Sixth Canadian Workshop on Fusarium Head Blight

Sixième Colloque Canadien sur la Fusariose

Ottawa Marriott Hotel
Ottawa, Ontario
November 1 - 4, 2009

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Proceedings compiled by Thérèse Ouellet, Don Leger and Jennifer Thompson, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada.

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Editor's note: Papers or abstracts in these proceedings may be cited as follows:
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6th Canadian Workshop on Fusarium Head Blight / 6ième Colloque Canadien sur la Fusariose

November 1 - 4, 2009, Ottawa, Ontario

Dear Friends and Colleagues,

Thank you for joining us to participate in the 6th Canadian Workshop on Fusarium Head Blight/6ième Colloque Canadien sur la Fusariose (6th CWFHB/6ième CCF) in Ottawa, Ontario. With over 35 invited speakers and 70 posters, the biennial Workshop will cover a broad range of approaches to understanding and combating FHB in Canada and around the globe. We have endeavoured also to provide participants many opportunities for discussion and networking through shared meals and social events. The 6th CWFHB/ 6ième CCF Proceedings contain abstracts for invited oral and submitted poster presentations, as well as a few comprehensive reports. They will be available on-line after the Workshop at www.cwfhb.org. Some of the abstracts also will be published in the Canadian Journal of Plant Pathology. We hope that you will find this Workshop experience informative and enlightening, providing the catalyst and opportunities for future collaboration and innovation.

I would like to acknowledge the various sponsors who have been very generous in their financial support of the 6th CWFHB/6ième CCF. We thank them for their continued assistance and participation. I also would like to thank all the members of our National and Local Organizing Committees who have provided so many hours of their time to aid and ensure the smooth and successful delivery of this Workshop. Finally, thank you to all of our participants for agreeing to present and share their research to make the 6th CWFHB/6ième CCF possible.

Sincerely,

Linda Harris
Chair, *6th CWFHB/6ième CCF*

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Further information on BASF is available at:
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Bayer CropScience AG, a subsidiary of Bayer AG with annual sales of EUR 6.4 billion (2008), is one of the world's leading innovative crop science companies in the areas of crop protection, non-agricultural pest control, seeds and plant biotechnology. The company offers an outstanding range of products and extensive service backup for modern, sustainable agriculture and for non-agricultural applications. Bayer CropScience has a global workforce of about 18,000 and is represented in more than 120 countries.

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Une Société opérationnelle de Bayer AG, Bayer CropScience AG a réalisé en 2008 un chiffre d'affaires de 6,4 milliards d'euros qui la classe parmi les plus grandes entreprises innovantes du secteur mondial de la protection des récoltes. Elle opère dans les domaines du contrôle phytosanitaire, de la lutte antiparasite non agricole ainsi que des biotechnologies végétales et des semences. L'entreprise dispose d'une gamme de produits exceptionnels et offre de nombreux services à ses clients destinés à l'agriculture moderne et durable et aux applications non agricoles. Présente dans plus de 120 pays, Bayer CropScience emploie quelque 18,000 personnes dans le monde.

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Canadian Grain Commission | Commission canadienne des grains

The Canadian Grain Commission protects the rights of Canadian grain producers when they deliver their grain to licensed grain handling companies and grain dealers. Through its activities, the Canadian Grain Commission supports a competitive, efficient grain sector and upholds Canada's international reputation for consistent and reliable grain quality.

Further information on CGC is available at:
www.grainscanada.gc.ca/index-eng.htm

La Commission canadienne des grains protège les droits des producteurs de grains canadiens lorsqu'ils livrent leur grain aux exploitants de silos agréés et négociants en grains. Grâce à ses activités, la Commission canadienne des grains appuie le secteur des grains concurrentiel et efficace et maintient la réputation internationale du Canada pour la qualité constante et fiable de ses grains.

Canadian National Millers Association

The Canadian National Millers Association is a national, not-for-profit industry association representing the Canadian cereal grain milling industry. CNMA member companies are processors of wheat, oats and corn and producers of milled grain products. The CNMA is among Canada's longest established trade organizations, founded over eighty-five years ago. CNMA serves as the principal voice for the grain milling industry in consultation with government departments and agencies concerning regulatory issues and public policy. The association is an active participant in industry-government dialogue in the areas of food regulation, nutrition and health, food safety, transportation, international trade, occupational safety and health and environmental protection.

Further information on CNMA is available at: www.canadianmillers.ca/

Gold…

Genome Prairie is the leading organization for support and management of large-scale genomics and proteomics research projects in Manitoba and Saskatchewan. With its partners, Genome Prairie has supported more than $188 million of research activity in plant, animal and human genomics, bioinformatics, instrumentation development and bioethics since 2000. Genome Prairie works collaboratively with all levels of government, universities and industry as well as Genome Canada, a not-for-profit organization implementing a national strategy in genomics and proteomics research to benefit all Canadians. With offices in Winnipeg and Saskatoon, Genome Prairie continues to work with researchers in both provinces to facilitate genomics and proteomics projects in areas such as agriculture, environment, human and animal health.

Further information on Genome Prairie is available at: www.genomeprairie.ca/

The Ontario Wheat Producers' Marketing Board represents 17,000 Ontario wheat producers by providing strategic leadership initiatives that promote and improve Ontario wheat. Over 2 million tonnes of wheat are produced in Ontario. Ontario produces four commercial classes of wheat including soft red, soft white and hard red winter and hard red spring wheat along with two classes in limited production – hard white spring and durum. One area of research that is a high priority for our Board and producers is fusarium control. Great strides have been made by the Ontario wheat industry and government not only in variety development but also in management techniques and crop protection programs to address this disease concern for Ontario producers.

Further information on OWPMB is available at: www.ontariowheatboard.com/

Gold…

The Canadian Wheat Board (CWB) is a farmer-controlled marketer of the more than 20 million tonnes of wheat and barley grown in Western Canada. With annual sales revenues of $4 to $6 billion and customers in over 70 countries, the CWB is the world's largest single marketer of wheat and barley. With a marketing strategy that concentrates on high quality products and with a clear mandate to help maximize farmer income, the CWB is very supportive of finding solutions to Fusarium Head Blight (FHB). Through CWB direct funding of FHB research projects and through the Western Research Foundation Check-off deduction from CWB payments, Western Canadian farmers provide considerable financial support to breeding research for FHB resistance.

Further information on CWB is available at: www.cwb.ca/public/en/

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Program
6th Canadian Workshop on Fusarium Head Blight
Ottawa Marriott Hotel

Monday, November 2, 2009

0830h **Welcome address.**
Michèle Marcotte, Agriculture and Agri-Food Canada

0850 - 1010h **Plenary Session**
Chair: Linda Harris, Agriculture and Agri-Food Canada

0850h **The impact of FHB on grain grading and handling in Canada.**
Randy Clear, Canadian Grain Commission

0930h **Genetics of fusarium head blight resistance in wheat.**
Hermann Buerstmayr, Institute for Agrobiotechnology

1010h **Health break**

1040 - 1200h **Breeding & Markers A**
Chair: George Fedak, Agriculture and Agri-Food Canada

1040h **Progress in breeding for FHB resistance in Canadian Spring Wheat – 2009.**
Steve Fox, Agriculture and Agri-Food Canada

1100h **Breeding for FHB resistance in winter wheat; from public and industry perspectives.**
Mark Etienne, Hyland Seeds

1120h **Introgression of two QTLs from exotic sources in adapted European wheat.**
Viktor Korzun, KWS Lochow GmbH

1140h **Developing a YIPEE wheat cultivar.**
Ron DePauw, Agriculture and Agri-Food Canada

1200h **Lunch**

1330 - 1520h	**Disease Management A** *Chair: Jeannie Gilbert, Agriculture and Agri-Food Canada*

1330h **The way we deal with head blight at Moulins de Soulanges Mill.**
Elisabeth Vachon, Les Moulins de Soulanges

1350h **Incidence of *Fusarium graminearum* and *F. poae* from a 2-year wheat monitoring: factors that promote infection and mycotoxin contamination.**
Susanne Vogelgsang, Agroscope Reckenholz-Taenikon ART

1410h **Integrated management of fusarium head blight - Research and outreach.**
Marcia McMullen, North Dakota State University

1430h **Realistic expectations of foliar fungicides for management of FHB.**
Luc Bourgeois, Bayer CropScience

1450h **Glyphosate associations with *Fusarium* diseases of cereal and pulse crops.**
Myriam Fernandez, Agriculture and Agri-Food Canada

1520h **Health break**

1600 - 1800h **Poster Session**

Tuesday, November 3, 2009

0830 - 1010h	**Biology of the Disease – Pathogen** *Chair: Keith Seifert, Agriculture and Agri-Food Canada*

***Plenary lecture*:**

0830h **What has been learned from six years of genomic research on the fusarium head blight pathogen?**
Corby Kistler, United States Department of Agriculture

0910h **Biogeography of trichothecene chemotypes, and the origins of North American 3ADON populations.**
Todd Ward, United States Department of Agriculture

0930h **Alternate route to attain resistance against *Fusarium graminearum.***
Gopal Subramaniam, Agriculture and Agri-Food Canada

0950h **Molecular identification and databases in *Fusarium*.**
Dave Geiser, Penn State University

1010h **Health break**

1040 - 1200h **Biology of the Disease – Plant**
Chair: Jas Singh, Agriculture and Agri-Food Canada

1040h **Involvement of phytohormone signalling in FHB.**
Paul Nicholson, John Innes Centre

1100h **Expression profiling of FHB resistant wheat: what we have learned from it so far.**
Thérèse Ouellet, Agriculture and Agri-Food Canada

1120h **Transgenic field trials for FHB resistance and related research in wheat and barley.**
Lynn Dahleen, United States Department of Agriculture

1140h **Yeast genomic screening to identify cellular targets of *Fusarium graminearum* fungal toxins.**
Anne Hermans, Agriculture and Agri-Food Canada

1200h **Lunch**

1330 - 1530h **Breeding & Markers B**
Chair: Ron DePauw, Agriculture and Agri-Food Canada

1330h **Update on improving fusarium head blight resistant Canadian barley.**
William Legge, Agriculture and Agri-Food Canada

1350h **An overview of oat FHB studies in Canada.**
Weikei Yan, Agriculture and Agri-Food Canada

1410h **Breeding for resistance to ear rot and tolerance to mycotoxins in European maize.**
Thomas Miedaner, University of Hohenheim/Stuttgart

1430h **Breeding - Round-table discussion.**
Lana Reid (Agriculture and Agri-Food Canada); Cécile Tétreault (Nevico); Curtis Pozniak (University of Saskatchewan); George Fedak (Agriculture and Agri-Food Canada)

1530h **Health break**

1600 - 1700h **Disease Management B**
Chair: Jeannie Gilbert, Agriculture and Agri-Food Canada

1600h **Biological management of fusarium head blight of wheat with *Clonostachys rosea* strain ACM941.**
Allen Xue, Agriculture and Agri-Food Canada

1620h **Biological control as a viable and commercially sustainable alternative to dealing with the fusarium head blight (FHB) problem from a small company perspective.**
Gordon Genge, ICUS Canada

1640h **Biocontrol and bioprotection fungi against FHB and mycotoxin accumulation in cereals.**
Vladmir Vujanovic, University of Saskatchewan

Wednesday, November 4, 2009

0830 - 1005h **Mycotoxins**
Co-chairs: Marc Savard & Barbara Blackwell, Agriculture and Agri-Food Canada

0830h **Fusarium mycotoxins: biosynthetic pathways and role in virulence.**
Nancy Alexander, United States Department of Agriculture

0855h **A tale of contrasting economic reaction to mycotoxin in maize and wheat in the same agricultural-system in Ontario, Canada.**
Victor Limay-Rios, University of Guelph

0915h **Identification of a biomarker for deoxynivalenol risk assessment and regulation.**
Chidozie Amuzie, Michigan State University

0935h **Alberta Seed Testing Laboratory perspectives: *Fusarium graminearum* testing methodology and detection**.
Holly Gelech, Biovision Seed Labs and Kevin Zaychuk, 20/20 Seed Labs

1005h	**Health break**
1030h	**AAFC Pest Management Centre Risk Reduction Strategy Mini-session.** *Chair: Cezarina Kora, Agriculture and Agri-Food Canada*
1030h	**Mycotoxin Network Meeting.** *Chair: Marc Savard, Agriculture and Agri-Food Canada*
1200h	**Workshop closing**

List of Oral Presentation Abstracts and Summaries

Biology of the Disease - Plant

Breeding & Markers B

Disease Management B

Mycotoxins

Oral Presentation Abstracts and Summaries

T01. The impact of FHB on grain grading and handling in Canada

R. Clear and T. Nowicki. *Grain Research Laboratory, 1404-303 Main St., Winnipeg, MB, Canada, R3C 3G8.*

Abstract

Since fusarium head blight first became a Canadian issue in 1980, it has influenced the grading and handling of cereals. With increasing research and practical experience we have been able to relax the initially very tight tolerances while improving grower returns, marketability, and meeting ever more stringent and numerous government regulations. Handling of infected grain has run the gamut from an early outright ban on exports, to the present system where blending, advanced cleaning equipment, and regular monitoring manages the quality and safety of our exports. The development of semi-rapid toxin testing methods has given another tool to the industry, and is frequently employed to monitor and select grain for DON-sensitive industries and customers. While the grain industry has developed ways of handling infected grain, producers have had less success reducing disease levels. Development of improved varieties has been slow, and better management practices have limited effect. In the worst affected areas, entire classes of grain have essentially been lost to producers. In the first 20 years the area affected by *Fusarium graminearum* grew steadily, mainly in the eastern prairies. The recent change in the pathogen population may result in increasing toxin levels in the grain, which in turn might require lowering grading tolerances.

Introduction

In the early 1980's, a disease that had been known to occur only sporadically and at low levels in Canada suddenly emerged as an important issue in the grain trade. Fusarium head blight (FHB) was not a new disease to the small grain industry in Canada, but the rise in disease levels was due to the emergence of *Fusarium graminearum* Schwabe as the principal causal agent. The detection of deoxynivalenol (DON) in the 1980 Ontario winter wheat and Quebec spring wheat crop elevated the disease from solely a plant pathology issue to a food safety one as well. Subsequently, the combination of yield and quality reduction and mycotoxin concerns of FHB have cost Canadian producers and the grain trade over a billion dollars over the last 30 years and is still far from being controlled. As early as 1998, the total estimated loss in just Manitoba and Quebec was 520 million dollars. Since then the area affected by *F. graminearum* has increased as have disease levels. As the frequency and severity of FHB has increased over the years, changes to the grading and handling system have also occurred, but in a direction one may not have expected. In this presentation we will use the Canada Eastern White Winter (CEWW) and Canada Western Red Spring (CWRS) wheat classes as examples of the evolution of grading tolerances.

Grading

When pink kernels appeared in the 1980 eastern wheat crop, initial tests found high levels of trichothecenes. Later they were identified as deoxynivalenol, also known as DON or

vomitoxin. Early toxicological literature on DON was too scanty to permit establishment of a tolerable level in foods intended for human consumption and suggested that levels as low as 0.7 ppm could cause vomiting, feed refusal, weight loss and reduced performance in livestock. Concern for public health and the lack of data on human toxicity resulted in a 1980 Health Canada recommendation that feed grade Ontario winter wheat not be used for human consumption and an initial ban on exports. The estimated cost to Ontario growers was 17 million dollars. After scientists in Ottawa found a reasonably good correlation between colour intensity and DON levels, very tight tolerances for these pink kernels were adopted, and the same tolerances used for 'heated' kernels were then applied for grading (Fig.1). For feed, it was recommended that Ontario wheat with $\leq 0.1\%$ pink kernels be used at a maximum of 25% of the feed ration, whereas Quebec wheat, which had higher DON levels, be used at no more than 10% of the feed ration. In September 1982, Health Canada's Health Protection Branch (HPB) issued a policy statement on vomitoxin that stated that as a precautionary measure, Ontario soft winter wheat should not be used in the preparation of infant foods and that DON levels in flour and bran to be used in the processing of non-staple foods must not exceed 0.3 ppm. In October 1982, based on new information on the effect of processing, the policy statement was modified to permit use of uncleaned soft wheat with a level of DON up to 0.6 ppm and to clarify that blending with other Ontario or western soft wheat would be permitted to achieve this level.

In May, 1983, as more research became available, the HPB set limits for uncleaned soft wheat of 1.0 ppm for baby food and 2.0 ppm for other uses, and in October of 1986 they established a temporary safe limit of 1 ppm of vomitoxin (DON) in unprocessed durum wheat for human consumption in Canada. In 1991, Health Canada set 0.5 ppm as the "level of interest" for the level of DON in the final blend of cleaned eastern wheat for use in processing of staple foods. No other limits for DON have so far been set in Canada, although they are presently being considered. During this time the term 'tombstone' was adopted to refer to the damaged kernels, and the definition was broadened to include shrunken, chalky-white kernels, both with and without pink discolouration. It wasn't until 1996 that the term was changed from tombstone to *Fusarium* damaged kernel (FDK), a more accurate and palatable term. The final two changes to the CEWW tombstone tolerances were in 1987 and 1990 (Fig. 1). Because the CEWW tolerances in 1990 were based on safety considerations and DON regulation, not quality, the tombstone tolerances were the same for all numerical grades, the ones typically used for human consumption. The ability of the grading system in managing DON levels in CEWW exports can be seen in Fig. 2. Most CEWW is used locally, or shipped directly to the USA, and usually only a small portion is exported. The number of CEWW cargoes tested ranged from 1 to 89. The limited area from which CEWW is drawn and the limited number of cargoes, results in large annual fluctuations in DON levels.

In western Canada, the first reported incidence of FHB caused by *F. graminearum* and DON contamination occurred in the Red River Valley (RRV) of southern Manitoba in 1984. The two affected wheat samples were from a single farm. It was considered that the pathogen posed little threat on the prairies as the conditions were usually too dry for extensive disease development. This conclusion was supported by historic data that found

F. graminearum had been rarely isolated from seed or soil in western Canada for several decades. HPB surveys for DON in western wheat began in 1980 and until 1984 found the crops free of DON. In 1984 the HPB found DON on about 25% of producer samples from the RRV at a maximum of just over 0.2 ppm, and in 1985 found DON in just under 50% of samples from the RRV (maximum of just over 1.0 ppm) but no DON in wheat from outside the RRV. The Canadian Grain Commission (CGC) began testing cargoes of the numerical grades of CWRS (not feed wheat) for DON in 1984 (Fig. 3), and since 1985 the CGC has conducted surveys on the frequency and distribution of *F. graminearum* in western Canada. Even then it was clear that the more susceptible classes of wheat, the amber durum (CWAD) and the recently introduced semi-dwarf wheat of the Canada Prairie Spring (CPS) class, were especially vulnerable to the disease. For example, in 1986 CGC results found mean tombstone levels of only 0.12% in CWRS, but 0.68% in CWAD and 1.09% in CPS. Grading tolerances were established in western Canada in September, 1985, and as in the early years in eastern Canada, they first mirrored the tolerances for heated kernels. However, by October of 1986, less rigid tolerances for western wheat were set. For the 3 grades of CWRS, they were higher at 0.25%, 0.5%, and 1.0% (Fig. 4). Tolerances for CWRS were altered several times more to reflect new research and marketplace acceptance, with the last change made in 1999 (Fig. 4). 1993 was a watershed year in terms of dealing with FHB in western Canada. Although the severe epidemic was primarily limited to the RRV, the impact was far reaching. Besides screaming headlines in local newspapers, the epidemic spawned serious efforts at introducing FHB resistance into western grain, and research into all aspects of the problem blossomed. Losses were exacerbated because of the widespread adoption of the susceptible CWRS variety Roblin which was grown on almost half the bread wheat acreage in Manitoba that year, and tolerances that were much stricter than they are today.

Growers in the affected area suffered enormous financial loss, but because the area affected was only a small portion of the western wheat area, the industry was able to blend some of the wheat and DON levels remained low in CWRS exports (Fig. 3). The grading system would also have diverted the worst affected samples away from the numerical grades, further protecting our exports and grain used for domestic consumption. That year also saw the increasing use of ELISA based quick tests for DON determination. By the following July, market acceptance of the grading systems ability to manage DON levels in the grain and continued research allowed for an increase in tombstone tolerances for the 1994 wheat crop (Fig. 4). The development of economic levels of *F. graminearum* in south-eastern Saskatchewan in 1998, the first time this occurred outside of Manitoba, did not cause an alteration to FDK tolerances, but in 1999 studies on the quality impact of FDK required that the tolerance for a #2 CWRS be lowered from 2.0% to 1% (Fig. 4), a level unchanged to this day.

Adjustments to the tolerances for FDK and an increasing area affected likely contributed to a general rise in DON levels in the CWRS in the mid 1990's onwards. In 2001 and 2002, the highest mean level of FDK in western wheat yet recorded is reflected in the highest average DON levels in the CWRS wheat exports (Fig. 3). Because FDK tolerances in #2 and #3 CWRS could potentially allow for DON levels in cargoes that

would exceed the recently instituted European Union maximum of 1.25 ppm, extensive DON testing by ELISA is done on wheat transported from Thunder Bay to terminal elevators along the St. Lawrence, as well as on exports from both the east and west coast. The CGC uses GC/MS to both monitor the ELISA tests as well as perform analyses on several hundred cargoes taken at random each year. Increasing regulations and a more demanding consumer place additional burdens on exporting countries. Until a sufficiently rapid, robust, reliable and economical test for DON is developed, the bulk handling system will continue to rely primarily on the grading system to manage the safety and quality of the grain.

Handling

In eastern Canada, opportunities for blending are limited as the growing area is small and the disease is endemic to the entire agricultural region. However, in western Canada blending naturally occurs as grain is collected and moved to export terminals. With approximately 80% of western wheat exported, localized problems can be mitigated by combining with other grain. The area in western Canada where FHB is at economic levels is just over 15%, and unlike many degrading factors in wheat, the most affected kernels can sometimes be removed. Grain elevators in the most FHB prone areas of Manitoba have installed extra cleaning equipment (gravity tables) to maximize their ability to clean out FDK. In 1997 the Canadian Wheat Board introduced their 'Fusarium Program' in the interests of improving returns to producers in the FHB affected area. In select classes of wheat, growers with wheat that grades higher in all respects but contains high levels of FDK, are eligible to receive the higher grade. The amount by which the FDK level exceeds that grade's maximum is considered as dockage and the price is adjusted accordingly. Implementation of the ad hoc program depends on several factors, including DON levels and the availability of blending opportunities.

With the advent of rapid DON tests in the early 1990's, it has become possible for DON-sensitive industries, such as malting and feed companies, to use DON as a selection tool and price determinant. Sometimes this results in intermittent alterations to the normal grain handling system. For example, in years when FHB is a problem in Manitoba, some grain from the western prairies is shipped to Manitoba to supply the DON-sensitive hog industry, while affected Manitoba grain is shipped west to the DON-tolerant cattle industry.

In both eastern and western Canada, FHB has also had profound effects on the variety and class of grain grown. In FHB prone areas, acreage of more susceptible classes of grain have greatly declined over the years. In the early 1990's, growers in Ontario began to switch to soft red winter (CESRW) wheat from CEWW, initially in a desire to try a bread making wheat. The better sprout resistance and FHB tolerance of the red wheat has resulted in a steady decline in the acreage of CEWW to where little CEWW is now grown. Even now, 8 of 10 CEWW but only 7 of 15 CESRW varieties are listed as susceptible or highly susceptible. In Manitoba, durum wheat acreage has declined precipitously since 1987 (Fig. 5), to the point where in 2008 only 0.3% of Manitoba acres were planted to durum. Acreage planted to malting barley varieties, especially 6-row varieties, has also declined in Manitoba in the last decade (Fig. 6), and little of it is now

accepted for malting. The burden of lower grades and reduced crop selection falls hardest on the producers. Although the handling system has demonstrated an ability to protect our wheat exports, further expansion of the affected area and changes to the pathogen population will reduce its effectiveness. Increased toxin production by the new population of *F. graminearum* could pressure FDK tolerances downward, countering the general trend of the last 20 years. Combined with ever increasing demands and regulations in the marketplace, the urgency for improved varieties is stronger than ever. However, improvement has been slow. In Manitoba, the number of varieties rated as poor to very poor for FHB resistance is actually increasing (Fig. 7), and they presently account for a sizeable percentage of the planted acreage. In 2008, 53% of the CWRS acreage in Manitoba was planted to varieties rated as poor and very poor for FHB. This no doubt contributed to over 50% of the Manitoba CWRS crop being degraded due to FDK in 2008.

Figure 1. Tombstone/*Fusarium* damaged kernel tolerances

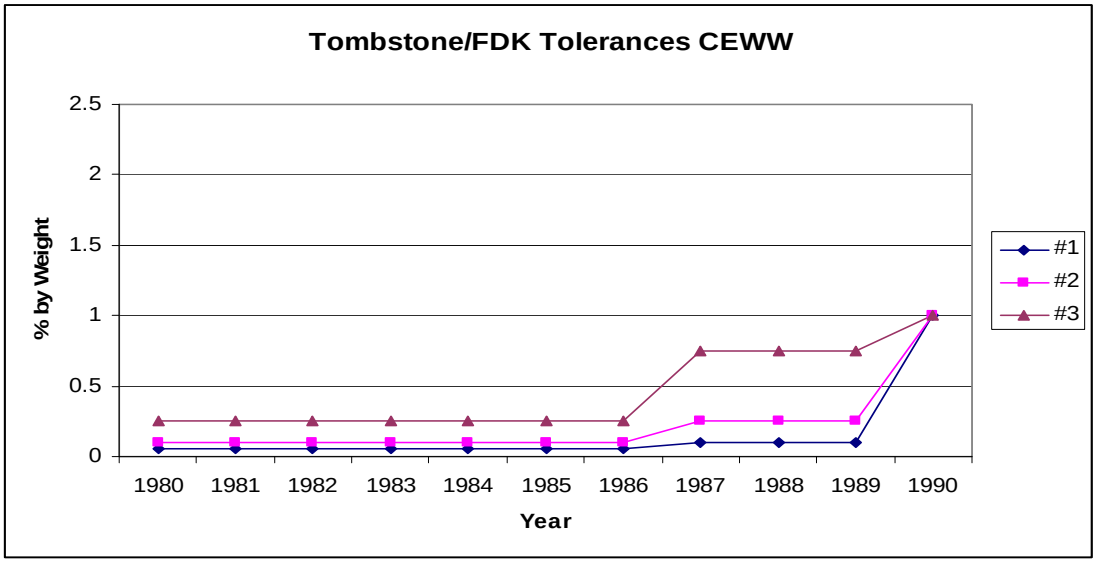


Figure 2. Deoxynivalenol levels in Canada Eastern White Winter wheat

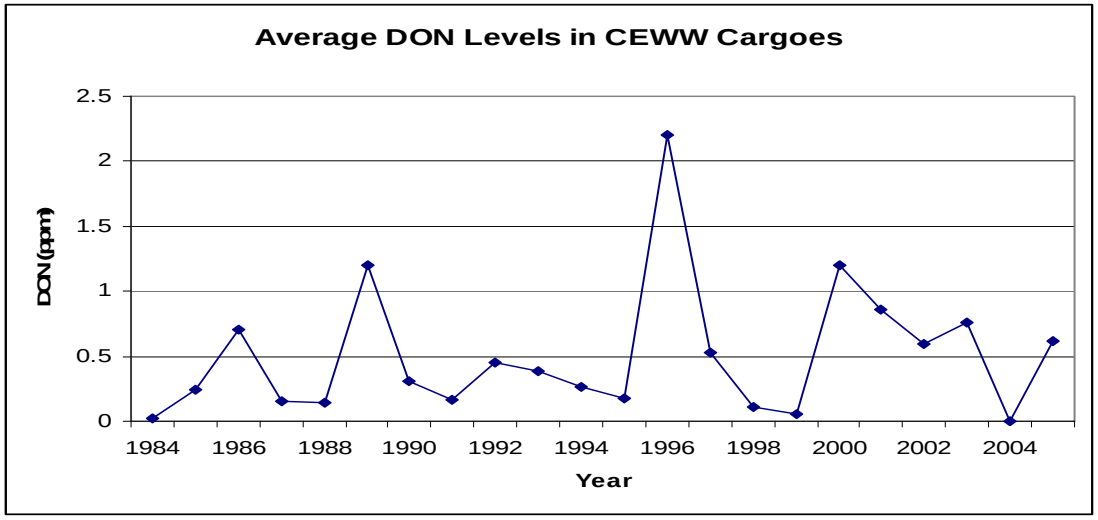


Figure 3. Deoxynivalenol in Canada Western Red Spring wheat

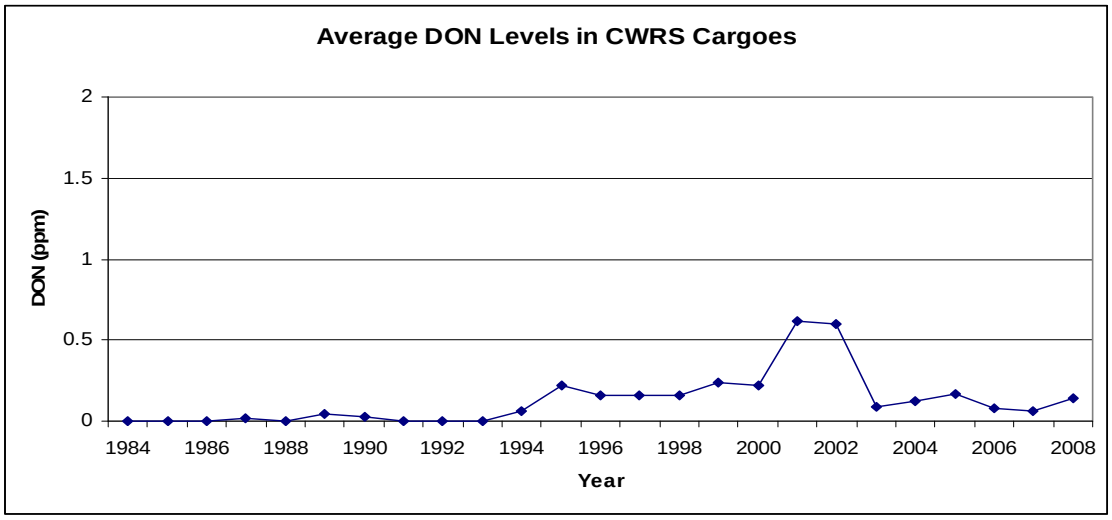


Figure 4. Tombstone/*Fusarium* damaged kernel tolerances

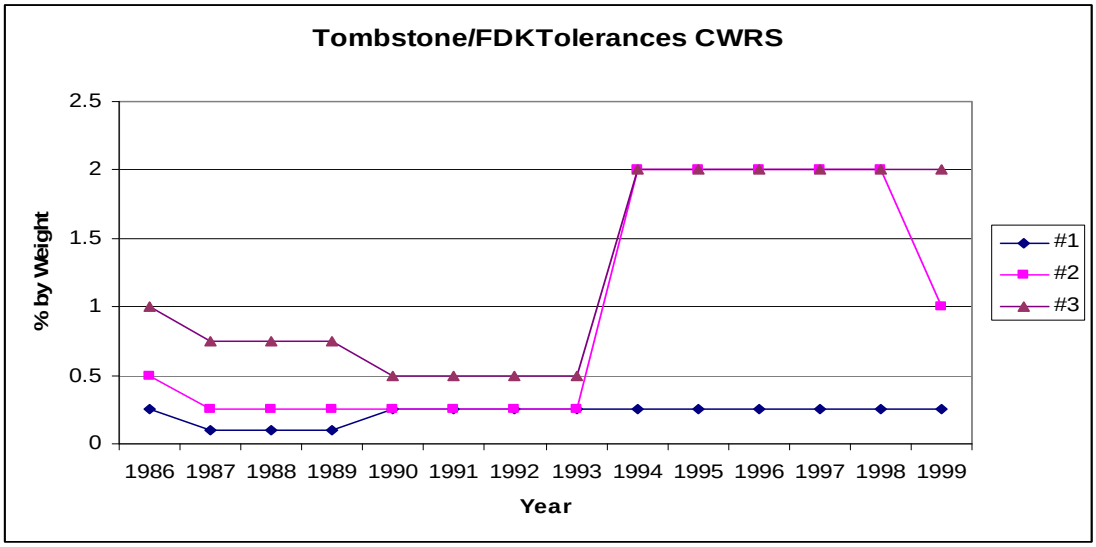


Figure 5. Hectares planted to amber durum wheat in Manitoba

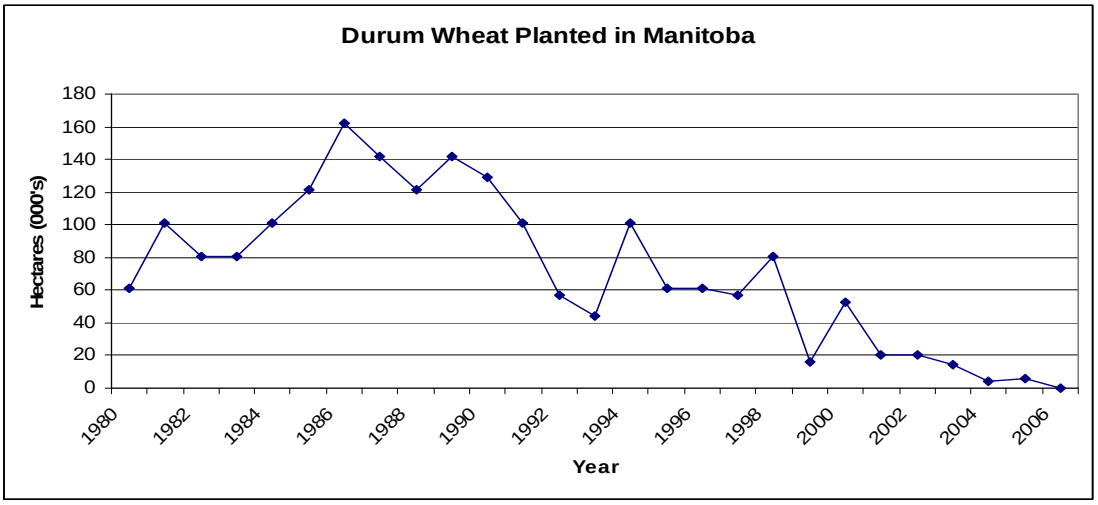


Figure 6. Hectares planted to barley in Manitoba

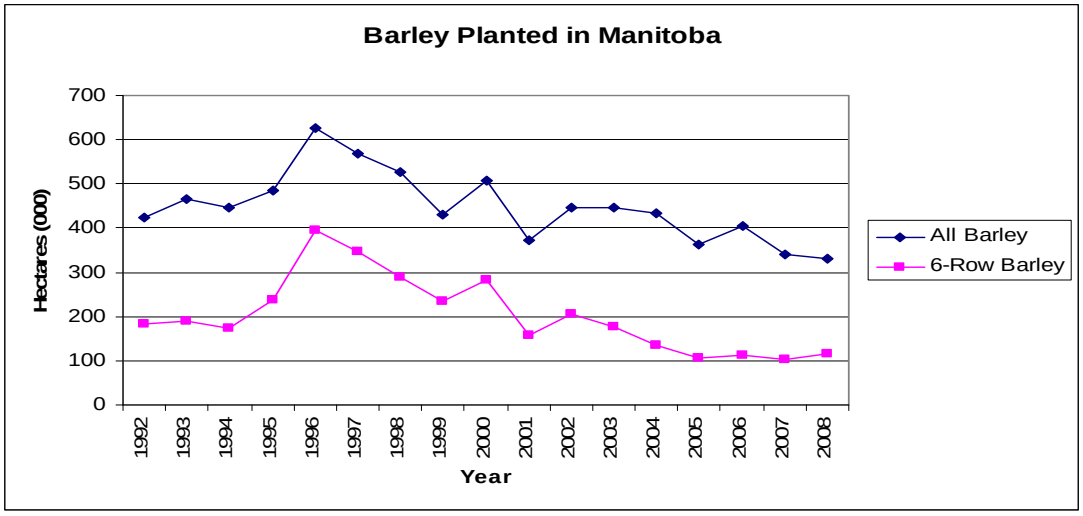
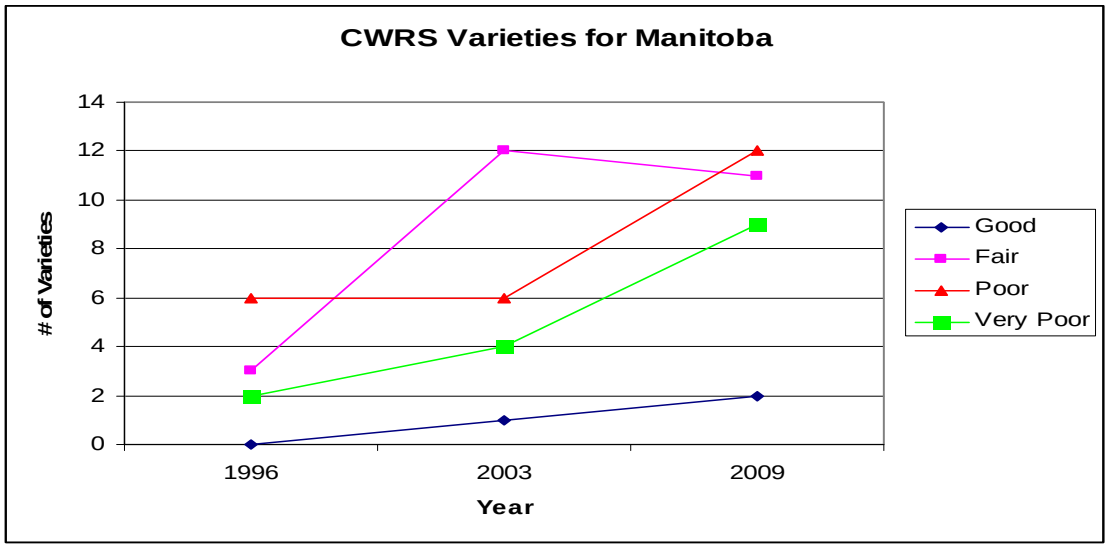


Figure 7. Fusarium head blight ratings of wheat varieties in Manitoba



T02. Genetics of fusarium head blight resistance in wheat

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In the wheat gene pool significant variation for resistance to fusarium head blight is evident. Resistance to fusarium head blight is a quantitative trait controlled by polygenes. During the past decade numerous studies have been performed to decipher the inheritance of fusarium head blight resistance in wheat. We summarize the relevant findings from 51 quantitative trait loci (QTL) mapping studies, 9 research articles on marker assisted selection and 7 on marker assisted germplasm evaluation. QTL for FHB resistance were reported on all wheat chromosomes except 7D. Some QTL were found in several independent mapping studies indicating that such QTL are stable and appear therefore useful in breeding programs. We summarize and update current knowledge on the genetics of fusarium head blight resistance in wheat and review breeding strategies based on the available information and DNA markers. A detailed review on FHB resistance QTL has been published recently by Buerstmayr *et al*. (2009) Plant Breeding 128:1-26.

In addition some information on ongoing research work at the group of IFA-Tulln will be presented, especially on mapping novel FHB resistance from *Tritcum macha*, *T. dicoccum* and *T. dicoccoides*, as well an ongoing work on the identification of candidate FHB resistance genes based on differential gene expression experiments.

T03. Progress in breeding for FHB resistance in Canadian spring wheat – 2009

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Breeding for resistance to fusarium head blight caused by *Fusarium graminearum* continues to be a primary objective for spring wheat breeding programs in western Canada, and the breeding effort is more relevant now due to the recognition that this pathogen is present in Alberta. Progress in the various spring wheat market classes since 2005 is evident in several indices including increased frequency of lines with potential resistance, increased frequency of lines with characterized FHB genes/QTLs and the recent successful registration of three cultivars that are moderately resistant to FHB. However, no FHB resistant cultivars have been registered.

Advancement in the art of breeding for FHB resistance is apparent in improvements in field nursery operation resulting in reliable annual results, ease of marker assessment due to fine mapping of FHB QTL regions and rapid throughput marker platforms, merging research information into breeding programs and identification of elite lines with intermediate, uncharacterized background resistance. Several breeding technology improvements are required including a review of the fine mapping around the FHB 6B QTL, utilization of other resistance sources, rapid assessment of FDK, expansion of DON testing and the impact of stacking known QTL's for FHB resistance into elite backgrounds.

T04. Introgression of two QTLs from exotic source in adapted European wheat

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At the beginning of the 21st Century, mankind faces the challenge of feeding a growing population with increasing demands in the quality of food. Land and water resources are limited and threatened by degradation worldwide. Forecasts of climate change indicate additional uncertainty and place even more pressure on our natural resources to provide enough food for everyone. Fusarium head blight (FHB) is one of the most important wheat diseases that causes yield and quality losses as well as contamination with deoxynivalenol (DON). This study presented here was aimed at the marker-based introduction of two previously mapped QTL from the exotic resistance source Sumai 3 into an elite background of two German winter wheat varieties. Phenotypic selection is a current method to improve FHB resistance in wheat breeding in Europe by exploiting the existing variability of a lot of loci with small effects in adapted and elite germplasm. Markers for several QTL have been published that may be useful for the development of FHB resistant cultivars in the future. The aim this study is (1) to validate the effectiveness of the resistance from the exotic Sumai 3, and (2) to examine the amount of linkage drag on agronomic traits. Detailed results of this study will be presented and discussed.

T05. Developing a YIPEE wheat cultivar

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Recombining multi-genes that confer resistance to fusarium head blight (caused by *Fusarium* spp.) with the myriad of genes for "on-farm" production traits, resistance to rusts and smuts, and high grain yield of premium end-use suitability has been a major challenge. Carberry represents a "step change" in variety development in the Canada Western Red Spring market class. Carberry sets a new standard for resistance to fusarium head blight coupled with high grain yield, high protein concentration, semidwarf stature with strong straw, and resistance to leaf rust, stem rust and common bunt. Based on 36 trials over three years, Carberry yielded similar to Superb, the highest yielding check. Carberry had protein concentration 0.5 units higher than Superb. These attributes were achieved without extending maturity beyond that of Superb, the latest maturing check. Carberry had significantly shorter straw than the semidwarf Superb, and had significantly higher volume weight than Superb. Carberry derives from parents Alsen and Superb. Access to FHB disease nurseries to phenotype large populations has been fundamental to shift the frequency of desirable gene combinations, to identify superior recombinants and to overcome negative associations. Doubled haploid technology accelerated production of inbred lines to be assessed against the battery of target trait requirements in the development of Carberry.

T06. Incidence of *Fusarium graminearum* and *F. poae* from a 2-year wheat monitoring: factors that promote infection and mycotoxin contamination

S. Vogelgsang, N. Bischofberger, F. Wettstein, and H.R. Forrer. *Research Station Agroscope Reckenholz-Taenikon ART, Reckenholzstrasse 191, 8046 Zurich, Switzerland.*

In a 2-year investigation, wheat samples and respective information on cultivation techniques were collected from Swiss growers. Wheat kernels were examined for *Fusarium* species incidence and mycotoxin content. From a total of 248 samples originating from 16 out of 26 cantons, three *Fusarium* species were dominant: *F. graminearum*, followed by *F. poae* and *F. avenaceum*. The average deoxynivalenol (DON) content was 1.0 ppm and thus barely below the European limit for unprocessed cereals (1.25 ppm). With respect to pre-crop maize, conservation tillage or ploughing resulted in an average DON content of 3.2 ppm or 0.6 ppm, respectively. Recently, we started to measure the content of other trichothecenes and zearalenone (ZON). Preliminary data (124 samples) suggest that nivalenol (NIV) and ZON production are correlated to similar production factors (NIV: average of 33.9 and 16.8 ppb for the two cropping systems; ZON: 73.5 and 10.7 ppb). However, since no correlation was found between *F. poae* incidence and both the NIV content and the two cropping systems, we assume the presence of *F. graminearum* NIV chemotypes. Ongoing toxin measurements, chemotype investigations of fungal isolates as well as in-depth analyses of the cultivation data should contribute to elucidate factors that influence the occurrence and toxin contamination by the most prevalent *Fusarium* species on wheat.

T07. Integrated management of fusarium head blight – Research and outreach

M. McMullen. *Department of Plant Pathology, North Dakota State University, Walster Hall, Fargo, ND, USA, 58102.*

Fusarium head blight (FHB) is a difficult disease to manage, and under environmental conditions favorable for FHB development, use of just a single management strategy may result in management failure. In the past decade, more FHB management research has focused on use of multiple techniques, and today, most published FHB management recommendations encourage an integrated approach. In 2007, the US Wheat and Barley Scab Initiative (USWBSI) began a multi-state research effort to examine use of two or more strategies in managing this disease; the goal - demonstrate to grain producers the value of an integrated approach. For example, research in North Dakota in 2007 showed an incremental improvement in reducing FHB parameters and increasing yield, by stacking strategies. The primary strategies examined in these USWBSI sponsored integrated studies have been host resistance, crop rotation and fungicide use. Disease levels have varied considerably across years and locations, but integrated strategies have proven reliable, regardless of disease level. An update on results of these USWBSI sponsored integrated management studies, as well as information on a new US outreach effort called *Scab Smart* will be presented. (www.scabsmart.org)

T08. Realistic expectations of foliar fungicides for management of FHB

L. Bourgeois and S. Henry. *Bayer CropScience, 5-160 Research Lane, Guelph, ON, CA N1G 5B2*

The performance of fungicide to manage FHB can be affected by fungicide type, crop variety, application quality and application timing. Current fungicide options for FHB management are limited to the triazole family and provide disease suppression averaging 25% for Folicur and 40% for Proline. The second factor is related to the responses to varieties to fungicides/disease interaction.

The susceptibility of varieties amongst cultivar is well documented but more research is needed in evaluating the interaction of the fungicides and the cultivars. Finally application quality and timing are essential for optimum performance of the fungicides. Coverage peaks at about 30% using the best spray technology available. Application timing is fleeting with drop of efficacy of fungicides prior to and after the optimum timing. Therefore fungicides require to be integrated with other FHB management tools in order to produce a quality crop with low *Fusarium* damage.

T09. Glyphosate associations with *Fusarium* diseases of cereal and pulse crops

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A summary of multi-year field studies on the effects of tillage systems and glyphosate use on the development of fusarium head blight (FHB) and common root rot (CRR) in wheat and barley in eastern Saskatchewan will be presented. Although environment was the most important factor determining FHB development, previous glyphosate use and tillage practice were among the production factors with the greatest association with FHB. Overall, disease was highest in crops under minimum-till management. Previous glyphosate use was consistently associated with higher FHB levels caused by *F. avenaceum* and *F. graminearum.* *Cochliobolus sativus*, the most common CRR pathogen, was negatively associated with previous glyphosate use, while other fungi, including *F. avenaceum* and *F. graminearum*, were positively associated, suggesting that glyphosate might cause changes in fungal communities.

Previous glyphosate applications were also correlated positively with *F. avenaceum* and negatively with *C. sativus* on crop residues. These studies established a relationship between previous glyphosate use and increased *Fusarium* colonization in wheat and barley. Because of the close association between noncereal crops, reduced tillage and glyphosate use, it was not possible to completely separate the effects of these factors on disease development. Research aimed at elucidating the nature of the association between previous glyphosate use and *Fusarium* crop infections is underway.

T10. What has been learned from six years of genomic research on the fusarium head blight pathogen?

H.C. Kistler. *USDA ARS Cereal Disease Laboratory, 1551 Lindig Street, University of Minnesota, St. Paul, MN, USA, 55108.*

Since its first public release in May, 2003, the genome sequence of *Fusarium graminearum* (*Fg*) has guided a broad range of research on fusarium head blight disease and toxicology. Using genomic information, over two dozen genes essential for full pathogenicity of the fungus have been identified. The basic genetic framework involved in pathogenicity for *Fg* consists of elements highly conserved with other fungi as well as pathways specific to *Fg* and closely related species. Microarrays have been designed based on the *Fg* genome sequence and these tools have led to a greater understanding of developmental processes essential for disease initiation and reproduction of the fungus such as spore germination and perithecium formation. Fundamental studies on the regulation of mycotoxin accumulation also have been aided by whole genome sequence information.

My talk will summarize these recent advances in genome biology and prospects for rational disease control and toxin reduction based on fundamental knowledge of pathogen biology.

T11. Biogeography of trichothecene chemotypes, and the origins of North American 3ADON populations

T. Ward. *Agricultural Research Service, United States Department of Agriculture, 1815 N. University St., Peoria, IL, USA. 61604,*

Previously, we documented a 14-fold increase in 3ADON-producing *F. graminearum* between 1998 and 2004 in western Canadian provinces. Significant population structure associated with trichothecene chemotype differences was observed, and isolates from the 3ADON populations were found to accumulate significantly more trichothecene and had higher fecundity and growth rates. Expanded molecular surveillance based on 4,266 *F. graminearum* isolated from seven Canadian provinces between 2005 and 2007 demonstrated the trichothecene chemotype distribution in Canada was characterized by two longitudinal clines. The frequency of 3ADON isolates continued to increase significantly in western Canada between 2005 and 2007. However, similar changes in chemotype frequency among isolates from eastern Canada were not observed.

These data suggest a difference in selective pressure acting on FHB pathogens in eastern and western Canada or an uncoupling of the 3ADON chemotype from the trait or traits under selection in some eastern Canadian FHB populations. Analyses of the global population structure of *F. graminearum* revealed a very close genetic relationship between a Japanese 3ADON population and the novel 3ADON populations in North America. Additional evidence of transcontinental movement of populations followed by limited genetic exchange between resident and introduced populations is presented.

T12. Alternate route to attain resistance against *Fusarium graminearum*

G. Subramaniam, C. Nasmith, L. Wang, and W. Leung. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Breeding programs have yielded many tolerant varieties of wheat over the years against fusarium head blight (FHB). Although, this line of defense against FHB will continue, it is imperative that we understand the underlying molecular mechanisms such that we can develop crop varieties that are durable in their resistance against FHB. Of the two types of resistance that are present in plants, resistance against *Fusarium* utilizes the ancient innate immunity pathway. This resistance pathway has evolved in plants to recognize the patterns or signatures of microorganisms in order to mount an effective response. In the long term, this type of response is also more durable. In order to gain insight into this pathway, our laboratory is involved in characterizing the patterns contained in *Fusarium* that will trigger the innate immunity pathway. A detailed characterization of one such pattern in the pathogen has allowed us to evoke the innate immunity pathway and provide resistance against FHB, even in the susceptible wheat cultivar Roblin.

T13. Molecular identification and databases in *Fusarium*

D.M. Geiser, S. Kang, and K. O'Donnell. *Department of Plant Pathology, The Pennsylvania State University, University Park, PA, USA, 16802. (K.O.) Microbial Genomics and Bioprocessing Research, National Center for Agricultural Research, US Department of Agriculture, Peoria, IL, USA, 61604.*

DNA sequence based methods for identifying *Fusarium* isolates have become standard as molecular technologies are increasingly available worldwide, and as more data are available in sequence databases for comparative purposes. Unfortunately, the use of BLAST against GenBank as an identification tool is in some ways becoming less reliable, as GenBank cannot possibly curate all submitted sequences and annotation for accuracy, and isolates associated with sequences are not necessarily available to researchers. In 2004, we released the first version of FUSARIUM-ID, a translation elongation-factor 1-alpha database of *Fusarium* on a BLAST server. All sequences in the database are from isolates that are available for distribution from major culture collections.

An updated version of the database was released in late 2008, with additional features including cross-reference to strain information and downloadable sequence data from multiple loci. In this talk, I will discuss these resources, and outline our ultimate goals to create a comprehensive bioinformatics resource that facilitates collaboration and integrates culture collection resources and phenotypic and genotypic data.

T14. Involvement of phytohormone signalling in FHB

P. Nicholson. *John Innes Centre, Norwich Research Park, Colney Lane, Norwich, United Kingdom, NR4 7UH.*

Studies with wheat near-isogenic lines have demonstrated that dwarfing alleles at the *Rht-B1* and *Rht-D1* loci enhance susceptibility to initial infection (Type I resistance). However, these mutations confer enhanced Type II resistance making their relative impact on overall disease dependent upon the infection pressure and relative importance of the two components in particular environments. Both *Rht-B1* and *Rht-D1* encode homologues of DELLA proteins that are negative regulators of GA signalling. The GA-insensitive semi-dwarf '*1b*' alleles encode stabilized versions of these proteins suggesting DELLA stabilization leads to enhanced resistance to DON-induced cell death.

Studies on interactions between *F. graminearum* and *Arabidopsis thaliana* indicate that ethylene and jasmonate pathways play contrasting roles in *F. graminearum* resistance. Abolition of ethylene synthesis/signalling resulted in reduced fungal growth and disease development whereas JA signalling plays a positive role in defence against *F. graminearum*. Partial silencing of *EIN2* (ethylene insensitive2) lines of Bobwhite wheat significantly enhanced resistance to FHB and suggests that *F. graminearum* exploits ethylene signalling to compromise resistance in monocot and dicot hosts.

We propose that genes involved in these signalling pathways may be suitable candidates for allele-mining in cereal crops for resistance to FHB. Identification of a new resistance gene in forward genetic screens in Arabidopsis will also be discussed.

T15. Expression profiling of FHB resistant wheat: what we have learned from it so far.

T. Ouellet, J. Hattori, S. Gulden, M. Balcerzak, H. Rocheleau, L.Wang, J. Singh, N.A. Foroud, G. Fedak, R. Pandeya, D. Somers, F. Eudes, and N. Tinker. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6. (D.S.) Cereal Research Center, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (N.A.F., F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403-1st Avenue South, Lethbridge, AB, Canada, T1J 4B1.*

To gain a better understanding of the response of wheat to *Fusarium graminearum*, expression profiling experiments have been performed by some laboratories in the last few years. These experiments have focused on three aspects: the changes occurring in gene expression after *F. graminearum* attack, the differences between susceptible and resistant cultivars (with or without fungal infection), and the effect of DON on the wheat response. The presentation will summarize the work done by our group of collaborators and relate them to observations by other teams.

T16. Transgenic field trials for FHB resistance and related research in wheat and barley

L. Dahleen, R. Dill-Macky, J. Shah, G. Muehlbauer, R. Skadsen, M. Manoharan, T. Abebe, and J. Jurgenson. *USDA-Agricultural Research Service, 1307 18th St N. Fargo, ND, USA, 58105. (R.D.-M.) Departmentt of Plant Pathology, University of Minnesota, St. Paul, MN, USA. (J.S.) Department of Biological Sciences, University of North Texas, Denton, TX, USA. (G.M.) Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN, USA. (R.S.) USDA-Agricultural Research Service, Madison, WI, USA. (M.M.) Department of Agriculture, University of Arkansas-Pine Bluff, AR, USA. (T.A., J.J.) Department of Biological Sciences, University of Northern Iowa, Cedar Falls, IA, USA.*

Transgenic wheat and barley lines expressing genes with the potential to reduce FHB and DON have been tested in field trials in MN since 1997 and in ND since 2001 (barley only). Replicated trials are planted, grown, and harvested to meet containment regulations of the Animal and Plant Health Inspection Service (APHIS) to prevent accidental escape of the transgenic organisms, and trials are routinely inspected. Harvested seed is evaluated for DON contamination in labs at MN and ND. Early transgenes used were typically genes for antifungal compounds or genes induced by pathogen infection. More recently, genes selected as specifically influencing FHB reaction in model systems have been inserted into wheat and barley. As homozygous lines are developed, they are tested in these field trials, providing direct comparisons with resistant and susceptible lines from wheat and barley breeding programs.

T17. Yeast genomic screening to identify cellular targets of *Fusarium graminearum* fungal toxins

A. Hermans, L. Mitchell, K. Baetz, W. Bosnich, L.J. Harris, and S. Gleddie. *Eastern Cereal and Oilseed Research Center, Agriculture and AgriFood Canada, 960 Carling Ave., Ottawa, ON, Canada, K1A 0C6. (L.M., K.B.) Ottawa Institute of Systems Biology, University of Ottawa, Ottawa, ON, Canada, K1H 8M5.*

The extensive homology between yeast and higher eukaryotic biochemical pathways means that high-throughput chemogenomic profiling in yeast has become a very powerful method to find drug and toxin targets. The yeast *Saccharomyces cerevisiae* genome contains about 6000 genes of which ~1000 are essential for survival. The yeast community has produced haploid non-essential gene deletion collections, heterozygous deletion collections, and over-expression collections (http://www.yeastgenome.org). These collections can be arrayed on agar plates in the presence of drugs or other compounds to provide extremely useful predictions concerning drug mode(s) of action, receptors, ligands, metabolic fate, and/or turnover. We have been using these resources to screen for cellular targets of the major mycotoxin produced by the cereal pathogen *Fusarium graminearum*. This poster will highlight our results of four genome-wide screens of the heterozygous deletion collection against sub-lethal doses of the mycotoxin deoxynivalenol (DON) which contributes to the virulence of this pathogen against cereals.

From these screens, we have selected a number of yeast strains whose gene-deletions have resulted in either increased or decreased sensitivity to DON. The effect of these gene deletions on yeast cell tolerance to DON was validated by sensitive real-time liquid growth assays in the presence of toxin for a 24 hrs period. These data confirmed the screening of yeast on solid medium as a predictor of mycotoxin sensitivity/resistance. DON is known to bind to the eukaryotic ribosomal protein L3 (RPL3), inhibiting protein synthesis. This screen has identified genes expected to be associated with DON tolerance/sensitivity such as those involved in protein synthesis, folding, transport as well as some novel targets. We will discuss the implications for the generation of knowledge for the development of *Fusarium* tolerant plants, and the elucidation of the targets and mode of action of this mycotoxin.

T18. Update on improving fusarium head blight resistant Canadian barley

W.G. Legge. *Brandon Research Centre, Agriculture and Agri-Food Canada, PO Box 1000A, RR 3, Brandon, MB, Canada, R7A 5Y3.*

Fusarium head blight (FHB) caused by *Fusarium graminearum* is the most important disease of barley in Canada. Good progress has been made over the past decade by Canadian barley breeding programs in developing FHB resistant cultivars and germplasm with low deoxynivalenol (DON) accumulation. The two-row feed barley cultivar CDC Mindon, registered in 2007, has set the standard with about half the DON content of AC Metcalfe over many years of testing. Two new cultivars, Norman and HB705, resulting from *in vitro* selection using *Fusarium* mycotoxins during doubled haploid production were registered in 2009. Norman is a two-row malting cultivar selected from CDC Kendall with 25-30% lower DON content than its parent, while HB705 is a two-row hulless cultivar with malting quality potential and lower DON content than other hulless cultivars. Progress has lagged in six-row germplasm. Most programs are in the second or third breeding cycle, and have better parents available for crossing purposes to enhance FHB resistance. In 2009, the FHB project in western Canada will replace selection based on visual symptoms for most advanced breeding lines with preliminary selection for DON content using near infrared reflectance (NIR) spectrometry to identify lines for further DON testing with standard methods. Funding constraints may pose a serious challenge to future progress.

T19. An overview of oat FHB studies in Canada

W. Yan. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave. Ottawa, ON, Canada. K1A 0C6.*

Studies on fusarium head blight (FHB) in oats are new and limited. In this paper we will provide an overview on the current activities related to oat FHB research and breeding in Canada under the following subtitles:

1) assessment of the FHB problem in oats,
2) identification of FHB-tolerant germplasm,
3) breeding for FHB-tolerant genotypes, and
4) proposed genetic studies to facilitate marker assisted selection.

It is now clear that oats are not immune to FHB as previously believed. In fact, oats can be highly susceptible to FHB under excessive inoculation and inductive conditions, suggesting that FHB can be a potential problem for oat production. In natural conditions, the phototoxic level produced by FHB in oats is generally low, but in some areas in some years, the DON level, averaged across multiple genotypes, was higher than 3 ppm, presenting a real problem for the oat industry. There are inheritable differences in oats in FHB tolerance. Tolerant lines have been identified and used in breeding programs. Some genetic studies on oat FHB have also been initiated, which may facilitate breeding for FHB tolerant oat cultivars.

T20. Breeding for resistance to ear rot and tolerance to mycotoxins in European maize

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Maize was grown in the European Union (EU27) in 2008 on 14 million hectares, 37% thereof as silage maize (DMK 2009). In the northern part of the growing region, maize is exclusively used for feeding, predominantly as silage maize and to a lower extent as grain maize, in the southern part only grain maize is grown for feeding and human food. Three maturity groups according to the agroclimatical conditions are described: "Early" (*dent x flint*) for Denmark, The Netherlands, Germany, Northern France; "Mid-late" (*dent*) for Southern France, Hungary; and "Late" (*dent*) for Spain, Italy, the Balkan States, Romania. In the northern parts of Europe, ear rot is primarily caused by *Fusarium graminearum* (FG), but 12 other *Fusarium* species, including *F. verticillioides* (FV), were isolated from naturally infected maize in Germany with the prevalent species changing between years according to weather (Görtz *et al.* 2008). In Southern Europe, *F. verticillioides* prevails, but *F. subglutinans* and *F. proliferatum* also occur frequently (Bottalico *et al.* 1998. Logrieco *et al.* 2002). Ear rot reduces both yield and quality of grain and leads to mycotoxin contamination. FG produces among others the mycotoxins deoxynivalenol (DON), zearalenone (ZEA) and their derivatives, whereas FV is a producer of fumonisins (FUM). All these mycotoxins can cause severe diseases in animals and humans and legal limits for human food exist within the European Union (Table 1).

Table 1. Maximum levels for *Fusarium* toxins in foodstuffs in maize and maize products in the European Union (EC 1126/2007)

Mycotoxin	Foodstuff	$\mu g\ kg^{-1}$
Deoxynivalenol	Unprocessed (except for wet milling)	1750
	Human food	750
	Breakfast cereals	500
	Baby food for infants and young children	200
Zearalenone	Unprocessed (except for wet milling)	350
	Refined maize oil	400
	Human food incl. snacks, breakfast cereals	100
	Baby food for infants and young children	20
Fumonisins (B_1+B_2)	Unprocessed (except for wet milling)	4000
	Human food	1000
	Snacks, breakfast cereals	800
	Baby food for infants and young children	200

For feed of animals recommended levels vary between 2,000 – 8,000 μg kg^{-1} for DON and FUM and 250 – 500 μg kg^{-1} for ZEA depending on animal species and its age. These levels can be largely exceeded when natural infection occurs (Logrieco *et al.* 1995). In Europe, no fungicide for control of infection has been released in maize. Lower mycotoxin concentrations of US and Canadian maize due to higher ear rot resistance have been reported for both *Fusarium* spp. (Reid *et al.* 1996b; Robertson *et al.* 2006). Breeding and growing resistant varieties is the only alternative to fulfill legal limits and recommendations. Our overall aim was to estimate basic population parameters for optimizing resistance selection to FG and FV.

Three maturity groups (early, mid-late and late) were evaluated for resistance to FV across two years. The early maturity group consisted of 50 flint and 90 dent inbred lines, the mid-late and late group of 147 and 148 dent lines, respectively. Additionally, the same genotypes of the early maturity group were grown in adjacent but separate trials to evaluate resistance to FG. From grain of each of 60 (early) and 50 lines (mid-late, late) mycotoxin contents were analysed from all trials and from the same lines testcrosses with each of two testers of the respective opposite heterotic group were grown. All lines were current breeding lines of the KWS SAAT AG. Locations for evaluation of the early maturity group were Einbeck (EIN; Central Germany), Gondelsheim (GON; South Germany) and Chartres (CHA; North France). The mid-late group was tested in Alzonne (ALZ; South France) and Murony (MUR; Hungary) and the late maturity group in Monselice (MCE; North Italy). All genotypes were grown in one-row plots 4 m long with 0.75 m distance. Inoculum production and inoculation were done according to Reid *et al*. (1996). Primary ears of ten inbred lines or testcrosses per plot were silk-channel inoculated with 1 or 2 ml of inoculum, respectively, with concentrations of 1×10^5 and 1×10^6 conidia ml^{-1} for FG and FV, respectively. The percentage of mycelium coverage on the ear was rated at harvest (0-100 %). Inoculated ears of selected genotypes were harvested, dried, shelled and the kernels were milled. Samples were analyzed with the immunoassay RIDASCREEN® FAST-DON, FAST-ZEA and FAST-FUM (R-Biopharm, Darmstadt, Germany). Single plot data were used for analyses of variance (ANOVA), correlations were calculated with mean values of each trial. For details please refer to Löffler *et al*. (2009).

Inoculation was superior to natural infection because of higher disease severities and heritabilities although natural FV infection in southern Europe often resulted in similarly high ear rot severity than artificial inoculation. Broad ranges and significant ($P<0.01$) genotypic variation allow breeding inbred lines resistant to ear rot and mycotoxin accumulation for all maturity groups and both *Fusarium* species (Table 2). Selection is complicated by significant ($P<0.01$) genotype × environment interactions. Heritabilities of ear rot rating were similar or higher than those of mycotoxin concentrations.

Table 2. Means, variance components for genotype (V_G) and genotype x environment interaction ($V_{G\ x\ E}$), heritabilities and genotypic correlations (r_G) between ear rot rating and the respective mycotoxin concentration for three maturity groups with each of 60 (early) and 50 inbred lines

Maturity group	*Fusarium* species	Trait	Mean	V_G	$V_{G\ x\ E}$	Herita-bility	r_G rot-toxin
Early	FG	Ear rot (%)	49.2	392.6**	188.9**	0.90	-
		DON (mg kg^{-1})[a]	438.1	0.95**	0.52**	0.89	0.99++
		ZEA (mg kg^{-1})[a]	26.8	0.21**	0.20**	0.82	0.97++
	FV	Ear rot (%)	11.5	0.21**	0.16**	0.72	-
		FUM (mg kg^{-1})[a]	68.6	0.07**	0.04**	0.74	0.97++
Mid-late	FV	Ear rot (%)	9.9	0.97**	0.39**	0.91	-
		FUM (mg kg^{-1})[a]	20.4	0.59**	0.23**	0.92	0.87++
Late	FV	Ear rot (%)	11.7	0.53**	0.25**	0.69	-
		FUM (mg kg^{-1})[a]	43.4	0.11**	0.06**	0.58	0.95++

**, Significant at P<0.01. ++, Estimate exceeded twice its standard error.
[a] Natural log (FG) and fourth root (FV) transformed data, respectively.

High genotypic correlations between ear rot and corresponding mycotoxin concentrations (Table 2) suggest that quantitatively resistent lines had lower mycotoxin contents than susceptible ones. This allows the breeder frequent identification of lines with reduced mycotoxin concentrations by superior ear rot rating. Among the mycotoxin concentrations tested high correlations were found between DON and ZEA produced by *F. graminearum* but also between these two mycotoxins and FUM produced by *F. verticillioides*. Correlation between inbred line and testcross performance for 60 (early) and 50 (mid-late, late) entries was moderate in early and mid-late maturity group but poor in the late group as preliminary seen from one-year data.

In early maturing lines, FG caused significantly (P<0.01) higher ear rot severity than FV. The distribution of ear rot rating after FG inoculation reflected a normal distribution but ratings after FV inoculations were generally skewed to lower ear rot severity (Fig. 1). Both are in good agreement with studies from Canadian or US maize for FG and FV, respectively (Clements *et al*. 2004; Reid *et al*. 2002; Robertson *et al*. 2006). The lower aggressiveness of FV might be caused by its dual nature as an endophyte with symptomless infection or a pathogen causing symptoms more randomly (Bacon *et al*. 2008). Correlation between FG and FV severity was moderate in flints and dents (r=0.59 and 0.49, respectively), but lines resistant to both fungi clearly exist.

Chances for selecting improved European elite maize material is promising by multi-environ-mental inoculation trials. Separate testing of FG and FV is necessary since genotypes resistant to DON or ZEA were not necessarily resistant to FUM accumulation. Regarding the high costs of mycotoxin quantification and low costs of ear rot rating, indirect selection on ear rot rating is more effective than direct selection on reduced mycotoxin concentrations assuming a fixed budget. Testcrosses must be tested separately because the resistance of inbreds alone does not allow a secure prediction. Breeding for resistant varieties is an urgent need in view of the high mycotoxin contents found even in less infected samples.

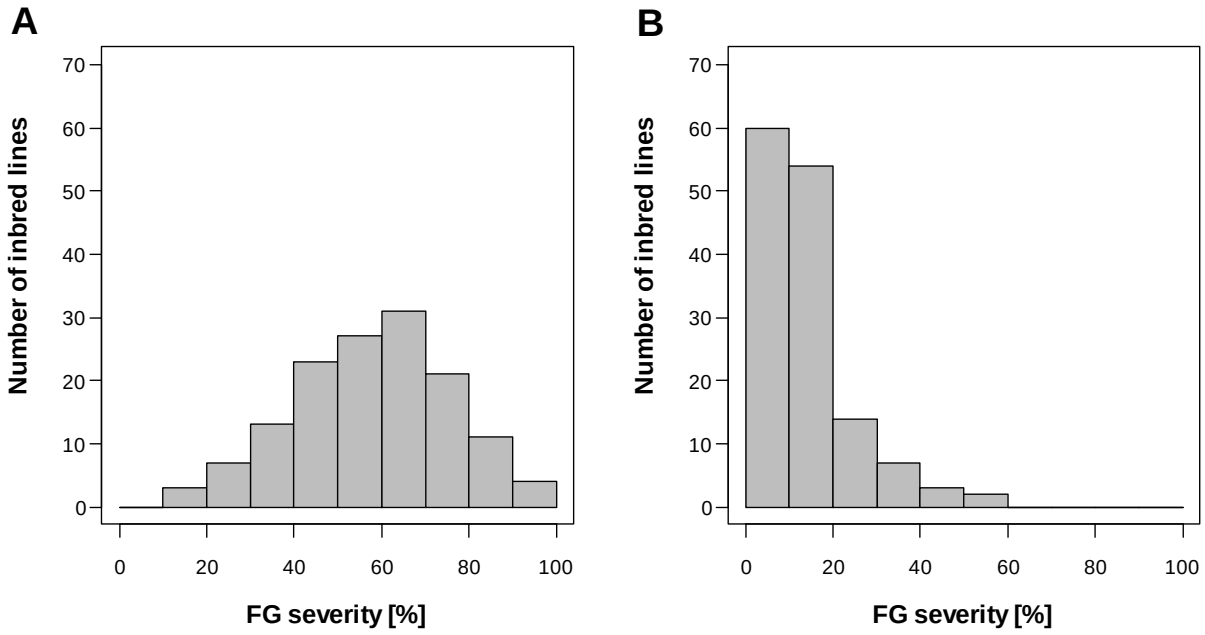

Fig. 1. Frequency distribution of 140 inbred lines inoculated by *Fusarium graminearum* (A) and *F. verticillioides* (B) in a factorial design at six environments in the early maturing group

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T21. Biological management of fusarium head blight of wheat with *Clonostachys rosea* strain ACM941

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Fusarium head blight (FHB), caused by *Gibberella zeae*, is a harmful disease of wheat. A strain of *Clonostachys rosea*, ACM941 (ATCC #74447) was evaluated for its ability to inhibit perithecial production of *G. zeae* and for the control of FHB and deoxynivalenol (DON) contamination under greenhouse and field conditions, in comparison to the registered fungicide Folicur® (tebuconazole). ACM941 reduced *G. zeae* perithecial production by >99% in a leaf disk assay and by >60% under field conditions. In the greenhouse trials, ACM941 significantly reduced infected spikelets (IS) by 64% and *Fusarium* damaged kernels (FDK) by 65%. Under simulated disease epidemic conditions during 2005-2007, ACM941 significantly reduced the FHB index by 58%, IS by 46%, FDK by 49%, and deoxynivalenol (DON) in kernels by 21%. ACM941-CU, a formulated product of ACM941, was evaluated in two field trials in 2008. Over the average of the two trials, ACM941-CU reduced FHB index by 31.4%, FDK by 43.8%, and DON by 37.1%. These effects were significant but less than those achieved by tebuconazole in the same trials, suggesting that ACM941 is a promising bioagent against *G. zeae* and may be used as a control measure in an integrated FHB management program.

T22. Biocontrol and bioprotection fungi against FHB and mycotoxin accumulation in cereals

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In North America, fusarium Head Blight (FHB) causes severe yield losses in wheat and barley, thus greatly impacting Canadian agriculture. Saskatchewan prairies represent approximately 40% of the overall cultivated land in Canada and are one of most important cereal crop regions worldwide. Fungi can counteract *Fusarium* species causing FHB and hence prevent the appearance of the disease and accumulation of associated mycotoxins (deoxinivalenol, nivalenol and zearalenone). Optimal biocontrol seems to rest within *Fusarium*-specific mycoparasites, whereas bioprotection mostly depends on plant-specific endophytes of wheat and barley. The overall aim is the creation of a new generation of beneficial bioinoculants efficient against FHB. These are considered environmentally friendly or green solutions for feed and food safety, as well as economic profitability of cereal crops.

T23. *Fusarium* mycotoxins: biosynthetic pathways and role in virulence

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Fusarium head blight (FHB) of wheat and barley is a devastating disease that has reached global proportions. Not only does this disease result in lower yields, but the mycotoxins produced by the fungus affect the quality of the grain. *Fusarium* sp. can produce a number of mycotoxins. The trichothecenes most commonly found in infested grain are deoxynivalenol (DON) and nivalenol (NIV), the former of which has been identified as a virulence factor for FHB. The identification of genes involved in the formation of trichothecenes has lead to the description of a complex biosynthetic pathway. The physical location of the genes involves at least three chromosomes yet expression of many of the toxin biosynthetic genes is coordinated by the products of at least two genes located within the core cluster of twelve genes. The identification of genes involved in the formation of the mycotoxins butenolide and culmorin has been aided by the production of expressed sequence tag (EST) libraries. Searching the libraries for genes expressed at high levels during mycotoxin formation provided a number of candidate genes. Gene knock-out studies, as well as transgenic expression studies, provided the proof of function of these genes. Studies are underway to determine if culmorin and/or butenolide are virulence factors for FHB.

T24. A tale of contrasting economic reaction to mycotoxin in maize and wheat in the same agricultural system in Ontario, Canada

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In 1996 Ontario experienced the worst *Fusarium* epidemic in recent history in wheat. This event precipitated a gathering of industry stakeholders to design strategies to collectively prevent further harm to the sector by supporting and developing an integrated management system. In 13 years this strategy yielded significant progress on: breeding for more tolerant varieties, an improved variety registration system, implementing fungicide recommendations and improving application technologies, developing a pre-harvest forecasting system (DONcast) as well as setting up surveillance strategies and mycotoxin testing. In contrast, more than 20 years have passed since the first severe epidemic of *Fusarium* in corn in 1986 and very little progress has been achieved toward the development of an integrated approach as was seen in the severe epidemic of 2006. This paper makes comments on why we believe there is a contrasting approach between these two cropping systems within the same region. In summary, the corn hybrid registration requirements were abandoned approximately 10 years ago. There is a distinct difference between the two commodities in their end use and the associated mycotoxin regulations (feed/industrial for corn versus food for wheat) and a different approach to grain trading giving more opportunities to mitigate mycotoxin problems by dilution in the corn market. The advent of transgenic corn and the logistical challenges of handling seed inventory led to a narrow genetic pool of widely adapted high yielding hybrids, perhaps at the expense of some disease resistance.

T25. Identification of a biomarker for deoxynivalenol risk assessment and regulation

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Deoxynivalenol (DON) is the most commonly detected trichothecene fungal metabolite in cereal grains and processed food globally. Upon exposure, DON is rapidly distributed in animal tissues and induces proinflammatory cytokines (< 2 h). Longer term (> 2 wk) DON exposure reduces weight gain in many species through a less understood mechanism, thus creating uncertainties in human safety assessment. We hypothesized that DON-induced weight reduction is preceded by a dysregulation of growth pathway-related proteins. Models of acute and chronic DON exposure were used to test this hypothesis. We determined that DON acutely induces hepatic suppressors of cytokine signaling (SOCS). The effect of SOCS on growth pathway was evaluated by measuring forms of insulin-like growth factor acid-labile subunit (IGFALS), a growth-related protein. Acute DON exposure (0.1-12.5 mg/kg) impaired growth hormone-induced IGFALS mRNA by 60-80%. Furthermore, dietary DON (20 ppm for 8 wk) suppressed IGFALS mRNA (65%), circulating IGFALS (66%), weight gain and elevated plasma DON (≤ 63 ng/ml). In obese mice, dietary DON (10 ppm) also suppressed circulating IGFALS by 42 %. Together, these data indicate that dietary DON consistently suppressed IGFALS in lean and obese mice, while elevating plasma DON. Therefore, circulating IGFALS is a potential biomarker for DON's effect, and might be used for epidemiological surveillance and human risk assessment.

T26. Alberta Seed Testing Laboratory perspectives: *Fusarium graminearum* testing methodology and detection

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The 1999 declaration of *Fusarium graminearum* in the Alberta Agricultural Pest Act resulted in the 2002 release of AAFRD's Alberta *Fusarium graminearum* Management Plan. A crucial component of the plan is the requirement for all cereal grains intended for seed use to be tested for the presence of *F. graminearum.*

Since the inception of the plan in 2002, tens of thousands of cereal grain seed samples have been tested in Alberta using one (or both) of 2 testing methodologies (the whole seed PDA plate culture or a PCR method). Between the 2001/2002 and the 2008/2009 crop years, the number of wheat samples testing positive for *F. graminearum* has increased from 4.6% to over 15% in southern Alberta whereas infection in cereal samples submitted from northern Alberta is rare. PCR data, which analyzes large samples with high sensitivity, suggest that the percentage of wheat samples testing positive across Alberta has doubled annually since 2006. Plate tests, used to quantify the number of seeds infected with *F. graminearum* within a sample (≥0.5%), further shows that the highest level of infection detected in the 2008/2009 crop year from southern Alberta was 15.5%, with an average infection leve1 of 1.86%.

F. graminearum is detected in other cereals in Alberta to a much lesser extent.

T27. AAFC Pest Management Centre Risk Reduction Strategy Mini-session

C. Kora. *Pest Management Centre, Agriculture & Agri-Food Canada, Building 57, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

The FHB has been identified as a top priority issue for wheat industry in every wheat growing region of Canada. The AAFC's Pesticide Risk Reduction Program initiated preliminary stakeholder consultations on the issue and potential solutions since 2006, but the strategy development process was initiated in February 2009. A working group, with a cross sector representation of stakeholders (including industry groups, growers associations, AAFC and university researchers, and provincial specialists) was established to discuss key pest management gaps and propose reduced risk solutions to the issue. An action plan is being prepared based on the specific management gaps and solutions identified by the working group members. This plan will reflect the areas where PRRP has been and will be actively supporting for developing and implementing reduced risk solutions to address FHB. The goal is to minimize the use of, or improve efficiency of currently available fungicides with the use of newly developed reduced risk tools and practices. The need for alternative solutions becomes more important as a replacement in the event that any of the old chemistries may be lost as a result of the PMRA's re-evaluation process.

List of Poster Abstracts

Breeding & Markers

Disease Management

Biology of the Disease

Mycotoxins

Poster Abstracts

P01. Molecular mapping of resistance to fusarium head blight derived from three triticum species. M. Buerstmayr, K. Huber, A. Alimari, J. Heckmann, M. Lemmens, B. Steiner, and H. Buerstmayr. *University of Natural Resources and Applied Life Sciences Vienna, Department IFA-Tulln, Konrad Lorenz Str. 20, A-3430 Tulln, Austria.*

While many reports on genetic analysis of *Fusarium* resistance in bread wheat have been published during the past decade, only limited information is available on FHB resistance derived from wheat relatives, and from tetraploid wheats so far. In this contribution we report about genetic analysis of FHB resistance derived from three *Triticum* sources: 1) *Triticum macha* (Georgian spelt wheat), 2) *Triticum dicoccum* (cultivated emmer) and 3) *Triticum dicoccoides* (wild emmer). Back-cross derived recombinant inbred line populations were developed from crosses of the resistance donors with adapted cultivars. The populations were evaluated for *Fusarium* response in well replicated experiments with artificial inoculation. The same lines were genetically analysed using SSR and AFLP markers. Map construction based in the backcross derived RIL populations was done with Carthagene and QTL mapping in Qgene. Several novel QTL were identified. In *T. macha* five new QTL were found on four chromosomes (2A, 2B, 5A, 5B), the largest effect QTL overlapped with the *Q-locus* (spelt type) on 5A. In *T. dicoccum* the largest QTL mapped to chromosome 4B (overlapping with *RhtB1*). In wild emmer (*T. dicoccoides*) significant QTL were detected on chromosomes 3A and 6B.

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P02. Effectiveness of marker assisted selection for resistance to fusarium head blight in a wheat backcross breeding program. W. Cao, G. Fedak, D. Somers, C. McCartney, A.G. Xue, M.E. Savard, and H.Voldeng. *Eastern Cereal and Oilseed Research Center, Agriculture and Agri-Food Canada, Ottawa, ON, Canada K1A 0C6. (D.S. and C.M.) Cereal Research Center, Agriculture and Agri-food Canada, 195 Dafoe Rd, Winnipeg MB, Canada, R3T 2M9.*

Marker assisted selection (MAS) has been considered as a useful tool in wheat breeding programs. The objective of this study was to determine the effectiveness of MAS relative to conventional visual selection (CVS) for resistance to FHB in a spring wheat backcross breeding program. BW301, an elite line but susceptible to FHB, was crossed in 2002 with HC374, a line resistant to FHB and carrying three QTLs, and the F_1 backcrossed to BW301. A MAS population (MAS BC_2F_5) has been developed through an F_2-derived method, while two conventional visual selection populations (CVS BC_1F_6 and BC_2F_5) have been developed though single seed descent. Seven lines with all three FHB QTLs were selected from the MAS population, while the top 10 resistant lines were selected from CVS BC_1 and CVS BC_2 populations, respectively, based on FHB symptoms in the FHB nursery of 2005. The 27 lines, plus two parents, were further evaluated for FHB resistance in a four replicate field experiment in 2006 and 2007. A combined analysis from two years data showed that the means for FHB incidence, severity and FHB index and deoxynivalenol (DON) content for the MAS population were 39.2%, 27.3%, 11.2% and 4.6 ppm. For the CVS BC_1 population these values were 43.0%, 30.8%, 14.4% and 6.1 ppm and for the CVS BC_2 population they were 50.6%, 41.2%, 21.6% and 9.1 ppm. Marker profiles showed that the lines in the CVS BC_1 population carried from 0 to 2 QTLs whereas none of the lines in CVS BC_2 population carried the FHB resistance QTLs. The results confirmed that MAS is more effective than CVS for improving FHB resistance in wheat backcross breeding programs, and indicated that in a CVS program, the more backcrosses, the greater the risk of loss of resistance QTLs. The results also suggested that minor genes have an important role in providing resistance to FHB of wheat.

P03. Application of MAS for development of white seeded wheat resistant to fusarium head blight. W. Cao, G. Fedak, D. Somers, H. Voldeng, A.G. Xue, M.E. Savard, and S.S. Miller. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave. Ottawa, ON Canada, K1A 0C6. (D.S.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9.*

Fusarium head blight (FHB) is an important disease of wheat, and Sumai 3, a Chinese wheat cultivar, has been used as a source of resistance to FHB in almost all wheat breeding programs. We are attempting to develop a white seeded wheat with a high level of FHB resistance using marker assisted selection (MAS). Snowbird, a FHB susceptible white hard white seeded wheat registered by the CRC in Winnipeg, was crossed to Sumai 3 as a female parent. Twenty thousand F_2 plants were produced and grown in the greenhouse. One thousand and five hundred white seeds were visually selected from the F_2 population. This population was advanced to F_5 by single seed descent. At the seedling stage of F5, a MAS was performed for three FHB QTLs on chromosome 5A, 3B and 6B. Two hundred and fifty F_5 lines were selected with two or three resistance QTLs and grown in a FHB nursery in 2008. Fifteen F_6 lines were selected based on FHB resistance and agronomic performance. Seed of the 15 lines was increased in the greenhouse in the winter of 2009. These 15 lines and two parents Sumai 3 and Snowbird, plus AC Vista as a check were planted in the FHB nursery with three replications and in a preliminary yield trial with two reps in the summer of 2009. The results showed that several white seeded wheat lines had high levels of resistance to FHB; significantly higher than Snowbird and matured earlier than Sumai 3. Quality is also improved significantly compared to Sumai 3, based on the Glutomatic test.

P04. Systemic germplasm development delivers more fusarium head blight resistance together with maximum test weight in bread wheat. A. Comeau, F. Langevin, Y. Dion, S. Rioux, H. Voldeng. *(A.C., F.L.) Soils and Crops Research and Development Centre, Agriculture and Agri-Food Canada, Québec, QC, Canada, G1V 2J3. (Y.D.) CÉROM, Saint-Mathieu-de-Beloeil, QC, Canada J3G 2E0. (S.R.) CÉROM, Québec, QC, Canada, G1P 3W8. (H.V.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, K1A 0C6.*

In breeding trials in eastern Canada, a test weight (TWT) (syn. specific weight) above 82 kg/hL is seldom obtained. Going back from 2003 to 2008, in most years, less than 1% of the TWT data of individual registration candidate lines at any given yield trial site went above 82. In 2009, in artificially inoculated fusarium head blight (FHB) resistance trials in Lévis, cv. SS Blomidon had a TWT near 73; bread-making check cultivars had TWT values near 74-76, and yet many progenies of the systemic selections from the cross AB143 had a TWT above 83. This cross had been selected annually (from F_1 to F_7) for multiple disease and stress resistance, including very high FHB resistance, through a systemic methodology. High TWT is not totally equivalent to FHB resistance; grain shape and composition also alter TWT, and any stress during grain fill also reduces it. However, although so many factors can reduce TWT, whenever FHB plays a role, maximal values above 83 can be attained only if the resistance-tolerance factors include very high FHB resistance. Selection for TWT is not costly. This trait is very important for the wheat industry. A stringent use of TWT selection can be recommended to increase FHB resistance and also obtain stable cultivar performance.

P05. Approaches to improving the FHB resistance of triticale. G. Fedak, W. Cao, J. Gilbert, A.G. Xue, A. Comeau, F. Eudes, and A. Singh. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave. Ottawa, ON, Canada, K1A 0C6. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (A.C.) Soils and Crops Research and Development Centre, Agriculture and Agri-Food Canada, 2560 Hochelaga Rd. Quebec, QC, Canada, G1V 2J3. (F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403 1-Ave S, PO Box 3000, Lethbridge, AB, Canada, T1J 4B1. (A.S.) Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, Swift Current, SK, Canada, S9H 3X2.*

We are using several approaches to improve the fusarium head blight (FHB) resistance of triticale. A screening of genebank accessions has identified TMP 16315 with a good level of resistance to Type I, Type II and Type III. This accession has been crossed to AC Ultima and the production of a mapping population by means of microspore culture is in progress. A second approach is to produce new combinations of primary triticale using FHB-resistant Brazilian rye landraces such as Boller and Vacaria. These have been crossed onto Canadian durum cultivars such as Strongfield and Brigade and the amphiploids have been produced. Another approach is to enhance the FHB resistance of durum wheat as a prerequisite to the production of new combinations of primary triticale. Firstly, after extensive screening, a strain of *Triticum carthlicum* (4x) with good type II resistance was identified. Resistance QTLs were mapped on chromosome 6B, while a QTL for resistance in Strongfield was mapped to chromosome 2B. This same population was used to map a QTL for Type I resistance on chromosome 5A. An attempt was made to introgress the FHB resistance QTL of Sumai3 into Strongfield durum. Preliminary observations indicate that a few durum lines from this combination have enhanced levels of FHB resistance. Finally a durum line with enhanced FHB resistance was isolated from hybrids with tritordeum (AABBHH).

P06. Two new sources of F HB resistance in bread wheat. G. Fedak, W. Cao, J. Gilbert, A.G. Xue, M.E. Savard, and H. Voldeng. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave. Ottawa, ON, Canada, K1A 0C6. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9.*

Our studies on the introgression of FHB resistance from alien sources into bread wheat and durum are continuing. In one phase of these studies, two bread wheat lines have been produced with resistance from two different wild wheat species. The resistance of M321 is derived from *T. monococcum*, while the resistance of S184 is inherited from *Ae. speltoides*. Both have an AC Superb genetic background. Some of the linkage drag that was present in segregating progenies included late maturity, excessive plant height and other undesirable agronomic traits. As a result of selection against these factors, these two lines have minimal linkage drag. For example, in terms of grain yield, days to heading, test weight and thousand kernel weight the two lines are very similar to AC Barrie, the check cultivar. In terms of plant height, M321 was slightly shorter than AC Barrie, while S184 was slightly taller. The protein content of the two lines was nearly identical to that of AC Barrie. The grain samples of AC Barrie gave a flour yield of 66.8%, S184 gave 67.2% and M321 gave 57.5%. The FHB resistance of the two lines has remained stable. In the latest test M321 and S184 had DON contents of 1.6 and 0.7 ppm respectively compared to AC Barrie at 4.5 and Roblin at 10.4 ppm. Molecular markers, to facilitate the pyramiding of these sources of resistance with other known sources, are being developed. Thus far a unique QTL for resistance has been mapped on chromosome 5A of M321.

P07. The effect of three spray inoculation protocols on fusarium head blight infection of cultivars of common wheat and of durum wheat. A.T. Guerrieri, A. Brûlé-Babel, W.G.D. Fernando, and J. Gilbert. *Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada R3T 2M9.*

Fusarium head blight (FHB) caused by *Fusarium graminearum* Schwabe is a serious disease of wheat capable of causing severe losses in yield and quality. Reliable measurement of cultivar FHB reaction is key to successful resistance breeding. The objective of this study was to compare the effect of three commonly used field macroconidial inoculation protocols on expression of resistance in eight cultivars of spring wheat and two cultivars of durum wheat that differ in reaction to FHB. For the three protocols, plots were inoculated at: 1) 50% anthesis and three days later; or 2) first anthesis and then at three day intervals until the end of anthesis; or 3) the beginning of heading and at three day intervals until the soft dough stage. An uninoculated control was also included. Disease incidence and severity were measured at three day intervals from the onset of first symptoms through to senescence. Although there were absolute differences in disease incidence and severity among the protocols over two years of testing, relative ranking of cultivars was consistent and highly correlated across inoculated protocols. As anthesis is the stage of maximum susceptibility, differences in time to anthesis between cultivars required that comparisons of cultivar disease reactions be made at a consistent time from anthesis.

P08. An alternative path to fusarium head blight (FHB) resistant wheat cultivars: expression rather than introgression. S. Haber, J. Gilbert, and D.L. Seifers. *(S.H., J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Rd., Winnipeg, MB, Canada, R3T 2M9. (D.L.S.) Kansas State University-Hays, Hays, KS, USA, 67601-9228.*

Decades of sustained effort attest to the difficulty of generating FHB-resistant spring wheat cultivars. The common assumption underlying these efforts is that discrete genes conditioning superior FHB resistance must be introgressed from sources such as Sumai 3 into elite germplasm. An alternative path could start from a very different assumption. FHB-susceptible near-isogenic lines of Sumai 3, like Sumai 3 itself, carry pathogenesis-related genes that are induced by *Fusarium graminearum* Schwabe. This suggests that the key to FHB resistance is the control of expression of critical genes that are already present. A scheme that might generate variation in expression was suggested when we observed that progeny derived by selfing of plants under pressure from systemic virus infection could vary visibly from type. We devised an iterative protocol which, even within small populations, selects such variants and identifies by their expression in subsequent generations those whose altered traits are heritable. Promising individuals are then advanced as founders of lines for testing. Within three years we have thus derived lines from the doubled haploid cultivar 'McKenzie' that express traits not seen in their progenitor: short stature, near-immunity to wheat streak mosaic virus, and improved resistance to leaf spot diseases and FHB. These new characteristics have been stably expressed over multiple generations.

P09. Fusarium head blight in barley: identification of the causal *Fusarium* species in Europe and testing of FHB resistance using artificial inoculation. P. Holzknecht, P. Bury, M. Lemmens, and H. Buerstmayr. *University of Natural Resources and Applied Life Sciences Vienna, Department IFA-Tulln, Konrad Lorenz Str. 20, A-3430 Tulln, Austria. (P.B.) Syngenta Seeds Limited, Guildford, Surrey, UK, GU2 7YH.*

Fusarium head blight (FHB) in barley can be caused by different *Fusarium* spp. producing various mycotoxins. Breeding for resistance requires 1) resistance sources and 2) a reliable screening technique. We wanted to investigate FHB resistance of a barley nursery as a basis for future breeding programs. We also compared different inoculation methods and the resistance to DON/NIV and T2/HT2-producing *Fusarium* spp. We started with the isolation, purification and identification of *Fusarium* isolates from infected barley kernels originating from France, Germany and UK. In total 63 isolates were identified belonging to 8 different *Fusarium* spp. Most frequently detected isolates in Germany was *F. poae*, in France *F. cerealis* and *F. graminearum* and in the UK *F. avenaceum*. FHB resistance was tested with spray inoculation and with the kernel spawn method. Five different *Fusarium* spp. were used for inoculation. Scored was disease incidence and severity. ANOVA analyses showed highly significant differences between genotypes and treatments. Resistance data obtained with both inoculation techniques and with most *Fusarium* spp. were significantly related (r=0.57-0.81). Correlation coefficients between disease incidence and severity data were highly significant (r=0.93-0.99). We could not find any evidence for specific resistance against a particular type of toxin producer.

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P10. Sources of type II *Fusarium* resistance for triticale breeding. F. Langevin, F. Eudes, A. Comeau, Y. Dion, S. Rioux, H. Randhawa, G. Fedak, W. Cao, J. Gilbert, C. Lachance, and D. Salmon. *Soils and Crops Research and Development Centre, Agriculture and Agri-Food Canada, Québec, QC, Canada, G1V 2J3. (F.E., H.R.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, T1J 4B1. (Y.D.) CÉROM, Saint-Mathieu-de-Beloeil, QC, Canada, J3G 2E0. (S.R.) CÉROM, Québec, QC, Canada, G1P 3W8. (G.F., W.C.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C6. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada, R3T 2M9. (D.S.) Field Crop Development Centre, Alberta Agriculture, Food and Rural Development, Lacombe, AB, T4L 1W8.*

Triticale lines have been assessed in field and indoors trials for *Fusarium* resistance since 2006. In artificially inoculated field trials, Pronghorn had rather consistent MR FHB reaction; however, its lateness also leads to escape, and the presence of type I (resistance to infection) R genes interferes with the observation of other R types. Replicated trials were conducted indoors using point inoculations with *F. graminearum* on 62 triticale lines, to obtain clear evidence of type II R (resistance to spread). Quite strong type II R was found in a few triticale lines (e.g. Trit-910, Trit-875, Trit-1317 and a few others). However, Pronghorn rated only MRMS for type II R. Breeding for type II R should be an easy task, using point inoculation. However, this does not suffice. The challenge in order to get acceptable overall resistance in natural FHB epidemics is to increase also the type I R, and to improve pericarp resistance (a third mechanism), which is inadequate in most current germplasm. Sources of genes for those three mechanisms exist. Gene pyramiding assisted by a systemic approach (including FHB inoculations by spawn, spray and point-inoculation methods) may be ideal for the task, since the desired end result, toxin-free grain, is necessarily under polygenic control.

P11. Progress in developing fusarium head blight resistant two-row malting barley at Agriculture and Agri-Food Canada's Brandon Research Centre. W.G. Legge, J. Tucker, B. Bizimungu, M. Banik, A. Tekauz, R.A. Martin, T.M. Choo, and M.E. Savard. *Brandon Research Centre, Agriculture and Agri-Food Canada, PO Box 1000A, RR. 3, Brandon, M, Canada, R7A 5Y3. (B.B.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403 1st Avenue South, Lethbridge, AB, Canada, T1J 4B1. (M.B., A.T.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (R.A.M.) Crops and Livestock Research Centre, Agriculture and Agri-Food Canada, 440 University Avenue, Charlottetown, PE, Canada, C1A 4N6. (T.M.C., M.E.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C6.*

Fusarium head blight (FHB) caused by *Fusarium graminearum* continues to be the most destructive disease of barley in Canada. Development of FHB resistant cultivars with low deoxynivalenol (DON) accumulation has been an important objective of the two-row malting barley breeding program at Agriculture and Agri-Food Canada, Brandon, MB, for the past decade. To hasten progress, doubled haploid lines were produced with many subjected to *in vitro* selection (IVS) using culture media containing *Fusarium* mycotoxins. TR05915, an IVS selection from CDC Kendall showing 25-30% lower DON content than CDC Kendall while maintaining its desirable quality profile, was recently registered as Norman. HB705, an IVS selection from the CDC Freedom/Rivers cross, was also registered as a two-row hulless cultivar with malting quality potential. HB705, which combines reduced DON content relative to other hulless cultivars with high malt extract, may be attractive to the malting and brewing industry. Using exotic parents, such as two-row Chinese accession Harbin, to enhance FHB resistance has been attempted with limited success. A second breeding cycle is currently underway; TR08203, a promising line tracing back to Harbin, was advanced to a second year in the 2009 Western Cooperative Two-row Barley Registration Test. Numerous FHB resistant breeding lines at various stages of development will be evaluated further in 2009.

P12. Correlation between ear rot rating and deoxynivalenol, zearalenone, and fumonisin concentrations in European maize. M. Löffler, B. Kessel, M. Ouzunova, and T. Miedaner. *State Plant Breeding Institute, Universitaet Hohenheim (720), Fruwirthstrasse 21, D-70599 Stuttgart-Hohenheim, Germany. (M.L. and T.M.) KWS SAAT AG, Grimsehlstrasse 31, D-37555 Einbeck, Germany.*

Fusarium graminearum and *F. verticillioides* cause ear rot in maize and contaminate the grain with deoxynivalenol (DON), zearalenone (ZEA), and the fumonisins (FUM). Indirect selection for mycotoxin concentrations by visual rating would allow larger selection intensities due to high costs of mycotoxin analyses. Our objectives were to estimate quantitative-genetic parameters and associations between ear rot severity and mycotoxin concentrations. Three maturity groups each comprising about 50 elite inbred lines were tested in Germany, France, Italy and/or Hungary according to their maturity group. Ten plants per plot were silk channel or kernel inoculated and percentage of the ear area covered with mycelium was rated and mycotoxins analyzed with an immunotest. Analysis of variance revealed significant ($P<0.01$) genotype and genotype by environment interactions for ear rot and mycotoxin concentrations allowing selection but requesting multi-environmental trials. Correlations between ear rot rating and mycotoxin concentrations were high ($r>0.75$). Relative efficiencies of indirect selection based on ear rot rating were >1.0 for DON and ZEA indicating that indirect selection is more effective than direct selection, but <1.0 for FUM. Assuming a fixed budget indirect selection by visual rating should be effective also for FUM. In later selection stages, lines should be analyzed for mycotoxin concentrations, particularly for FUM.

P13. Relationship between incidence of fusarium head blight, tombstone kernels and deoxynivalenol concentration of spring wheat as affected by planting and harvesting dates, and nitrogen fertilization. B.L. Ma, K.D. Subedi, A.G. Xue, and H. Voldeng. *Eastern Cereal and Oilseed Research Center, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Proper crop management is an important strategy to reduce deoxynivalenol (DON) concentrations in the grain of spring wheat (*Triticum aestivum* L.). A field experiment was conducted at two sites in Ottawa, Canada for three growing seasons to investigate DON concentration, visual assessments and their relationships as affected by planting and harvesting dates and nitrogen (N) application. The cultivar 'AC Brio' was seeded at three planting dates with five N treatments. At early dough stage, each plot was surveyed for fusarium head blight (FHB) infestation. From physiological maturity (PM), plant samples were taken at weekly intervals, and grain and plant moisture contents determined. The *Fusarium*-damaged tombstone kernels (FDK) were counted and the kernel samples ground and analyzed for DON concentration. For both soil types, later planting significantly increased the FDK count and DON concentration in the grain. The effect of late planting was greater in coarse-textured sandy loam than in a fine-textured loam or clay loam soil. DON concentration was significantly increased when harvest was delayed from two to four weeks after PM. There was often weak or no correlation between FHB incidence and grain DON concentration. Our data indicate that under the eastern Canadian conditions, some precautionary practices such as early seeding and timely harvest will reduce the incidence of FDK and DON content.

P14. Association of morphological and developmental traits with fusarium head blight in a population of wheat derived from a cross of wheat cultivar 'Brio' and a *Triticum timopheevii* line. A. Malihipour, J. Gilbert, A. Brûlé-Babel, G. Fedak, and W. Cao. *Department of Biological Sciences, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (A.M., J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada, R3T 2M9. (A.B.) Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (G.F., W.C.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C6.*

Fusarium head blight (FHB), caused mainly by *Fusarium graminearum*, is one of the most devastating diseases of wheat in many parts of the world. In a population of recombinant inbred lines derived from the cross of bread wheat cultivar 'Brio' and a *Triticum timopheevii* line, the association of morphological and developmental traits (number of days to anthesis, height, and presence/absence of awns) with disease-related variables [disease incidence, disease severity, disease index, and *Fusarium* damaged kernels (FDK)] was investigated. The evaluations were done in single-floret-inoculated trials in the greenhouse and spray-inoculated field experiments in two locations. Results showed that the number of days to anthesis had significant negative correlation with disease incidence, disease severity, and disease index in 2006 (-0.2543, -0.4119, and -0.3572, respectively at $\rho \leq 0.01$) but positive correlation with FDK (0.3080 at $\rho \leq 0.01$). In the greenhouse, where only disease severity was assessed, correlation of the number of days to anthesis and plant height with disease severity was relatively low (0.1762 at $\rho \leq 0.01$) or not significant, respectively. The presence of awns in plants significantly reduced the level of disease in both field (disease incidence, severity, index, and FDK) and greenhouse (disease severity) conditions. These results generally support the results of earlier FHB studies and should be considered when breeding wheat genotypes for resistance to FHB.

P15. Linkage mapping of genes associated with dehydrodiferulic acid mediated cross-linking in maize cell walls and resistance to *Fusarium graminearum* (Schwabe). C.J. Martin and K.P. Pauls. *Department of Plant Agriculture, University of Guelph, 50 Stone Road East, Guelph, ON, Canada, N1G 2W1.*

The fungus *Fusarium graminearum* Schwabe [sexual state: *Gibberella zeae* (Schweinitz) Petch] causes gibberella ear rot in maize and fusarium head blight in wheat. The most devastating effect of these diseases is the deposition of mycotoxins in the grain. Molecular markers may allow rapid introgression of stable genetic resistance. Previous work has shown that there is a negative correlation between severity of disease and the concentration of cell wall bound dehydrodiferulic acid (DFA) in the pericarp of the grain. Furthermore, some chromosomal regions that are important for resistance are also important for DFA content. Since DFA is a derivative of the phenylpropanoid pathway, we have adopted a candidate gene approach to identify the genes that are responsible for the resistance/DFA QTL. Candidate genes were roughly mapped *in silico* using maize genetics resources prior to molecular analysis. Polymorphisms have been discovered in putative phenylalanine ammonia lyase (PAL), caffeoyl-coenzyme A 3-O-methyltransferase (CCoAOMT), and hydroxycinnamoyl-coenzyme A shikimate/quinate hydroxycinnamoyltransferase (HCT) genes. They have been converted to molecular markers and mapped. Preliminary results indicate that these genes are located between the flanking markers of various resistance/DFA QTL. PAL and HCT are tightly linked in a significant resistance/DFA QTL region that was detected across environments. This research should provide a foundation for gene-derived marker assisted resistance breeding in elite maize adapted to the northern United States and Canadian corn growing areas.

P16. Strategic application of proteomics and plant genotyping toward improved FHB resistance in Canadian wheat. C. Rampitsch, D. Somers, L. Tamburic-Ilincic, G. Fedak, T. Ouellet, G. Humphreys, and R. DePauw. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (L.T-I.) Department of Plant Agriculture, University of Guelph, Ridgetown Campus, Main St East, Ridgetown ON, N0P 2C0. (G.F., T.O.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa ON, Canada, K1A 0C6. (R.D.) Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, PO BOX 1030, Swift Current, SK, Canada, S9H 3X2.*

This poster describes progress from a bilateral project between Canada and Germany, funded under the GABI-Canada collaboration. The project combines traditional breeding, molecular breeding and mapping with '-omics' sciences to mitigate the impact of fusarium head blight and its associated mycotoxins in spring and winter wheat. FHB is a major concern in both countries. It is imperative to apply genomics-based science to safeguard Canadian and European wheat. The outcome of this project will be the reduction of mycotoxins in grain by introducing new FHB-resistant wheat cultivars to sustain the multi-billion/yr Canadian and German wheat industries and to ensure food and feed safety. In this project three sources of FHB resistance (Asian, Brazilian, European) were pyramided and examined phenotypically and biochemically. The major goals included: introgression of European winter wheat FHB resistance into elite Canadian wheat; introgression of Asian resistance into a broader-based Canadian spring wheat genetic platforms using marker-assisted selection; creating a new biochemical understanding of FHB resistance in wheat through comparative proteomics and genomics. This poster summarizes progress of the project, principally on the Canadian side, to date.

P17. QTLs for *Gibberella* stalk rot resistance in maize population CG62 x CO387.

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Gibberella stalk rot caused by *Fusarium graminearum* is prevalent in cool maize growing regions. The disease can cause extensive crop damage by initiating premature plant death, interfering with the translocation of water and nutrients during grain filling and promoting crop lodging. Previously, a maize recombinant inbred line (RIL) population from the cross CG62 x CO387 was used to map quantitative trait loci (QTLs) for *Gibberella* ear rot resistance and seed phenolics. The objective of the current study was to identify QTLs for *Gibberella* stalk rot resistance in the same population by the means of selective phenotyping. Forty RILs (selected based on the seed ferulic acid content) and parental genotypes (CG62 and CO387) were evaluated for *Gibberella* stalk rot and physical/chemical characteristics of the stalk in Ottawa in 2008 and 2009. The RILs were segregating for the stalk rot disease severity ratings and ranged from 4.67 to 7.00 in 2008. Twelve molecular markers were associated with the 2008 disease severity ratings. There was no significant correlation between the 2008 *Gibberella* stalk rot and the previously determined ear rot disease severity ratings. Chemical and physical characterization of the stalk (height, diameter, lodging, phenolics) is underway. Initial analysis of the stalk free phenolics indicated that there is no correlation with the *Gibberella* stalk rot disease severity ratings. After completing the 2009 experiment, the data will be used to map stalk rot QTLs. The associated markers can be used in the future to facilitate introgression of resistance into elite germplasm.

P18. Cross tolerance to *Fusarium* spp. in FHB-tolerant genotypes of spring wheat. Y.F. Ruan, R. Babonich, C.J. Pozniak, P. Hucl, G. Hughes, and J.M. Clarke. *Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8.*

Fusarium head blight (FHB), caused by *Fusarium* species, is of interest due to its negative impact on wheat quality and yield. A majority of FHB resistance genes in wheat have been identified based on reaction to *F. graminearum*. To better understand tolerance to other *Fusarium* spp. four wheat genotypes with contrasting resistance to *F. graminearum* (*Fg*) were screened with five *Fusarium* spp. (*F. culmorum, F. poae, F. graminearum, F. avenaceum* and *F. sporotrichioides*) in the greenhouse. Plants were artificially inoculated with 2 or 3 isolates from Manitoba, Saskatchewan and Alberta for each of *Fusarium* species. The development of FHB was rated as disease severity (DS) on a 0-5 scale every two days from day 4 to day 22 after inoculation. FHB severity over time was summarized as the area under disease progress curve (AUDPC). Regardless of wheat cultivar, *F. culmorum* resulted in the greatest disease development. Cultivars resistant to *F. graminearum* were also resistant to *F. culmorum*. The *Fg* tolerant wheat cultivars showed better resistance than the susceptible cultivars, regardless of *Fusarium* spp. These results suggest that the current tolerance genes deployed in spring wheat provide cross tolerance to all *Fusarium* spp. currently present in western Canada.

P19. Mapping of QTL for tolerance to fusarium head blight in the tetraploid wheat population TG3487/2*DT735. Y.F. Ruan, C.J. Pozniak, P. Hucl, and J.M. Clarke. *Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8.*

Most durum wheat (*Triticum turgidum* L var. durum) is susceptible to fusarium head blight (FHB). To date, few tolerance genes have been identified in durum and none provide adequate tolerance under endemic infection. This study was conducted to identify FHB resistance in tetraploid wheat and map quantitative trait loci (QTL) associated with FHB tolerance. A backcross recombinant inbred line population was developed from the tetraploid cross TG3487/2*DT735. A total of 160 lines were evaluated for type II resistance to *Fusarium graminearum* in greenhouse experiments and in endemic nurseries at Carman, Manitoba in 2008 and 2009. The population was genotyped with approx. 100 microsatellite markers and QTL for FHB severity (greenhouse) and disease incidence, severity and index (field) were identified using single marker analysis. In field and greenhouse studies, TG3487 was tolerant to FHB, with reactions equal to the hexaploid line ND2710. DT735 expressed tolerance better than AC Morse and similar to AC Barrie. In 2008, severity and incidence in the field were not correlated with heading time, but the correlations were significant in 2009. Xwmc349, Xbarc167, Xwmc445, and wmc201 were associated with greenhouse and field tolerance in both years. Efforts are underway to validate these QTL and to further backcross and pyramid these QTL into commercially adapted backgrounds.

P20. Development and evaluation of winter wheat breeding lines carrying fusarium head blight QTLs from spring wheat. A. Salameh, B. Almaghrabi, and H. Buerstmayr. *(A.S.) Current address: Hebron University, Palestine; (B.A.) Current address: BOKU-University of Natural Resources and Applied Life Sciences, Vienna, Department for Applied Plant Sciences, Institute for Plant Protection, Vienna; (A.S., B.A., H.B.) BOKU-University of Natural Resources and Applied Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production, Konrad Lorenz Str. 20, A-3430, Tulln, Austria.*

A series of BC_2 derived lines were developed from crosses of CM-82036 (FHB resistant spring wheat) with 11 winter wheat lines or cultivars as recurrent parents. BC_2 derived lines were chosen with either two QTL (*Fhb1*_3B, *Qhfs.ifa-5A*) one of these or no QTL by use of linked SSR markers and evaluated for FHB severity in replicated field trails. Lines based on moderately resistant recurrent parents showed lower average FHB severity compared to lines based on susceptible recurrent parents. When comparing the FHB severity of related lines descending from the same recurrent parent but carrying different combinations of the two QTL, the general trend was that FHB severity decreased most when both spring wheat derived QTL (*Fhb1* and *Qfhs.ifa-5A*) were present and less when only one QTL was present. BC_2 derived lines with the winter wheat alleles at both QTL regions showed generally more disease severity than lines with one or both QTL from spring wheat. Presence of *Qfhs.ifa-5A* increased plant height by 5-15 cm. All in all, introduction of two large effect QTL from spring wheat had a significant effect in reducing FHB susceptibility of winter wheat breeding lines.

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P21. Evaluation of fusarium head blight resistance and DON level in winter wheat germplasm from Germany and Canada. L. Tamburic-Ilincic, A. Brûlé-Babel, L. Hartl, J. Haeberle, E. Ebmeyer, and J. Durand. *(L.T.-I.) Department. of Plant Agriculture, University of Guelph, Ridgetown Campus, ON, Canada, N0P 2C0. (A.B.-B.) Department. of Plant Science, University of Manitoba, 66 Dafoe Road, Winnipeg, MB, Canada, R3T 2N2. (L.H., J.H.) Bayerische Landesanstalt für Bodenkultur und Pflanzenbau, Vöttingerstrasse 38, D-85354 Freising, Germany. (E.E.) Lochow-Petkus GmbH, Bollersener Weg 5, D-29303 Bergen, Germany. (J.D.) Semican, Princeville, QC, Canada, G6L 4K7.*

Fusarium head blight (FHB) is an important wheat disease with significant economic impact on the grain industry. *Fusarium graminearum* (Schwabe) is the predominant species that causes FHB and produces the mycotoxin deoxynivalenol (DON) in grain. The most practical way to control FHB is through the development of resistant cultivars. The objective of this study was to evaluate the reaction of winter wheat germplasm from Germany and Canada to FHB resistance and deoxynivalenol (DON) accumulation and to determine whether resistant host cultivars were stable in performance across the environments. Forty winter wheat breeding lines from Germany and Canada were screened for FHB resistance at three sites in Canada (Ridgetown, Carman, Princeville) and three sites in Germany (Herzogenaurach, Freising and Seligenstadt) in 2008. Disease levels were calculated as fusarium head blight index (FHBI), which was the product of the percent heads infected (incidence) and the percent spikelets infected (severity), divided by 100. The harvested grain in Canada was quantified for DON accumulation using EZ-Quant® Vomitoxin ELISA kit from Diagnostix (www.diagnostix.ca). In general, all environments produce similar genotype rankings and results. FHB visual symptoms were significantly correlated (r=0.77) between the continents. RCUOGDHACF110902D (Canadian) and DSV720500 (German) had the lowest overall FHB ratings, while 32C*17 (Canadian) and DSV720500 had the lowest DON level among all lines tested. RCATL33 (FHB resistance derived from both Sumai 3 and Frontana) was rated amongst some of the most resistant entries in the test. FHB index in Germany ranged from 1%-72% and in Canada from 3%-55%. DON level in Canada ranged from 0.3 ppm - 41.3 ppm. In Germany overall mean for FHB index was 32%. In Canada overall mean for FHB index and DON level was 26% and 9 ppm, respectively. The resistant host cultivars were stable in performance across the environments.

P22. Testing the WCORT oat entries for reaction to fusarium head blight. A. Tekauz and J. Mitchell Fetch. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9.*

The Western Cooperative Oat Registration Test (WCORT) is grown annually to evaluate advanced oat breeding lines for their agronomic, disease, and quality attributes. This test supplies the data necessary for support by the 'Prairie Recommending Committee for Oat and Barley', and subsequent official registration by the Canadian Food Inspection Agency. Disease data generated include that for stem rust, crown rust, smut and BYD. Information on fusarium head blight (FHB), now regarded as an important disease of oat in western Canada, has hitherto not been available or considered. The 36 entries in the 2008 WCORT were grown in the irrigated CRC FHB Nursery at Portage la Prairie, MB, and exposed to ground applied *Fusarium graminearum*-infested corn kernel inoculum to assess their FHB reactions, based on levels of deoxynivalenol (DON) in the whole grain. A severe FHB epidemic ensued in 2008. DON levels ranged from 8.0 to 40.2 ppm; 'Leggett', a check cultivar, had the lowest DON. Preliminary resistance designations (MR or moderatetely resistant, etc.) were applied to the lines. Entries from individual breeding programs were scattered in several resistance categories. These preliminary designations need to be verified or amended in 2009 and in future years, to provide valuable new information on overall oat cultivar performance for western Canada and beyond.

P23. Evaluation of oat germplasm for resistance to fusarium head blight. A. Tekauz, J. Mitchell Fetch, and M.E. Savard. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M. (M.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Cananda, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Fusarium head blight (FHB) is an important disease of cereals in western Canada. In oat FHB is difficult to recognize, and its presence and severity requires sampling of harvested grain for the causal *Fusarium* spp. and/or deoxynivalenol (DON), a mycotoxin associated with infection. The food and feed industries both require oat with a low content of DON. Differences in DON occur among registered Canadian oat cultivars, but better resistance to FHB is needed and may be available in exotic *Avena* germplasm. A selection of oat lines from various international genebanks has been evaluated at the irrigated CRC Portage la Praire FHB Disease Nursery for the past three years. In 2008, conditions for infection following artificial inoculation with *Fusarium graminearum* were particularly favourable resulting in high levels of disease. DON measured 26-57 ppm in wheat and barley checks included in the nursery. Among 150 oat accessions tested, DON levels in the most susceptible tier of 25 averaged 29 ppm, with seed-borne *F. graminearum* as high as 76%. However, DON levels in the 9 best lines were considerably lower, averaging 7.4 ppm, and were accompanied by relatively low *F. graminearum* (29% kernel infection). These low DON oat accessions are promising parents for use in crosses to breed for enhanced resistance to FHB.

P24. Cytogenetic analysis of an intergeneric amphiploid × triticale hybrid: a new source of resistance to fusarium head blight. Y. Yang, G. Fedak, W. Cao, D. Chi, J. Zeng, A.G. Xue, and F. Han. *Eastern Cereal and Oilseed Research Center, Agriculture and Agri_Food Canada, 960 Carling Ave., Ottawa, ON, Canada, K1A 0C6.* (J.Z.,Y.Y.) *Triticeae Research Institute of Chengdu Science Academy, Sichuan Agricultural University, Wenjiang District, 611130, Sichuan, China.* (F.H.) *Division of Biological Science, University of Missouri-Columbia, Columbia, MO, U.S.A. 65211-7400.*

Fusarium head blight (FHB), caused by *Fusarium graminearum* Schwabe, is a ravaging disease of cereal crops worldwide. The *Thinopyrum* species carry important traits such as biotic and abiotic stress tolerance. The amphiploid 8801 (AABBEE) with the E genome from *Thinopyrum elongatum*, derived from the hybrid *Triticum turgidum* (AABB*)* /*Thinopyrum elongatum* (EE), is an excellent sources of FHB resistance. We produced a hybrid between the amphiploid 8801 and the triticale line T182 (AABBRR). In this study, 29 F_3 plants were analyzed by genomic *in situ* hybridization. The chromosome numbers of the plants ranged from 28 to 45. The results showed that one plant with 42 chromosomes and one plant with 45 chromosomes each contained a pair of E/R Robertsonian translocation chromosomes and one E/R translocation chromosome was also found in each of five additional F_3 plants. Among these five plants, there was a unique plant with 28 chromosomes which contains an E/R chromosome translocation. One R genome chromosome and seven E genome chromosomes were also found in three F_3 plants with 35 chromosomes. Meiotic studies are being conducted on the above materials and their progenies.

P25. A new source of resistance to fusarium head blight from wheat- *Elymus repens* introgressions. J. Zeng, W. Cao, G. Fedak, P. Hucl, Y. Yang, A.G. Xue, and D. Chi. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave. Ottawa, ON, Canada, K1A 0C6. (J.Z., Y.Y.) Triticeae Research Institute of Chengdu Science Academy, Sichuan Agricultural University, Wenjiang District 611130, Sichuan, China. (P.H.) Department of Plant Sciences, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8.*

Elymus repens (L.) Gould (2n = 6x = 42, **StStStStHH**) is a hexaploid wild grass species, distantly related to bread wheat (*Triticum aestivum* L. em Thell; 2n = 6x=42, **AABBDD**). It has a high level of resistance to fusarium head blight (FHB). The objective of this study was to transfer genes for resistance to FHB from *E. repens* to common bread wheat. The cross Crocus/*E. repens* was made in Crop Development Center experimental field, Department of Plant Sciences, University of Saskatchewan. The F_1 plants were backcrossed to Crocus, then seeds from the BC_1F_1 plants were bulked and advanced to the BC_1F_7 generation. Sixteen lines were selected and evaluated for FHB reaction in the nursery in Ottawa in 2007 and 2008. Two lines, P1142 and P1131 (F8), were re-selected based on agronomic traits and FHB resistance performance. The results showed that the line P1142 was still segregating, with chromosome numbers ranging from 42 to 56, while the line P1131 with 56 chromosomes was stable morphologically. Cytological study and *in situ* hybridization analyses indicated that we obtained several wheat-*E. repens* addition and translocation lines, and two partial amphiploids. The results of greenhouse FHB evaluation by point inoculation showed that all the lines had a high level of resistance to FHB with only one spikelet infected (6%), compared to the check Roblin (100%) and the parent Crocus (85%). This new resistance source will be useful for the improvement of FHB resistance in wheat.

P26. Effect of glyphosate treatment to previous crop on fusarium head blight in wheat and barley. M.-È. Bérubé, A. Vanasse, S. Rioux, N. Bourget, Y. Dion, G. Tremblay, and G. Bourgeois. *Département de phytologie, Faculté des sciences de l'agriculture et de l'alimentation, Pavillon Paul-Comtois, Université Laval, Québec, QC, Canada, G1V 0A6. (S.R. and N.B.) Centre de recherche sur les grains, 2700, rue Einstein, Québec, QC, Canada, G1P 3W8. (Y.D. and G.T.) Centre de recherche sur les grains, 740 chemin Trudeau, Saint-Mathieu-de-Beloeil, QC, Canada, J3G 0E2. (G.B.) Horticulture Research and Development Centre, Agriculture and Agri-Food Canada, 430 Gouin Boulevard, Saint-Jean-sur-Richelieu, QC, Canada, J3B 3E6.*

The objective of the study was to determine, under three different tillage practices (conventional, minimum, and no-till), the effect of glyphosate applied the year preceding a wheat or barley crop on the incidence of fusarium head blight (FHB). In 2007 and 2008, six field experiments (two species × three tillage practices) were conducted in Saint-Augustin-de-Desmaures and Saint-Mathieu-de-Beloeil, QC. The herbicide treatments, glyphosate (G) and a herbicide (NG) chosen according to weed species were applied as main plots on a RoundUp Ready™ (RR) soybean. The following year, three wheat and three barley cultivars with different levels of FHB resistance were seeded in their respective experiments as subplots in the main herbicide plots. In 2007, in any of the 12 experiments, there were no significant herbicide x cultivar interaction nor herbicide effects on visual symptoms, DON content, and amount of inoculum of *Fusarium graminearum* (Fg) coming from soybean residues. In 2008, only the barley-minimum-till trial of Saint-Augustin showed a DON content significantly (P=0.046) higher in G (1.5 ppm) than in NG (1.1 ppm) treatments, but there was no significant herbicide effect on Fg inoculum production. All results collected indicate that under Quebec cropping conditions, glyphosate used on a RR soybean as previous crop has no or low impact on FHB whatever the tillage practices used.

P27. Efficacy of ACM941-CU, a formulated product of *Clonostachys rosea* strain ACM941 on the control of FHB in wheat. Y.H. Chen and A.G. Xue. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Previous studies demonstrated that *Clonostachys rosea* strain ACM941 was an antagonist against *Gibberella zeae,* the causal agent of fusarium head blight (FHB) of wheat in Canada. This research was to evaluate the efficacy of ACM941-CU, a formulated product of ACM941, on the reduction of perithecial production and control of FHB in comparison with registered fungicide Folicur® (tebuconazole). Over the average of two field trials in 2009, ACM941-CU reduced the daily perithecial reproduction over a period of 42 days by 90% on corn residues, 90% on soybean residues, and 71% on wheat residues. These effects were better than those of tebuconazole in the same trials. Five concentrations of ACM941-CU, ranging from 10^4 to 10^8 CFU/mL, were tested for the control of FHB in 2009 and ACM941-CU at 10^8 CFU/mL was the most effective treatment, reducing the area under the disease progress curve (AUDPC) by 79%, infected spikelets (IS) by 30%, and *Fusarium* damaged kernels (FDK) by 91% in the greenhouse experiments. Under the field conditions, ACM941-CU significantly reduced the AUDPC by 43%, FHB index by 52%, IS by 45% and FDK by 43%. These effects were less but not significantly different from those achieved with tebuconazole, suggesting that ACM941-CU is a promising biocontrol product and may be used for managing FHB in wheat.

P28. Effects of planting date and earliness on the level of deoxynivalenol contamination in barley under natural epidemic conditions. T.M. Choo, R.A. Martin, M.E. Savard, and B. Vigier. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, KlA 0C6. (R.M.) Crops and Livestock Research Centre, Agriculture and Agri-Food Canada, 440 University Avenue, Charlottetown, PE, Canada, CIA 4N6.*

Fusarium head blight is a destructive disease of barley in many countries. *Fusarium* species produce mycotoxins such as dexoynevalenol (DON) in the grains. An integrated management strategy is needed to mitigate the level of DON contamination in barley. Therefore, a study was initiated to determine if early planting and earliness can be used to avoid severe contamination of DON under natural infection conditions in eastern Canada. Twelve six-row barley cultivars were planted in a split-plot design with two planting dates (early vs. late) as main-plot units and cultivars as sub-plot units at the Harrington Farm in Prince Edward Island in 2007 and 2008. Seed samples from all experimental plots were contaminated with DON and one contained as much as 10.5 mg/kg. The amount of rainfall in August 2008 was very high (224 mm) and consequently DON contamination was more severe in 2008 than in 2007. For the majority of the 12 cultivars, early planting resulted in less DON contamination in comparison with late planting. Days to heading was positively correlated with DON concentration only at late planting in 2007. The results suggest that six-row barley should be planted early in eastern Canada to avoid a high level of DON contamination and a reduction in grain yield.

P29. Potential utility of RT-PCR to detect and quantify *Fusarium* spp. causing FHB in oat. T. Grewal, W. Yan, W. Yajima, X.M. Zhang, A. Beattie, and B. Rossnagel. *Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8. (T.G.) Saskatchewan Research Council, 125-15 Innovation Boulevard, Saskatoon, SK, Canada, S7N 2X8. (W.Y.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Potential significant economic losses from a widespread outbreak of fusarium head blight (FHB) in oat necessitate development of effective disease management strategies. The absence of visual FHB symptoms on oat requires the use of relatively expensive toxin analysis or time-consuming culturing of fungi on appropriate growth media for disease diagnosis. The Crop Development Centre, University of Saskatchewan has initiated a research project (partially funded by the Western Grains Research Foundation) in which a PCR-based assay to detect and quantify the presence of DNA specific to *Fusarium* species responsible for FHB on oat is being developed. Advantages of such an assay include simplicity, sensitivity and speed. Here we report the standardization of a real time PCR assay using TaqMan technology to detect and quantify *Fusarium graminearum* Schwabe in oat plants infected in controlled environments. Subsequent DON analysis revealed a relatively strong positive correlation ($r = 0.75$) between DON accumulation and abundance of *F. graminearum* DNA on infected oat. Primer and probe combinations that can specifically detect and quantify *F. poae, F. culmorum, F. avenaceum,* and *F. sporotrichioides* have also been generated. Results of a 2009 survey of Saskatchewan oat fields to determine prevalence and severity of *Fusarium* species using the RT-PCR assay are also presented.

P30. Strategy to prevent lodging in a barley nursery for fusarium head blight. M. Lacroix, S. Marchand, K. Smith, and F.J. Belzile. *Université Laval, Québec, QC, Canada, G1K 7P4. (K.S.) University of Minnesota, St. Paul, MN, USA.*

In the context of FHB nurseries, lodging can represent a tremendous hazard as it can lead to an improper assessment of the tolerance of certain genotypes. Once lodged, practically all genotypes will show severe symptoms and higher mycotoxin content as lodging creates conditions conducive to the rapid development of *Fusarium gramineaum* on the spike. Furthermore, once the plants have lodged, it can become extremely difficult to walk through and take notes in the nursery as the plants from different plots/rows are intermingled. In addition, some standard practices in FHB nurseries can promote lodging such as the use of irrigation. It provides conditions favorable to the normal development of the pathogen but can concomitantly contribute to excessive plant height and lodging. To prevent lodging in our nursery, we have implemented a system initially developed at the University of Minnesota. Plastic mesh netting is installed on top of the plots when plants are still in the vegetative state. As the plants start to head, they simply grow through the netting, which thus provides additional support to the stem. The netting can be raised as the stem elongates over the course of the season. We have implemented this system over the last two seasons (2008 and 2009) and have found it to be quite inexpensive, easy to install and extremely effective in preventing lodging. In this poster, we provide information on how to install the netting and detail the numerous benefits accruing from the use of such a system.

P31. Fighting *Fusarium* with *Fusarium:* Priming for FHB protection in wheat. C. Nasmith, G. Subramaniam, and L. Wang. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Attempts to control *Fusarium graminearum,* the causal agent for fusarium head blight (FHB), have involved chemical and biological approaches, along with resistance breeding. Priming, a more recently characterized control strategy, involves innate plant immunity through microbe perception. In this study priming is tested with 3 targeted gene disrupted mutants including NADPH oxidase, *MGV1* map kinase, and the trichothecene regulator *Tri6*. All three genes have diverse functions, but mutants display similar non-virulent disease phenotypes on wheat. The *Tri6* mutant was first tested for various parameters associated with priming including, priming load, priming window, and challenge load, on the FHB susceptible wheat variety Roblin. Wheat heads were first inoculated (primed) with the *Tri6* mutant, and then after 24 hrs, the wild-type progenitor strain was inoculated (challenged). Following 21 days the *Tri6* mutant drastically reduced disease symptoms compared to mock controls. The NADPH oxidase mutant reduced symptoms as well, but the *MGV1* mutant showed no difference in disease reduction compared to mock controls. Priming works and is presently being investigated both as a biological control, and as the basis for continuing wheat expression studies, as a novel procedure to characterize FHB resistant wheat germplasm.

P32. Site-Specific DONcast. I. Nichols. *Weather INnovations Incorporated, PO Box 23005, 7159 Queen's Line, RR 5, Chatham, ON, Canada, N7L 0B1.*

Site-Specific DONcast is a tool designed for wheat producers to provide a means of predicting deoxynivalenol toxin concentration (DON in ppm) produced by *Fusarium* sp. This internet based platform, developed by Weather INnovations Incorporated in conjunction with the University of Guelph, uses actual, forecasted and historical weather data along with field-specific agronomic data to accurately predict DON concentrations in wheat at harvest. Producers can use this tool to make informed management decisions on whether to apply a fungicide at heading for reducing potential DON concentrations in mature grain; help strategize which fields with elevated DON potential should be harvested first; provide an advance warning of DON concentrations toward alternative market destinations before harvest.

DON predictions become more reliable as the forecasted weather data is updated with observed data. The predictions are based on a determination of the heading date, Zadok's Stage 59 (75% of the heads in a canopy completely emerged from the flag leaf) for the calculation. DON predictions for individual fields vary based on the wheat variety, previous crops, tillage practices, heading dates, and local weather conditions. DONcast has been delivered by Weather INnovations Incorporated, since 2002, with the assistance of The Ontario Wheat Producers' Marketing Board and more recently, Bayer CropScience.

P33. Rapid optical sorting of *Fusarium* infected wheat. D.A. Prystupa. *Spectrum Agricultural Inc., Box 883, 402 Ara Mooradian Way, Pinawa, MB, Canada, R0E 1L0.*

A colour sorter for identifying and removing *Fusarium* damaged kernels (FDK) in wheat has been developed. The device singulates wheat kernels, makes a series of measurements at two wavelengths, classifies kernels as "infected" or "healthy" and directs infected kernels into a separate stream. The classification accuracy is 92.4% for "healthy" kernels and 93.4% for "infected" kernels. The deoxynivalenol concentration is reduced by an average of 84% by removing the "infected" kernels, in all cases to less than 1 ppm. The process is economic at $10 per tonne.

P34. Fungicides control of fusarium head blight symptoms and deoxynivalenol (DON) level caused by 15-ADON and 3-ADON *Fusarium graminearum* isolates in wheat in Ontario. L. Tamburic-Ilincic, A. Muckle, and A. Schaafsma. *University of Guelph, Ridgetown Campus, Ridgetown, ON, Canada, N0P 2C0.*

Fusarium graminearum (Schwabe) causes fusarium head blight (FHB), an important wheat disease. Deoxynivalenol (DON) is the most important mycotoxin produced by *F. graminearum*; 15-acetyl DON (15-ADON) and 3-acetyl DON (3-ADON) analogs are also produced. A shift in the presence of two *F. graminearum* chemotypes, 15-ADON and 3-ADON, has been reported in North America. The shift may influence current FHB management strategies including the use of fungicides. FOLICUR (tebuconazole) and PROLINE (prothioconazole) are two fungicides commonly used for FHB control in Ontario, while PROSARO has active ingredients from both fungicides. The objectives of this study investigated: 1) the effect of the fungicides on FHB symptoms and DON level after inoculation with 15-ADON and 3-ADON *F. graminearum* isolates in inoculated, misted wheat plots, and 2) the mycelium growth of different isolates of *F. graminearum* on PDA medium with and without fungicides. In 2008, both FHB index (%) and DON level were lower in cv. "Alsen" (moderately resistant) compared to cv. "Roblin" (highly susceptible) in all fungicide treatments and the untreated control, confirming that host resistance plays an important role in host-pathogen-fungicide interaction. Among all fungicide treatments, PROSARO and PROLINE produced the lowest FHB index DON concentration in the variety "Alsen", respectively. In addition, PROSARO resulted in the highest reduction of mycelium growth of both chemotypes compared to other fungicides.

P35. Interaction between *Cochliobolus sativus* and *Fusarium graminearum* on seed of barley. A. Tekauz and E. Mueller. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M.*

Cochliobolus sativus and *Fusarium graminearum* both are important pathogens of barley in Manitoba, and can often be isolated from the same seed. Their presence is indicative that spot blotch and/or fusarium head blight (FHB), respectively, affected the crop. The finding that *C. sativus* appeared to be inhibitory to *F. graminearum* and reduced the severity of FHB on barley spikes in a controlled environment prompted an examination of their isolation frequency from seed putatively infected by both species. Seed of the barley cvs. 'Harrington', 'CDC Stratus' and 'Stander', grown at two test locations in southern Manitoba, was plated onto PDA medium, PDA amended with Benlate® (benomyl, to which *C. sativus* is insensitive), and PDA amended with PCNB (quintozene, to which *Fusarium* spp. are insensitive). Averaged over the three cultivars and two test sites, isolation frequencies of *C. sativus* and *Fusarium* spp. (mainly *F. graminearum*) on PDA, PDA + Benlate®, and PDA + PCNB were 46.7 and 25.0, 64.7 and 0.3, and 3.8 and 36.3%, respectively. The results suggest that when both pathogens are present, their isolation frequency on a 'neutral' medium is reduced (here by 31% for *Fusarium* spp. and 22% for *C. sativus*) compared to when one or the other is absent, and is indicative of a mutually inhibitory effect.

P36. Comparison of the fungicide sensitivity of Alberta and Prince Edward Island isolates of *Fusarium graminearum* producing either 3- or 15-acetyl deoxynivalenol.
T.K. Turkington, R. Clear, J. Gilbert, T. Nowicki, K. O'Donnell, A. Tekauz, T. Ward, A.P. Rooney, H. Klein-Gebbinck, and R.A. Martin. *(T.K.T. and H.K.-G.) Lacombe/ Beaverlodge Research Centre, Agriculture and Agri-Food Canada, Lacombe, AB, Canada, T4L 1W1. (J.G. and A.T.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (R.C. and T.N.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main Street, Winnipeg MB, Canada, R3C 3G8. (K.O., T.W. and A.P.R.) United States Department of Agriculture, Peoria, IL, USA. (R.A.M.) Charlottetown Research Centre, Agriculture and Agri-Food Canada, 440 University Avenue, Charlottetown, PE, Canada, C1A 4N6.*

Fusarium graminearum Schwabe of the '3ADON' chemotype is now displacing '15ADON' isolates in Canada. One concern regarding this shift in chemotypes is related to potential differences in fungicide sensitivity. This could have significant implications as fungicide application is an important strategy to reduce disease severity and mycotoxin contamination. Fungicide sensitivity was assessed for a total of 12 isolates of *F. graminearum* (*Fg*); three 3ADON and three 15ADON from each of Alberta and Prince Edward Island. Spezieller Nährstoffarmer agar (SNA) plates were amended with 0, 0.78, 2.16, 4.16, 9.00, 16.78, and 30.62 μg/ml of commercial grade tebuconazole. Plates were inoculated with mycelial plugs and after 72 hours colony diameters were measured along two transects. Measurements were averaged for each plate, with results expressed as a percentage of the diameter of the unamended control. There were significant effects due to isolate, fungicide rate and their interaction. Contrasts indicated no significant differences due to chemotype, while there were significant linear, quadratic and cubic effects for mean response over fungicide concentrations. These preliminary results suggest that the 3ADON and 15ADON isolates tested had similar sensitivity to tebuconazole. Further research is investigating the potential of a microplate technique to assess fungicide sensitivity of *Fg*.

P37. Diversity and density of fungal and bacterial populations in wheat rhizosphere under monoculture and rotation cropping systems. D. Zhao, Y. Xu, G. Zhou, B.L. Ma, M. Lin, H. Qu, L. Chun, F. Yang, Y. Fu, and Y. Zhang. *Horticultural Branch, Heilongjiang Academy of Agricultural Sciences, 666 Haping Road, Harbin, Heilongjiang, China, 150069. (D.Z.) Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, 138 Haping Road, Harbin, Heilongjiang, China, 150081. (G.Z., B.M.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave., Ottawa, ON, Canada, K1A 0C6.*

Soil fungi and bacteria are major components of rhizosphere environment and important factors affecting crop growth. A field study was conducted to determine the diversity and density of fungi and bacteria in wheat rhizosphere in the wheat monoculture and wheat-corn-soybean rotation cropping systems. Soil samples were collected at seedling, jointing, flowering and ripening growth stages of wheat. Petri dish culture and PCR-DGGE were used to study fungal and bacterial populations and diversity. The fungal population density was higher in wheat monoculture than in the rotation system at all growth stages, but the difference was significant ($P < 0.05$) only for the flowering stage. The fungal population density increased for both cropping systems during the first three stages, and decreased during the ripening stage. Both the diversity and the population density of bacteria were higher in wheat monoculture than in wheat rotation for all four growth stages, but the difference were significant ($P < 0.05$) for only the jointing and ripening stages. Bacterial population showed a single peak at the wheat flowering stage for both cropping systems. Our data suggested that the dynamics of fungi and bacteria in wheat rhizosphere might affect the resistance of wheat crop to fusarium head blight and other diseases, and the importance to further investigate the relationship between rhizosphere microorganism and disease incidence and yield formation.

P38. Investigation of chemotype diversity in *Fusarium graminearum* isolates collected from wheat and corn fields in Ontario. C. Amarasinghe, A. Ratnayaka, L. Tamburic-Ilincic, A. Brûlé-Babel, and W.G.D. Fernando. *Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2L9. (L.T.-.I.) Department of Plant Agriculture, University of Guelph, Ridgetown Campus, Ridgetown, ON, Canada, N0P 2C0.*

Fusarium head blight (FHB) is a devastating fungal disease that infects a number of crops in Canada including wheat, barley, oats, rye and corn. *Fusarium graminearum* (= *Gibberella zeae*) is considered as the principle pathogen. FHB often results in indirect loss due to grain contamination with potent mycotoxins especially deoxynivelenol (DON). Three chemotypes were found in *F. graminearum*. They produce a C-4 oxygenated derivative of DON, nivalenol (NIV); an acetyl ester derivative of DON at 15-position oxygen (15ADON); and an acetyl ester derivative of DON at 3-position oxygen (3ADON). Recent research shows that the higher DON producer 3ADON is replacing the 15ADON chemotype populations in certain parts of Canada. The objective of this study was to investigate the variations of 15ADON and 3ADON chemotypes of *F.graminerum* isolates collected from wheat and corn fields in Ontario. Fifty three isolates from twelve different wheat fields and 65 isolates from five different corn fields in Ontario were used for chemotype analysis. DNA was extracted from pure cultures of *F.graminearum* and the fungus chemotypes were identified using the multiplex PCR primers with specific primers for each chemotype. Ninety three percent of the isolates collected from wheat fields were 15ADON and only 7% were 3ADON. All the investigated isolates from corn fields were 15ADON and no 3ADON isolates were found.

P39. Phenotypic evaluation of wheat transgenic lines expressing 10R and MsrA2 antimicrobial peptides. A. Badea, F. Eudes, A. Laroche, N.A. Foroud, and S. Misra. *Lethbridge Research Centre, Agriculture and Agri-Food Canada, PO Box 3000, Lethbridge, AB, Canada, T1J 4B1. (S.M.) Department of Biochemistry and Microbiology, University of Victoria, Victoria, BC, Canada, V8W 3P6.*

Antimicrobial peptides (AMPs) are potent natural antibiotics and their activity has been demonstrated against various pathogens. We hypothesized that control of fusarium head blight (FHB) in wheat could be accomplished through *in vivo* expression of two synthetic AMPs (MsrA2 and 10R) that were previously reported active *in vitro* against a variety of *Fusarium* species and strains. Three tissue specific promoters were chosen for regulating the AMPs expression in the wheat epidermis (*GstA1),* lemma/palea (*Lem1)* and epicarp *(Ltp6)*. Biolistic transformation and direct somatic embryogenesis were used to generate transgenic plants from cv. Fielder. The transgenic plants were evaluated in the greenhouse for resistance to FHB and powdery mildew. When compared to the non-transformed control Fielder, the transgenic lines showed on average a 49% and 58% reduction in FHB and powdery mildew susceptibility, respectively. Moreover, the transgenic plants showed on average a 34% reduction in DON accumulation compared to cv. Fielder.

P40. Expression profiling of trichothecene induced genes in wheat. M. Balcerzak, S. Gulden, and T. Ouellet. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, K1A 0C6.*

Trichothecene mycotoxins synthesized by *Fusarium graminearum* (Fg) are thought to be virulence factors in the infection of plants by Fg since they are necessary for the spread of the disease in wheat spikes. Loss of function mutations of the Fg *tri5* gene, the first step in the biosynthesis of trichothecenes, result in the loss of DON biosynthesis and reduced virulence on wheat. However, the effect of DON on the wheat host response to Fg infection is largely unknown. DON is a potent eukaryotic protein synthesis inhibitor and might affect the plant's ability to respond to Fg infection. RNA profiling has been performed using the Affymetrix GeneChip Wheat Genome Array (representing approximately 54,000 expressed sequences), comparing the response of the susceptible wheat cultivar Roblin when inoculated with either a wild type Fg (DON+) strain or a *tri5* knockout (DON-) strain. To confirm the microarray expression profiles, quantitative PCR assays were performed on some differentially expressed genes. We identified several DON+ up-regulated genes that belong to plant signaling pathways: shikimate, JA, SA and gibberellin pathways. They were designated as basal defense response genes which induction was largely independent of DON accumulation. We also detected several genes that were specifically induced in infection with the DON+ Fg strain, including transcription factors, ubiquitination-related proteins and stress induced proteins. Some genes, such as cytochrome b561, a leucine-rich repeat protein and shikimate kinase were identified as significantly down-regulated in DON+ Fg infection. Our results suggest that wheat response to trichothecene accumulation can be separated from the basal host response to *F. graminearum* infection.

P41. Differential gene expression of wheat backcross lines with *Fusarium* resistance QTL. W. Chen and P. Schweizer. *Leibniz Institute of Plant Genetics and Crop Plant, Correnssstrasse 3, 06466 Gatersleben, Germany.*

Fusarium head blight (FHB) is a devastating disease of several species of wheat and other cereals. Resistance to FHB is governed by quantitative trait loci (QTL) distributed on different chromosomes, but the genes at these QTL and their function in resistance are still unknown. Winter wheat backcross lines carrying two resistance QTLs (on chromosomes 3BS and 5A) against FHB were used to investigate QTL-specific differential gene expression between resistant and susceptible lines following single spikelet inoculation with *Fusarium culmorum* spores. Transcript profiling was carried out by using a barley 13K cDNA macroarry, which contains EST clones from different pathogen-attacked tissues. By analysing differential gene expression from 72 hybridization experiments, seven candidate genes were found to be significantly up regulated at 48 hours after inoculation in a resistant line pool carrying both 3BS and 5A QTL, but not in a susceptible line pool, in which both QTL were absent. A functional test system based on virus induced gene silencing (VIGS) in adult wheat plant was established and ready for testing the function of those 7 candidate genes.

P42. Results of field inoculation of barley lines by two chemotypes of *Fusarium graminearum*. R. Clear, J. Tucker, D. Gaba, S. Patrick, and W G. Legge. *Grain Research Laboratory, 1404-303 Main St., Winnipeg, MB, Canada, R3C 3G8. (J.T., W.L.) Brandon Research Centre, Agriculture and Agri-Food Canada, Box 1000A, 18th & Grand Valley Rd, R.R. #3, Brandon, MB, Canada, R7A 5Y3.*

In 2008, autoclaved corn kernels colonized by *Fusarium graminearum* Schwabe were spread between lines of barley in an FHB nursery at a 2:1 ratio of 15 ADON to 3 ADON isolates. This was the first use in this field of a 3 ADON isolate after 9 years of artificial inoculation with exclusively 15 ADON isolates. *F. graminearum* infected an average of 88.5% of 1800 seeds tested, with little difference across the nursery. However the chemotype distribution of *F. graminearum* recovered from infected kernels, and DON levels, showed a several fold difference from the eastern (67% 3 ADON isolates and 46 ppm) to the western end of the field (16% 3 ADON isolates and 13 ppm). Highest DON and 3 ADON levels were associated with the highest frequency of the 3 ADON chemotype, but not without exception. The difference in results between the eastern and western end of the nursery suggest that resident inoculum on previous crop residue played an important role in the disease. Although the 3 ADON isolates were recovered from 44% of the infected grain, significantly higher than the expected recovery rate, the possible impact of resident inoculum makes it difficult to draw firm conclusions at this time regarding differences in competitive ability between the two chemotypes.

P43. Investigating the role of the production of auxins by *Fusarium graminearum.* C. DesRoches, M. Balcerzak, B. Blackwell, and T. Ouellet. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Auxins are well known as key plant hormones regulating multiple facets of development in various cell types, including cell division, elongation and differentiation. The production of auxins is however not limited to plants, some bacteria and fungi are also able to synthesize these hormones but their role is not as well defined, ranging from pathogenesis to phytostimulation. In recent years many studies have focused on the role of the production of auxins, particularly indole-3 acetic acid, during infection; data suggests that some pathogens may synthesize IAA or manipulate plant auxin biosynthesis to promote virulence and disease. In contrast to the broad insight of IAA biosynthesis pathways in plants and bacteria, little is known about IAA and other auxins biosynthesis in fungi. The ability of *Fusarium graminearum* to efficiently convert exogenously added tryptophan to auxin and the subsequent large accumulation of auxin levels upon infection suggests a potential role for pathogen-induced auxin as an infection strategy for this pathogen. These initial results are leading to further research in order to investigate this prediction.

P44. Priming induced systematic resistance and susceptibility to fusarium head blight in wheat. N.A. Foroud, B.E. Ellis, and F. Eudes. *(N.A.F., F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403-1st Avenue South, Lethbridge AB, Canada, T1J 4B1. (N.A.F., B.E.E) Michael Smith Laboratories, University of British Columbia, 2185 East Mall, Vancouver, BC, Canada, V6T 1Z4.*

Two major forms of resistance to fusarium head blight have been identified in wheat: type I resistance (resistance to initial infection) and type II resistance (resistance to disease spread). In a recent microarray study, we observed that *F. graminearum* and deoxynivalenol induced upregulation of few defense-related transcripts in the uninoculated spikelets of a type II resistant 'Sumai 3'-derived line, GS-1-EM0168 ('CM82036'/'Superb'*2) compared with type I resistant GS-1-EM0040 ('CIMMYT 11' /'Superb'*2). In order to understand the impact of the induced defense-signaling responses on systemic resistance in these lines, we point inoculated a single floret of GS-1-EM0040, GS-1-EM0168 and 'Superb' with different inocula, including water, a trichothecene non-producing *Fusarium graminearum* mutant (Tri5-), and deoxynivalenol. Eight hours after point inoculation, the plants were spray inoculated with an aggressive *F. graminearum* strain (Tri5+), and evaluated for disease severity at 7, 9, 12 and 18 days after inoculation. Priming of wheat spikes with Tri5- and deoxynivalenol increased type I resistance in GS-1-EM0040, had no impact on resistance in GS-1-EM0168, and increased susceptibility in 'Superb'. Based on the defense pathways differentially regulated in the microarray study, we believe the susceptible response is due to increased salicylate production and the type I resistance response to increased jasmonates production. We are currently testing this hypothesis.

P45. Fungal trichothecene-genotypes play a role in fusarium head blight disease spread and trichothecene accumulation in wheat. N.A. Foroud, S. McCormick, T. MacMillan, B.E. Ellis, D.F. Kendra, and F. Eudes. *(N.A.F., T.M., F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403-1st Avenue South, Lethbridge AB, Canada, T1J 4B1. (N.A.F., B.E.E.) Michael Smith Laboratories, University of British Columbia, 2185 East Mall, Vancouver, BC, Canada, V6T 1Z4. (S.M., D.F.K.) Agricultural Research Service, United States Department of Agriculture, 1815 N University Street, Peoria, IL, United States, 61604. (T.M.) University of Lethbridge, 4401 University Drive, Lethbridge, AB, Canada, T1K 3M4.*

In the current study, we evaluated the impact of the observed North American evolutionary shift in the *Fusarium graminearum* complex on disease spread, kernel damage, and trichothecene accumulation in resistant and susceptible wheat genotypes. Four inocula were prepared using composites of *F. graminearum* strains with either 3-ADON (Fg3ADON), 15-ADON (Fg15ADONα and Fg15ADONβ), or NIV (FgNIV) genotypes. Isolates used in Fg15ADONβ are believed to be related to the 3-ADON population, and some of these strains have been shown to produce higher levels of DON than those used in Fg15ADONα. Stable resistance or susceptibility to disease spread, as well as *Fusarium*-damaged kernel (FDK) scores, were observed in highly-resistant or highly-susceptible wheat genotypes. Trichothecene genotype-dependent interactions with disease spread and FDK were observed in moderately or intermediate genetic sources of resistance/susceptibility: susceptibility to disease spread increased in wheat infected with Fg3ADON or Fg15ADONβ, and decreased in wheat infected with FgNIV. Unexpectedly, the amount of FDK and trichothecene observed in the grain were lowest in Fg3ADON- and FgNIV-infected wheat, even though Fg3ADON was much more aggressive than FgNIV. Our results indicate that *F. graminearum* trichothecene genotypes differ in their aggressiveness in colonizing wheat tissues, consistent with earlier reports identifying trichothecenes as an aggressiveness factor.

P46. Differences in transcript and protein accumulation provide insight into the different mechanisms of FHB-resistance in wheat. N.A. Foroud, T. Ouellet, B. Oosterveen, M. Jordan, A. Laroche, B.E. Ellis, and F. Eudes. *(N.A.F., A.L., F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403-1st Avenue South, Lethbridge AB, Canada, T1J 4B1. (N.A.F., B.E.E.) Michael Smith Laboratories, University of British Columbia, 2185 East Mall, Vancouver, BC, Canada, V6T 1Z4. (T.O.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6. (B.O., M.J.) Cereal Research Center, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9.*

The defense-response pathways of three wheat genotypes of varying resistance/ susceptibility with cv. 'Superb' pedigree in response to individual components of fusarium head blight (FHB) were evaluated by differential gene expression and protein accumulation studies. The variables evaluated include *F. graminearum* (trichothecene-producing and non-producing), aggressiveness factors and the trichothecene toxin deoxynivalenol. Uninoculated spikelets of point-inoculated heads were harvested at multiple time points within 24 hours after inoculation (hai) to identify temporal changes in transcript accumulation and at 72 hai for differential protein analysis. The type II resistant 'Sumai 3'-derived line, GS-1-EM0168 ('CM82036'/'Superb'*2), showed the highest variability in defense-related transcript accumulation in response to the different components evaluated. Among the total observed differences in transcript and protein accumulation, only a handful were associated with the plant-defense response, suggesting that resistance to disease spread is not a response induced in distal tissues. The most extensive transcript up-regulation for defense-response pathways was observed in type I resistant GS-1-EM0040 ('CIMMYT 11'/'Superb'*2), in which up-regulation of lignin biosynthesis genes was also detected. We propose that cell wall lignification within the spikelet glumes may improve type I resistance by preventing fungal penetration, whereas up-regulation of salicylic acid production in 'Superb' may divert phenylpropanoid substrate away from the lignin biosynthesis pathway, ultimately contributing to susceptibility in this cultivar.

P47. A comparison of the effects on disease ratings of 3-ADON and 15-ADON chemotypes of *Fusarium graminearum* on spring wheat. V. Gauthier, A. Brûlé-Babel, W.G.D. Fernando, and J. Gilbert. *Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada, R3T 2M9.*

Fusarium graminearum is the most common causal agent of fusarium head blight (FHB) of wheat in Canada. Presence of the pathogen results in losses in yield and quality. Production of the mycotoxin deoxynivalenol (DON) by the pathogen also limits the uses for feed and food. Recently, it has been shown that the higher DON producer, 3-acetyl DON (3-ADON) is replacing the 15-acetyl DON (15-ADON) chemotype populations in western Canada. The objective of this study was to evaluate the interaction between 25 *F. graminearum* isolates which differ in chemotype production and three spring wheat genotypes which differ in reaction to *F. graminearum*. In 2008 and 2009, each trial was a split plot with three replicates. *F. graminearum* isolate was the main plot effect and wheat genotype was the sub plot effect. Each plot was inoculated with a macroconidial suspension containing 50,000 macroconidia ml^{-1}at anthesis and three days later. Data from both years showed significant differences among isolates and genotypes for area under the disease progress curve. In 2008, there were significant differences for the isolate x genotype interaction, but not in 2009. Disease progressed similarly for all 3-ADON isolates but there were differences among the 15-ADON isolates. In general, 3-ADON isolates produced higher levels of disease than the 15-ADON isolates.

P48. Recovery of *Fusarium graminearum* chemotypes from the 2008 FHB nursery at Glenlea, MB. J. Gilbert, R. Clear, and D. Gaba. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 3E5. (R.C., D.G.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main St. Winnipeg, MB, Canada, R3C3G8.*

Fusarium head blight (FHB) is a serious threat to the Canadian grain industry. Most isolates of *Fusarium graminearum* Schwabe, the principal cause of FHB in North America, produce the mycotoxin deoxynivalenol (DON) and one of its acetylated derivatives, 3- or 15-ADON. In North America, a rapid shift from the 15-ADON to 3-ADON chemotype has been documented. While the 3-ADON isolates are not more aggressive than the 15-ADON isolates, they produce significantly more DON. The wheat FHB screening nursery at Glenlea, MB was inoculated with a macroconidial suspension of both chemotypes in equal ratio. The objective of this study was to determine if isolates of 3- or 15-ADON were recovered in the same ratio as applied. A set of 6 check cultivars/lines, planted throughout the nursery, was sampled after harvest in 2008. For each check variety, 100 seeds were surface-sterilized and plated on potato dextrose agar. The first 40 isolates of *Fusarium graminearum* recovered per check were single-spored and analysed for chemotype by PCR. The ratio of 3-ADON to 15-ADON isolates recovered from seed was on average 4:1, respectively, for all 6 checks. The 3-ADON chemotypes appear to effectively out-compete 15-ADON isolates for space and resources on wheat heads.

P49. The use of microsatellite loci in strain-typing of *Fusarium graminearum*. T. Gräfenhan, C.T. Lewis, R. Clear, J.T. Chapados, and K.A. Seifert. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave., Ottawa, ON, Canada, K1A 0C6. (R.C.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main Street, Winnipeg, MB, Canada, R3C 3G8.*

Microsatellites are composed of tandemly repeated DNA sequence motifs up to five nucleotides long. These simple sequence repeats (SSRs) are commonly found in pro- and eukaryotic genomes and typically are highly polymorphic within species and populations. Based on DNA sequences of highly variable SSRs (hvSSRs), we developed a multilocus genotyping system (MLGT) to better estimate the number of strains/populations in *Fusarium graminearum*. The process first involved a microsatellite discovery technique that mined the public domain *F. graminearum* genome sequence data of the North American strain PH-1 for short tandem repeats and their 500–600 bp flanking regions (MFRs). Subsequently, single nucleotide polymorphism (SNP) rates were assessed by sequencing ~68,000 bp from 64 SSR loci of a European isolate (BBA 67639). In comparison to the PH-1 data, SNP rates of the MFRs were highly variable among loci, with nucleotide diversity ranging from a low of zero to a high of 0.083 (one change per 12 bp). Based on nucleotide diversity estimates, six hvSSRs from three chromosomes were selected as SNP genetic markers (total of 6,600 bp). Preliminary results of the MLGT for >120 strains of *F. graminearum* from various locations and hosts indicate this will be an effective tool for population genetic and evolutionary studies.

P50. Competitive ability of 3 and 15 ADON chemotypes of *Fusarium graminearum* after spray inoculation of spring wheat. A.T. Guerrieri, R. Clear, A. Brûlé-Babel, W.G.D. Fernando, and J. Gilbert. *Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (R.C.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main St., Winnipeg, MB, Canada, R3C 3G8. (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada, R3T 2M9.*

Fusarium head blight of wheat caused by *Fusarium graminearum* Schwabe is of great significance to western Canadian grain producers, processors and consumers. A shift in population structure of *F.graminearum* has occurred from the 15 ADON chemotype to the more toxigenic 3 ADON chemotype. The objective of this study was to elucidate the cause of the recent and rapid shift in population structure. Five wheat lines were spray-inoculated in the field with an equal number of macroconidia of two 15 ADON isolates and two 3 ADON isolates. An uninoculated control was also included. Four hundred isolates were recovered from the harvested seed, grown in pure culture, and the chemotype determined by PCR. Chi-square tests of pooled results for single spore isolations failed to fit a 1:1 ratio for 3 ADON: 15 ADON, with 3 ADON being the more abundant chemotype. The results suggest that the 3 ADON chemotype is considerably more competitive on the wheat spike than the 15 ADON chemotype. This difference could be one reason for the rapid population shift occurring in Canada.

P51. *Thinopyrum elongatum* as a novel source of FHB resistance. S. Gulden, E. Watson, S.S. Miller, G. Fedak, and T. Ouellet. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6*.

Fusarium graminearum is the major causal agent of fusarium head blight (FHB) of wheat, a fungal disease causing significant yield losses and reduction in grain quality due to the production and deposition of mycotoxins in the seed. Very few sources of resistance to FHB have been identified in wheat and other cereal crops. *Thinopyrum elongatum* (2n=14, EE genome), a wild relative of wheat, was identified as carrying a strong resistance to FHB on its chromosome 7E. In this report, we have used disease rating data, fluorescence microscopy and a modified strain of *F. graminearum* expressing a green fluorescent protein to characterize the resistance carried by chromosome 7E when expressed in the susceptible wheat background of Chinese Spring (CS). Our observations of infected wheat heads showed that inoculated spikelets were less infected in the addition line carrying the ditelo of the long arm of *Th. elongatum* chromosome 7E, than in the parental line CS. Even more striking was the progression of the fungus from the inoculated spikelet to the adjacent node and rachis tissues: the fungus spreads easily and extensively from the inoculated spikelet into the node and adjacent spikelets in susceptible CS heads, but is effectively blocked from spreading in the addition line. Microscopic data clearly showed fungal growth was inhibited within the inoculated spikelet in heads of the addition line and that the fungus was completely blocked from spreading by the node tissue. Taken together, these observations suggest that *Th. elongatum* chromosome 7E carries a novel allele for resistance to spread (Type II resistance) of *F. graminearum*. Details of our observations will be presented.

P52. Shotgun proteomics gives insight into reversible redox modifications in double *Nox* mutants of *Fusarium graminearum*. M. Joshi, C. Rampitsch, and G. Subramaniam. *Cereal Research Center, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (R.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6*.

Shotgun proteomics, a strategy integrating protein digestion into peptides and sequencing through mass spectrometry, has become the method of choice for identification of key proteins in complex mixtures. We have used a shotgun redox proteomic technique to identify the reversibly modified cysteine groups in double mutants of *Fusarium graminearum.* The fungus is mutated for NADPH oxidase (*Nox*) A and B genes. The free cysteines groups were blocked with IAA followed by reduction of oxidised cysteines with DTT and biotin-affinity chromatography was used to isolate a "redox subpeptidome" which permits selective targeting of redox sensitive cysteines that undergo reversible modification. The reversible post translational modifications of thiol groups influence the protein's structure and function and control a broad spectrum of biochemical processes, including redox regulation, protection against oxidative stress and cell signalling. In this study, we are using a gel-free approach to detect redox sensitive cysteines within key proteins in double *Nox* mutants of *F. graminearum* in order to elucidate the mechanism involved in disease progression through the reduction/ oxidation of disulfide bonds in a pathway that includes NADPH oxidase A and B enzymes.

P53. Development of transgenic maize plants expressing GFP, a marker gene and DON detoxifying gene, *Tri101* for *Fusarium* resistance. P. Kant, B. Nash, and K.P. Pauls. *Department of Plant Agriculture, University of Guelph, Guelph, ON, Canada, N1G2W1.*

Fusarium graminearum (teleomorph *Gibberella Zeae*) causes fusarium/gibberella ear rot disease in maize. This is a major disease problem for maize production in North America, Europe and China. The current report describes progress in our efforts to develop resistance to Gibberella ear rot in maize by transformation with genes that may provide resistance against *F. graminearum*. An *Agrobacterium*-mediated transformation system was used to transform maize multiple shoot meristems cultures initiated from apical shoot meristems of seedlings. The method of transformation was demonstrated using the GUS and GFP markers. Multiple shoot meristems are good starting materials that overcome the need to use of immature embryo explants and can be produced throughout the year. Presently we have developed transgenic corn plants expressing DON detoxifying gene, *Tri101* encoding trichothecenes 3-o-actetyltrasferease, whose product converts DON to a less toxic derivative, 3- acetyldeoxynivalenol (3-ADON). The gene was obtained from our collaborators in Hebrew University of Jerusalem, Rehovot, Israel and cloned into the plant expression vector PBI121-*Tri101*, replacing the *Gus* gene. Transgenic plants developed were confirmed for the presence and expression of *GFP* or *Tri101* gene with southern analysis and reverse-transcriptase PCR, respectievly. All transgenic plants were fertile and set seeds. Tests of the ability of the transgenic kernels to detoxify DON are currently being performed and studies of the enzymatic activity of the TRI101 protein will be carried out. Our work suggests that corn transformation with genes that interfere with the disease or detoxify DON are good approaches to enhancing the resistance of maize to *F. graminearum*.

P54. A comparison of the aggressiveness and deoxynivalenol content of Canadian 3-acetyl and 15-acetyldeoxynivalenol producers of *Fusarium graminearum* in field-grown spring wheat. C. Knopf, V. Gauthier, L. Tamburic-Ilincic, A. Brûlé-Babel, W.G.D. Fernando, R. Clear, T. Ward, and T. Miedaner. *(C.K., T.M.) State Plant Breeding Institute, Universitaet Hohenheim (720), Fruwirthstrasse 21, 70599 Stuttgart, Germany. (V.G., A.B.-B., W.G.D.F.) Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. (L.T.-I.) Department of Plant Agriculture, University of Guelph, Ridgetown, ON, Canada, N0P 2C0. (R.C.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main Street, Winnipeg, MB, Canada, R3C 3G8. (T.W.) Microbial Genomics and Bioprocessing Research Unit, Agricultural Research Service, United States Department of Agriculture, 1815 N. University St., Peoria, IL, USA. 61604.*

Twenty four isolates of *Fusarium graminearum* of Canadian origin half of which were 3-acetyldeoxynivalenol (3-ADON) and half 15-acetyldeoxynivalenol (15-ADON) producers, were tested for their ability to cause fusarium head blight (FHB), as measured by FHB index and production of deoxynivalenol (DON) in spring wheat. Objectives were to determine (i) whether 3-ADON producers differ in aggressiveness and DON production from 15-ADON producers under field conditions, (ii) whether resistant host cultivars were stable across isolates. Field tests of all isolates were conducted with three replications at each of two locations in Canada and Germany in 2008, with three host genotypes differing in FHB resistance level. Mean FHB indices and DON content were analysed. Mean FHB indices across locations ranged from 5.48 - 34.42%. The resistant host genotype showed resistance regardless of the isolate or location. The differences between mean FHB indices of 3-ADON and 15-ADON chemotypes were not significant. In contrast, DON production by the 3-ADON isolates was significantly ($P < 0.05$) higher at two locations (13.0 *vs*. 7.6 mg kg^{-1}). Acetylated forms of DON accounted for only 2.5% (3-ADON) and 0.4% (15-ADON) of the total DON concentration across the two German locations. 3-ADON isolates may produce more DON depending on location than 15-ADON producers, but their mean aggressiveness is quite similar.

P55. Differences in rachis characteristics of Chinese Spring wheat, and CS-7EL, an addition line derived from Chinese Spring that is resistant to *Fusarium graminearum*. S.S. Miller, E. Watson, and J. Lazebnik. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C6.*

A strain of *Fusarium graminearum* transformed to express green fluorescent protein was used for point inoculations of Chinese Spring (CS) wheat, which is very susceptible to FHB, and CS-7EL, a resistant wheat line derived from CS which contains a portion of the long arm of the 7E chromosome from *Thinopyrum elongatum*. The resistance displayed by CS-7EL appears to be multi-component, involving both physical and chemical factors. In CS, the fungus quickly destroys the inoculated floret (by 4 days after inoculation) and by 7 days, progresses through the node into the rachis in both directions. In CS-7EL, growth within the inoculated floret is much slower (10 days to floret browning), but ultimately the inoculated floret is destroyed also. Typically very little fungus progresses through the node into the rachis. In the research reported here, we have targeted the rachis for more extensive study. Preliminary results suggest that the internodes of the rachis are longer in the resistant line, thus increasing the physical distance from the infected floret to other potential sites for spread of the infection. Microscopy reveals early and extensive deposition of brown matter in the node of the inoculated floret of CS-7EL which fills cells and vessels in both parenchyma and vascular tissues. These deposits appear to block the spread of the fungus beyond the infected floret. In CS, although early browning of the tissue is visible to the naked eye as well as through the microscope, the cells and vessels are not filled with brown matter as they are in CS-7EL. Tests to determine the nature of browning in both varieties are ongoing.

P56. Transcriptional profiling of maize kernels infected with *F. graminearum*. M. Mohammadi, W. Bosnich, D. Schneiderman, A. Johnston, P. Couroux, N. Tinker, and L.J. Harris. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Fusarium graminearum is the causal agent of gibberella ear rot in maize. Transcriptional profiling enriches our knowledge of the molecular response of maize kernels to *F. graminearum*. Two maize inbred lines (B73 and CO441), differing in their susceptibility to *F. graminearum* infection, were selected. Comparisons of transcript abundance were conducted by hybridizations of maize long oligomer microarrays (representing 58K transcripts) with RNA isolated from *F. graminearum*-treated kernels and (i) untreated kernels and (ii) mock-treated kernels. By comparing the results obtained with those previously reported for trichothecene-responsive transcripts in wheat and barley, we have divided our discussion into two sections: (a) genes for which previous evidence exists showing that they are more likely induced upon trichothecene accumulation (e.g. glucosyltransferases, transporters); and (b) genes that are putatively induced independent of trichothecenes and as a result of pathogen invasion per se (e.g. pathogenesis-related proteins, peroxidases, and proteins involved in plant hormonal responses, cell wall modifications, and secondary metabolism biosynthesis).

P57. Salicylic acid inhibits the germination and growth of *Fusarium graminearum* and improves the resistance of wheat to fusarium head blight. P.F. Qi, T. Ouellet, Y.M. Wei, and Y.L. Zheng. *(P.Q., Y.W., Y.Z.) Triticeae Research Institute, Sichuan Agricultural University, Yaan, Sichuan 625014, PR China. (P.Q., T.O.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C.*

Salicylic acid (SA) is one of the key signal molecules in regulating the resistance of plant to diverse pathogens. It is predominantly associated with resistance against biotrophic and hemibiotrophic pathogens, and triggering systemic acquired resistance (SAR). However, whether SA directly affects *Fusarium graminearum* (Fg) and how SA influences the defence efficiency of wheat against fusarium head blight (FHB) are still poorly understood. Here we show that the germination of Fg spores can be significantly reduced by a concentration of SA between 600-800μM in a modified SNA media, and 1mM SA can stop Fg germinating; Growth of Fg mycelia can be significantly inhibited under a concentration higher than 1mM, and can be stopped by 5mM SA; Point inoculation of 10μl solution containing 400μM-1mM SA and 1000 spores can protect a very susceptible *Triticum aestivum* cultivar 'Roblin' away from Fg infection, and further work is needed to differentiate the direct effect of SA from SAR induced by exogenously applied SA. The above results clearly demonstrate that SA has a significant and direct impact on Fg through the reduction of germination and growth rate, and SA could improve the resistance of wheat to FHB.

P58. Comparative gene expression profiling of major plant hormone pathways during infection by *Fusarium graminearum*. H.J. Rocheleau, W. Zheng, S. Gulden, R. Xu, L. Wang, and T. Ouellet. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Plant host defence involve many changes in hormone signalling pathways (jasmonic acid, salicylic acid, gibberellin, auxin, shikimate) for the majority of plant infection diseases. In this study, we examine the molecular basis of the host wheat resistance against *Fusarium graminearum* (Fg) by comparing RNA profiles of key genes of these plant hormone signalling pathways. Differences in RNA transcript levels of three wheat varieties: FHB-susceptible Roblin, moderately resistant NuyBay and very resistant Wuhan were determined by the Affymetrix GeneChip Wheat Genome Array and then validated by quantitative PCR (qPCR). RNA was extracted from head tissues sampled at 0, 1, 2, 4 days post-inoculation (dpi) with water or Fg spores (100 spores/floret). Each cDNA sample was tested for its relative expression level and normalized using the averaged expression of three reference genes. We observed a major trend among most genes validated: expression varies during the development of infection, is strongly associated with the fungal biomass and also consistent with DON accumulation. The responsiveness of the very infected, susceptible variety Roblin contrasted strongly in our study with the modest response observed in the resistant variety Wuhan. This suggests that, for the pathways analysed, the level of RNA accumulated is not the major determinant of resistance in Wuhan.

P59. Differential effect of chemotypes of *Fusarium graminearum* on spring wheat. Y.F. Ruan, C.J. Pozniak, P. Hucl, and J.M. Clarke. *Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8.*

Two trichothecene chemotypes of *Fusarium graminearum*, 3-ADON and 15-ADON, are the most prevalent chemotypes in Canada. Few studies have examined the interaction of wheat genotype and *Fusarium* chemotype, which would be valuable in designing resistance breeding strategies. In this study, four tetraploid and three hexaploid wheat genotypes representing different FHB resistance levels were evaluated in replicated greenhouse trails. Spikes were inoculated with one of four isolates (two 3-ADON; two 15-ADON), and two mixtures containing one isolate of each chemotype. Disease severity was scored at day 7, 14 and 21 after inoculation and recorded as percentage of infected spikelets. Spread of FHB within the heads was assessed by the area under disease progress curve (AUDPC). Compared with the 15-ADON chemotype, a higher level of disease development was observed in hexaploid wheat inoculated with the 3-ADON chemotype. The opposite was observed in tetraploid wheat except for the susceptible control, and suggests different genetic mechanism(s) of resistance in tetraploid wheat. For all genotypes, a mixture of 3-ADON and 15-ADON showed similar pathogenicity to individual inoculations with 3-ADON or 15-ADON chemotype. Our results also indicated that there was some influence of isolate on the pathogenicity within chemotype.

P60. Overexpression of the *Arabidopsis* GLK1 transcription factor in wheat and its effect on reprogramming gene expression networks in disease resistance. J. Singh, M. Jordan, R. Pandeya, G. Allard, M. Zirino, A. Kalikililo, J. Douglas, and P. Couroux. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6. (M.J. and M.Z.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9.*

GLK1 is a GARP domain transcription factor that has been shown to regulate chloroplast development in *Arabidopsis* (Waters *et al,* 2009). Constitutive overexpression of GLK1 in *Arabidopsis* shows accumulation of transcripts encoding defense response proteins and resistance to *Fusarium graminearium* in the leaves (Savitch *et al,* 2007). To assess if GLK1 is able to impart tolerance to fusarium head blight in wheat, cv. Fielder was transformed to constitutively express *Arabidopsis* (*At*)GLK1. Transgenic wheat *At*GLK1OX plants showed improved resistance to FHB compared to the wild type after the heads were spray inoculated with *F. graminearum*. To gain insights into the nature of *At*GLK1 induced resistance, transcript profiles of the transgenic wheat plant leaves were interrogated with the 61K Affymetrix Wheat Microarray. Transcripts of 1200 genes constitutively accumulated greater than 2 fold with P-values of less than 0.1 in the transgenic plants compared to the WT. The most upregulated transcripts encoded genes that are directly or indirectly involved in plant defense against biotic and abiotic stresses. These include UDP-glycosyl transferases (50-150x), AAA-ATPase (50-70x), glutaredoxins (10x), cellulases, thioredoxins, glutathione peroxidases and other genes involved in management of RO and pathogen responses. The significance of the reprogramming of the gene expression networks by GLK1 to produce defenses against biotic stress will be discussed.

P61. Gene expression analysis of related wheat lines with contrasting levels of head blight resistance after *Fusarium graminearum* inoculation. B. Steiner, A. Limmongkon, K. Schiessl, M. Lemmens, H. Jia, G. Muehlbauer, A. Posekany, D.P. Kreil, and H. Buerstmayr. *BOKU-University of Natural Resources and Applied Life Sciences Vienna, Department IFA-Tulln, Institute for Biotechnology in Plant Production Konrad Lorenz Str. 20 A-3430 Tulln Austria. (A.P. and D.K.) BOKU-University Vienna, Dept of Biotechnology. (H.J., G.M.) Department of Agronomy and Plant Genetics, University of Minnesota, 411 Borlaug Hall, 1991 Upper Buford Cir., St. Paul, MN, USA, 55108-6026.*

Eight spring wheat genotypes with contrasting phenotypes for FHB resistance were used in this study: the highly resistant line CM82036, the highly susceptible cultivar Remus, four BC_5F_2 near isogenic lines (NILs) for *Fhb1* and *Qfhs.ifa-5A* and two doubled haploid (DH) lines from a CM82036/Remus mapping population differing in *Fhb1* and *Qfhs.ifa-5A*. At anthesis the flowering ears of the plants were single floret inoculated by *F. graminearum* or water. The inoculated spikelets were harvested at several time points after inoculation and dissected into the generative and vegetative parts for RNA preparation. Differential gene expression was monitored with two complementary methods: 1) cDNA-AFLPs or 2) using the Affymetrix wheat GeneChip. At early time points (8-24 hpi) after inoculation only few genes were differentially expressed, at later time points (48-72 hpi) an increasing number of differentially expressed transcripts was evident. A comparative analysis of the data on identified candidate genes gained by the two complementary approaches will be presented.

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P62. Preliminary observations on diversity in several fusarium head blight-causing *Fusarium* species from Manitoba, Ontario and Québec. A. Tekauz and D. Gaba. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M. (D.G.) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main Street, Winnipeg, MB, Canada, R3C 3G8.*

Several *Fusarium* species are routinely isolated from seed of oat and barley crops affected by fusarium head blight (FHB). These species, *F. avenaceum* (Fa), *F. graminearum* (Fg), *F. poae* (Fp) and *F. sporotrichioides* (Fs), incite the disease in field trials or controlled environment settings on both oat and barley. Recent work shows that the population of *F. graminearum* from wheat in Canada comprises two deoxynivalenol (DON) chemotypes, 3-AcDON and 15-AcDON, which can differ in their total DON production. It is not known whether the Canadian populations of the other species which cause FHB are physiologically uniform or diverse, or differ in their mycotoxin profiles. To assess their diversity, 20-29 isolates of each of Fa, Fg, Fp and Fs, obtained from three Canadian provinces, were cultured on agar media under uniform conditions to observe their rates of growth. Within each species colony growth varied significantly among isolates. Regional origin, more than the host source (wheat, oat or barley) of the isolate, influenced colony growth. A first test for trichothecene mycotoxins on single isolates from oat, grown on sterilized rice medium, indicated that Fa produced no toxin (moniliformin not assayed), Fg produced DON and 3-AcDON, Fp produced nivalenol, and Fs produced HT-2 and T2.

P63. Determining the structure of DNA integrated into *Fusarium graminearum* by transformation: Input DNA forms tandem repeats. R.J. Watson, S. Burchat, J. Bosley, and S. Wang. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Transformants of *Fusarium graminearum* were derived using plasmid DNA linearized using different restriction enzymes. The plasmids were designed to replace the trichodiene synthase gene, a cutinase gene or a xylanase gene with a hygromycin-resistance marker cassette by homologous recombination between 1-kbp segments of flanking DNA. The transformants did not exhibit the DNA structure expected to arise if the marker cassette was introduced by double recombination at the homologous ends. Instead, they contained complete linearized plasmids joined end-to-end and integrated into the genome. Transformant types included ectopic integrations and integrations at the target site with or without removal of the targeted gene. We have analyzed a large number of transformants using cloning, PCR and DNA sequencing to determine the structures of their integrated DNA. The data indicate that 1-3 copies of input DNA are first joined end-to-end to produce either linear or circular structures, probably mediated by the nonhomologous end-joining (NHEJ) system. The end joins typically have 1-5 nucleotides in common and are near or within the original cleavage site of the plasmid. For ectopic integrations the linear DNA is joined to two ends of genomic DNA with the same join characteristics. Integration at the target site involves replication around circularized input DNA, beginning and ending within the flanking homologous DNA, resulting in the integration of multiple tandem copies of the entire structure. This results in deletion or duplication of the target site, or leaves one copy at either end of the integrated multimer.

P64. New insights into the hydrogen-bonding behavior of T-2 toxin and inferences on its role in trichothecene toxicity. P. Chaudhary, J.T. Goettel, T. Montina, N.A. Foroud, P. Hazendonk, and F. Eudes. *Department of Chemistry and Biochemistry, University of Lethbridge, 4401 University Drive West, Lethbridge, AB, Canada, T1K 3M4. (N.A.F. and F.E.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, 5403-1st Avenue South, Lethbridge, AB, Canada, T1J 4B1.*

The structure of T-2 toxin in the solid-state is limited to x-ray crystallographic studies, which lack sufficient resolution to provide information on its hydrogen-bonding interactions. Furthermore, its solution-structure is not well described and also provides little insight into its hydrogen-bonding behavior. Hydrogen-bonding interactions are often an important part of biological activity. In order to study these interactions, the structure of T-2 toxin was compared in solution- and solid-state using NMR. It was determined that the solution and solid-state structure differ dramatically as indicated by differences between their carbon spectra. These differences are further supported by the hydrogen coupling constants observed in the solution-state. Furthermore, exchange dynamics was observed between the hydroxyl hydrogen on C-3 and water molecules in solution. In addition, the NOESY technique showed that the hydrogens on C-3 and C-4 were bound to at least one water molecule. This indicates a strong preferential hydrogen bonding interaction on the side of the molecule with the reactive epoxide ring, which is known to be essential for trichothecene toxicity. These results infer that these hydrogen-bonding interactions must play an important role in the biological function of T-2 toxin and that further detailed studies of this whole class of toxins, namely trichothecenes, should be pursued using this methodology.

P65. Intracellular expression of a single-domain antibody reduces cytotoxicity of 15-AcDON. P.J. Doyle, H. Saeed, A. Hermans, S. Gleddie, G. Hussack, M.E. Savard, B.A. Blackwell, C. Seguin, C.R. MacKenzie, and J.C. Hall. *Department of Environmental Biology, University of Guelph, Guelph, ON, Canada, N1G 2W1. (H.S., S.G., M.S., B.B., C.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, Canada, K1A 0C6. (G.H., C.M.) Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, Canada, K1A 0R6.*

15-Acetyl-Deoxynivalenol (15-AcDON) is a low molecular weight sesquiterpenoid trichothecene mycotoxin associated with fusarium ear rot of maize and fusarium head blight of small grain cereals. The accumulation of mycotoxins such as 15-AcDON and deoxynivalenol (DON) within harvested grain is subject to stringent regulation as both toxins pose dietary health risks to humans and animals. These toxins inhibit peptidyl transferase activity which in turn limits eukaryotic protein synthesis. To assess the ability of intracellular antibodies (intrabodies) to modulate mycotoxin-specific cytotoxocity, a gene encoding a camelid single-domain antibody fragment (V_HH) with specificity and affinity for 15-AcDON was expressed in the methylotropic yeast *Pichia pastoris*. Mycotoxin-mediated cytotoxicity was assessed by continuous measurement of cellular growth. At equivalent doses, 15-AcDON was significantly more toxic to wild-type *P. pastoris* than was DON which, in turn, was more toxic than 3-AcDON. Intracellular expression of toxin-specific V_HH within *P. pastoris* conveyed significant ($p = 0.01$) resistance to 15-AcDON cytotoxicity at doses ranging from 20 to 100 $\mu g \bullet mL^{-1}$. Interestingly, we documented a biochemical transformation of DON to 15-AcDON which explained significant attenuation to DON at 100 and 200 $\mu g \bullet mL^{-1}$. The "proof of concept" established in this work suggests that *in planta* V_HH expression may lead to enhanced tolerance to mycotoxins and thereby limit *Fusarium* infection of commercial agricultural crops.

P66. *Fhb1* protects wheat against nivalenol and deoxynivalenol. M. Lemmens, A. Koutnik, B. Steiner, H. Buerstmayr, F. Berthiller, R. Schuhmacher, F. Maier, and W. Schäfer. *University of Natural Resources and Applied Life Sciences Vienna, Department IFA-Tulln, Konrad Lorenz Str. 20, A-3430 Tulln, Austria. (F.B., R.S.) Department for Agrobiotechnology - IFA-Tulln, Center for Analytical Chemistry. (F.M., W.S.) Center of Applied Molecular Biology of Plants, Department of Molecular Phytopathology and Genetics, University of Hamburg, Germany.*

The main goal in this contribution was to investigate whether *Fhb1* also governs resistance towards NIV, a trichothecene structurally related to DON. Using mutants with a disrupted trichodiene synthase, it is confirmed that NIV and DON production are fungal virulence factors important for the spread of symptoms in the wheat ear but not for the induction of fusarium head blight. Both toxins influence the probability that symptoms spread. It is demonstrated that NIV is phytotoxic on wheat ears and that the purified toxin can induce symptoms identical to those described for DON. *Fhb1* protects the wheat line against both NIV and DON. The mechanism of NIV resistance is not known but is probably different from the detoxification mechanism of DON.

P67. Coordinated regulation by Tri10 and Tri6 in *Fusarium graminearum.* W. Leung, C. Nasmith, L. Wang, A. Johnston, L.J. Harris, and G. Subramaniam. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6.*

Fusarium graminearum is a globally distributed fungal pathogen that causes disease in cereal crops such as wheat and barley and it also contaminates host plants with secreted mycotoxins such as deoxynivalenol. Consumption of infected or contaminated cereal products is hazardous to animals, as a result, fungal infection and contamination causes major economic losses in agriculture industries worldwide. Gene expression profiling of *Tri10Δ* and *Tri6Δ* mutant strains under DON-inducing conditions *in culture* provided evidence that the regulatory roles of the two studied genes extend beyond the trichothecene biosynthesis pathway. They are potential regulators of another clustered secondary metabolic pathway - butenolide biosynthetic pathway. Furthermore, *Tri10* and *Tri6* also regulate genes that might contribute to virulence of *F. graminearum*. In addition to the previously proposed model where *Tri10* and *Tri6* are independently regulated, we have evidence to show that *Tri6* auto-regulates its own expression.

P68. Protein phosphorylation and DON synthesis: Candidate phosphopeptides from wild-type and map kinase (mgv1)-deficient *Fusarium graminearum*. C. Rampitsch, G. Subramaniam, N.V. Bykova, and S. Djuric-Ciganovic. *Cereal Research Center, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, Canada, R3T 2M9. (R.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6. (N.B.) Department of Biology, Memorial University of Newfoundland, St. John's NL, Canada, A1B 3X9.*

The steps comprising the synthesis of deoxynivalenol (DON) and its derivatives by *Fusarium graminearum* are well understood, but their regulation at the molecular level is not. We are investigating a potential role for protein phosphorylation in initiating DON synthesis during nitrogen starvation *in vitro*. We have previously used multidimensional separations and analyses, MUDPiT and GeLCMS, to establish a phosphoproteome map of wild-type *F. graminearum*, and are now probing the phosphoproteome of MAP-kinase (MGV1)-deficient *F. graminearum* after the onset of DON synthesis. This mutant produces only about 20% of the DON levels seen in wild-type *in vitro*. Phosphopeptides were enriched by immobilized metal affinity chromatography and titanium dioxide affinity chromatography. Enriched peptides were analyzed by LC-MS using collision induced dissociation (CID) and electron transfer dissociation (ETD) fragmentation of peptides. The Mascot search engine was used to query a complex fungal sequence database to assign tentative protein identities based frequently on single peptides. All phosphorylation sites were confirmed manually. The biological role of some of these proteins in regulating DON synthesis will be assessed *in vivo* by producing *F. graminearum* mutants and measuring both their virulence and ability to produce DON.

P69. Concentration of *Fusarium* toxins in naturally contaminated maize and processing co-products derived from ethanol production.* A. Schaafsma, V. Limay-Rios, and J.D. Miller. *University of Guelph, Ridgetown Campus, Ridgetown, ON, Canada, N0P 2C0. (J.D.M.) Department of Chemistry, Carleton University, Ottawa, ON, Canada, K1S 5B6.*

Three matrices [corn meal, distiller's dried grains with solubles (DDGS), and condensed distiller's soluble (CDS)] were sampled in sequence from a continuous dry milling processing plant located in Chatham, Ontario, for the determination of mass balance of DON. LC-MS/MS was used as a confirmatory method. DON concentrations in the CDS and the final DDGS co-product were significantly higher ($P \leq 0.01$) than in the starting material (corn grain). Toxin concentration increased by a factor of 3 on a dry weight basis in DDGS compared to the starting corn, and by 4 in CDS. Mass balance calculations show that CDS is the main source of contamination of DON comprising ca. 70% of the toxin found in the final product (DDGS). The presence of mycotoxins in DDGS and CDS affects their utility as animal feed supplements. Our data indicate that concentrations in the grain corn entering ethanol plants should be close to the dietary values recommended for swine in Canada and the United States for DON (1 mg kg^{-1}). Small amounts of acetyl-DON, DON-3-glucoside (D3G) and zearalenone were found in corn, DDGS and CDS. Unlike the situation for DON, the D3G was not concentrated into DDGS and CDS. This indicates that some D3G may have been hydrolyzed during the fermentation process.
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P70. Disruption of genes involved in butenolide and culmorin synthesis in *Fusarium graminearum.* D.T. Schneiderman, S. McCormick, N.J. Alexander, A. Johnston, and L.J. Harris. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON, Canada, K1A 0C6. (S.M., N.J.A.) Mycotoxin Research Unit, National Center for Agricultural Utilization Research, Agricultural Research Service, U.S. Department of Agriculture, Peoria, IL, USA, 61604.*

Butenolide (4-acetamide-4-hydroxy-2-butenoic acid γ-lactone) and culmorin (a tricyclic sesquiterpene diol) are two less-studied mycotoxins produced by several *Fusarium* species, including *Fusarium graminearum*. A putative butenolide biosynthetic eight-gene cluster in *F. graminearum* includes *fg08080* which encodes a zinc-finger protein and may function as a regulatory gene for the cluster. Gene disruption of *fg08080* resulted in loss of butenolide biosynthesis and the down-regulation of other genes within the cluster. *Fusarium* infection in wheat revealed no difference in virulence between the *fg08080*-disrupted and wild-type strains. *fg08080* is required for butenolide biosynthesis in *F. graminearum* and may control butenolide biosynthesis through regulation of the gene cluster.

A terpene synthase gene, *fg10397*, was observed to be induced under trichothecene-inducing conditions and during plant infection (based on EST library representation, Northern and microarray analysis). Transformed yeast cultures expressing FG10397 produced longiborneol, a terpene with the same tricyclic structure as culmorin, which was not produced by the progenitor yeast strain. The *fg10397* gene was disrupted in *F. graminearum* strain 9F1, a wild-type strain that produces a significant amount of culmorin *in vitro*. No culmorin was produced by 9F1 strains with a disrupted *fg10397* while wild-type and *fg10397* add-back strains produced culmorin in liquid cultures. Thus, *fg10397* encodes a longiborneol synthase that is required for culmorin synthesis.

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