFGHIJKLMNOPQRSTUV	74	Harvest	6.82	LMNOPQRSTUVWXYZ	74	CDC Stanley	38.94	(2)FGHI
75	CDC Teal	39.75	FGHIJKLMNOPQRSTUV	75	AC Foremost	6.68	MNOPQRSTUVWXYZ	75	Helios	38.32	(2)GHIJ
76	Enterprise	38.99	GHIJKLMNOPQRSTUV	76	Napoleon	6.59	NOPQRSTUVWXYZ	76	5700PR	38.07	(2)HIJ
77	AC Foremost	38.77	GHIJKLMNOPQRSTUV	77	5604HR CL	6.51	OPQRSTUVWXYZ	77	CDC Teal	37.82	(2)IJK
78	Stettler	35.61	HIJKLMNOPQRSTUV	78	Stettler	6.47	OPQRSTUVWXYZ	78	AC Barrie	36.00	(2)JKL
79	CDC Go	34.45	IJKLMNOPQRSTUV	79	5700PR	6.30	PQRSTUVWXYZ	79	Lillian	35.88	(2)JKL
80	Snowstar	33.59	JKLMNOPQRSTUV	80	CDC Go	6.10	QRSTUVWXYZ	80	AC Morse	35.40	(2)KLM
81	Glenn	32.06	KLMNOPQRSTUV	81	Waskada	5.65	RSTUVWXYZ	81	CDC Origin	35.05	(2)LM
82	5700PR	31.57	LMNOPQRSTUV	82	Snowstar	5.45	STUVWXYZ	82	Commander	34.81	(2)LM
83	Carberry	30.51	MNOPQRSTUV	83	AC Avonlea	5.33	TUVWXYZ	83	Kyle	34.69	(2)LM
84	Waskada	29.30	NOPQRSTUV	84	Strongfield	5.07	UVWXYZ	84	5600HR	33.76	(2)LMN
85	AC Superb	29.12	OPQRSTUV	85	Journey	4.75	VWXYZ	85	Somerset	33.52	(2)LMNO
86	AC Avonlea	28.40	PQRSTUV	86	CDC Zorba	4.64	VWXYZ	86	CDC VR Morris	33.29	(2)MNOP
87	Muchmore	25.83	QRSTUV	87	AC Superb	4.42	WXYZ	87	5601HR	31.58	(2)NOPQ
88	Journey	24.41	RSTUV	88	Transcend	4.33	XYZ	88	Snowbird	31.36	(2)OPQ
89	CDC Zorba	22.92	STUV	89	Carberry	4.22	YZ	89	Goodeve VB	31.14	(2)PQ
90	Transcend	20.90	TUV	90	Glenn	4.18	YZ	90	Strongfield	30.91	(2)PQ
91	Strongfield	19.10	UV	91	Muchmore	4.16	YZ	91	CDC Zorba	30.58	(2)Q
92	Eurostar	14.31	V	92	Eurostar	2.77	Z	92	Eurostar	23.04	(2)R

[a] Percent sclerotia by weight = 100 * weight sclerotia/(weight grain + sclerotia)
[b] Sclerotia/spike = number of sclerotia / number of spikes
[c] Sclerotium weight (mg) = 1000 * (weight of the sclerotia / number of sclerotia)
[w] SE = 5.598, [x] SE = 0.896, [y] SE = 0.157
[z] Means with different letters are considered to be significantly different from each other

Percent of Total Inoculated Florets That Were Aborted

Florets aborted			
	Cultivar	**Estimate^2 [x]**	
1	Eurostar	39.06	A [z]
2	Napoleon	31.02	AB
3	CDC Origin	28.41	ABC
4	CDC Zorba	27.67	ABCD
5	Kyle	20.70	ABCDE
6	Navigator	20.25	ABCDEF
7	Prodigy	19.89	ABCDEF
8	Commander	19.62	ABCDEF
9	Red Fife	16.00	ABCDEFG
10	CDC Verona	15.68	ABCDEFG
11	Strongfield	15.44	ABCDEFGH
12	AC Morse	13.99	ABCDEFGHI
13	Transcend	12.39	ABCDEFGHI
14	Brigade	12.32	ABCDEFGHI
15	PT559	12.11	ABCDEFGHI
16	AC Avonlea	12.04	ABCDEFGHI
17	Unity VB	11.42	ABCDEFGHI
18	NRG003	11.16	ABCDEFGHI
19	5604HRCL	11.09	ABCDEFGHI
20	CDC Go	10.24	BCDEFGHI
21	Vesper	10.18	BCDEFGHI
22	Sadash	9.99	BCDEFGHI
23	NRG010	9.99	BCDEFGHI
24	Marquis	9.86	BCDEFGHI
25	Fieldstar VB	9.55	BCDEFGHI
26	Journey	9.49	BCDEFGHI
27	CDC Plentiful	9.42	BCDEFGHI
28	Eatonia	9.36	BCDEFGHI
29	AC Abbey	8.53	BCDEFGHI
30	CDC Abound	8.12	BCDEFGHI
31	CDC Kernen	7.95	BCDEFGHI
32	Snowbird	7.78	BCDEFGHI
33	Alvena	7.62	BCDEFGHI
34	AC Crystal	7.40	BCDEFGHI
35	Stettler	7.40	BCDEFGHI
36	Muchmore	7.34	BCDEFGHI
37	Enterprise	7.34	BCDEFGHI
38	Infinity	7.24	BCDEFGHI
39	5702PR	7.18	BCDEFGHI
40	Lillian	6.92	CDEFGHI
41	Lovitt	6.86	CDEFGHI
42	5601HR	6.76	CDEFGHI
43	Bhishaj	6.76	CDEFGHI
44	CDC Imagine	6.66	CDEFGHI
45	CDC Merlin	6.60	CDEFGHI

46	5700PR	6.55	CDEFGHI
47	Goodeve VB	6.40	CDEFGHI
48	CDC Teal	6.30	CDEFGHI
49	Katepwa	6.25	CDEFGHI
50	AC Andrew	5.76	CDEFGHI
51	AC Taber	5.76	CDEFGHI
52	CDC Osler	5.66	DEFGHI
53	859CL	5.62	DEFGHI
54	AC Barrie	5.62	DEFGHI
55	Harvest	5.57	DEFGHI
56	5600HR	5.52	DEFGHI
57	CDC Utmost	5.43	DEFGHI
58	CDC Alsask	5.43	EFGHI
59	AC Foremost	5.38	EFGHI
60	Minnedosa	5.34	EFGHI
61	CDC Rama	5.29	EFGHI
62	McKenzie	5.06	EFGHI
63	Carberry	4.80	EFGHI
64	Snowstar	4.75	EFGHI
65	Waskada	4.49	EFGHI
66	AC Vista	4.49	EFGHI
67	KANE	4.49	EFGHI
68	Glenlea	4.45	EFGHI
69	CDC Stanley	4.45	EFGHI
70	CDC Bounty	4.45	EFGHI
71	AC Conquer VB	4.33	EFGHI
72	SY985	4.24	EFGHI
73	Somerset	4.12	EFGHI
74	AC Cadillac	4.08	EFGHI
75	Glenn	4.08	EFGHI
76	AC Elsa	4.08	EFGHI
77	5603HR	4.08	EFGHI
78	Shaw	3.96	EFGHI
79	AC Superb	3.92	EFGHI
80	CDC VR Morris	3.80	EFGHI
81	CDN Bison	3.65	EFGHI
82	CDC Thrive	3.31	EFGHI
83	5500HR	3.24	EFGHI
84	AC Splendor	3.20	EFGHI
85	Glencross VB	3.06	EFGHI
86	CDC Walrus	3.03	EFGHI
87	Roblin	2.89	EFGHI
88	Burnside	2.46	FGHI
89	5701PR	2.10	GHI
90	AC Intrepid	1.69	GHI
91	5602HR	1.00	HI
92	Helios	0.98	I

[x] SE = 0.479

[z] Means with different letters are considered to be significantly different from each other

Honeydew Production and Sclerotia Size Ratings

Honeydew Production				Sclerotial Size			
	Cultivar	Estimate [x]			Cultivar	Estimate [y]	
1	AC Conquer VB	4.9	A [z]	1	Marquis	2.96	A
2	Unity VB	4.9	AB	2	Carberry	2.94	A
3	KANE	4.9	AB	3	NRG003	2.93	A
4	AC Taber	4.7	ABC	4	CDC Utmost	2.91	A
5	AC Crystal	4.7	ABC	5	Bhishaj	2.90	A
6	Infinity	4.6	ABCD	6	Minnedosa	2.90	A
7	5603HR	4.6	ABCDE	7	CDC Plentiful	2.90	A
8	McKenzie	4.6	ABCDE	8	AC Conquer VB	2.89	A
9	AC Foremost	4.6	ABCDEF	9	5603HR	2.88	A
10	Sadash	4.6	ABCDEF	10	Prodigy	2.88	A
11	CDC Merlin	4.5	ABCDEFG	11	Eatonia	2.88	A
12	Somerset	4.5	ABCDEFGH	12	AC Crystal	2.88	A
13	Harvest	4.5	ABCDEFGHI	13	Glenn	2.87	A
14	Brigade	4.5	ABCDEFGHI	14	CDC Merlin	2.87	A
15	CDC Osler	4.4	ABCDEFGHIJ	15	AC Elsa	2.87	A
16	AC Abbey	4.4	ABCDEFGHIJK	16	Unity VB	2.87	A
17	Fieldstar VB	4.4	ABCDEFGHIJK	17	Katepwa	2.86	AB
18	5500HR	4.4	ABCDEFGHIJK	18	NRG010	2.86	AB
19	AC Andrew	4.2	ABCDEFGHIJKL	19	Lovitt	2.86	AB
20	Lillian	4.2	ABCDEFGHIJKLM	20	Vesper	2.84	AB
21	Katepwa	4.2	ABCDEFGHIJKLM	21	AC Superb	2.84	AB
22	CDC VR Morris	4.2	ABCDEFGHIJKLMN	22	AC Vista	2.84	AB
23	Shaw	4.2	ABCDEFGHIJKLMNO	23	Muchmore	2.82	AB
24	Muchmore	4.1	BCDEFGHIJKLMNOP	24	Burnside	2.81	AB
25	AC Navigator	4.1	BCDEFGHIJKLMNOPQ	25	McKenzie	2.81	AB
26	AC Intrepid	4.0	CDEFGHIJKLMNOPQ	26	Brigade	2.81	AB
27	AC Cadillac	4.0	CDEFGHIJKLMNOPQ	27	CDC Osler	2.81	AB
28	Bhishaj	4.0	CDEFGHIJKLMNOPQ	28	AC Splendor	2.80	AB
29	CDC Imagine	4.0	CDEFGHIJKLMNOPQ	29	Infinity	2.80	AB
30	5701PR	4.0	CDEFGHIJKLMNOPQ	30	Glenlea	2.78	ABC
31	Roblin	4.0	CDEFGHIJKLMNOPQ	31	Harvest	2.78	ABC
32	Marquis	3.9	DEFGHIJKLMNOPQR	32	AC Taber	2.78	ABC
33	CDC Stanley	3.9	DEFGHIJKLMNOPQRS	33	5604HR CL	2.78	ABC
34	CDC Thrive	3.8	EFGHIJKLMNOPQRST	34	Journey	2.75	ABCD
35	859CL	3.8	EFGHIJKLMNOPQRST	35	5500HR	2.75	ABCD
36	CDC Utmost	3.8	EFGHIJKLMNOPQRST	36	AC Abbey	2.75	ABCD
37	AC Avonlea	3.8	FGHIJKLMNOPQRSTU	37	Stettler	2.75	ABCD
38	5600HR	3.8	FGHIJKLMNOPQRSTU	38	Roblin	2.74	ABCD
39	5700PR	3.7	GHIJKLMNOPQRSTUV	39	CDC Bounty	2.74	ABCD
40	5702PR	3.7	HIJKLMNOPQRSTUV	40	KANE	2.74	ABCD
41	Snowstar	3.7	HIJKLMNOPQRSTUV	41	AC Cadillac	2.73	ABCD
42	Goodeve VB	3.7	IJKLMNOPQRSTUV	42	CDN Bison	2.73	ABCD
43	AC Superb	3.7	IJKLMNOPQRSTUV	43	Sadash	2.73	ABCD
44	CDC Plentiful	3.7	IJKLMNOPQRSTUVW	44	Shaw	2.73	ABCD
45	Waskada	3.6	JKLMNOPQRSTUVW	45	Red Fife	2.71	ABCDE
46	AC Splendor	3.6	KLMNOPQRSTUVW	46	CDC Abound	2.71	ABCDE

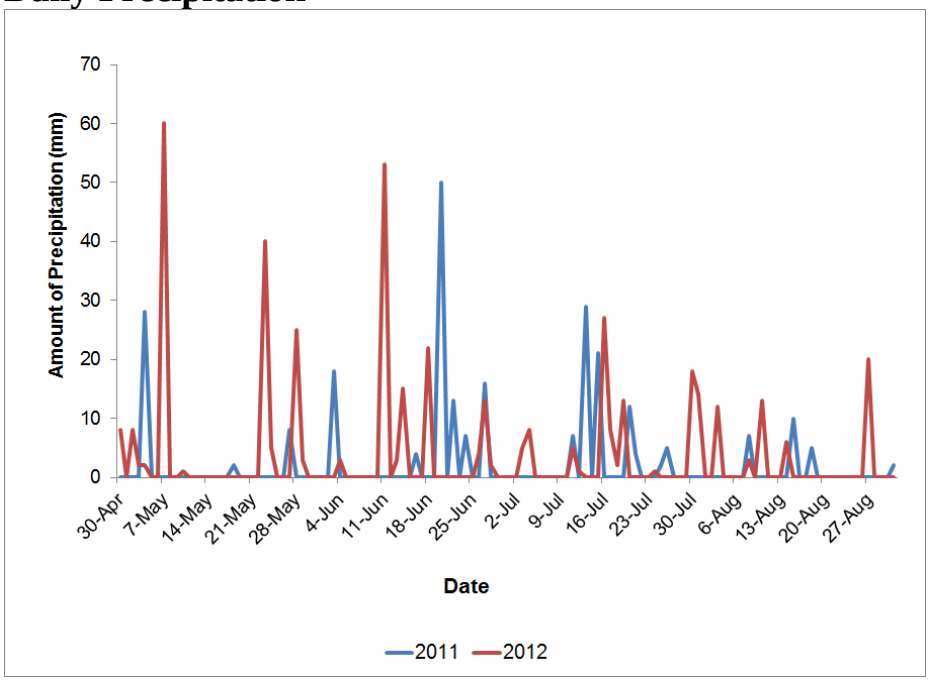
47	Snowbird	3.6	KLMNOPQRSTUVW	47	CDC Kernen	2.70	ABCDE
48	Enterprise	3.6	LMNOPQRSTUVWX	48	CDC Rama	2.69	ABCDE
49	Glencross VB	3.5	LMNOPQRSTUVWX	49	CDC Stanley	2.69	ABCDE
50	AC Elsa	3.5	LMNOPQRSTUVWXY	50	Fieldstar VB	2.69	ABCDE
51	SY985	3.4	LMNOPQRSTUVWXY	51	AC Foremost	2.68	ABCDEF
52	CDC Kernen	3.4	LMNOPQRSTUVWXY	52	CDC Walrus	2.67	ABCDEF
53	Napoleon	3.4	LMNOPQRSTUVWXY	53	859CL	2.67	ABCDEF
54	CDC Bounty	3.4	LMNOPQRSTUVWXYZ	54	CDC Go	2.66	ABCDEF
55	CDN Bison	3.4	MNOPQRSTUVWXYZ	55	PT559	2.65	ABCDEF
56	CDC Alsask	3.4	MNOPQRSTUVWXYZ	56	Lillian	2.65	ABCDEF
57	CDC Verona	3.4	NOPQRSTUVWXYZ	57	Somerset	2.63	ABCDEFG
58	5601HR	3.3	OPQRSTUVWXYZ	58	CDC Thrive	2.63	ABCDEFG
59	Helios	3.3	OPQRSTUVWXYZ	59	CDC Alsask	2.63	ABCDEFG
60	PT559	3.3	PQRSTUVWXYZ	60	AC Intrepid	2.62	ABCDEFG
61	Glenlea	3.3	PQRSTUVWXYZ	61	Waskada	2.62	ABCDEFG
62	AC Barrie	3.3	QRSTUVWXYZ	62	AC Barrie	2.61	ABCDEFG
63	Eatonia	3.3	QRSTUVWXYZ	63	CDC Imagine	2.58	ABCDEFG
64	CDC Go	3.3	QRSTUVWXYZ	64	5602HR	2.58	ABCDEFG
65	CDC Teal	3.3	QRSTUVWXYZ	65	5702PR	2.57	ABCDEFG
66	Kyle	3.2	RSTUVWXYZ(2)A	66	CDC VR Morris	2.56	ABCDEFG
67	5602HR	3.2	RSTUVWXYZ(2)A	67	CDC Teal	2.55	ABCDEFG
68	Transcend	3.2	RSTUVWXYZ(2)A	68	Snowbird	2.55	ABCDEFG
69	NRG003	3.1	RSTUVWXYZ(2)A	69	Goodeve VB	2.54	ABCDEFG
70	Lovitt	3.1	RSTUVWXYZ(2)A	70	Glencross VB	2.52	ABCDEFG
71	NRG010	3.1	RSTUVWXYZ(2)A	71	Alvena	2.51	ABCDEFG
72	CDC Walrus	3.1	RSTUVWXYZ(2)A	72	5701PR	2.50	ABCDEFG
73	CDC Abound	3.1	RSTUVWXYZ(2)A	73	5600HR	2.48	ABCDEFG
74	5604HR CL	3.1	RSTUVWXYZ(2)A	74	SY985	2.47	ABCDEFG
75	Carberry	3.1	RSTUVWXYZ(2)A	75	AC Andrew	2.44	ABCDEFGH
76	Minnedosa	3.1	STUVWXYZ(2)A	76	Helios	2.42	ABCDEFGH
77	Strongfield	3.1	STUVWXYZ(2)A	77	Snowstar	2.40	ABCDEFGH
78	AC Vista	3.0	TUVWXYZ(2)A	78	5601HR	2.38	ABCDEFGHI
79	Burnside	3.0	UVWXYZ(2)A	79	Enterprise	2.21	ABCDEFGHIJ
80	Red Fife	3.0	UVWXYZ(2)A	80	CDC Verona	2.21	ABCDEFGHIJ
81	Stettler	3.0	UVWXYZ(2)A	81	5700PR	2.11	BCDEFGHIJK
82	Alvena	2.9	VWXYZ(2)A	82	AC Navigator	2.03	CDEFGHIJK
83	Journey	2.9	WXYZ(2)A	83	AC Morse	2.01	DEFGHIJK
84	CDC Rama	2.9	WXYZ(2)A	84	AC Avonlea	1.97	EFGHIJK
85	Vesper	2.8	XYZ(2)AB	85	CDC Origin	1.94	FGHIJK
86	Glenn	2.7	YZ(2)AB	86	Napoleon	1.92	FGHIJK
87	Prodigy	2.6	Z(2)AB	87	Kyle	1.88	GHIJK
88	AC Morse	2.4	(2)ABC	88	Transcend	1.88	GHIJK
89	Commander	2.4	(2)ABC	89	Commander	1.70	HIJK
90	CDC Origin	2.0	(2)BCD	90	CDC Zorba	1.64	IJK
91	CDC Zorba	1.6	(2)CD	91	Eurostar	1.53	JK
92	Eurostar	1.3	(2)D	92	Strongfield	1.43	K

[x] SE = 0.15, [y] SE = 0.12

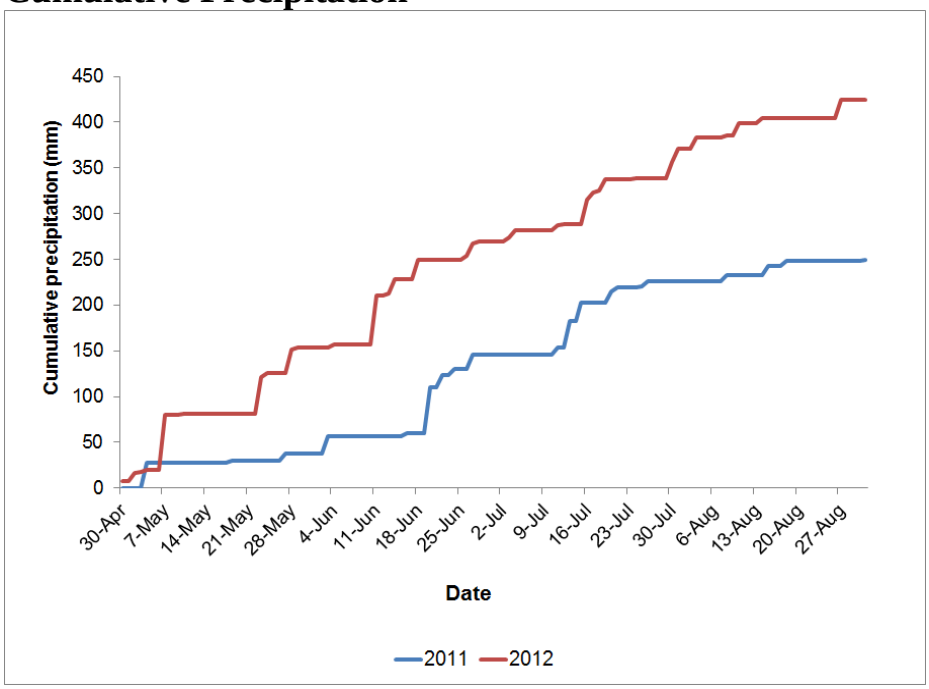
[z] Means with different letters are considered to be significantly different from each other

Appendix E. Precipitation (measured in mm) for the 2011 and 2012 cropping seasons at the Crop Development Center Research Station (University of Saskatchewan, Saskatoon, SK).

Daily Precipitation



Cumulative Precipitation



Appendix F. Daily maximum and minimum temperatures (in degrees Celsius) recorded at Saskatoon, SK throughout the growing season.