

3rd Canadian Workshop on Fusarium Head Blight/ Colloque Canadien Sur La Fusariose

Delta Winnipeg
Winnipeg, Manitoba
Dec. 9th to 12th, 2003

National Committee:

Jeannie Gilbert (Chair)
Brent McCallum (Secretary)
Shaffeeq Ali
Randy Clear
André Comeau
George Fedak
Dilantha Fernando
Mark Jordan
David Kaminski

Richard Martin
Iris Meck Communications
Therese Ouellet
Penny Pearse
Art Schaafsma
Andy Tekauz
Albert Tenuta
Harvey Voldeng
Allen Xue

Proceedings Compiled by Randy Clear, Grain Research Laboratory, Canadian Grain
Commission, 1404-303 Main St., Winnipeg, Manitoba, R3C 3G8

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Please note: These Proceedings have been altered somewhat from those in the hard
copies handed out at the meeting. There have been some changes to a few abstracts and
the participant list has been updated. In addition, the comments from the discussions held
at the 8 Breakout Sessions and the recommendations reached at the Issues and Priorities
meeting have been added.

This workshop is brought to you by the following:

- *Alberta Agriculture*
- *Manitoba Agriculture, Food and Rural Initiatives Saskatchewan Agriculture, Food and Rural Revitalization*
- *Ontario Ministry of...*
- *Ridgetown College, University of Guelph*
- *Canadian Grain Commission*
- *Agricultural Adaptation Council*
- *Agriculture and Food Council*
- *CARDS*
- *Manitoba Rural Adaptation Council Inc.*
- *AAFC*
- *University of Manitoba*
- *Fusarium Action Canada*
- Funding was provided jointly by the Agricultural Adaptation Council in Ontario, Agriculture and Food Council in Alberta, Canadian Adaptation and Rural Development Saskatchewan and the Manitoba Rural Adaptation Council under Agriculture and Agri-Food Canada's Canadian and Adaptation and Rural Development Fund. Created in 1995, the Canadian Adaptation and Rural Development (CARD) Fund has invested over \$450 million in more than 5,000 national and regional initiatives to help stimulate progressive change in the industry and rural communities across Canada.

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***3rd Canadian Workshop on Fusarium Head Blight
3^{ème} Colloque Canadien sur la Fusariose***

Dear Workshop Participant

Welcome to the 3rd Canadian Workshop on Fusarium Head Blight/Colloque Canadien sur la Fusariose, (CWFHB/CCF) Winnipeg, Manitoba, Canada, December 9-12, 2003.

The summaries of reviews and the abstracts in the following pages represent both our current understanding of fusarium head blight (FHB) and the latest research findings on a disease that has cost the Canadian grain industry millions of dollars. Since biennial meetings were initiated in 1999, the main priority of the CWFHB/CCF has been to provide a forum for information exchange among researchers, producers, and industry. The 3rd CWFHB/CCF continues this tradition with sessions devoted to Industry and Consumer Issues, Breeding and Genetics, Disease Management, Epidemiology, and Molecular Breeding and Biotechnology.

Many persons, institutions, and companies have worked together to bring this meeting to fruition. I would like to acknowledge the Canadian Adaptation and Rural Development (CARD) fund and our industry sponsors for their generous financial support, and the in-kind support of both federal and provincial governments and universities across Canada. Thanks also are extended to all those who have agreed to chair sessions and moderate in the Breakout Sessions, and everyone who took the time to prepare their presentations and posters that provide a backdrop against which to discuss the needs and priorities for research in the coming years.

This year marks the inception of Fusarium Action Canada (FAC), a federally incorporated, not-for-profit corporation whose objective is to help facilitate meetings such as the CWFHB/CCF. Through FAC, we gratefully acknowledge the able assistance of Iris Meck Communications Inc. for working with our industry sponsors and for help in conference registration coordination.

Last but not least, I wish to recognize the members of the local and national organizing committees whose willing and prompt responses to all the demands placed on them have made this workshop possible. It has been a pleasure working with such a diligent and supportive team.

Welcome to the 3rd CWFHB/CCF. I trust you will take away memories of a meeting that brought together researchers, producers and industry members in a spirit of mutual learning and information exchange, to further our understanding of the many facets of FHB, and to identify the direction required of future research.

Sincerely
Jeannie Gilbert
Chair 3rd CWFHB/CCF

3rd CWFHB

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Saskatchewan Wheat Pool/AgPro

AGENDA

Tuesday, December 9, 2003

6:00-9:00 pm Registration

7:30-9:30 pm Reception and Poster viewing in Ballroom

Wednesday, December 10, 2003

7:00 am Registration

7:00 Breakfast

8:00 Welcome from 3rd CWFHB Chair

Jeannie Gilbert, Cereal Research Centre, and Agriculture and Agri-Food Canada, Winnipeg, MB

8:05 Welcome address

Dr. Barry Todd, Acting Deputy Minister, Manitoba Agriculture, Food and Rural Initiatives, Winnipeg, MB

8:10-10:00 Session I: Overview and toxins

Chair: Andy Tekauz, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

8:10 History and status of Fusarium Head Blight

Robert Stack, North Dakota State University, Fargo, ND, USA

8:40 Why the tolerances for DON are going down

David Miller, Carleton University, Ottawa, ON

9:05 Mycotoxin analysis, present and future

Marc Savard, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON

9:25 Consumer health and safety: monitoring food and feed.

Helen Page, Canadian Food Inspection Agency, Winnipeg, MB

9:40 Fusarium Head Blight – Alberta's Initiative

Shafeek Ali, Alberta Agriculture, Food and Rural Development, Edmonton, AB

10:00-10:30 Refreshment Break

10:30-12:00 **Session II: Industry and consumer issues**
Chair: Randy Clear, Canadian Grain Commission, Winnipeg, MB

10:30 Methods and issues regarding detection of Fusarium/DON in commercial grain in Canada

Art Schaafsma, Department of Plant Agriculture, University of Guelph, ON

10:45 Current impact and emerging issues for *Fusarium* head blight in the malting and brewing industries.

Richard Joy, Rahr Malting (Canada) Ltd., Alix AB

11:00 Current impact and emerging issues for FHB in grain handling from farm to delivery

Al Morris, Agricore United, Winnipeg, MB

11:15 Current impact and emerging issues for mycotoxins in feedstock

James House, University of Manitoba, Winnipeg, MB

11:30 Opportunities for using distillers dried grain with solubles (DDGS) in pig diets

Ian Seddon, Manitoba Agriculture and Food, Winnipeg, MB

12:00-1:30 Lunch served

1:30-3:00 **Session III: New sources of and breeding for FHB resistance**
Chair: Harvey Voldeng, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON

1:30 Progress in spring wheat

Stephen Fox, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

1:45 Progress in winter wheat

Radhey Pandeya, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food, Canada, Ottawa, ON

2:00 Progress in improvement of *Fusarium* resistance of durum wheat.

John Clarke, Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, Swift Current, SK

2:15 Progress in breeding for *Fusarium* head blight resistance in barley.

Bill Legge, Agriculture and Agri-Food Canada Research Centre, Brandon, MB

2:30 *Fusarium* head blight of oat - current status in western Canada

Andy Tekauz, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

2:45 Haplotype diversity at *Fusarium* head blight resistance QTLs in wheat

Curt McCartney, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

3:00-3:30 Refreshment Break

3:30-5:00 **Session IV: Host resistance genetics**
*Chair: Mark Jordan, Cereal Research Centre, Agriculture and Agri-Food
Canada, Winnipeg, MB*

3:30 From QTL mapping to breeding to production

Daryl J. Somers, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

4:00 Transformation to provide new genes for FHB resistance: A summary of current US public research

Lynn Dahleen, USDA, Fargo, ND

4:30 Genomics studies to understand the interactions between *Fusarium graminearum* and its cereal hosts

Thérèse Ouellet, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON

5:00-6:00 Poster session (Authors present)**6:30-7:00 Cocktails****7:00-9:30 Evening Banquet**

Thursday, December 11, 2003

7:00 am Registration

7:00-8:30 Breakfast

8:30-10:00 Session V: Epidemiology
Chair: Allen Xue, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON

8:30 Predicting deoxynivalenol in wheat for Ontario.

David Hooker, University of Guelph, Guelph, ON

8:45 Progress in forecasting: FHB risk forecasts in Manitoba.

David Kaminski, Manitoba Agriculture, Food, and Rural Initiatives, Carman, MB

9:00 Infection of tolerant and susceptible wheat and barley varieties using GFP-Fusarium.

Shea Miller, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON

9:15 Mechanisms of resistance and tolerance to FHB.

André Comeau, Agriculture and Agri-Food Canada, Ste-Foy, PQ

9:30 Biological control methods to manage fusarium head blight disease of wheat: is it a short or long term solution to the problem?

W.G. Dilantha Fernando, University of Manitoba, Winnipeg, MB

9:45 Sources and dispersal of *Gibberella zeae*/*Fusarium graminearum* inoculum

J. Gilbert, Agriculture and Agri-Food Canada, Winnipeg, MB

10:00-10:30 Refreshment Break

10:30 – 12:00 SESSION VI: DISEASE MANAGEMENT
Chair: Penny Pearce, Saskatchewan Agriculture, Food and Rural Revitalization, Regina, SK

10:30 The grower's perspective: Managing fusarium head blight on the farm

Ray Mazinke, Grain Producer and Agri-retailer, Morris, MB, and Peter Johnson, Grain Producer and Cereal Specialist, London, ON

11:00 Sprayer technology: How best to apply fungicides

Tom Wolf, Saskatoon Research Centre, Agriculture & Agri-Food Canada, Saskatoon, SK, and Helmut Spieser, Ontario Ministry of Agriculture & Food, Ridgetown, ON

11:30 Integrated Fusarium head blight management: Employing all the tools

Albert Tenuta, Ontario Ministry of Agriculture & Food, Ridgetown, ON

12:00-1:30 Lunch served

1:30-2:30 Poster Session (Chair: André Comeau)

Discussion of specified posters. Questions to authors from floor.

2:30 -5:30 Breakout Sessions

See the end of Proceedings for your session times

- 1. Mycotoxins and FDK- global and local - health, trade, detection, consumer issues**
Moderators: Art Schaafsma (2:30), David Miller (3:30), and Marc Savard (4:30) (Randy Clear)
Room: **Campaign B**
- 2. - Milling and brewing**
Moderator: Jim Dexter (3:30)
Room: **Campaign A**
- 3. - Livestock, industrial end-use, ethanol production/Seed production and trade**
Moderator: Jim House (2:30) (Shaffeeq Ali)
Room: **Strathcona**
- 4. - Breeding for FHB resistance - wheat / oats**
Moderator: Brent McCallum (2:30), George Fedak (3:30), Harvey Voldeng (4:30)
Room: **Ballroom C**
- 5. - Breeding for FHB resistance - barley/corn**
Moderator: Brian Rossnagel (4:30)
Room: **Kildonan**
- 6. - Molecular breeding and biotechnology**
Moderator: Mark Jordan (2:30), Therese Ouellet (3:30) (Daryl Somers)
Room: **Victoria**
- 7. - Disease management**
Moderator: Penny Pearse (2:30), Richard Martin (3:30), David Kaminski (4:30)
Room: **Ballroom A**
- 8. - Epidemiology (including the role of infected seed)**
Moderator: Allen Xue (3:30), Kelly Turkington (4:30) (Jeannie Gilbert)
Room: **Colbourne**

Refreshment break

Evening Open

Friday, December 12, 2003

7:00-8:00 am Breakfast

8:00-9:00 International Perspective on FHB

Chair: Jeannie Gilbert, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

8:00 Fusarium diseases of wheat and barley: an Australian perspective

Steven Simpfendorfer, Plant Pathologist, Crop Protection, NSW Agriculture, Tamworth Agricultural Institute, Tamworth, NSW, Australia

8:20 Studies for enrichment of wheat germplasm with fusarium head blight (FHB) resistance in Japan.

T. Ban, Biological Resources Division, Japan International Research Center for Agricultural Sciences, Tsukuba, Ibaraki, Japan

8:40 International approach to breeding for fusarium head blight resistance.

Maarten Van Ginkel, Head Fusarium Research and Gene Bank, CIMMYT International, Mexico, D.F. Mexico

9:00 Reports from Breakout Sessions, 8 X 10 min each (Moderated)

10:30 Refreshment break

11:00 Issues and priorities for next two years

Chair: Kelly Turkington

12:00 Closing remarks

Jeannie Gilbert, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB

End of conference

3rd Canadian Workshop on Fusarium Head Blight

Speaker Profiles

Shafeek Ali, Head, Pest Risk Management Unit, Alberta Agriculture, Food and Rural Development, Edmonton, Alberta

Shafeek provides leadership in proactive measures to prevent the establishment of new pests in Alberta and administers the Provincial Pest Legislations and is Chair, Alberta's Fusarium Action Committee and the person responsible for the implementation of the Plan in Alberta. Shafeek will present Alberta's *Fusarium graminearum* Management Plan and its impact in managing the spread of this pest in Alberta.

André Comeau, Agriculture and Agri-Food Canada, Ste-Foy, Quebec

André is known as a senior scientist, cereal breeder and "germplasm developer". André has a breadth of general knowledge about what Fusarium head blight can do in different environments, through his collaboration with Brazil. André Comeau is developing disease resistant wheat germplasm at Agriculture Canada Ste-Foy (Quebec) and has discovered interactions between FHB and other diseases, and is now active in collaboration with scientists across Canada in the development of better sources of resistance.

Lynn Dahleen, Research Geneticist, USDA-Agricultural Research Service, Fargo, North Dakota

As the former chair of the Biotechnology Research Area of the US Wheat and Barley Scab Initiative, Lynn is familiar with the transformation research being conducted in the US and has contacts with all the researchers. Lynn conducts Biotech research on barley, including gene mapping, gene and promoter isolation, and transformation with antifungal and antitoxin genes.

Lynn will present a summary of some of the US transformation research in wheat and barley to reduce FHB and DON contamination.

Dilantha Fernando, Associate Professor, Department of Plant Science, University of Manitoba, Winnipeg, Manitoba

Dilantha is a university professor of plant pathology at the undergraduate and graduate level, and is a research leader on epidemiological and biological control processes in Canada. Dilantha works with farmers and scientists to develop sustainable disease management strategies. He has been working with the FHB disease since 1994 (10 years) and has made some significant contributions to the understanding of this pathogen's epidemiology which have been published in several peer reviewed journals. Dilantha is the principal investigator on a major research grant looking at novel methods of FHB disease management, and also co-investigator on a new NSERC strategic grant specifically looking at how weather and FHB impact wheat quality as an end product. He will be presenting along with Dr. Jeannie Gilbert, some of the novel approaches that are taken in North America to reduce the impact of FHB on yield and toxin production in wheat.

Jeannie Gilbert, Research Scientist, Plant Pathologist, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, Manitoba

Jeannie has been working on FHB on wheat for 14 years and has collaborated on more than 30 funded projects since 1996; Jeannie was project leader on 15. The work has improved the understanding of the effect of weather on FHB of wheat, effect of the disease on yield and quality, duration of pathogen survival on wheat kernels, refined disease screening protocols etc. Jeannie investigates control and management strategies for fusarium head blight (FHB) of wheat.

Jeannie and Dilantha Fernando will present current knowledge of control and management strategies for FHB excluding the areas of chemical control and breeding and will include rotations, tillage practice, biocontrol, under-storey cover crops, irrigation and stubble management etc.

David C. Hooker, Ph.D., Ridgetown College, University of Guelph, Guelph, Ontario

David's current position is Post Doctoral Fellow. His expertise is in pest modeling and forecasting, statistics, and cropping systems. During the past five years, Dave has co-developed and successfully implemented the DON toxin-Fusarium forecasting model in Ontario called DONcast. His academic achievements are supplemented with extensive work at the farm level.

Jim House Ph.D., Associate Professor, University of Manitoba, Winnipeg, Manitoba

Jim House received his Ph.D. in nutrition and metabolism from the Department of Animal & Poultry Science at the University of Guelph in 1995. He is currently an Associate Professor in the Department of Animal Science at the University of Manitoba, where he is involved in research to determine efficient strategies for the removal of deoxynivalenol (DON) from contaminated grains.

Peter Johnson, Provincial Cereals Specialist, Wheat's Wild Warrior, Ontario Ministry of Agriculture and Food, London, Ontario

Peter provides agronomy extension on cereal crops in Ontario, plus has a small farm. He maintains research plots that determine the impact/benefit of various management techniques, including the comparisons between the fungicides *Folicur*, *Headline* and *Tilt*; timing trials; application comparisons; and much more. Peter has access to a large database of information and a great deal of hands-on experience both on my own farm and on other growers' farms. Peter will present farm trial data, techniques growers use to minimize fusarium risk, and explain why growers end up with wheat in high risk situations in his presentation.

Richard W. Joy ,Manager of Quality Control & Technical Services, Rahr Malting (Canada) Ltd., Alix, Alberta, (formerly Westcan Malting)

Richard was awarded his Ph.D. in Developmental Plant Physiology from the University of Calgary and was previously a Research associate with the Plant Biotechnology Institute, National Research Council of Canada in Saskatoon. Richard spend 3 years studying and conducting research at Tohoku University, Sendai, Japan, and received many Canadian and international awards and scholarships during his distinguished academic career.

David A. Kaminski, MPM, P.Ag., Plant Pathologist, Manitoba Agriculture & Food, Soils & Crops Branch, Carman, Manitoba

David continues to oversee and provide interpretation for the posting of a Fusarium Head Blight Risk Forecast for Manitoba that is generated from data collected by the ACE Weather Network. The forecast is employed by growers and agronomists in the assessment of risk and fungicide management decisions.

Curt McCartney, Post-doc, Agriculture and Agri-Food Canada, Cereal Research Centre, Winnipeg, Manitoba

Curt is a geneticist working on wheat improvement, primarily using micro satellite markers for genetic mapping. Curt will be presenting the genetic diversity in a diverse collection of Fusarium head blight resistant and susceptible wheat lines as assessed by micro satellite markers.

Ray Mazinke, farmer and Ag-retailer (Rosenort Agro Ltd.), Morris, Manitoba

Ray is the owner and operator of a large grain farm in the Red River Valley and has had ample experience in dealing with Fusarium head blight in small grain cereals. Since 1990, Ray has also been involved in an independent agronomic business.

Shea Miller, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, Ontario

Shea Miller does her research at ECORC in Ottawa, with expertise in the microstructure and microchemistry of cereals. She has been using fluorescence microscopy to elucidate the infection process in resistant and susceptible varieties of maize, wheat, and barley, with a strain of *Fusarium graminearum* that has been transformed to express a green fluorescent protein (GFP) from jellyfish. Fungal growth is observed directly on fresh material, and also in greater detail in sectioned material.

Al Morris, Agricore United, Winnipeg, Manitoba

Al Morris has spent over 31 years in the Malting / Grain Industry and has held positions of Senior Merchant - Malting Barley, and manager of Food Grains and Quality Control with Agricore United; and several positions with malting companies in barley selection, purchasing and laboratory technician.

Andrej (Andy) Tekauz, Plant pathologist, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, Manitoba

Andy studies fusarium head blight (FHB) and leaf spot diseases of barley and oat re. their management by resistance and other means. He is the Head of Cereal Diseases at the Cereal Research Centre and did the initial studies on FHB in wheat, barley, and most recently, in oat, during the current and ongoing FHB epidemic in western Canada dating back to 1986. Andy will describe the status of FHB in oat in western Canada, cultivar responses to the disease, and the prospects of breeding for resistance in his presentation.

Helmut Spieser, Agricultural Engineer, Ontario Ministry of Agriculture and Food, Ridgeway, Ontario

Ontario has been one of the leaders in FHB spray coverage technology over the past three years. Helmut has used water sensitive paper, powdered copper, and U.V. dye to determine the quality and quantity of spray coverage in his work. Helmut also educates producers, ag-business, and researchers on sprayer application technology through articles, videos, demonstrations and hands-on workshops using real sprayers as well as demonstration with a wind tunnel and patternator. Helmut's presentation will cover the importance of uniform fungicide application on wheat heads; and impact on travel speed, nozzle configuration, etc on whole head coverage. Different variables can impact the amount and quality of coverage, what are the limiting factors and how best to maximize coverage.

Daryl J. Somers, Research Scientist, Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, Manitoba

Daryl manages the primary AAFC lab for wheat molecular breeding technology development that includes genetic mapping, QTL analysis, structural genomics and technology implementation and has extensive experience and knowledge in the area of molecular breeding in wheat and specifically in FHB resistance. Daryl will present an overview of molecular breeding and structural genomics efforts for FHB resistance and how this relates to AAFC efforts.

Dr. Ian Seddon B. Sc. M.Sc. Ph.D., Swine Specialist, Animal Industry Branch, Manitoba Agriculture and Food, Winnipeg, Manitoba

Dr. Seddon received his Ph.D. in Swine Nutrition from the University of Guelph and works with all aspects of the pork production industry. He recently has been investigating opportunities for the use of co-products from ethanol production in pig diets.

Arthur Schaafsma BSc MSc PhD, Associate Professor, Department of Plant Agriculture, Field Crop Protection, University of Guelph, Ridgetown, Ontario

Art Schaafsma has experience in Research and Regulatory Affairs, Crop Protection Industry across Canada, and has been working on applied integrated systems approaches to managing Fusarium and DON in wheat and corn for about 15 years. Art also has been a consultant many groups including: the crop protection industry on Fusarium issues; the Ontario Wheat Producers Marketing Board on wheat grading issues and processes related to fusarium; and the Food and Agricultural Organization on systems approaches to managing DON in wheat based products.

Marc Savard, Research Scientist, Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, Ontario

Marc conducts research on mycotoxins, especially Fusarium toxins, focusing on structure determination, analysis, resistance and control. His lab performs over 15 000 toxin analyses per year for AAFC cereal breeders. Marc will present information that will focus on analytical methods for mycotoxins, both present methods and potential future methods.

Thérèse Ouellet, Research Scientist, Eastern Cereal and Oilseed Research Centre (ECORC), Agriculture and Agri-Food Canada, Ottawa, Ontario

Thérèse does genomics studies to understand how 1) wheat and maize defend themselves against the fusarium head blight pathogen, *Fusarium graminearum*, and 2) what means *Fusarium* uses to attack its cereal hosts. She has been a spokes person for the leading group in this research area in Canada for the last 6 years. Thérèse presentation will be an overview of the progresses in characterizing the molecular interactions between *Fusarium* and its cereal hosts, at the Canadian and international level. The plant and pathogen sides will be both addressed.

Albert Tenuta, Extension Field Crop Plant Pathologist, Ontario Ministry of Agriculture and Food, Ridgetown, Ontario

Albert is the lead technology transfer expert on plant pathology and nematodes for Ontario's field crop commodities, responsible for developing training and resource materials on field crop diseases and nematodes. In addition, Albert provides leadership in the coordination and organization of field-testing, demonstrations and development of practical applications of recent research to determine suitability for Ontario conditions.

Albert's presentation will focus on how to develop an Integrated Fusarium Head Blight Management Program and what "tools" are available to producers, ag-business and extension personal.

Oral Presentation Abstracts and Summaries

(those with an * have chosen to have an abstract published in the Canadian Journal of Plant Pathology)

Session 1: Overview and Toxins

23...History and Status of Fusarium Head Blight. R.W. Stack

25...Why the tolerances for DON are going down. David Miller

25...Mycotoxin analysis, present and future. Marc Savard

26...Consumer health and safety: monitoring food and feed. Helen Page and Lynne Underhill.

27...Fusarium Head Blight – Alberta’s Initiative. Shaffeeq Ali

Session 2: Industry and Consumer Issues

29...*Methods and issues regarding detection of Fusarium/DON in commercial grain in Canada. Art Schaafsma.

48...Current impact and emerging issues for *Fusarium* head blight in the malting and brewing industries. Richard W. Joy IV

49...Current impact and emerging issues of FHB in grain handling from farm to delivery. Al Morris

50...Current impact and emerging issues for mycotoxins in feedstocks. James D. House

51...Opportunities for using Distillers Dried Grain with Solubles (DDGS) in Pig Diets. Ian R. Seddon

Session 3: New Sources of, and Breeding for, FHB Resistance

52...Progress in spring wheat. S. Fox

53...Progress in national winter wheat Fusarium research and development. Radhey Pandeya

66...*Progress in Improvement of *Fusarium* Resistance of Durum Wheat. J. Clarke

67...Progress in breeding for Fusarium head blight resistance in barley. W.G. Legge

75...Fusarium head blight of oat - current status in western Canada

A. Tekauz

82...Haplotype diversity at Fusarium Head Blight resistance QTLs in wheat. C.A.

McCartney

Session 4: Host Resistance Genetics

83...From QTL mapping to breeding to production. D.J. Somers

83...Transformation to provide new genes for FHB resistance: A summary of current US public research. L.S. Dahleen.

84...Genomics studies to understand the interactions between *Fusarium graminearum* and its cereal hosts. Thérèse Ouellet

Session 5: Epidemiology

85...*Predicting deoxynivalenol in wheat for Ontario. D.C. Hooker

85...*Progress in forecasting: FHB risk forecasts in Manitoba. David Kaminski

86...Infection of tolerant and susceptible wheat and barley varieties using GFP-Fusarium. S. Shea Miller

88...*Mechanisms of resistance and tolerance to FHB. A. Comeau

106...*Biological control methods to manage fusarium head blight disease of wheat: is it a short or long term solution to the problem? W.G. Dilantha Fernando

107..Sources and dispersal of *Gibberella zeae*/*Fusarium graminearum* inoculum
J. Gilbert

Session 6: Disease Management

113..The Grower's Perspective: Managing Fusarium Head Blight On The Farm
Peter Johnson

113..Sprayer Technology: How Best to Apply Fungicides. *T.M. Wolf and H. Spieler.*

117..Integrated Fusarium head blight management: Employing all the tools
Albert Tenuta

International Perspectives on FHB

116..*International Approach to Breeding for Fusarium Head Blight Resistance.
Maarten van Ginkel.

116.. *Studies for enrichment of wheat germplasm with fusarium head blight (FHB) resistance in Japan. *T. Ban*

117..*Fusarium diseases of wheat and barley - an Australian perspective. *S. Simpfendorfer*

Poster Abstracts

(those with an * have chosen to have the abstract published in the Canadian Journal of Plant Pathology)

Session 1: Overview and toxins

118..Prediction of deoxynivalenol (DON) in barley using near-infrared spectroscopy.

Arganosa, G.C., Rossnagel, B.G., Legge, B.G., Savard, M., Zatorski, T and Tucker, J.

118.*Toxin content in wheat seeds in Quebec in 2002. *Y. Dion, S. Rioux and M. Lauzon.*

119.*Fusarium head blight in the Atlantic region in 2003. *R. A. Martin*

119.*Fusarium- A serious threat to the Australian wheat industry. *V. Mitter , O.A.*

Akinsanmi, S. Simpfendorfer, D. Backhouse, D. Yates and S.Chakraborty

120.* Fusarium head blight in northern Quebec. *D. Pageau, J. Lajeunesse, M. Savard, C. Coulombe and Y. Lefebvre.*

120.*Deoxynivalenol production by *Fusarium graminearum* isolates in four winter wheat cultivars. *L. Tamburic-Ilicic and A. W. Schaafsma.*

Session 2: Industry and consumer issues

121.*Interactions of weather factors and fusarium head blight, and its effect on wheat grain quality. *W.G. Dilantha Fernando, X.W. Guo, P. Bullock, H. Sapirstein, J. Dexter and T. Nowicki.*

121..Fusarium toxins in infant cereal foods and adult breakfast cereals from the Canadian retail market. *Gary A. Lombaert, Peter Pellaers, Veronica Roscoe, Meena Chettiar, David Kitchen, Susan Kotello, Thomas Krakalovich, Don Lavallee, Greg Sliva, Robert Trelka, Gary Neumann, and Peter M. Scott.*

122.*Survival of *Fusarium graminearum* through the digestive tract of cattle – fact or fiction? *D. L. McLaren, S. L. Scott, T. K. Turkington, Y. Wang and T. McAllister*

122.*Fusarium spp. from commercial seed samples in Saskatchewan in 2001 and 2002. *R.A.A. Morrall, P.G. Pearse, B. Carriere, D. Schmeling, and C. Pearse.*

123.*A medium and procedure for identifying *Fusarium graminearum* in cereal seed. *S. Pouleur, L. Couture, R. Clear, and A. Comeau.*

123.. *Fusarium spp. infection of wheat grains in the Czech Republic and its relation to bread-making quality parameters *L. Tvaruzek*

Session 3: New sources of, and breeding for, FHB resistance

124..Reaction of intergeneric and synthetic spring wheat lines to *Fusarium graminearum*. A. Breker, P.J. Hucl, and G. Hughes

124..*Critical control points for large-scale fusarium head blight field screening trials. A.L. Brûlé-Babel, W.G.D. Fernando, P. Hucl, G. Hughes, S. Fox, R. DePauw, M. Fernandez, J. Clarke, R. Knox, J. Gilbert, G. Humphreys and D. Brown

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125..*Two seeding date system in Fusarium head blight nurseries. G. Butler, H.D. Voldeng, S. Rioux, A.G. Xue, M.E. Savard, J. L. Gale, P.H. Matthew, C. Danjou, F.E. Sabo, Y. Chen, R. Stanley.

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134.*Enhancement of spring wheat FHB resistance through pyramiding of genes from different sources. *Wenguang Cao¹, George Fedak¹ and Jeannie² Gilbert*

134.*Progress in developing cultivars and germplasm with FHB resistance in Eastern Canada. *Comeau A, Dion Y, Rioux S, Butler G, Langevin F, Martin R.A., Nass H., Fedak G, Xue A, Voldeng H, Gilbert J, Dubuc J.P.*

135.*A progress report on the incorporation of Fusarium head blight resistance into Canadian wheat cultivars using an *in vitro* selection technique. *François Eudes, Sadash Sadasivaiah, Robert Graf, Sylvie Rioux, André Comeau, François Langevin and Nathalie Lanoie.*

135.*Impact of trichothecene on Fusarium head blight type II resistance in six cereal species. *François Eudes, François Langevin and André Comeau.*

136.* Androgenic ability of FHB resistant barley accessions. *G. Fonquerne, I. Clermont, L. Laroche, S. Marchand, and F.J. Belzile.*

136.* Tissue specific biochip microarray analysis of genes differentially expressed in *F. graminearum*-challenged wheat heads. *Saber Golkari, Suvira Prashar, Jeannie Gilbert and J. Douglas Procunier*

137..Isolation, characterization and physical mapping of differential clones from a SSH library for Fusarium Head Blight (FHB) resistance. *Fangpu Han, George Fedak, Therese Ouellet and Daryl Somers.*

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Session 5: Epidemiology

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141..*Inhibition of *Fusarium* sp. by hen egg white lysozyme. *Y. Gao, S. Krentz and S. Smith.*

142..*The effect of *Trichoderma harzianum* on the production of perithecia of *Gibberella zeae* on wheat residue. *S. Inch and J. Gilbert*

142..*A method to observe barrage zone formation in *Fusarium graminearum* (*Gibberella zeae*). *B.D. McCallum, A. Tekauz, and J. Gilbert*

143..*Molecular genetic variation, geographical differentiation and hybridisation in populations of *Fusarium graminearum* in Canada. *P. K. Mishra, J. P. Tewari, R. M. Clear and T. K. Turkington.*

143..Determination of *Fusarium graminearum* chemotype based on upstream sequences of the *Tri5* gene. *M.K. Tan, S. Simpfendorfer, D. Backhouse and G.M. Murray.*

144..Moisture retention of cereal spikes and fusarium head blight risk. *T. K. Turkington and K. Xi*

144..*Assessment of the environmental suitability of the western Prairie region of Canada for fusarium head blight caused by *Fusarium graminearum*. *T.K. Turkington, O.O. Olfert, R. Weiss, R.M. Clear, K. Xi, and J.P. Tewari.*

145..Fusarium* spp. associated with cereal corn and grass residue in Alberta.** *T. K. Turkington, A. Kuzyk, R. Dunn, D.D. Orr, K. Xi, R. Lange, D. Benard, K. Kumar, J. Calpas, S. Ali, and B. Chapman.*

145..*Histological study of stem infection in barley and wheat by *Fusarium graminearum*. K. Xi⁺ and T.K. Turkington.

146..Effect of harvesting time on incidence of seedborne *Fusarium* spp. in spring wheat in eastern Ontario. A.G. Xue, J. Frégeau-Reid, J. Rowsell, C. Babcock, G.J. Hoekstra, E. Sparry, Y. Chen, and F. Sabo.

146..Pathogenicity of *Fusarium* species causing head blight in barley. A.G. Xue, K.M. Ho, C. Babcock, Y. Chen, F. Sabo, and M. Kuc

Session 6: Disease management

148..*Crop rotation as a tool for fusarium head blight disease management. H. U. Ahmed, J. Gilbert, W. G. D. Fernando, A.L. Brûlé-Babel, and M.H. Entz.

148..*Effect of fungicides on fusarium head blight and leaf pathogens in winter wheat. A.L. Brûlé-Babel and W.G.D. Fernando

149..The impact of rotation and fertility on root rot pathogens of wheat in the Brown soil zone of western Canada. M.R. Fernandez, and R.P. Zentner

149..* Dry heat treatments to control *Fusarium graminearum* in infested wheat seed. J. Gilbert, A. Tekauz, S.M. Woods, and T.K. Turkington.

150..* Fungicide efficacy for control of FHB in large-scale wheat plots. A. Tekauz, B. Hellegards and M. Savard

150..* Eradication of *Fusarium graminearum* from infested barley seed by heat treatment. A. Tekauz, T.K. Turkington and J. Gilbert

Oral Presentation Abstracts and Summaries

Session 1: Overview and Toxins

History and status of Fusarium Head Blight. R.W. Stack. *Plant Pathology Dept., North Dakota State Univ., Fargo, ND 58105, USA.*

Fusarium Head Blight (FHB) was recognized as a fungal infection about 120 years ago. Within two decades after it was first described it had been found throughout eastern North America, strongly suggesting it had already been widespread but unrecognized. The causal fungus *Gibberella zeae* (Schw.)Petch was found in 1822 in the eastern USA, and there is evidence that it is indigenous throughout North America. Repeated severe epidemics of FHB occurred from 1915 onward through the 1920's, especially in 1919, when it was first reported in Canada. Again in the 1940's, FHB erupted in eastern Canada and the eastern and central USA. FHB was among the plant diseases investigated for potential biological warfare during World War II, but apparently it was too environmentally dependent for that purpose. After the war's end, the military research on FHB was allowed to be published, as one of the first (and arguably still one of the best) quantitative epidemiological studies of this disease.

FHB was less frequent during the 1950's, 1960's, and 1970's, but in the early 1980's there were large outbreaks of FHB in eastern Canada, in Manitoba, and in the US wheat states from North Dakota to Kansas. This revived interest in the disease. The concern was not just loss of yield or damage to physical properties of the grain, but the presence of mycotoxins. In 1981 CPS sponsored a symposium on FHB and mycotoxins. The principal papers of this symposium were published in the journal the next year. The epidemics of 1980-1982 led to increased interest and research in Canada, and at a few US institutions. The disastrous epidemics of the 1990's in spring wheat and barley as well as in the winter wheat regions brought everyone in North America to the recognition that the FHB problem must be solved.

Historical outbreaks of FHB can be traced to several causes: widespread planting of highly susceptible cultivars; presence of colonized residue from previous crops; presence of corn in rotation with small grains; and weather favorable for infection. At present, interest in solving the FHB problem is high, but can it be sustained? Tools such as changes in residue management or crop rotations are not likely to add much to FHB control and chemicals offer limited benefit and at often noneconomic cost, quite apart from concerns about environmental quality and food safety that such wide fungicide use might raise.

Partial resistance to FHB, which plant breeders and others sometimes call "tolerance", has been recognized since early days. As far back as 1915, H.K. Hayes in Minnesota reported the existence of lines that were less damaged by FHB. In more recent times, lines with partial resistance have been recognized: In Canada, 'Neepawa' and some of its offspring, particularly 'AC Barrie', possess useful levels of FHB resistance; among American spring wheats, 'Stoa' and more recently 'Parshall' (North Dakota), and

'Marshall' (Minnesota), were known to show less FHB than other lines. Likewise in barley, variation in response allowed R.G. Shands in the 1930's to select 'Chevron' as a line of substantial resistance. The occasional appearance of such lines is believed to be from the fortuitous recombination of many genes each with minor effects. Systematic attempts to breed directly for FHB resistance were given a low priority for most of the twentieth century and the extended programs needed to develop acceptable resistant cultivars were largely lacking.

Since the FHB outbreaks of the 1990's, accelerated breeding efforts promise FHB resistant cultivars acceptable for commercial production in the future. The FHB effort in Canada has been extensive and prolonged. Millions of dollars have been spent since 1980. In the USA, the USWBSI Scab initiative started in 1997 as a special appropriation and has provided some 23 million US dollars through 2003. About 40% of that amount was allocated directly to germplasm testing, breeding and variety development. Even before the start of that national program, several states, notably North Dakota and Minnesota were funding enhanced FHB research and breeding. Will all this investment pay off with varieties farmers will actually want to grow? The answer had better be "yes!" As one example of a success story, in North Dakota the moderately resistant cv 'Alsen' was released in 2000. By 2002 and 2003, Alsen was grown on 0.9 and 1.1 million ha, respectively -- evidence that farmers will grow FHB resistant wheats if such varieties are of acceptable yield and quality.

Not since the stem rust situation in the 1950's has such a concentration of effort been marshaled against one crop disease. One reason past efforts at FHB have failed, was because scarce resources were re-assigned before the job was done. The FHB crisis of the 1990's may have made sufficient impression on governmental and business leaders who hold the purse strings that the effort to solve FHB may be continued for long enough to get the job done.

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Why the tolerances for DON are going down. J. David Miller, *Professor & NSERC Research Chair, Fungal toxins and allergens, Carleton University, Ottawa, Ontario K1S 5B6*

In response to a recommendation from the FAO/WHO/UNEP conference on mycotoxins in Tunisia, the Joint Expert Committee on Food Additives & Contaminants (JECFA) held a special meeting on mycotoxins and produced re-evaluations of a number of toxins in 1999 AND 2001. For the first time the JECFA produced PMTDIs for fumonisin and deoxynivalenol. The JECFA is the committee of reference in the WTO for food additives and contaminants. The background documents on DON were written by a team of both European and North American experts which indicates that a transatlantic consensus was reached. Exposure assessments indicate excess exposure to DON in the GEMS "European" diet which includes the US and Canada. Uncertainties in the analysis and consumption patterns are taken into account in simulations suggesting that children especially are at some risk of consuming more than the PMTDI for DON.

The PMTDI is based on a chronic Canadian study in mice demonstrating feed refusal resulting from the impressive neurotoxicity of DON. Aside from excursions over the TDI on a population basis, there remains concern about the safety factor between mice and humans on the basis of this neurotoxicity. From work done in the former Canadian mycotoxin program, there is a strong understanding of what does not cause the neurotoxicity but the cause is not known. One of the key recommendations of an ILSI EU meeting on DON held in September was that further research on this question was needed. It is possible that the answer will change the safety factors in the TDI.

The Germans have the strictest provisional guidelines for DON in food products and the EU action levels are already in principle lower than those in the US and Canada. Because of an aggressive registration in Germany reinforced by the farmers union, Germany has made progress in reducing DON content in Canadian grain. This is probably not true for Canada and this question is important to the evaluation underway of DON in infant food by Health Canada.

Mycotoxin analysis, present and future. Marc Savard, *Eastern Cereal and Oilseed Research Centre, Ottawa, Agriculture and Agri-Food Canada, ON K1A 0C6 Canada*

Ever since the identification of mycotoxins in the 1960's, we have been challenged with finding faster and more accurate ways of measuring their concentration in various foodstuffs, but mainly in raw cereals. While the analysis of packaged foods can be done in the lab with expensive equipment, those who receive raw grain at elevators, for instance, need a quick and accurate way of measuring toxin concentrations and cannot afford to use LC/MS to get that information.

The methods used in the past have been based on chromatography or visual detection. In the present, the same methods are still used, but new methods use what we have learned from biological systems. The future may hold much of the same in addition to new developments in material sciences. These present and future methods will be discussed.

Consumer health and safety: monitoring food and feed. Helen Page and Lynne Underhill. *Canadian Food Inspection Agency, Winnipeg, MB R3C 1B2 and (L.U.) Canadian Food Inspection Agency, Ottawa, ON K1A 0Y9.*

The Canadian Food Inspection Agency and Health Canada are responsible for the regulatory oversight of feed and food safety in Canada. This presentation describes the roles of the two departments, current and future legislation regarding mycotoxins, current regulatory programs, and finally some challenges and opportunities in research and regulation. Foods and feeds are regulated according to the Food and Drugs Act and Regulations, and the Feeds Act and Regulations. Both pieces of legislation generally state that no person shall manufacture or sell a food or feed that is harmful. Health Canada's authority relates to sale of food, and CFIA has broader authority over manufacture, import or sale of feed. Health Canada sets standards for food safety and the CFIA delivers inspection programs to ensure compliance with the standards. Health Canada also conducts safety assessments for incidents of food contamination, and advises CFIA on risk to human health. The CFIA uses Health Canada's advice to determine the appropriate follow up action. For livestock feed, CFIA is the responsible authority for standard setting, safety assessment, and the delivery of inspection activities. In addition to feed inspection, the CFIA is responsible for pre-market approval of feed ingredients. Additives for mold prevention in feeds are regulated as feed ingredients by CFIA, while products intended to bind or adsorb mycotoxins are regulated as drugs by Health Canada. No mycotoxin-binding agents have been approved in Canada. Both the Feeds Act and Regulations and the Food and Drugs Act and Regulations contain some standards for contaminants, for example aflatoxins, but there are no specific regulatory maximum levels for *Fusarium* mycotoxins. In cases of *Fusarium* mycotoxin contamination of food or feed, the CFIA can take enforcement action, if the contamination is "harmful." Both the CFIA and Health Canada rely on guidelines based on scientific literature. The CFIA analyses about 300 feed samples for mycotoxins annually. Results of our monitoring show a high incidence of vomitoxin in grains in Central and Eastern Canada, albeit at generally low levels. Food sampling for mycotoxins is implemented when forecast information indicates the potential for increased mycotoxins. Health Canada does some survey work, and recent results from a survey of *Fusarium* mycotoxins in infant formula and breakfast cereals are being presented at this workshop. A survey of Zearalenone and Zearalenol in soy- and milk-based infant formula is planned. Ongoing challenges in the regulatory oversight of mycotoxins include the absence of regulatory maximums, the safety assessment of mixtures of mycotoxins, and the time lag between sampling and results. New challenges include the increased use of food and industrial by-products as livestock feeds, and the way to make best use of limited resources for mycotoxin inspection programs. One regulatory initiative underway at CFIA is to amend the Health of Animals Regulations to allow for regulatory control of animals, where animals are suspected or known to have been contaminated by toxic substances. This initiative will strengthen the CFIA's regulatory control over the entire food continuum.

Fusarium Head Blight – Alberta’s Initiative. Shaffeek Ali, *P.Ag. Alberta Agriculture, Food and Rural Development, Edmonton, AB.*

Abstract: Caused by the fungus, *Fusarium graminearum*, Fusarium Head Blight (FHB) is the most destructive fungal disease of barley and wheat in Canada. *Fusarium graminearum* infection greatly decreases yield, seed quality and produces mycotoxins (deoxynivalenol (DON) and zearalenone). The occurrence and high severity of FHB in cereal crops in western Manitoba and eastern Saskatchewan are reasons for concern. It is estimated that Manitoba presently loses \$50-\$100 million per year in wheat and barley due to yield loss, access to malt and hog feed markets, increased transportation costs associated with sourcing mycotoxin-free grain and other impacts on end-use processing. Further movement westward of this disease could be disastrous for the grain producing regions of Alberta.

Alberta is currently at trace levels of *Fusarium graminearum* and actions have been taken to prevent or at least slow the spread of this disease. If this pest is allowed to establish in the Province, there can be significant economic losses to Alberta’s cereal and animal feeding industries.

Through the leadership of the Fusarium Action Committee and stakeholder public consultations, the **Alberta *Fusarium graminearum* Management Plan** was established. The Plan was approved by the Deputy Premier/Minister of Agriculture, Food and Rural Development on August 30, 2002 and took effect October 1, 2002.

The **Alberta *Fusarium graminearum* Management Plan** defines the prevention and control strategies to reduce the risk of *Fusarium graminearum* becoming established in Alberta. It provides a set of guidelines for the management of this pest and yet able to maintain a viable agricultural industry.

TEAM MEMBERS

The team members (Fusarium Action Committee) comprised key stakeholder representatives from all levels of Government, private industry and growers. They bring to the team scientific and technical skills, industry needs and mediation and conflict resolution skills. The team members and the organizations they represent are listed below:

Shaffeek Ali, Chair
AAFRD
304, 7000 – 113 Street,
Edmonton, AB. T6H 5T6.
(780) 422 – 4909

Jim Anderson
Agricore United
P.O. Box 2700, Station M
505 2nd Street S.W.
Calgary, AB. T2P 2P5
(403) 216 - 0116

Ron Axelson
 Alberta Cattle Feeders Association
 200, 1603 – 10th Avenue S.W.
 Calgary, AB. T3C 0J7
 (403) 250 - 2509

Brenda Brindle
 Alberta Grain Commission
 305, 7000 – 113 Street
 Edmonton, AB. T6H 5T6
 (780) 427 – 3077

Audrey Gall
 Northern Sunrise County
 Box 480
 Nampa, AB. T0H 2R0
 (780) 322 – 3831

Andrew Horton
 AAFRD
 100, 7000 – 113 Street
 Edmonton, AB. T6H 5T6
 (780) 422 – 0153

Paul King
 Camrose County
 4728 – 41 Street
 Camrose, AB. T4V 1X7
 (780) 672 – 4765

Richard Nordstrom
 Alberta Barley Commission
 Box 237
 Viking, AB. T0B 4N0
 (780) 336 – 4875

Len Vogelaar
 Alberta Cattle Commission
 216, 6715 – 8th Street N.E.
 Calgary, AB. T2E 7H7

Terry Young
 Alberta Barley Commission
 237, 2116 – 27 Avenue N.E.
 Calgary, AB. T2E 7A6
 (403) 347 - 7664

Dee Ann Bernard
 Alberta Research Council
 P.O. Bag 4000
 Vegreville, AB. T9C 1T4
 (780) 632 – 8612

James Calpas
 AAFRD
 304, 7000 – 113 Street
 Edmonton, AB. T6H 5T6
 (780) 422 – 4911

Erin Holowach
 Alberta Grain Commission
 305, 7000 – 113 Street
 Edmonton, AB. T6H 5T6
 (780) 422 – 0878

Mark Kaun
 Cdn Seed Growers Assoc. – AB Branch
 Box 417
 Penhold, AB. T0M 1R0
 (403) 886 – 4562

Ralph Lange
 Alberta Research Council
 P.O. Bag 4000
 Vegreville, AB. T9C 1T4
 (780) 632 – 8226

Kelly Turkington
 Agriculture & Agri-Food Canada
 6000 C & E Trail
 Lacombe, AB. T4L 1W1
 (403) 782 – 8138

Bill Witbeck
 Alberta Seed Cleaning Plants Assoc.
 5030 50 Street
 Lacombe, AB. T3L 1W3

Session 2: Industry and Consumer Issues

Methods and issues regarding detection of Fusarium/DON in commercial grain in Canada. A.W. Schaafsma, M.E. Savard, and R.M. Clear, (AWS) Department of Plant Agriculture, Ridgetown College, University of Guelph, ON, N0P 2C0; (MES) Eastern Cereal and Oilseeds Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave., Ottawa, ON., K1A 0C6; (RMC) Grain Research Laboratory, Canadian Grain Commission, 1404-303 Main St., Winnipeg, MB, R3C 3G8

Abstract

Lower guidelines for deoxynivalenol (DON) in wheat are imminent, challenging the detection limits for those methods used in trade. More producers market wheat directly with DON limits specified in contracts. Pooled-grain uses the Fusarium damaged kernels (FDK) system for grading, but it does not accurately predict DON in specific lots. Of the detection methods reviewed for the grain trade, image analysis (IA) and near infrared (NIR) are the most attractive because they are non-destructive, simple, and have multiple uses; however adoption may be restricted by capital cost and their limit of detection. Both methods can estimate DON and other Fusarium parameters, but neither is commercially proven. The problem with NIR is that it is not a measurement of DON, but rather other factors associated with the presence of DON, but which are not exclusive to the presence of DON, such as kernel hardness, colour, etc. IA has the advantage of looking at the factor that it is designed to measure, ie FDK (good for grading), but also suffers from the inaccuracy of FDK to estimate DON levels. Immuno-methods specific to DON and its metabolites are sensitive to within proposed DON guidelines, but require laboratory facilities, more time and technical skill to operate, and may not account for other Fusarium factors that affect quality. They require low capital investment, are reasonably priced, and widely accepted in the trade. Immuno-methods that target the fungus can be specific to genus or species, and while qualitative tests can identify the presence of the fungus, they cannot be used to estimate toxin presence or levels. Quantitative methods are better able to estimate toxin levels and quality effects resulting from the growth of *Fusarium* spp. At least 2 companies, Adgen and D² Biotechnologies, have commercially available qualitative and quantitative versions of their DNA-based kits for detecting *F. graminearum*. Methods chosen will be determined by DON guidelines, here and abroad, and the needs of each sector of the grain industry. More than one grading method may be appropriate.

Introduction

Epidemics of fusarium head blight (FHB) caused by *Fusarium graminearum* Schwabe, in wheat (*Triticum aestivum* L.) are becoming more frequent around the world (Buerstmayr 2000; Francl et al. 1999; Gilbert and Tekauz 1995; and Schaafsma et al. 2001). The disease reduces grain yield and quality, often causing grain to be unsuitable for human consumption because of the production of the trichothecene toxin deoxynivalenol (DON) (Dexter et al. 1996; 1997; Young and Fulcher 1984). In some regions of Europe, other species of *Fusarium*, such as *F. culmorum*, (also a DON

producer), and *F. avenaceum* (a producer of moniliformin), are important pathogens causing FHB (Magan et al. 2002; Golinski et al. 1999). In Canada these species are of minor importance for FHB but can produce an array of mycotoxins often more potent than DON. However, world wide, the most troublesome epidemics resulting in DON-contaminated wheat, are caused by *F. graminearum*.

The World Health Organization regards DON as a teratogen, a neurotoxin, an embryotoxin, and an immuno-suppressant (WHO 2001, FAO/WHO 2002, Mirocha 1999). Population sectors at the greatest risk include woman of child bearing age and young children that obtain a high proportion of their diet from wheat. Canada's guidelines for DON in raw soft wheat are 2 and 1 ppm, respectively, for wheat destined for the normal population and for wheat destined for baby food production (Scott et al 1985). These guidelines are currently under review in Health Canada and will likely extend to spring wheat. In the USA the Food and Drug Administration has issued an advisory of 1 ppm DON in finished wheat products such as flour, bran and germ destined for human consumption (Cheesemore 1993). There is pressure in developed nations to lower these guidelines in view of recent toxicological findings (FAO/WHO 2002).

In Canada, the visible effect of FHB on the seed of small grain cereals is a degrading factor established by the Canadian Grain Commission via the *Official Grain Grading Guide* (Canadian Grain Commission, 2003). Here it states: *Fusarium*-damaged wheat is typically characterized by thin or shrunken, chalk-like kernels. *Fusarium*-damaged kernels (FDK) have a white or pinkish fibrous growth, which may be visible only under a magnifying lens. The minimum portion of grain for analysis is 10 g and the optimum portion as well as the portion used in grading for export is 100 g. The FDK determination procedure involves dividing the grain sample using a Boerner-type divider. The kernels showing any evidence of *Fusarium* damage, including any kernels with chalk-like appearance, are separated. A 10-power magnifying lens is used to confirm evidence of a white or pinkish mould or fibrous growth. Only those kernels with the white or pinkish mould or fibrous growth are to be used in the determination; and the determination is given as a percentage w/w. In Canadian Western Red Spring Wheat) 0.25, 1.0, 2.0 and 5.0 % FDK are allowed in the No. 1, 2, 3 and CW Feed classes, respectively. In contrast, all Eastern Canadian wheat classes allow only 1.0 % FDK in grades 1 through 3, and 5.0% in CE Feed wheat.

Aside from the direct toxicological effects of DON on human health, other factors associated with *F. graminearum* can affect the quality of wheat grain and processed wheat products. *Fusarium* spp. can reduce test and thousand-kernel weight in commercial wheat grain (Jones and Mirocha 1999). The presence of FDK adversely affected flour refinement (ash and color), glutenin content, dough handling quality and loaf volume in hard red spring wheat (Dexter et al. 1996). The reduction in bread loaf volume was due to the presence of proteolytic enzymes, associated with FDK, which decreased dough consistency and resistance to extension (Nightingale et al 1999). Dexter et al. (1997) noted that the presence of as little as 2% FDK had a negative effect on pasta color in pasta made from lower grade durum wheat confirming that strict tolerances for FDK in premium durum wheat are warranted (No.1 CWAD (0.25%) and No.2 CWAD (0.5%)). These quality issues are reported for hard spring wheat, but similar problems have not been reported for products made from soft wheat.

Functionally, the regulation of FDK in western, hard spring wheat is more directed toward processing quality, while in eastern soft wheat, because of the DON guidelines from Health Canada that are in effect for soft wheat, the emphasis regarding *Fusarium* is more on the content of DON. The emphasis on DON is expected to expand to hard wheat classes along with the anticipated extension of DON guidelines to these classes.

Most of the winter wheat produced in Eastern Canada is destined for domestic consumption or local export to the US. The trade of soft winter wheat in the east is tied closely to the trade occurring directly across the border. The *Fusarium* problem is managed in the USA strictly by DON content as enforced by the USA limit of 1 ppm in finished wheat products (Cheesemore 1993), and by the trade limit of 5 ppm set by the Chicago Board of Trade (2003). Market forces pass these tolerances down the processing chain to grain suppliers. In fact, some industry sectors have tightened the tolerances, such as 0.5 ppm DON in soft wheat grain for the North American breakfast cereal market (Schaafsma 2002); and tolerances in international wheat tenders and contracts ranging from 0.03 to 3 ppm (Dexter and Nowicki 2003).

Because FDK and DON are both important to trade, and wheat grading is based on FDK, it is important to understand the relationship between FDK and DON. This relationship was reviewed by Nowicki (2001) and then discussed in more detail by Dexter and Nowicki (2003) who state that the DON to FDK relationship is not straightforward and is easily obscured. There is no doubt that these measurements are positively correlated, with correlations ranging from $r = 0.28$ to 0.99 with a mean of 0.73 . Dexter and Nowicki (2003) suggest that using a ratio of DON to FDK (DF-ratio) is a more meaningful way to describe the relationship resulting in less variability; and that these ratios may vary amongst wheat classes, and within wheat classes from year to year.

Dexter and Nowicki (2003) report several factors that lead to variations in correlation. One is the tendency for the DF-ratio to be high at FDK levels less than 0.5 %, and to decrease as the percentage FDK increases. Some samples may contain FDK but be negative for DON because the *Fusarium* species is a non-DON producer. This occurs primarily in western Canadian durum wheat when FHB is caused by *F. avenaceum*. *F. avenaceum* is not an important cause of FHB in other Canadian wheat classes, likely due to the greater level of resistance to FHB in the varieties composing the other wheat classes and to the lower pathogenicity of this species compared to *F. graminearum* and *F. culmorum*. Some kernels test positive for DON but show no sign of *Fusarium* damage and therefore are not included in the FDK sample. Some asymptomatic or atypical kernels can have relatively high levels of DON (5 ppm in asymptomatic kernels, Sinha and Savard, 1997) and because these kernels are more plump and dense they would contribute more to the FDK percentage, than do the lighter, chalky-white kernels that contain most of the toxin in a sample.

In the end, Dexter and Nowicki (2003) concluded that the relationship between DON and FDK was insufficiently robust to precisely predict the level of DON in an individual sample, but was useful and sufficiently reliable to manage the risk of DON contamination at country elevators to minimize the DON levels in bulked samples at terminal elevators. The use of FDK may continue to be appropriate when working with pooled grain lots, but may become inappropriate when grain is direct-marketed outside of

the marketing board to end users as is the growing practice in Ontario (Ontario Wheat Producers Marketing Board 2003).

Dexter and Nowicki (2003) also stated that the definition of FDK was critical to the DF-ratio and applying the definition requires training and patience. A typical FDK determination by an official trained grain inspector takes from 10 to 20 min per sample (Jim Lowe Canada Grain Commission). At the elevator, when trucks are lined up, grain graders may not take the required effort to make the determination. In Ontario, in 2000, the University of Guelph presented the same 0.5-kg harvest sample of soft red winter wheat to a total of eleven grain elevators across three counties, representing five different companies. The University determined the FDK to be 0.7 % and the Canadian Grain Commission in Chatham graded the sample at 0.9 % FDK. However the determinations by the grain elevators ranged from 0.4 to 1.8 % with a mean of 1.1 %. There were four results less than 1% and seven greater than 1 %. The variation in the interpretation of FDK, led to frustration and lack of trust amongst producers toward grain handlers.

In light of changing marketing strategies and opportunities, the tightening restrictions on DON in wheat, and the inherent differences between Eastern and Western grain handling and marketing, the grading practices for *Fusarium* and/or DON need to be discussed. This paper summarizes the several DON/FDK grading tools available to the wheat industry and suggests which of these may be most appropriate for each sector. DON/*Fusarium* detection methods can be divided into two main categories, those that are destructive to the grain samples and those that are non-destructive.

1. Destructive/Invasive methods of detection

The majority of destructive methods require some level of wet chemistry, and each method varies in the requirements for technical investment. Several reviews of wet analytical methods for DON have been published (Richard et al. 1993; Gilbert 1995; Gilbert 2000 and Mirocha et al. 2003). Of the methods available today, gas chromatography (GC) and high performance liquid chromatography (HPLC) (Schothorst and Jekel 2001; Yoshizawa 2001; and Walker and Meier 1998), often coupled to mass spectrometry (Plattner 1999) are the most accurate but the least appropriate for analyzing large numbers of samples. These highly technical methods give reliable and precise results but can take 24 hr for 100 samples (Schaafsma et al 1998). The grain handling industry requires high throughput, limited capital investment, short turn-around time and reasonable cost per sample, which these methods do not offer.

The remaining destructive wet methods include thin-layer chromatography (TLC), (Yoshizawa 2001; Sherma 2000; and Trucksess et al. 1987); solution fluorometric detection (Malone 2001; and Malone et al. 1998), polymerase chain reaction (PCR) for *Fusarium* DNA (Knoll et al 2002; Schnerr et al 2001; Niessen and Vogel 1998; Koopman et al 1997; Nicholson et al. 1998), and enzyme-linked immunosuppressive assays (ELISA) (Maragos and Plattner 2002). These will be discussed and assessed for utility in the grain-handling sector. Some other methods that are not used at this time but show promise will also be discussed.

1.1. Methods for detecting DON

1.1 a. Thin layer chromatography

Thin-layer chromatography requires careful sample preparation and extraction similar to that for more technical methods of chromatography (Yoshizawa, 2001). The time required to run a batch of 100 samples, is about 16 hr (Schaafsma et al 1998), which is unacceptable when dealing with trucks waiting to be unloaded. However, TLC is about half the cost of the more technical wet methods, when capital depreciation is included in the calculation, and about 25 % of the cost of ELISA kits. The advantages for TLC are its low cost in equipment and materials, the limit of detection of 0.2 ppm for DON, and the wide range over which DON concentration can be determined (Schaafsma et al 1998). The disadvantages are the slow turn-around time, the requirement for skilled technicians, and the requirement for a wet laboratory. TLC would be inappropriate in a grain elevator setting, but may be useful in a large processing facility that does routine testing involving multiple samples. Economies of scale impact cost per sample with this method.

1.1 b. Fluorometric Methods

Malone et al. (1998) describe a one-step extraction, clean up and fluorometric analysis of DON which may have utility in grain grading facilities handling large numbers of samples, and this method is reviewed in detail by Malone (2001). This method can quantify DON at concentrations from 0.5 to 50 ppm without dilution. Individual analyses are conducted in less than 30 min, and 24 samples analyzed in 2 hr. This method requires access to a low level wet laboratory, with grinder, blender, evaporator and fluorometer. Its advantages include: the wide range of DON-determination without dilution; the flexibility to run individual samples and multiple samples without significant impact on the cost per sample; less skill required and less chance for error when compared with chromatographic methods; and the reasonable cost per sample. The disadvantages include: the requirement for some moderately-costly specialized equipment and laboratory space, the limit of detection of 0.5 ppm and the half hour time it takes to run a sample, although this time includes every step from grain sampling to result.

1.1 c. Enzyme-linked immunosorbent assay (ELISA) for DON.

ELISA tests are one of the most widely-used method in the grain industry around the world to screen for and quantify DON (Maragos and Plattner 2002). These tests are based on reactions where the toxin and a toxin-enzyme conjugate compete for reaction sites on monoclonal antibodies. The enzymes produce a colour when exposed to certain substrates and the concentration of toxin is inversely proportional to the amount of colour (Sinha and Savard 1996). This method can be quite specific to a target compound, such as DON, but cross-reaction with some DON-metabolites can occur depending on the antibody used (Maragos and Plattner 2002).

Several commercial ELISA kits are available (Diagnostix 2003; Neogen 2003; Romer Labs Inc. 2003). There are two types of ELISA kits. Some will only report that the DON concentration is above a set control concentration, while others use a series of standards to produce a standard curve from which the amount of DON can be quantified

more accurately. Quantification of DON by ELISA is limited by the design of each kit. Some kits have a range of detection of 0.5 to 5.0, 0.5 to 6.0 ppm, or 0.2 to 2.5 ppm. Samples containing DON outside these limits require concentration or dilution prior to retesting. Most manufacturers of these semi-quantitative kits report a reading time of 20 minutes, but neglect to include the sample grinding and extraction steps in the calculation. Running these tests typically takes a minimum of 30 minutes although once samples have been ground and extracted, a number of them can be analyzed at the same time. Moderate skill, and access to a wet laboratory with relatively low cost equipment are also required. The equipment needed includes grinder, blender, multi and single pipettors, and a microwell reader for more accurate determinations. The advantages of ELISA are: their sensitivity and specificity to DON and closely related metabolites, the relatively low investment in equipment required, accessibility and reasonable cost of kits for large numbers of tests, and the wide acceptance of this technology for this and other mycotoxins. The disadvantages of semi-quantitative ELISA include: the requirement in space, equipment and time for sample preparation and extraction; the inflexibility and inefficiency of running single samples; cross reactivity with analogues of DON that can result in overestimation of DON levels; and the moderate level of skill and steps required to run the tests. With changes in the health guidelines for DON the limit of detection may also become a problem, however Yang et al. (2003) report that ELISA is suitable to analyze baby food and cereal samples in accordance with the tightening European legislative limits for DON.

1.1 d. Lateral flow assays

Lateral flow assays are rapid immunologically-based assays where a drop of extract is allowed to flow laterally on a porous layer in conjunction with appropriate antibodies bound to microscopic gold particles. The target compound binds to the antibodies, which then cannot bind to a line of bound target. If the extract does not contain a sufficient amount of the target compound, the antibodies will reach the bound target and the gold particles will accumulate on this line to produce a red line.

De Saeger and Van Peteghem (1996) reported a dipstick immunoassay for T-2 toxin in wheat at a detection limit of 0.25 ng/mL, but it took about 45 min to obtain a result. More recently, the testing time for lateral flow strip assays have been cut to under 15 min (Orr 2002), including grinding and extraction steps.

1.1 e. Surface Plasmon Resonance

Surface plasmon resonance also makes use of antibodies. In this case, plane polarized light entering a glass prism is reflected off a gold or silver film. At a certain angle, there is total internal reflection. A component of the light called the evanescent wave will penetrate the gold film. If large molecules can be attached to this gold film, they will affect this evanescent wave and change the optimum angle of refraction of the light. To use this method to detect mycotoxins, one attaches some of the toxin to the metal film and then elicits a competition between this bound toxin and free toxin in a sample for antibodies in the liquid phase flowing over the gold film (Mullett et al. 2000). The effect on the angle of refraction of the light through the prism will be proportional to the amount of antibody getting attached to the bound toxin.

This method has been used for fumonisins (Mullett et al., 1998) and deoxynivalenol (Tudos et al, 2003; Schnerr et al, 2002), but the equipment cost and level of skill required for this method are prohibitive at this time.

1.1 f. Fluorescence polarization

With fluorescence polarization analysis of toxins, a tracer consists of the toxin bound to a fluorescent molecule instead of an enzyme as in ELISA. Instead of measuring the intensity of the fluorescence, the speed of rotation of the fluorophore is measured through its polarization. If the fluorophore is attached to a large molecule, the speed of rotation is reduced and the polarization increased. Therefore, by eliciting a competition between DON, for instance, in a sample and DON bound to fluorescein for an antibody, as the antibody attaches to the fluorophore conjugate, the rotation of the fluorophore is reduced and polarization increased (Maragos et al., 2002). The advantage of this method is that it can be used even in opaque solutions, and, as with ELISA, requires no clean-up. While good results have been obtained for analysis of wheat samples, corn samples did not yield as good a correlation with HPLC analysis (Maragos and Plattner, 2002).

1.1 g. Molecularly Imprinted Polymers (MIP)

Molecularly imprinted polymers are essentially artificial antibodies. By careful selection of the monomers and using specific polymerization techniques in the presence of the target molecule, these polymers are generated with cavities matching the target's shape. Removal of the target yields a polymer that can, in theory, be used to retain this target from a solution (Sellergen, 2003). This method is still under development and the results are currently unsatisfactory. While very large amounts of MIP can be obtained in a very short time compared to antibodies, the selectivity has so far only allowed a retardation of the movement of the targets. Therefore, their best use has been as alternative SPE (solid phase extraction) sorbents. Such MIPs have been obtained for DON (Weiss et al., 2003) and used as HPLC solid phases to test their ability to retain DON. Although DON was found to have a longer retention time than acetone, fusarenone-X and 3-acetyl-DON, the peak shape was poor. This method does show promise but will require more work in the selection of monomers, but especially in the polymerization techniques used to generate MIPs.

1.1 h. Designed membrane channels

There are some novel analytical techniques that show promise but have yet to be used for DON, or even mycotoxins. One of these, designed membrane channels, is based on the properties of certain membrane proteins to produce certain effects when encountering target molecules. These proteins, such as hemolysin, act as a channel through membranes, which upon contact with ions or small molecules will open, close or generate a small current. It is now possible to chemically alter these proteins to incorporate receptors inside their central cavity (Bayley, 1999). These receptors can then react with their target molecule and close off the channel. If for instance, the channel allows ions through, the flow of ions can be stopped and a signal generated. These channel proteins can be modified in many ways, and there are many ways in which they can generate a signal. Somebody will undoubtedly soon apply this methodology to toxins.

1.2 Detecting and quantifying *Fusarium* fungi

Focusing on detecting the fungus rather than DON takes into account the non-mycotoxin quality factors related to the presence of the fungus. There are two approaches to the quantification of *Fusarium* in grain samples. The first approach is destructive and involves the detection of proteins, either immunochemically as reviewed recently by Li et al (2000), or by DNA fingerprinting methods (PCR) which extract, amplify and detect DNA sequences specific to the species or genus of the *Fusarium* of interest (Knoll et al 2002). The second approach is non-destructive and detects fungal metabolites, other than mycotoxins, that are mainly volatile components. This approach will be discussed later.

1.2 a. Immunochemical methods

Immunochemical detection operates under the same principle as that described above, but with antibodies to fungal proteins instead of DON or other toxins (Suzhen et al 2000). These proteins can be unique to a species or genus of the fungus. The amount of specific protein would be proportional to the amount of fungal biomass, and this in turn may be correlated to the amount of factor or toxin. Gan et al (1997) demonstrated the ability to generate immunoassays to detect *Fusarium* spp. using chicken antibodies. There are two options for the delivery of these protein immunoassay systems (Stave 2002): the first is the ELISA test, which allows better quantification; and the second is the lateral flow strip test which is qualitative at a prescribed level of detection. These protein-based tests are sensitive at 2 ppb for the Cry9C Starlink protein (Diaz et al 2002; and Trucksess 2001) and are now used commonly in the corn and soybean trade (Orr 2002). The dip test technology is widely used in other sectors and is inexpensive (human pregnancy test retails at US \$3.50 per test (www.mistertest.com)).

1.2 b. DNA-PCR methods

PCR techniques can characterize strains of *Fusarium graminearum* (Ouellet and Seifert, 1993). Although previously *F. culmorum* and *F. graminearum* were distinguishable in co-cultures but not in plant tissue using PCR (Koopman et al 1997), commercial kits are now available that will distinguish several species of *Fusarium*, including *F. graminearum*, *F. culmorum*, and *F. avenaceum*, from seed and other plant tissue (Adgen, www.adgen.co.uk). Trichothecene-producing *Fusarium* spp. can be detected by PCR assays and the intensity of signals are well correlated to DON content in wheat samples (Niessen and Vogel 1998). *Fusarium*- specific PCR assays are routinely used in pathology experiments (Nicholson et al 1998; and Doohan et al 1999).

Typically, PCR techniques require DNA extraction and preparation steps, which take up to 6 hr to complete. The visualization step using gel electrophoresis adds another 2-3 hr to the process. Using the LightCyclerTM-PCR systems reduces the DNA preparation time by 1.5 hrs (Schnerr et al 2001). The use of DNA Detection Test StripsTM eliminates the need for electrophoresis and cuts the visualization step down to 20 min (Klepp 2000 and Knoll et al 2002b). Knoll et al (2002a) shaved the DNA extraction step to 5 min using sonication and a commercial extraction kit. Recently, a PCR-based commercial kit for the detection of single and multiple species of *Fusarium* (Adgen) and a DNA-based ELISA kit (D² Biotechnologies) have come onto the market. These kits are available in qualitative and quantitative versions, the PCR-based kit adding an ELISA final stage for quantitation.

Even with all the time saving new steps available, PCR reactions require skilled operators, in clean laboratories, with relatively unique and expensive equipment, and a minimum of several hours to complete. PCR reactions are not likely to find their way into industrial grain grading.

1.3 Mycotoxin Bioassays

Bioassays have proven very useful in the past to detect toxins, especially unknown toxins or to test the toxicity of toxins. Bioassays have been performed with whole organisms as well as cell cultures. For the detection of toxins, the organisms used have ranged from yeasts (Koshinsky and Khachatourians, 1992) to rats (Gelderblom et al., 1988). Bioassays for the purpose of quantifying toxins are limited to microorganisms since the results depend on the survival or death of large populations of animals that require very small doses. However, bioassays are usually impractical for commercial use as they require facilities to store and use organisms and a minimum of 24 hr to develop (Madhyastha 1994). Furthermore it is doubtful that they can detect DON at the limits currently proposed.

2. Non-Destructive/Non-Invasive methods of detection

Non-invasive or non-destructive methods are attractive because they avoid the time-intensive steps of sample preparation and/or extraction. The capital and labour investment for the sample handling and preparation step is generally expected to be low. However, the determination step, while quick, may involve costly capital investment. Most commercial applications of non-invasive methods seek the “desk-top”, “black-box” approach, removing the need for highly skilled operators. Three technologies for detecting DON/*Fusarium* have emerged: the electronic nose (Jelen et al 2003), near infrared spectroscopy (Pettersen and Aberg 2003), and image analysis of whole grain (Kokko et 1999).

2.1 Electronic nose

Electronic noses mimic the human olfactory system by sampling the headspace of a whole-grain sample with an array of non-specific sensors and then comparing the pattern of their responses to the headspace volatiles to differentiate samples (Dickinson et al., 1998; Jelen et al. 2003). These sensors can be made of silica or organic polymers and function in a number of ways, such as a frequency shift in a resonating crystal, a change in optical absorption of a dye, or a change in the electrical resistance of a polymer (Matzger et al. 2000).

One application for barley containing DON takes about 20 min including a two minute heating step, a 30 second aeration step a 90 second sensing step, and finally a 15 min sensor regeneration step between samples (Olsson et al 2002). More research in this field is slowly reducing the analysis time as the sensors are made to adsorb and desorb volatiles faster.

The equipment costs in the order of US\$10,000, and requires moderate technical training to run. The electronic nose can also be used to grade several odour-related factors in grains. The limited number of applications for the electronic nose, and the time factor hinder the adoption of the electronic nose at the grain elevator at this time. Miniaturization and new developments in polymer synthesis may eventually solve these

problems, but, for the time being, it may still be a useful and cost effective technology for millers and malsters for lot screening by eliminating several steps in the analytical process.

2.2 Near Infrared spectrometry

Infrared spectroscopy (IR) can be coupled with gas chromatography to give a stable spectral pattern to allow characterization and quantification of several mycotoxins (Young and Games 1994; Mossoba et al 1996). However this application is highly technical and would be inappropriate in an industrial setting. Interest in detecting fungal contaminants in whole grain through near infrared spectroscopy (NIR) alone, however, continues to grow since its modest beginnings a decade ago (Greene et al 1992). NIR identified single FDK as accurately as official grain inspectors (Dowell et al 1999). These single kernels contained at least 120 ppm DON and the standard error of prediction was around 100 ppm. Dowell et al (2002) were able to distinguish single corn kernels containing over 100 ppm fumonisin from those containing less than 10 ppm. Single kernel analysis slows sampling time down (Dowell and Maghirang 2002) and requires additional costly technology to run. Ruan et al. (2002) reported a limit of detection of 2 ppm in bulk barley samples and a good correlation between the NIR and GC/mass methods. Kos et al. (2002) reported no incorrect predictions of DON at 2.59 ppm, but 20 % false positive, and 18% false negative results at 1.17 ppm DON in maize. By grinding the samples and recording the spectra under standard pressure Kos et al. (2003) could detect DON as low as 0.3 ppm. In wheat, Pettersson and Aberg (2003) reported a limit of detection near 0.4 ppm in whole kernel samples. These results are above the proposed Netherland guidelines of 0.12 ppm for DON in whole wheat (Pieters et al 1999). NIR appears to predict DON as well or better than the current FDK system, although field trials with NIR may not be as favourable since the factors that NIR measures are not exclusive to the presence of DON. The price of NIR equipment is dropping and with improved calibration curves, this system may be a viable tool for screening samples to the same accuracy as currently afforded by the FDK grading system. NIR appears to be equally promising in barley (Arganosa et al 2003).

2.3 Image analysis

Imaging and computer technology advanced to the point in the mid 1980's that researchers began proposing discriminating between wheat classes and varieties by image analysis (Zayas et al 1986, Symons and Fulcher 1988a and 1988b; Neman et al. 1989a and 1989b). Shatadal et al. (1995a and 1995b) developed software to allow individual grain kernels to be recognized and analyzed within a sample of touching and overlapping kernels. Majumdar and Jayas proposed models to classify cereal grains by morphology (2000a), color (2000b), texture (2000c) and the combination of the three (2000d).

Luo et al. (1999 a and b) used machine vision and the non-parametric approach to differentiate between healthy and damaged wheat kernels at accuracies greater than 90%. Kokko et al. (1999) reported on the detection of FDK using image analysis and neural networks on individual seeds. Ruan et al. (2001) reported a correlation coefficient of 0.96 between estimated and actual FDK using machine vision and neural networks and a sample layer of kernels. Delwiche and Moon (2001) with the USDA in Beltsville are working on imaging to detect scab in wheat. Wiwart et al. (2001) reported a strong

relationship between color components, hue, saturation and intensity, and kernels per spike or one thousand kernel weights, suggesting the potential for image classification of *Fusarium* damage in triticale. Commercialization of machine vision for FDK classification is imminent through an image analysis system called Acurum™ (www.acurum.com) through DuPont Canada Inc., which samples a large number of individual kernels within seconds. Hinz Technologies markets a flatbed vision machine under the name of TrueGrade (www.hinztechnologies.com/truegrade.html), which is currently available to grade lentils, hay and noodles. This machine scans a planar bed of material rather than individual units and there is some interest to expand to FDK in wheat. Both machines are unproven to detect FDK precisely and accurately under commercial conditions. A better measurement of FDK levels in a sample will result in more accurate grading, but will still allow for only a surrogate estimate of the DON levels.

3.0 Conclusions and Recommendations

Regardless of the detection method chosen, the result is only as good as the sample taken. Gilbert (2000) listed sampling as a critical first step in analyzing for mycotoxins. Johansson et al. (2000) showed that the percentage of total variance for analysis of aflatoxins by HPLC for sampling, sample preparation and analysis was 77.8, 20.5 and 1.7%, respectively. Hart and Schabenberger (1998) clearly showed that DON is not uniformly distributed in truckloads of wheat. Care must be taken in all sampling steps to achieve a representative sample (Whitaker 2001; and Whitaker et al. 2000).

The selection of a *Fusarium*/DON detection method to steer grain handling depends on a number of factors. The need for a lower limit of detection is increasing due to changing health guidelines. The grain industry is averse to capital investment if the equipment is limited in its scope of utility. Ease of use, speed of result and low technical requirement are critical to the grain industry. Equally important are the degree of precision and accuracy afforded by each method. Using these criteria, four technologies arise as potential surrogates for FDK .

Both image analysis and NIR methods potentially have multiple uses in the grain handling and/or processing industry (Dexter and Marchylo 2000). Both are non-destructive, simple to operate, offer quick sample results and require no laboratory facilities. Their potential limitations are the capital cost for the equipment and the limit of detection for either DON or FDK. Both methods can be calibrated to take into account DON and other *Fusarium* quality parameters, and neither has been validated under commercial conditions.

Immunological methods that detect DON, whether dip tests for screening or ELISA tests for semi-quantification, are more sensitive than the image and NIR methods. They require some laboratory facility and technical skill for sample preparation and determination, and the results can take up to 40 minutes to obtain. They are specific to DON-related compounds but may not account for non-DON-related *Fusarium* factors. While these factors are not regulated they are important in the trade of grain. Their advantage lies in the relatively low capital investment required and per sample cost incurred, if economies of scale can be achieved; and their sensitivity within the proposed guidelines. They also are widely accepted in the grain trade.

Immunological methods that detect *Fusarium* fungi have all the advantages and disadvantages of those that detect DON. They can be highly specific to a single species of *Fusarium* or broadly specific to several *Fusarium* species. Their advantage is that they can account for all *Fusarium*-related toxins and non-toxin-related *Fusarium* factors that affect processing quality. No commercial product is currently available, neither has this method been commercially validated. Ultimately the choice of method will be largely determined by the DON guidelines set in Canada and in markets abroad and the needs of each sector of the grain industry. One size may not fit all.

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Current impact and emerging issues for *Fusarium* head blight in the malting and brewing industries. Richard W. Joy IV; *Rahr Malting Canada Ltd*

Barley in Canada is grown mostly throughout the prairie provinces with annual production averages in the area of 12 to 14 million metric tonnes. Of this production, the majority is brought into the feed stream (close to 80+%) with a further 15% annually being selected for malting quality. Approximately 50% of selected malting barley remains in Canada for use by the 4 major malting companies whereas the remaining 50% is exported to be malted overseas.

The importance of the yearly Canadian barley crop can easily be illustrated by events precipitated by the poor 2002 crop. Decreased volumes led to feed shortages bringing about increased prices for feed barley and left malting barley short due to being sold for feed. Prices also spiked due to other more commonly used cereals being in short supply, hence, an overall supply deficit in all sectors of barley related industries. Further, this was the first year that barley was imported into Canada for malting purposes. In the final analysis, all barley related sectors lost money due to the poor quality and quantity of the 2002 crop.

As with all cereals, barley too is susceptible to a number of diseases which can have long reaching and potentially disastrous effects, specifically, decreased volumes, grading issues, mycotoxin contamination, increased costs to all sectors..... *Fusarium* Head Blight (FHB), in particular, is the continuing incipient disease that is gradually creeping across the prairies. Its increasing presence has caused many barley related industries (producers, seed companies, grain handlers, malt companies, brewers.....) hardship, headaches and not to mention, money.

The impact of FHB on the malting and brewing industries is wide spread and varied. It ranges from small variations in quality control to large investments in intra and inter company infrastructure for the malting industry. For the brewing industry it may mean a paradigm shift which includes the eventual acceptance of different malt specifications. These events, however, will be determined by how FHB and its progression is handled in the future and by the many players currently involved. This talk will touch on issues as they relate to the malting and brewing industries such as impact, perception of severity, current and emerging problems, management strategies and potential progress and challenges into the future.

Current impact and emerging issues of FHB in grain handling from farm to delivery. Al Morris, *Agricore United, Winnipeg MB.*

The profile of the Western Canadian Grain Industry has changed over the past ten years. As of August 2003 there is only 382 elevators with 5.101 ('000mt) storage compared to just under 1400 elevators with a storage capacity of 7.000 ('000mt) in 1993. Average storage capacity, per elevator since 1993 has gone from 4900 mt. to 13500 mt. High throughput elevators of >10000 mt. increased from 60 to 165 presently. There is now increased competition. In 1994 the six major grain companies (SWP, Alberta Wheat Pool, UGG, Pioneer, Cargill, Manitoba Pool) had 91 % of the country elevation storage capacity in Western Canada. In 2003 the 6 major companies (Agricore United, SWP, Pioneer, Cargill, Paterson, Louis Dreyfus) have 78 % storage capacity. The number of licensed company buyer farmer groups has increased from 19 to 36 over the past decade with 22% of the country storage. Terminal facilities in 1994 were 9 with storage of 1.700 ('000mt). Today there 8 port terminals with storage capacity of 1.300('000mt). The major reason for the large rationalization of facilities was to take advantage of shipping multi car blocks. Over 80% of the business moves in 25 car blocks or higher.

Three major trends are driving the whole grain system.

- Concern for food safety and “quality” with growing focus on “traceability” and the entire production and handling process.
- Technology
- Competition

Due to the huge rationalization in the country the key to handling Fusarium Head Blight (FHB) through the grain handling system has changed from a push to the pull system. An intensive training process for producers on proper sampling to obtain a representative sample to evaluate all customer specifications is critical to manage a grain companies' liability.

Once all hot spots are flagged and severity of the problem is assessed, a logistical game plan needs to be developed to maximize blending in the country or terminal. The control process of monitoring all producer deliveries to be binned according to quality. Once all in store samples are tested and evaluated a bin blending process is done to ensure customer specifications are met. Depending on commodity, CGC could be onsite to grade at load to reduce a companies' liability on shipments directly to a customer.

The past 10 years the country handling system have gone through a tremendous change and costs in infrastructure to deal with FHB to meet customer satisfaction. As end-use customers become more concerned with safety and traceability. The country handling system over the next five years will continue to rationalize but need to invest money in processes such as ISO/HACCAP, training, equipment (testing/cleaning/grading) and storage. This will ensure Canada continues to be a leader of quality and a reliable supplier to the end-use customers.

Current impact and emerging issues for mycotoxins in feedstocks. James D. House. *Department of Animal Science, University of Manitoba, Winnipeg, MB, R3T 2N2.*

The westward migration of the incidence of *fusarium*-infected grains, from Manitoba through to Alberta, poses significant challenges to the expanding livestock sector, particularly swine, in these provinces. Of the mycotoxins monitored to date, the trichothecene deoxynivalenol (DON) has been identified as the primary fusarium toxin of concern from a livestock feeding perspective. The presence of DON in final livestock rations can depress feed intake in susceptible animals, with concomitant reductions in weight gain and immunosuppression. Current guidelines for maximal DON levels in finished feeds are 1 ppm for swine, horses, dairy cattle, and 5 ppm for beef cattle, sheep, and poultry. However, these guidelines are in need of re-evaluation. For swine, current research indicates that weanling pigs and grow-finish pigs may be able to tolerate higher levels of DON, when supplied as naturally-contaminated barley, in the finished ration (2-4 ppm) without significant effects on performance. These new data may provide an opportunity for pork producers to increase their utilization of locally-produced, DON-contaminated grains, thus taking advantage of lower transportation costs, and reducing their reliance on imported nutrients. However, greater assurances are required by regulators and the feed industry in order to increase the utilization of DON-contaminated grains in commercially-prepared livestock rations, in order to address issues related to liability under the Feeds Act. As such, candidate strategies are being investigated to decontaminate feed ingredients, or reduce the toxicity of DON in complete feeds. In general, these control strategies can be classified as physical, chemical or biological in nature. Physical methods reduce DON levels through mechanical techniques, including blending (eg. dilution of contaminated grains with clean grains), abrasive dehulling (eg. pearling), and sieving. Chemical methods reduce DON levels through modification of the chemical properties of the trichothecene molecule (eg. epoxide hydrolysis). Biological methods reduce DON levels through transformation of the trichothecene molecule through the action of enzymes working in isolation (eg. exogenous enzymes) or within a biological system (eg. bacteria, yeast, etc...), or through the development of resistant grains. Successful strategies must meet certain criteria, including a) evidence, through feeding studies, that toxicity is reduced; 2) cost-effectiveness, reflecting the tight margins within the livestock industry; 3) maintaining or improving the utilizable nutrient value of the feed ingredient; 4) meeting the requirements of federal regulations pertaining to livestock feeds; and 5) consumer acceptability. While attention in the prairies is currently focused on DON-contaminated cereal grains (wheat and barley), we must be mindful of the increasing acreage of other grains, including grain corn, and the potential for new mycotoxin concerns. Additionally, vigilance against the importation of mycotoxins on feedstocks coming from the U.S. must be heightened. In the end, a combination of some of the aforementioned strategies will likely prove to be the answer to increasing the utilization of mycotoxin-contaminated grains by the livestock sector.

Opportunities for using distillers dried grain with solubles (DDGS) in pig diets. Ian R. Seddon, *Ph.D., Swine Specialist, Animal Industry Branch, Manitoba Agriculture and Food, Winnipeg, Manitoba.*

Interest in a grain-based ethanol industry in western Canada has increased. This could result in the production of various co-products, in particular DDGS that would be suitable for feeding to livestock. In Manitoba, it is estimated that the most significant livestock market for DDGS would be the pork industry. Most research to date has focused on corn-based DDGS and has shown that this product can be successfully fed to pigs. DDGS can serve as a source of amino acids and energy for pigs however, the improved availability of phosphorus in DDGS, compared to its original grain source, is the most likely reason to use this product in pig rations.

At present, little DDGS of Manitoba origin is used in pig rations. However, imports of corn-based DDGS from the United States are not uncommon. The concern with the Manitoba origin DDGS is two-fold – a) consistency of product (i.e. constantly shifting blends of wheat and corn as substrates for ethanol and ethanol co-product production) and b) mycotoxin contamination of the DDGS. Research efforts are underway to determine the nutrient composition of wheat-based DDGS that will be produced in Manitoba as well investigating how the mycotoxin concentrations in DDGS can be ameliorated to allow for the successful introduction of DDGS in pig diets. Until these issues are resolved, utilisation of DDGS by the pork industry will be minimal.

Session 3: New Sources of, and Breeding for, FHB Resistance

Progress in spring wheat. *S. Fox, D. Brown, A. Comeau, R. DePauw, G. Fedak, P. Hucl, G. Humphreys, H. Nass, and H. Voldeng.*

Development of resistance to Fusarium head blight (FHB) in spring wheat has been a slow process as breeders introgress the multiple genes for resistance into regionally adapted cultivars that offer competitive agronomic and disease characteristics within the confines of end-use quality and kernel visual distinguishability (KVD) that define most of Canada's commercial production. Most spring wheat breeding programs currently have FHB resistance as a goal. Irrigated and inoculated FHB nurseries have allowed for screening of large amounts of germplasm and advanced breeding material. In western Canada, FHB resistant lines are now entering the registration system and can be expected with increasing frequency. The yield penalty of obtaining FHB resistance as was observed with BW278 has been overcome. KVD and end-use quality conflicts have been encountered as with BW330 and BW346. Combining FHB resistance with resistance to the orange blossom wheat midge as with BW346 has increased the complexity of cultivar development. However, work is required in regaining appropriate levels of resistance for the traditional diseases such as leaf rust. In eastern Canada, CRGB-O-623.4 was registered in 2002 and was recently recommended for cultivation in Quebec. Investigation of new germplasm has tended to occur more in eastern Canada. Expression of FHB resistance of lines outside their area of adaptation can be different; for example, HY644 is very resistant in most tests conducted in the prairies but is not so in Quebec.

Progress in National Winter Wheat Fusarium Research and Development

Radhey Pandeya¹, Robert Graf², Mark Etienne³, Peter Matthew, Andre Kalikililo¹ and Morley McLean¹

¹*Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Central Experimental Farm, Ottawa, Ontario K1A 0C6;* ²*AAFC, Lethbridge Research Centre, Highway #3, East Lethbridge, P.O. Box 3000, Main, Lethbridge, Alberta T1J 4B1;* ³*Hyland Seeds, W.G. Thompson and Sons Limited, Nairn Research Lab R#1AilsaCraig, Ontario N0M 1A0*

SUMMARY

The following is the progress report on Head Blight (FHB) research and development at Eastern Cereal and Oilseed Research Centre (ECORC) in collaboration with an industry partner, Hyland Seeds of W.G. Thompson and Sons Limited, Nairn Research Lab. The initial developmental research was conducted at ECORC; and four Fusarium resistant / tolerant winter wheat donor genetic stocks (with resistant gene from a Brazilian spring variety, Frontana) were developed. AAFC-WGT collaboration, established in 1995, has led to the development of winter wheat cultivar candidates and advanced lines in multi-location experimental trials. We registered the first soft red Fusarium resistant winter wheat cultivar, Wonder (genetic line OTH017-033: Table 3) in 2002. We have received approval from the Ontario Cereal Crops Committee (OCCC) for two soft white winter wheat lines (OTF 013-081 and TWF020-038: Tables 4 and 5) for registration; and the process to register these lines are underway in 2003. Large-scale breeder seed production and seed increase are in progress towards commercialization. The Fusarium resistant hard red cultivar, AC Morley developed by AAFC, has gained wide spread adoption by our producers. Progress towards Fusarium resistance breeding would have direct positive outcome in relation to all five elements of the National APF priorities (such as Risk Management, Food Safety and Quality and Safety, Environment, Innovation and Renewal, and International).

Introduction:

Wheat is Canada's most important agricultural crop. Wheat based products provide a significant portion of carbohydrate calories to Canadians and to people worldwide. On average, nearly 27 million tonnes of wheat are produced on 11.8 million hectares annually. Wheat production generates 9 to 10 percent of the Total Canadian Farm Cash Receipt of \$35.696 billion, and more importantly, represents 20 to 25 percent of the Total Crop Farm Cash Receipt. Although Canadian wheat is predominantly an export commodity, with winter wheat accounting for only 2.8 to 3.0 percent of total annual wheat acreage, it is a very significant component, as it forms the basis for significant domestic processing and manufacturing of many value-added products. Current estimates put the seeded acreage of winter wheat at well over 2 million acres, with over a million acres in both Ontario and western Canada. The total contribution of winter wheat to the Canadian agri-economy is well over \$1.5 billion.

Winter wheat has a wide range of usage in consumer food products. In Ontario, a large proportion of the soft red and soft white winter wheat that is produced is used in baby food, cakes, cookies and confectionaries for all of Canada. Hard red winter wheat produced in western Canada has many uses in traditional, hearth, and streamed breads, noodles, and is an expanding component of animal feed rations. In the future, the inherent yield advantage of winter wheat makes it an obvious choice for bio-ethanol production.

Spring wheat in Eastern Canada is relatively small but growing steadily. Winter wheat is the 5th or 6th most important field crop and second (following corn) most important cereal in Ontario. Its value added processing is considered to be well over a billion-dollar industry in Ontario. The Figures 1 and 2 depict the impact of improved genetics and crop husbandry practices in continuous climb in per unit yield of spring and winter wheat since 1881. **Wheat crop in Canada and USA has been suffering from continued FHB onslaught since past several years, which stands to nullify all previous gains.**

Figure 1

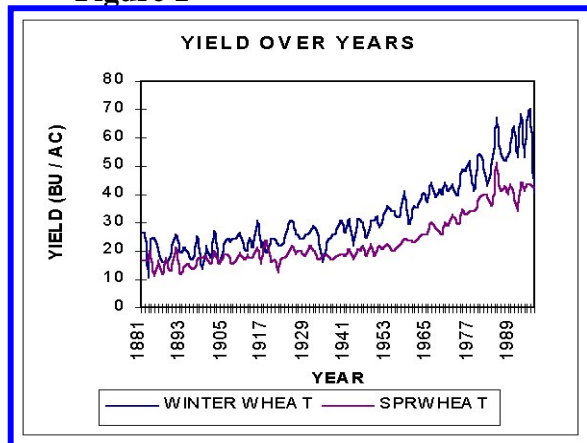
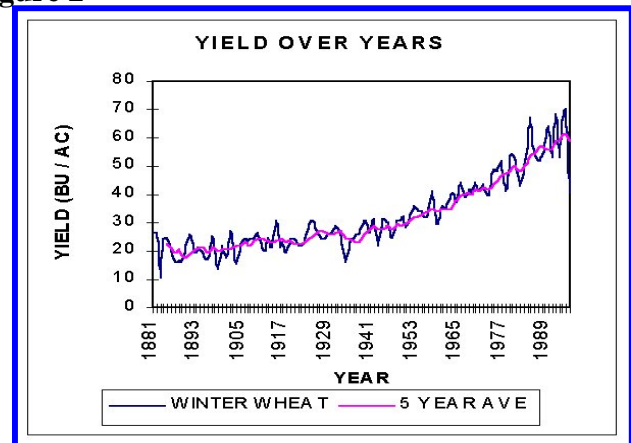
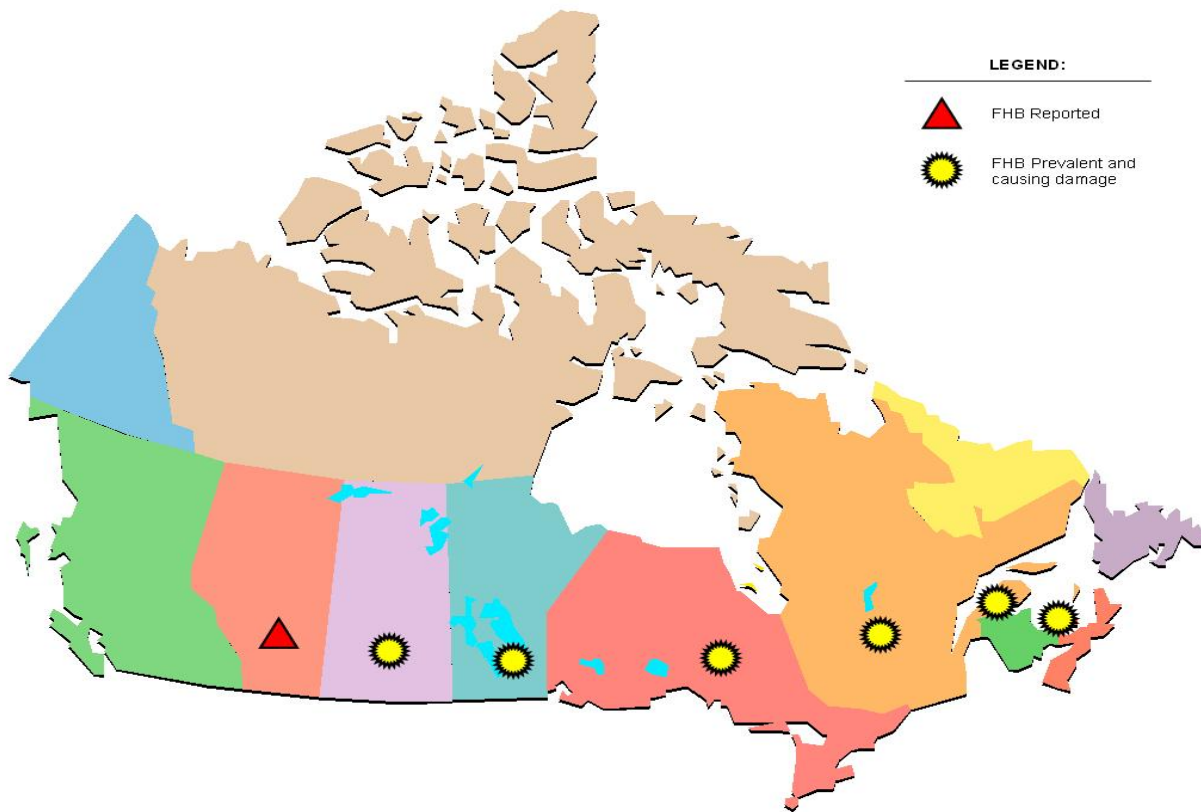


Figure 2



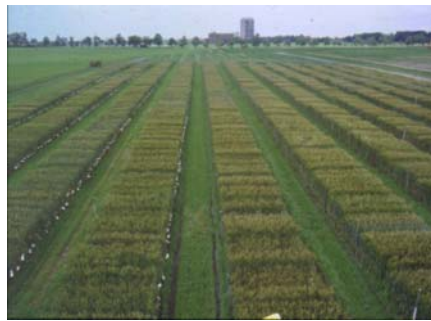
A sustained production of winter and spring wheat in Canada is threatened by Fusarium head blight, which had already devastated the Canadian wheat crop in eighties and nineties. Damage in 1981, 1986 and 1988 in Ontario was tolerable. In the 1995-96-crop season, Ontario crop suffered a great loss due to FHB epidemic. Only 42% of the expected production target (from 850,000 acres) of 1.4 million tons was achieved. Food-feed and seed quality was drastically impaired. Thus a billion dollar wheat agri-industry has been affected to a greater or lesser degree by Fusarium ever since. FHB, from being a disease of eastern Canada has acquired a national scope since early nineties (Fig.3)

Figure 3: Epidemiology of Fusarium Head Blight in Canada



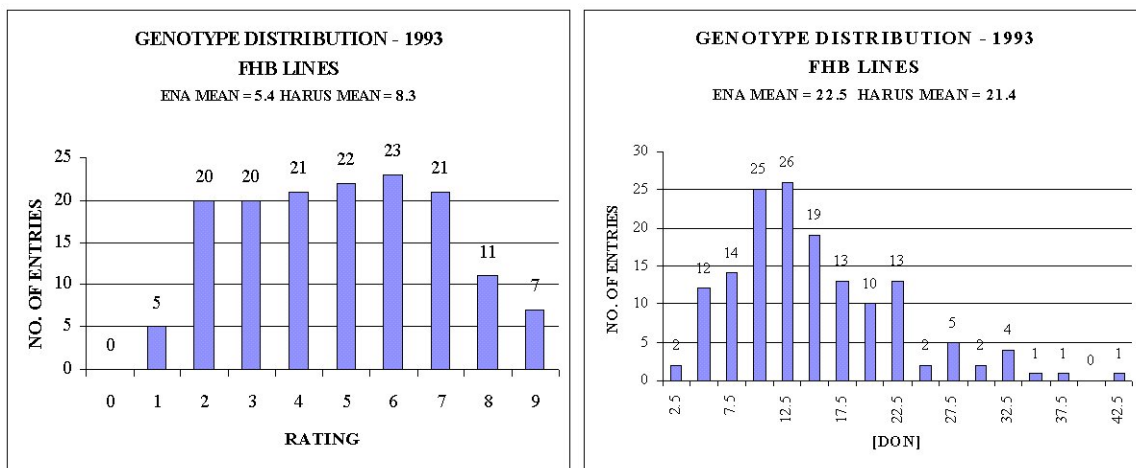
The Fusarium Head Blight (FHB) epidemic in the early eighties and a total lack of suitable chemical phytoprotection agents at the time placed an urgency to find genetic source of resistance and develop FHB resistant / tolerant cultivars. In 1984, Dr. Sampson selected a Saint-Hyacinthe accession of Frontana, a Brazilian Spring wheat variety as a source of FHB resistance donor, and made crosses with the standard winter wheat cultivars (Harus, Augusta and Fredrick) of the day. Segregating generations were grown and seed of F3 and F4 generations were preserved.

Screening procedure evaluated: ECORC established FHB screening nursery with misting provision to maintain required humidity for the disease development under epiphytotic conditions (Figure 4,5,6). We developed a suitable method of inoculation to assess the tolerance of different winter wheat lines. Injection and spray methods of inoculation were compared for pedigree derived and doubled haploid lines. Visual ratings for spray and injection methods exhibited a very high positive correlation ($r=0.92$). There were significant differences amongst wheat lines for visual rating. The correlation between DON and Visual rating was 0.39 to 0.6. DON values in two-crop season were different. However, the ratio between DON (from inoculated) and DON (from the control) were comparable. Subsequently, we used the spray method of inoculation at the anthesis period.

Figure 4: Field inoculation of genetic lines**Figure: 5 Epiphytotic nursery****Figure: 6 Aerial View of the Fusarium Nursery**

A pedigree population consisting of 625 lines (in F3-F4) was sprayed with Fusarium Headblight spores (50,000 spores per ml of suspension) under greenhouse epiphytotic nursery conditions in 1990 during the First Cycle of Selection. Lines were rated visually on a 1-10 scale (where higher numbers indicate susceptibility). Samples of selected lines with rating score of less than 4 were analysed for the mycotoxin Deoxynivalenol (DON) by GC-MASS SPEC method.

Based on the visual symptom ratings (VSR), (Fig. 7) with ratings below 4 were advanced for the next cycle of epiphytotic evaluation and selection in 1991-92 in field nursery. Further inoculation and selection were conducted; and a total of 176 lines were advanced for the third cycle of selection in 1992-93. We determined DON on the 1992-93 selections by monoclonal antibody base technique developed at ECORC (Figures 7).

Figure: 7 Third cycle of selection for symptom and DON

Over a three-year period, continuous selection pressure was applied in favour of low visual rating and / or DON. By 1992-93 crop season, a large number of lines were identified, with DON values ranging from 3 ppm to 43 ppm. We succeeded in transferring genes for resistance from a spring to winter wheat. The data distribution for

disease rating suggested multi-genic inheritance control. Correlation analysis revealed no definite associations between visual rating and DON contents. This indicated that symptom expression and DON are under separate genetic controls (Table 1). Low or no correlation between DON and VSR appeared to be compatible with the discrete class distribution (suggesting few major genes with modifiers may be controlling the characteristics).

Fusarium tolerant genetic stock identified Lines with LOW-LOW, LOW-HIGH, HIGH-LOW and LOW-HIGH combinations of DON and VSR were identified. Also inference that the DON and VSR were independently inherited appeared justified. Based on low DON and VSR, 27 lines were advanced for final evaluation in 1993-94

Table 1

VISUAL RATING	DON CONCENTRATION	NUMBER OF ENTRIES
HIGH	HIGH	25
HIGH	LOW	55
LOW	HIGH	41
LOW	LOW	53

Further full-sib of **FHB 143, 147, 148 and 161 (with Low Don and Low visual symptom rating)**, were evaluated under epiphytotic conditions and all sister lines regressed to the original parental line, suggesting that the four lines were approaching required level of homozygosity and stability of performance as far as Fusarium tolerance was concerned. Data distribution regressed towards the parent (Fig. 8). Thus the above four selected lines were chosen as donor parent for Fusarium resistance.

Collaboration established and development of Fusarium resistant/tolerant cultivar began: (R Pandeya and Leslie Shugar)

Radhey Pandeya of ECORC and Leslie Shugar of W.G. Thompson and Sons Limited received approval from the management of the two organisations for MII-funded collaborative project to develop Fusarium resistant soft winter wheat cultivar(s) in 1995. Techniques of haploidy, via wheat-maize pollination, embryo rescue and colchicine doubling were opted for developing large number of doubled haploid lines (DHS) from single, three-way, double and multiple cross combinations with one or all of the four designated donor parents and standard cultivars. Since 1995-96, well over 20,000 lines were developed. Almost 85 to 90% of the lines with poor agronomy and/or susceptibility to Fusarium are eliminated. Selected lines were increased for seed, and replicated tests are routinely conducted to assess yield potential, quality and agronomic types and FHB tolerance. FHB assessment is carried on all lines from the very beginning at the two experimental Centres. Don is determined on advanced replicated trials' entries. Figure 9 shows expression of Fusarium tolerance in the epiphytotic nursery.

Figure 8: Winter Wheat genetic stocks with Fusarium resistance

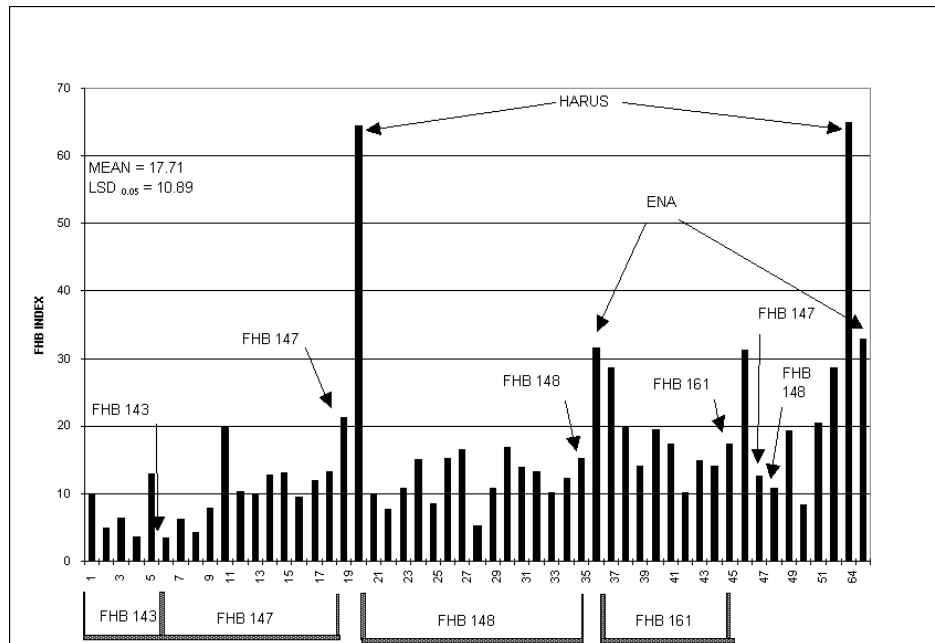


Figure 9: Field expression of Fusarium resistance



Progress to date:

- A DH line **OTH 017-033** has been advanced to the Ontario Winter Wheat Performance Trial in 2001-02 crop year. It has yield comparative to all checks and three to four times lower Fusarium Index values (35 to 40 for checks vs. 12 for the line). We registered the line in January 2002. Large-scale breeder seed production is underway for commercialisation.

- Two new lines (**TWF020-038** and **OTF013-081**) of soft white winter wheat are approved and proceeding for registration in 2003. Breeder seed multiplication has begun.

Table 2: lists the some of the selected lines with excellent FHB tolerance compared to checks. These have been already evaluated for FHB tolerance. These are in replicated trials for final yield and quality parameter determinations for eventual licensing of a selected few.

Table2: Fusarium index of doubled haploid lines compared to checks

NAME	FHB-INDEX	NAME	FHB
AUGUSTA	42.5, 40, 35	AUGUSTA	35, 45, 40, 30, 70, 30
AC RON	30, 40, 30	AC RON	45, 40, 40, 40, 70, 50
FREEDOM	32.50, 35, 40	FREEDOM	50, 40, 40, 50, 40, 40
P2540	20, 25, 15	P2540	25, 25, 20, 30, 30, 20
WISDOM	15	OTF025-079	10
OTH017-033	10	OTF008-025	20
OTF13-037	12.5	OTF012-019	10
OTF13-081	17.5	OTF029-052	25
OTF13-082	22.5	OTF029-072	20
OTF13-104S	12.5	OTF025-027	25
TWF013-043	15	OTF012-074	10
TWF013-045	22.5	OTF012-035	10
TWF013-065	10	OTF012-001	40
TWF020-038	15	OTF012-100	10
OTF022-074	27.5	OTF029-001	15
TWF007-003	10	OTF012-030	25
TWF007-066	55	OTF012-015	10
TWF013-060	25	OTF025-073	30
TWF019-018	10	OTF029-080	30
TWF026-059	10	OTF012-101	20
TWF028-011	15	OTF012-069	30
TWF032-014	50	OTF020-034	30
TWF032-029	25	OTF025-056	30
TWF032-039	10	OTF012-080	20
TWF036-007	20	OTF008-034	10
TWF036-052	20	OTF012-051	20
TWF037-001	40	OTF029-037	30
TWF037-006	10	WF034-055	20
		WF034-006	10

A few lines with high FHB Index are carried forward because of their excellent yield records.

- The **hard red winter wheat cultivar, AC Morley**, was registered in 2002, and has gained wide acceptance from the producers for its yield and Fusarium resistance;
- We have already begun the process of incorporating FHB resistance in winter wheat cultivars, adapted to the western Canadian regions. All FHB evaluations are to be conducted at ECORC. DH technology is deployed to develop pure breeding lines from the designated crosses.

New germplasm collected and new lines developed:

- Germplasm from Hungary, Austria, Ukraine, Russia, USA and China have been acquired.
- Five new Fusarium spring wheat lines (resistant to Fusarium) are developed at ECORC with Chinese source of resistance.

W.G. Thompson. Most of progress reported in this article in relation to cultivar development is from the AAFC-ECORC and W.G. Thompson joint project.

Table 3

Varieties	Fusarium	Fusarium										
	field % ##	Nairn 97- 98	Nairn 98- 99	Ott 99- 00	Inoculations ##		Nairn 00- 01	Nairn 00-01	Ott 00-01	Elora	Mean	Mean
	[3]	index %	index %	index %	index (%)	DON (ppm) ^	index %	DON (ppm)	index %	index % not in mean	index%	Mean DON (ppm)
											[6]	[2]
Augusta	22.0	25.0	62.5	47.5	65.0	6.00	30.0	0.64	30.0	n/a	—	3.32
AC Ron *	31.0	25.0	80.0	54.0	70.0	6.00	40.0	0.47	22.5	10.0	48.6	3.24
Freedom *	11.0	15.0	52.5	43.7	45.0	4.00	25.0	0.43	25.0	12.5	34.4	2.22
P2540 *	17.0	25.0	80.0	44.0	70.0	5.60	32.5	1.60	15.0	30.0	44.4	3.60
OTH 017- 033	2.0	5.0	20.0	10.0	25.0	1.90	7.5	0.23	12.5	not planted	13.3	1.07
check mean	11.0	15.0	52.5	43.7	45.0	4.00	32.5	0.43	25.0	12.5	37.7	2.22
LSD		12.8	22.9	n/a	15.2	1.40	9.4	0.80	8.7	10.8		
CV%		28.6	16.4	n/a	14.4	18.70	21.0	65.0	43.4	77.0		

[] for # of locations

Augusta is quality check

* AC Ron, Freedom and P2540 are yield, agronomy and disease checks, Freedom is Fusarium check

%: Fusarium index = % plot infection x % spikelet infection for inoculation data, for field (natural) and inoculation data

** 0 is none; 9 is worst

Note: Minimum Fus index for checks is 15 % in guidelines; sites with Freedom < 15%, not in means.

^ test kit only goes up to 6 ppm

Table 4: Disease data summary (2001-2002)
FUSARIUM

	COMPARATIVE DATA : 0TF013-081								
	2000	2001	2001	2002	2002	Nat Ind	Overall mean		Mean Natural infection reported from
	Nairn	Nairn	Ottawa	Nairn#	Ottawa		Inocu.		
Station yr	1	1	1	1	1		Mean		* Augusta is quality check
Varieties									** Yield checks
	FHBI	FHBI		FHBI			FHBI		Freedom is also the Fusarium check
Augusta*		30	42.5	21	27.5	2.4	33.3		
AC Ron**	68	40	30	3.5	30	3.5	42		FHBI= FHB index calculate d as:
Freedom**	35	32.5	32.5	9	20	2.3	31.25		(% head x %spikelets)/100
P2540**	58	45	20	22.5	25	4.5	34.5		Natural: 0= none, 9=worst
OTF 013-081	10	0	12.5	0	15	1.1	9.38		#Nairn 2002 data not included in mean
CV	12.62	19.22	44.1	16.8	29.4				DON value from 2002 Orthogonal test
LSD	14.9	13.7	11.83	51.97	8.6				

Table 5: Disease data summary (2001-2002) :COMPARATIVE DATA FOR TWF020-038
FUSARIUM

	2000		2001		2002		Overall mean	
	Inocu.	Inocu.		natural	Inocu.	natural	Inocu.	Natural
Station yr	3	3		1	6	5	12	6
Varieties								
Augusta	52.3	34.2		2.2	24	2.6	36.8	2.4
AC Ron	45.8	35		2.2	26.7	4.7	35.8	3.5
Freedom*	26.8	32.5		3	21.3	1.5	26.9	2.3
P2540	17.3	36.7		4.8	19.7	4.2	24.6	4.5
TWF020-038	19.8	11.3		1.2	7.1	1.5	12.7	1.4

Fusarium index of Ontario winter wheat cultivars

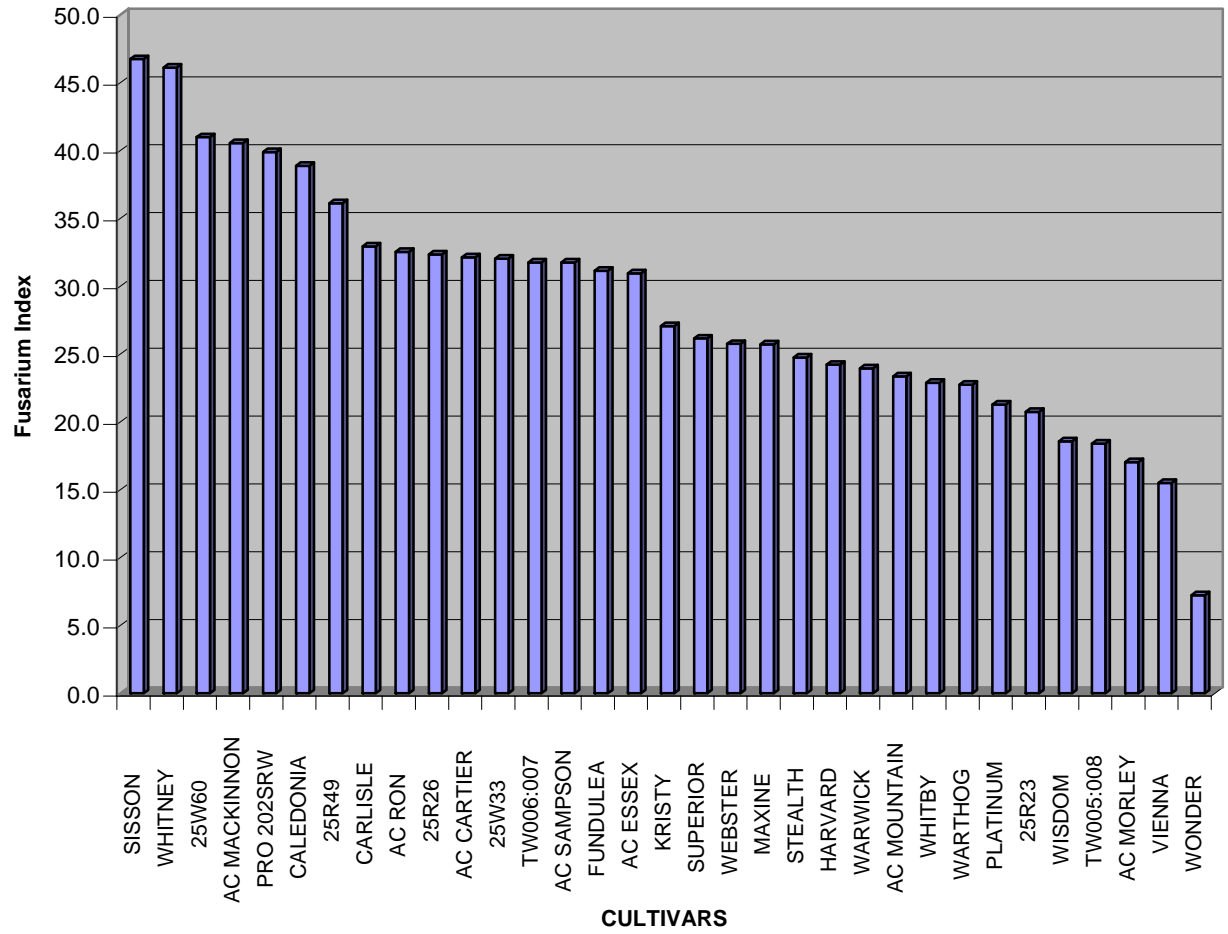


Figure 9

DON (ppm) of inoculated cultivars

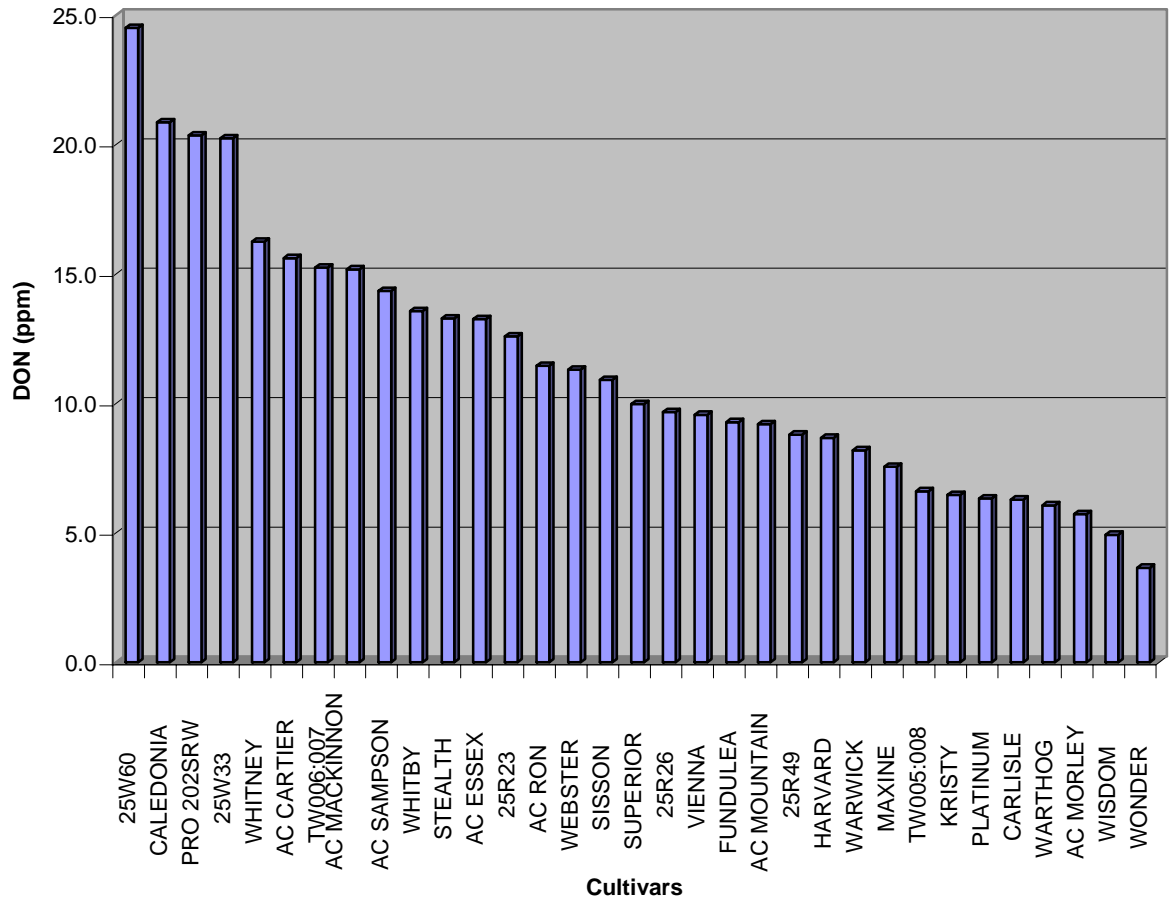


Figure 10

Progress in Improvement of *Fusarium* Resistance of Durum Wheat. J. Clarke,

J.Thomas, G.Fedak, D.Somers, J. Gilbert, Curtis Pozniak, M.Fernandez, and A. Comeau.
Agriculture and Agri-Food Canada, (J.C. and M.F.) Box 1030, Swift Current, SK S9H 3X2, Canada; (J.T., D.S. and J.G.), 195 Dafoe Road, Winnipeg, MB R3T 2M9, Canada; and (A.C) 2560, Boulevard Hochelaga, Sainte-Foy, QC G1V 2J3, Canada; (C.P.) Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK S7N 5A8.

Durum wheat (*Triticum turgidum* L. var *durum*) is reported to have greater susceptibility to *Fusarium* head blight (FHB) than common wheat (*T. aestivum* L.). Extensive surveys of durum germplasm have not identified any accessions with resistance approaching that of resistant common wheat. Our objective is to move FHB resistance from other wheat relatives into durum wheat, and to exploit the available resistance within durum wheat. The existing variation in FHB resistance within durum may be sufficient to reduce damage under the relatively light and sporadic disease pressure experienced in the major Canadian durum production area. Higher levels of resistance would increase this protection, and perhaps facilitate durum production in the eastern prairies where FHB is a greater risk. Sources of improved resistance include the tetraploid wheats *T. dicoccoides* and *T. carthlicum*, as well as hexaploid common wheat. The extensive research to identify quantitative trait loci (QTL) for resistance in common wheat and more limited research in *T. dicoccoides* is being used to facilitate transfer of the resistance to durum. Current work is transferring individual QTL from chromosomes 3AS, 3BS, 4BS, and 5AS into adapted durum. Other work in progress includes mapping of Type II resistance in a *T. carthlicum* X durum population.

Progress in breeding for Fusarium head blight resistance in barley. W.G. Legge¹, M.C. Therrien¹, J.R. Tucker¹, M. Banik¹, A. Tekauz², D. Somers², M. E. Savard³, B.G. Rossnagel⁴, E. Lefol⁴, D. Voth⁴, T. Zatorski⁴, B.L. Harvey⁵ and G. Scoles⁶

¹Agriculture and Agri-Food Canada, Research Centre, P.O. Box 1000A, RR #3, Brandon, MB R7A 5Y3, Canada; ²Agriculture and Agri-Food Canada, Cereal Research Centre, 195 Dafoe Road, Winnipeg, MB R3T 2M9, Canada; ³Agriculture and Agri-Food Canada, Eastern Cereal and Oilseed Research Centre, 960 Carling Avenue, Ottawa, ON K1A 0C6, Canada; ⁴Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK S7N 5A8, Canada; ⁵University of Saskatchewan, Box 5000 RPO University, 110 Gymnasium Place, Saskatoon SK S7N 4J8, Canada; and ⁶Department of Plant Sciences, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK S7N 5A8, Canada

Introduction

Much of the barley (*Hordeum vulgare* L.) breeding effort in western Canada from 2000 to the spring of 2003 to improve resistance to Fusarium head blight (FHB) has been conducted through a collaborative project funded by the Canada-Manitoba Agri-Food Research and Development Initiative (ARDI), Western Grains Research Foundation (WGRF) Barley Check-off and Endowment Funds, Saskatchewan's Agriculture Development Fund (ADF), and Agriculture and Agri-Food Canada's Matching Investment Initiative (MII) program. We have recently prepared final reports for the funding agencies and would like to share our main findings with you in this presentation. This will be followed by a brief description of our new project initiated last spring and an update on progress.

2000-03 FHB Project Report

Objectives:

The overall goal of the project was to improve FHB resistance of barley with the following specific objectives:

- 1) Evaluate current barley cultivars, all entries in registration trials, and advanced breeding lines from Agriculture and Agri-Food Canada (AAFC) Brandon and Crop Development Centre (CDC)/ University of Saskatchewan (U. of Sask.) programs for FHB resistance. The Alberta Agriculture, Food and Rural Development (AAFRD) Field Crop Development Centre (FCDC), Lacombe, may also enter some advanced breeding lines in the nursery.
- 2) Exchange elite germplasm with promising FHB resistance with other barley breeding programs, such North Dakota State University (NDSU), University of Minnesota (U. of Minn.), and Eastern Cereal and Oilseed Research Centre (ECORC), Ottawa.
- 3) Evaluate new putative FHB resistant parents from all possible sources.
- 4) Evaluate lines from crosses segregating for known sources of resistance, such as CI4196, or from crosses between two moderately resistant lines with different backgrounds. Select the most promising lines for further research and crossing.
- 5) Develop a protocol for *in vitro* selection of FHB resistant barley lines using deoxynivalenol (DON) or other mycotoxins in anther/microspore culture. Use this protocol to identify new sources of FHB resistance and develop recombinant lines resistant to FHB in all classes of barley.

- 6) Develop and evaluate special doubled haploid populations for determining the inheritance of FHB resistance.
- 7) An additional objective was added in 2002 to develop a calibration to determine DON content using near infrared spectroscopy (NIR). This is being done at the CDC, Saskatoon.

Procedure:

The project was initiated in 2000 with the establishment of a large nursery at Brandon, MB, in which thousands of barley lines from AAFC Brandon and CDC/ U. of Sask. breeding programs could be evaluated for FHB resistance. The total number of plots in the nursery, number of plots harvested, and number of DON samples sent to ECORC each year are shown in Table 1. We experimented with several different “plot” types, but settled on single 0.9-m rows to maximize efficiency.

Table 1. Number of plots sown and harvested and deoxynivalenol (DON) analyses for the Fusarium head blight (FHB) nursery at Brandon from 2000 to 2002.

Number:	2000	2001	2002
- Plots sown	12,416	16,200	14,400
- Plots harvested	3,406	7,882	9,728
- DON analyses	2,770	6,198	5,204
DON content, ppm¹:			
- AC Metcalfe	2.2 (142) ²	29.5 (173)	12.4 (174)
- Stander	8.3 (142)	46.6 (172)	18.7 (173)

¹Mean DON content of repeated nursery checks AC Metcalfe and Stander in ppm.

²Value in parentheses is the number of samples used to give the mean DON content.

The nursery was inoculated with grain spawn (i.e., corn seed infected with 3 isolates of *Fusarium graminearum* Schwabe) spread on the ground 3-5 times at weekly intervals starting before the earliest lines in the nursery headed, and irrigated to promote fungal development. In 2001 and thereafter, 1-2 blanket treatments of Tilt were applied to the nursery prior to the first application of corn inoculum to control *Cochliobolus sativus* (Ito & Kurib.) Drechs. ex Dast., a pathogen which may confound the results. All entries were rated visually on a 0-5 scale (0 = no symptoms, 5 = severe symptoms) about 3-3 ½ weeks after heading. A lodging score was also assigned at that time using a 1-9 scale (1 = erect, 9 = flat). Depending on the purpose of a test, the visual FHB scores were often used to select genotypes with the most promising FHB resistance for harvest and DON analysis.

Grain harvested from the nursery was cleaned, and a 20 g subsample was ground with 1 gram being sent to Dr. Marc Savard’s lab at ECORC, Ottawa, for the critical DON analyses using the ELISA technique (maximum of 5,000 determinations per year under the project). Along with 50 additional DON assays conducted on samples originating from a 1999 FHB nursery, several hundred samples from the current study and their corresponding DON data (determined by ELISA technique) were used by researchers at the CDC, Saskatoon, to develop a calibration for NIR to predict DON levels in barley.

The Brandon nursery was successful with DON levels varying from year to year as shown by the overall means for two of the repeated nursery checks (Table 1). It has been clear from the variation observed among checks that the expression of this disease has a strong environmental component and multiple years of testing are required for reliable conclusions.

In support of the Brandon nursery, the Cereal Research Centre (CRC) was to provide additional information and back-up for high priority material in a smaller FHB nursery previously established at Glenlea. Unfortunately, no useful data were obtained from this nursery at Glenlea over the three years of the project due to unfavourable environmental conditions there. The CRC did provide critical help in rating the Brandon nursery.

Over the last 3 years, we have used a FHB nursery in Hangzhou, China, to provide a second cycle of FHB screening each year over the winter months and an assessment of entries in a FHB nursery free of *C. sativus*. In 2000, we sent 1,000 entries, which was subsequently doubled in 2001 and 2002. Our Chinese colleagues determined days to heading, % diseased spikes, % infected seeds and a disease index. Overall correlations between the data from China and Brandon have generally been low, although similar patterns were observed among genotypes.

In vitro selection (IVS) in conjunction with doubled haploid (DH) production using anther culture was evaluated mainly as a tool for screening segregating populations for FHB resistance. It was applied to three types of parental germplasm: current varieties (two- and six-row, covered and hulless), FHB resistance sources, and populations segregating for FHB resistance. Several methods have been used to produce the different IVS groups, including variation in the types of mycotoxins applied to the selective growth media, the concentrations at which they were applied and the timing. In addition to DON, several other mycotoxins produced by *Fusarium* spp., such as 3-acetyl deoxynivalenol (ADON) and T2, were employed in some experiments. DON contents of IVS lines were compared in FHB nurseries to the parental genotypes or the control DH lines produced in the absence of mycotoxin.

Results and Discussion:

Although improving FHB resistance will be a long-term effort, good progress was made over the three years of the project. Valuable information was provided to producers through the Manitoba seed guides (and equivalent in other provinces) on the FHB resistance of current barley cultivars, allowing producers to make informed decisions. Without this project, the information would have been fragmented and incomplete. It also gave barley breeders a better idea of the FHB resistance of their cultivars so that they can plan their breeding efforts accordingly. One of the disappointing findings of the project was that the FHB resistance of some two-row malting cultivars, like AC Metcalfe and CDC Stratus, did not appear to be as good as we had thought at the outset of the project. Perhaps the resistance of these cultivars breaks down more under the heavier *F. graminearum* infection levels observed in 2001 and 2002. A number of two-row cultivars, such as Harrington, CDC Fleet and Conlon, appear to have lower DON content than AC Metcalfe but they tend to be early, highly susceptible to spot blotch or do not perform well in the FHB-affected areas. Few six-row cultivars surpassed CDC Sisler, although the six-row blue-

aleurone malting cultivars, Argyle and Tankard, appeared to be more resistant. However, there is no longer any market for this class of barley. Bedford and Bronco were the best six-row feed cultivars and they were comparable to CDC Sisler in DON content. Two-row hulless barley cultivars, like CDC Freedom and Phoenix, look very promising and may have the lowest DON content of all classes of barley, but unfortunately this class has been struggling for a market share in recent years. Perhaps producers and the livestock feeding industry should be taking another look at hulless barley. The six-row hulless cultivars, particularly AC Hawkeye, compare favourably with two-row cultivars in terms of DON content. The hulless trait may be partly responsible for this, but in looking at the six-row group in particular there are some hulless cultivars that have very high levels of DON.

The project also provided data on FHB resistance for potential cultivars in the western Canadian cooperative registration trials. Without this project, there would have been no data on these lines at all since the FHB nursery for barley at Glenlea failed all three years. Not many new cultivars were registered over the past 3 years with improved FHB resistance over AC Metcalfe (two-row check) or CDC Sisler (six-row check). The two-malting cultivars, Calder and CDC Goodale, may have lower DON content than AC Metcalfe, while the two-row feed cultivar Ponoka from Alberta also looks promising. One of the most promising lines in the two-row cooperative test during this time was TR361 which was supported but not registered due to high susceptibility to spot blotch, an important disease in Manitoba. From the hulless cooperative test, HB364 from the CDC was probably the most promising in terms of FHB resistance to be registered. It is expected that more lines with improved FHB resistance will be entering the registration tests over the next few years as a result of the research done under this project. In eastern Canada, the two-row feed cultivar Island with improved FHB resistance was recently registered, and is currently being evaluated in the Eastern Prairie Barley Test for its potential as a cultivar in western Canada.

Germplasm exchange has been occurring with researchers in eastern Canada, United States and ICARDA/ CIMMYT. Since these researchers have been working at this problem longer than we have in western Canada, we hope to identify FHB resistant lines that we can use for crossing purposes to improve FHB resistance. In 2002, we participated in the North American Barley Scab Evaluation Nursery (NABSEN) with American researchers for the first time. Although we only had 8 entries in the test, our lines performed reasonably well. These elite lines may have the potential to become new cultivars in western Canada or serve as parents for crosses. It is hoped that germplasm exchange and collaboration will build on this base and increase over the years ahead.

The best known sources of FHB resistance are CI4196 in two-row and Chevron in six-row types. Both are undesirable from an agronomic point of view, susceptible to other diseases and have unacceptable quality. Also, their FHB resistance will breakdown if infection levels are high enough. It would therefore be desirable to identify new sources of resistance to complement them or that are in a more desirable background. For this purpose, we have screened a large number of lines from Plant Gene Resources of Canada, as well as advanced breeding lines from European and Australian barley breeding programs. A number of lines have been identified with

promising FHB resistance, but additional testing will be needed to confirm the results and determine their suitability in terms of agronomic, disease resistance and quality traits. This is expected to be a long-term process.

All barley breeding programs in western Canada have at least some lines with moderate resistance to FHB surpassing AC Metcalfe or CDC Sisler. Most of the moderately resistant lines in the registration trials to date were identified in existing breeding material, but new lines specifically bred for FHB resistance are now being advanced through the breeding programs. Of particular note are 25 AAFC Brandon breeding lines from 10 crosses involving known sources of “exotic” FHB resistance including AC Sterling, Chevron, CI4196, Gobernadora, Harbin, Morrison, Siejo II, Symko, and Zhedar 1. Note that AC Sterling, Morrison and Symko are actually eastern Canadian varieties which we had not been using until recently. These 25 lines have shown consistently lower DON levels than AC Metcalfe over several years of testing in FHB nurseries. Initial results suggest that these gains may have been accompanied by trade-offs with resistance to other diseases like stem rust and spot blotch which are also important diseases in the FHB-affected areas. These lines will be grown in advanced yield tests at multiple sites in 2003 with the most promising being entered in registration tests in 2004.

Of particular note in the six-row and hulless barley breeding program at Brandon is the two-row hulless line EX645-3-6 which has low DON content and will be evaluated in the 2003 NABSEN test. A number of six-row lines, both hulless and covered, with low DON content have also been identified and will be advanced through the breeding program.

The CDC/ U. of Sask. barley breeding programs have identified and advanced many breeding lines with promising FHB resistance relative to AC Metcalfe and CDC Sisler. Some of these may become new cultivars in Manitoba and eastern Saskatchewan. Of particular interest are lines from the non-malting program from crosses involving CDC Freedom, CI4196 and HDE84194 as resistance sources. Some of these are being evaluated in yield tests.

It is expected that new two-row hulless, feed and malting barley cultivars with improved FHB resistance will be registered over the next 5 years. The order of the classes listed above is probably the order in which the new cultivars will be released with two-row malting barley being the most difficult and slowest because of quality constraints super-imposed on this problem. Improvements in FHB resistance, as indicated by lower DON levels, will probably be incremental in nature – no large reduction in DON content is expected in the near future. It may be 10 years before new six-row cultivars with significantly lower DON levels than CDC Sisler are registered. However, it is possible that our American colleagues, who have been working diligently on improving FHB resistance in six-row malting barley for many years now, may develop such a cultivar sooner which may also be adapted here.

We were successful in developing protocols for IVS that allowed us to regenerate DH plants, but the results from the field have been somewhat disappointing to date. It is possible that fine tuning our protocols, such as adjusting the concentration and composition of mycotoxins and length of exposure to the mycotoxins in culture, may improve results. Research is currently

underway for that purpose, and has been extended to isolated microspore culture since the breeding program now utilizes this technique for routine DH production. The IVS project was supported by the WGRF Endowment Fund with matching funds from ARDI. The WGRF has extended its support for another three years.

A number of DH populations have been developed for potential genetic studies on FHB resistance, but additional funding will be needed to proceed further. This is beyond the mandate of the current project.

We added a new objective in collaboration with the CDC to develop a calibration for NIR to determine DON content more rapidly, easily and cheaply than is possible with current methods. Results to date are encouraging, but the accuracy of the equation in predicting DON content will need to be improved using more samples containing a wide range in DON values from different varieties grown at various locations over a period of time. This calibration has been developed for ground samples. An equation based on whole grain also shows promise but many more samples must be added to improve accuracy. This would eliminate grinding and make initial screening prior to ELISA significantly more efficient. The number of samples that can be analyzed for DON content is a bottleneck in improving FHB resistance in barley.

A set of cultivars, consisting of two-row and six-row as well as covered and hulless cultivars, was evaluated over all three years of the project at Brandon to determine the relationships among various traits. Among the measures of FHB resistance, DON content (as determined by ELISA) over years gave the highest correlation ranging from 0.67 to 0.82. The correlation between 2002 DON content (ELISA) and DON content (as determined by NIR in 2002 only) was moderate at 0.76. The correlations between DON content (NIR) and other traits followed the same pattern as for 2002 DON content (ELISA). For FHB ratings, the correlation among years was somewhat lower ranging from 0.55 to 0.65. The correlation between FHB rating and DON content (ELISA) in a given year ranged from 0.54 to 0.73. Similar results were found in a cooperative study coordinated at ECORC. The moderate correlation coefficients for DON content (ELISA) and FHB ratings over years suggest that progress is possible but it will be a long-term effort.

In conclusion, this project has laid the foundation for developing new barley cultivars with improved FHB resistance. We have identified some of the more FHB resistant barley cultivars and advanced breeding lines currently available. Some of these advanced breeding lines may be under commercial production within 5 years, particularly for two-row barley. However, it may take up to 10 years for six-row cultivars with lower DON content than CDC Sisler to be available commercially. The development of highly resistant barley varieties using exotic sources of resistance will be a long-term effort due to unfavourable effects on other important traits.

New FHB Project

General:

In order to build on the progress described above, we initiated a new three-year project in the

spring of 2003 with funding from the WGRF Barley Check-off and special funds from interest earned on the WGRF Barley Check-off Reserve Fund, and the MII program. The new project will continue most of the objectives from the previous project but with a more national focus. Joining researchers from the previous project will be R. Martin from AAFC Charlottetown, PE; A. Choo, K.M. Ho and B. Vigier from ECORC, Ottawa, ON; K. Turkington from AAFC Lacombe, AB; and the AAFRD group also at Lacombe, AB, including J. Helm, P. Juskiw, J. Nyachiro, K. Xi, and J. Zantinge. The overall goal will be to develop barley germplasm with improved FHB resistance for all regions of Canada. The key is to continue our large nursery at Brandon in which barley lines from all breeding programs in Canada are evaluated for FHB reaction and DON content. Barley lines with promising FHB resistance at Brandon will then be evaluated in the Glenlea and Charlottetown FHB nurseries to confirm resistance and hasten the development of FHB resistant germplasm and cultivars. Access to the FHB nursery at Charlottetown is an important addition to the project.

Funding levels for the new project are somewhat reduced from the previous project so we have had to make a number of changes. We decided to discontinue sending germplasm to the FHB nursery at Hangzhou, China, because the relationship with Canadian data was not as high as we had hoped and no DON data are obtained. The funding will be utilized instead to support the FHB nursery at Charlottetown. The size of the FHB nursery at Brandon will be reduced to about 11,500 rows. The quota for DON testing by ELISA at ECORC will be increased to 6,000 samples with 5,000 samples for the Brandon nursery and 1,000 samples for Glenlea and Charlottetown combined.

Although we have increased our DON testing capabilities, this is still a major bottleneck for improving FHB resistance in barley. In collaboration with the CDC, Saskatoon, we will continue to work on developing a calibration for the NIR that can be used ultimately on whole grain samples. We may be able to use the NIR for an initial screening with the promising entries being sent to ECORC for a more accurate DON determination. The disadvantage of this would be the need to harvest many more rows from the FHB nursery. Over the past two years, we have hand-cut nearly 10,000 rows at Brandon which is not that much less than our reduced capacity of 11,500 rows. The reason for harvesting the large number of rows at Brandon relative to our DON quota was that we prepared composite samples for replicated entries where possible. Determining DON content on individual rows would be more desirable because of the variability in DON content due to environmental factors. However, harvesting more rows may not be feasible in future due to the reduction in labour at Brandon. It is also possible that some additional DON testing could be done at private laboratories if funding can be obtained from other sources.

There are no plans in the immediate future to use molecular marker assisted selection or other molecular techniques in our project. Researchers in the United States have been putting considerable effort into this in barley. One of the most interesting reports at the 2002 National Fusarium Head Blight Forum in Cincinnati was that a quantitative trait locus (QTL) for reduced DON in Chevron appeared to be syntenous with a major FHB resistance gene found on 3BS in wheat (Schmierer et al. 2002). In Canada, the group at ECORC has been working on identifying

molecular markers for FHB resistance in the Chevron/ AC Stephen DH population. There is also some work going on in Alberta. We hope to be able to make use of molecular techniques in the future. Cost, cross specificity of markers, number of genes involved (some of which are coincident with unfavourable traits), environmental effects, and the need to confirm results in the field with DON testing are some of the main limitations for adopting the technology.

2003 Results:

In the 2003 FHB nursery at Brandon, we planted 16,566 rows and harvested 9,679 of them, and will prepare 5,881 samples for DON testing. Infection levels were moderate to high with early results indicating that DON levels are somewhat higher than those in 2002. There appeared to be continuing problems with some of the susceptible six-row lines having lower DON content than expected relative to the more resistant two-row lines. We were able to handle more than 11,500 rows in the 2003 Brandon nursery because of funding carried forward from ADF. We were nearly 900 lines over our DON testing quota at ECORC, and will evaluate these lines with the NIR or possibly in a private laboratory.

The 2003 FHB nursery at Glenlea was successful in evaluating 800 entries for the project, although visual symptoms were lighter than at Brandon. Infection levels were high at Charlottetown for the 1000 entries evaluated. Nearly all rows in both nurseries were harvested with about 1,000 samples from both nurseries combined being sent to ECORC for DON testing. We look forward to receiving the data from these two nurseries.

Conclusion

We are making progress in improving FHB resistance in barley, but it will be a long-term effort.

Acknowledgments

We wish to acknowledge financial support for the 2000-03 project from the Canada-Manitoba ARDI, WGRF Barley Check-off and Endowment Funds, Saskatchewan's ADF, and AAFC's MII program. We also wish to acknowledge the WGRF Barley Check-off and special funds from interest earned on the WGRF Barley Check-off Reserve Fund and the MII program for providing the financial support to continue our project in 2003-06. We thank Kevin Moore at AAFC Brandon, Mel Ewen and Anic Perrier at ECORC, and technical support staff at all institutions involved in our project over the years for their technical assistance.

Fusarium head blight of oat - current status in western Canada

A. Tekauz, B. McCallum, N. Ames and J. Mitchell Fetch.

Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, R3T 2M9.

Introduction

Previous incidental observations had indicated that oats (*Avena sativa* L.) in Manitoba could be affected by fusarium head blight (FHB) (Clear et al. 1996; McCallum et al. 1999). However, as the disease is rarely visible or recognizable in a standing oat crop (in contrast to barley or wheat, in which damage to spikes, i.e., ‘blighting’, normally is distinct and can be quantified), FHB in oat largely has been overlooked since the contemporary, ongoing FHB epidemic in the eastern prairies began in 1993. Research at the Cereal Research Centre of Agriculture & Agri-Food Canada in Winnipeg during the past two years has attempted to define the scope and impact of FHB on the oat crop in Manitoba. The project has been funded in part by the Manitoba/Canada ‘Agri-Food Research and Development Initiative’, and involved a multi-disciplinary team. The goals have been to:

- 1) survey commercial fields to assess the occurrence and prevalence of FHB in the oat crop;
- 2) evaluate currently-registered oat cultivars for their reactions to FHB;
- 3) compare different methods of measuring FHB in oat; and
- 4) determine the fate of any deoxynivalenol (DON), when oats are processed (milled) for food use.

Materials and Methods

Surveys: Commercial oat fields were surveyed in 2002 and 2003 in southern Manitoba to assess FHB incidence and severity (to determine the FHB Index) by non-destructive sampling of panicles for visual symptoms, and subsequent collection of putatively affected spikelets/panicles for use in laboratory analyses. Seeds threshed from panicles were plated on potato dextrose agar (PDA) growth medium to detect the presence of *Fusarium* spp. and other fungi.

VPT Trials: Oat cultivars were seeded at three locations in southern Manitoba in 2001 and 2002, East Selkirk (producer site), Grosse Isle (MCVET site) and Rosebank (Agricore United site) as ‘Varietal Performance Trials’ (VPT). Plots, replicated three times, were 1m x 5m and comprised of four rows with 30 cm spacing. Following emergence, natural *Fusarium* inoculum was supplanted by spreading corn kernels infested with *F. graminearum* Schwabe, at about 40 g / sq. m. on the soil surface within and around the experimental plots. No irrigation or misting was applied. The total of 15 and 17 oat cultivars tested were as listed in the Manitoba Seed Guides for 2001 and 2002, respectively. Several cultivars of wheat and barley of known reaction to FHB (MR-MS or I, S and VS) also were included in the trials, to compare the relative levels of FHB in oat with that in these crops. Visual estimation of FHB levels in oat was attempted when the disease was evident on spikes of wheat and barley. At maturity, the two middle rows of each plot were hand-harvested and threshed, and the seed used to quantify levels of Fusarium damaged kernels (FDK), *Fusarium* spp., and DON.

Processing tests: A sub-sample of 100 mature kernels were surface-sterilized and plated on PDA, as above, to determine the proportion and identity of *Fusarium* spp. in selected samples from one of the 2001 VPT trials. These were subsequently used to measure the effects of processing on DON levels. Samples were de-hulled (= groats) and steamed and heated in the laboratory in a manner similar to that used by the industry to prepare oat flakes and other edible products. Whole oat samples and groats post-processing were ground to a fine powder and assayed for DON content using ELISA.

As appropriate, data were analyzed for significance using the Ryan-Einot-Gabriel-Welsch Multiple Range Test on arcsine- or log-transformed data, at the $P=<0.5$ level

Results

Based on visual symptoms of FHB on oat panicles, or on heads of wheat and barley, oat had considerably less FHB than the other crops sampled in southern Manitoba in 2002 (Table 1; Gilbert et al. 2003, Tekauz et al. 2003a, 2003b, 2003c). This also was observed in 2003 (Tekauz et al., unpublished data) when overall FHB levels were much lower than in 2002.

Table 1. Severity of FHB in cereal crops in southern Manitoba in 2002 and 2003.

Crop	FHB Index (%)	
	2002	2003
Barley	3.4	0.7
Oat	0.8	0.1
Wheat	Tr - 10.5	1.6
Winter Wheat	4.1	0.6

The *Fusarium* species found on cereals and their levels on kernels sampled from commercial fields in 2002 are shown in Table 2. In oat, *F. poae*, *F. graminearum* and *F. sporotrichioides* were those most commonly isolated. This was similar to what was found in barley, but different from wheat in which *F. graminearum* was the dominant species.

Table 2. *Fusarium* spp. isolated from Manitoba barley, oat and wheat kernels in 2002

<i>Fusarium</i> spp	Percent of fields		Percent of kernels		
	Barley	Oat	Barley	Oat	Wheat
<i>F. poae</i>	76.2	61.8	20.8	37.3	0
<i>F. graminearum</i>	69.1	52.9	44.5	32.3	95.9
<i>F. sporotrichioides</i>	52.4	64.7	26.4	23.4	2.7
<i>F. avenaceum</i>	19.1	26.5	3.5	5.5	0
<i>F. equiseti</i>	16.7	8.8	3.7	1.5	1.4
<i>F. culmorum</i>	4.8	0	1.1	0	0

Visual differentiation of FHB in the oat VPT trials was not possible as only a trace of putative FHB damage was observed both in 2001 and 2002. Usually only a single spikelet on a panicle appeared bleached and was either a light straw-colour, sometimes with an orange cast, when healthy spikelets were green; affected panicles were not found or were rare in any individual experimental plot.

In harvested grain samples, kernels having degrees of light to medium grey or brown discoloration could be found, but such kernels were not distinctive, as are the FDK or 'tombstone' in wheat, and thus were difficult to quantify as FHB-caused FDK. Counts were taken, but the grouping of discolored kernels (putatively FDK) and clean, healthy kernels was largely subjective (Tables 3, 4).

Components of FHB evaluated on harvested seed of 15 oat cultivars in the 2001 VPT trials are listed in Table 3 and contrasted with those of the wheat and barley checks. In oat, average levels of most components were lower than in wheat or barley. Levels of DON (5.6 ppm) were about half those in wheat or barley, and levels of *F. graminearum* (14.3%) and putative FDK (8.5%) lower yet. However, levels of *F. poae* were much higher in oat. In 2002, levels of *F. graminearum* and DON in the 17 oat cultivars tested were much reduced, while levels of *F. poae* and FDK remained similar (Table 4). In wheat and barley, levels of all components were much lower. In contrast to 2001, average levels of total *Fusarium* and DON in oat in 2002 were similar to those in wheat and barley.

Table 3. Fusarium head blight components in oat, and in wheat and barley checks, grown in southern Manitoba in 2001.

Cultivar	Total <i>Fusarium</i>	<i>F. gram.</i>	<i>F. sporo.</i>	<i>F. poae</i>	FDK	DON
Oat (n=15)	%	%	%	%	%	ppm
AC Medallion	37.4 a	17.2 ab	3.1	14.2	9.4 ab	4.6 ab
AC Pinnacle	37.3 a	23.7 a	2.5	8.3	11.6 a	6.0 ab
Dumont	34.8 a	17.2 ab	3.9	9.1	11.4 a	5.5 ab
CDC Pacer	34.1 a	13.5 ab	1.4	15.1	8.7 ab	5.3 ab
AC Gwen	32.9 a	12.8 ab	3.9	15.1	8.1 ab	5.9 ab
AC Ronald	28.8 ab	11.2 ab	1.2	14.3	7.8 ab	3.8 ab
Riel	28.8 ab	18.0 ab	2.8	6.2	9.1 ab	5.2 ab
CDC Boyer	27.8 ab	14.9 ab	0.8	9.2	9.0 ab	8.8 a
AC Rebel	26.6 ab	15.3 ab	1.9	7.5	8.8 ab	7.7 a
AC Preakness	26.0 ab	12.4 ab	2.3	8.9	8.6 ab	4.3 ab
Robert	25.4 ab	17.1 ab	1.7	4.8	9.0 ab	7.6 ab
SW Exactor	24.7 ab	12.5 ab	0.9	9.9	5.7 b	5.1 ab
AC Assiniboia	23.9 ab	11.6 ab	0.7	10.7	8.3 ab	5.6 ab
Triple Crown	22.7 ab	9.8 ab	1.5	10.4	5.2 b	6.3 ab
AC Belmont	13.8 b	7.0 b	0.9	5.3	6.7 ab	2.7 b
Average	28.3	14.3	2	9.9	8.5	5.6
Wheat (n=4)						
Roblin (VS)	54.2	47.7	3.1	1.4	29.1	11.5
CDC Teal (VS)	51.2	48.8	2.6	0.2	31.0	16.7
AC Barrie (I)	35.9	33.2	1.2	1.0	21.5	9.3
AC Cora (I)	30.3	29.6	0.1	0.4	22.5	7.0
Average	42.9	39.8	1.8	0.8	26	11.1
Barley (n=3)						
AC Lacombe (S)	54.9	48.8	1.9	1.8	24.5	9.3
Mahigan (VS)	53.2	45.6	3.3	1.0	14.1	20.2
CDC Stratus (I)	30.9	23.8	1.0	1.6	12.2	4.0
Average	46.3	39.4	2.1	1.5	16.9	11.2

- Values based on the mean of three environments.

- For total *Fusarium*, *F. graminearum*, FDK and DON in oats, values in a column followed by the same letter are not different from each other at $P < 0.05$.

Table 4. Fusarium head blight components in oat, and wheat and barley checks, grown in southern Manitoba in 2002.

Cultivar	Total <i>Fusarium</i>	<i>F. gram.</i>	<i>F. sporo.</i>	<i>F. poae</i>	FDK	DON
Oat (n=17)	%	%	%	%	%	Ppm
AC Ronald	23.1 a	3.0 ab	6.8	13.0	12.6 ab	1.2 ns
Dumont	22.4 ab	2.6 abc	4.6	13.4	9.8 abcd	1.2
CDC Pacer	19.6 abc	2.4 abc	2.5	13.6	7.9 bcd	1.3
Triple Crown	19.2 abc	2.2 abc	4.8	11.7	9.1 abcd	1.5
AC Rebel	19.1 abc	4.2 a	4.8	8.9	9.5 abcd	1.2
AC Preakness	18.7 abc	2.7 abc	3.2	11.4	12.3 ab	1.3
AC Medallion	17.0 abc	1.8 abc	3.2	10.2	10.1 abc	1.4
OT 2009	17.0 abc	2.8 ab	5.0	8.2	11.1 ab	1.4
SW Exactor	16.7 abc	2.7 abc	3.3	9.4	8.7 bcd	1.7
Riel	15.6 abc	2.7 abc	3.3	8.3	8.6 bcd	1.4
AC Assiniboia	15.1 abc	3.2 a	3.8	7.5	9.4 abcd	1.3
AC Pinnacle	14.9 abcd	2.4 abc	2.5	8.3	10.8 abc	1.2
Robert	13.9 abcd	3.5 a	3.3	6.3	15.9 a	1.2
CDC Boyer	13.6 bcd	4.0 a	2.7	6.6	9.2 abcd	1.2
OT 7008	11.7 cd	0.5 bc	3.7	7.0	5.0 d	1.5
AC Gwen	10.5 cd	0.3 c	2.4	7.3	8.7 abcd	1.4
AC Belmont	7.5 d	1.1 abc	0.5	5.8	6.6 cd	1.3
Average	16.2	2.5	3.6	9.2	9.6	1.3
Wheat (n=4)						
Roblin (VS check)	19.5	13.0	3.9	0.4	11.7	1.6
AC Barrie (I)	8.6	5.4	1.3	0.9	4.9	0.9
CDC Teal (VS)	8.3	4.9	1.8	0.7	7.3	0.8
AC Cora (I)	8	4.9	1.7	1.1	6.6	0.9
Average	11.1	7.1	2.2	0.8	7.6	1.1
Barley (n=3)						
Mahigan (VS)	21.9	14.6	5.4	1.2	10.2	1.6
AC Lacombe (S)	15.5	6.3	6.0	2.4	10.0	0.5
CDC Stratus (I)	6.4	2.0	1.3	2.9	6.7	0.3
Average	14.6	7.6	4.2	2.2	9.0	0.8

- Values based on the mean of three environments.

- For total *Fusarium*, *F. graminearum* and FDK in oats, values in a column followed by the same letter are not different from each other at $P < 0.05$; DON levels are ns.

Based on disease components measured in 2001 when FHB levels in the VPT trials were relatively high, allowing for more differentiation among lines, the hulless oat cultivar, AC Belmont, appeared to have the best ‘resistance’ (lowest DON and *F. graminearum* levels) of the oat cultivars tested. Several cultivars had higher levels of DON, *F. graminearum* or total *Fusarium* but without the apparent consistency to suggest that any of these are most susceptible to FHB. Levels of FHB in 2002 were relatively low and there were minimal apparent reaction differences among oat cultivars. This also was the

case with the wheat and barley check cultivars of known reactions ranging from ‘very susceptible’ (VS) to ‘intermediate’ (I or MR-MS).

The levels of *Fusarium* and DON in selected 2001 VPT samples (reps) of whole oats and heat-treated groats are shown in Table 5. Compared to whole oats, laboratory de-hulled oats (groats) when subsequently heat-treated had much lower levels of DON (average of 8.7 vs. 1.2 ppm). Individual levels were sometimes reduced to <1.0 ppm. There was an apparent lack of correlation between *Fusarium* levels and those of DON in whole oats. In individual samples, similar levels of *F. graminearum* (e.g., 22 and 25%) sometimes resulted in disparate levels of DON (3.5 ppm, 24.6 ppm).

Table 5. Levels of *Fusarium* and deoxynivalenol in whole oats and heat-treated groats from Grosse Isle MB in 2001.

Cultivar	Total <i>Fusarium</i>	<i>F. graminearum</i>	DON	DON
	(%)	(%)	(ppm)	(ppm)
			Whole oats	Heat-treated groats
AC Pinnacle	52	37	10.0	1.1
AC Gwen	38	22	3.5	1.7
CDC Boyer	36	27	24.6	1.7
Robert	33	31	18.5	0.7
AC Ronald	33	23	5.2	0.8
AC Medallion	26	17	7.2	1.4
Dumont	24	17	9.2	2.7
Riel	20	17	6.9	1.1
SW Exactor	20	14	5.6	0.6
AC Preakness	18	16	7.1	1.0
CDC Pacer	14	9	4.0	0.6
AC Rebel	13	10	6.1	0.8
AC Assiniboia	12	10	11.3	0.7
Triple Crown	11	5	9.7	1.5
AC Belmont	9	6	2.1	1.3
Average	23.9	17.4	8.7	1.2

Discussion

Fusarium head blight was found in most commercial crops of oat surveyed in southern Manitoba in 2002 and 2003 (Table 1; Tekauz et al. 2003b, Tekauz et al., unpublished data). Based on visual field symptoms, FHB did not affect oat as severely as wheat or barley and the low average values (FHB Index <1.0%) would suggest the disease does little damage to the crop. This also was the case in the VPT trials of 2001 and 2002, where visual plot symptoms were essentially non-existent; however, the levels of the various FHB components measured on harvested seed, while lower than in wheat or barley, indicated considerable FHB damage had occurred. This was particularly so in 2001 when resulting DON values were as high as 8.8 ppm. In the 2002 VPT trials, while FHB damage in oat was relatively light, it was similar to that in wheat and barley. These observations indicate that FHB is an important disease of oat in Manitoba, but one which can be grossly underestimated unless laboratory test are done.

Estimation of severity of FHB in oat can best be accomplished by evaluating levels of DON, and possibly those of *F. graminearum* on seed. The lack of definitive visual symptoms makes field estimation of severity undependable, and estimation using FDK unreliable. The unreliability of the latter is demonstrated by the similar average FDK values obtained for the VPT trials of 2001 and 2002, despite the large differences in objectively measurable FHB components such as *F. graminearum* and DON.

The role of *F. poae* as a causal or contributing agent of FHB in oat is unclear. This species was commonly isolated from harvested oat seed, often at higher levels than *F. graminearum*. In the VPT trials this occurred despite the application of supplemental *F. graminearum*-infested corn inoculum. In barley, *F. poae* can incite FHB, but is less pathogenic compared to *F. graminearum* (McCallum and Tekauz, 2003).

Heat treatment of groats reduced DON levels appreciably, suggesting that the combination of de-hulling whole oats and their subsequent 'processing' removes and/or destroys much of the mycotoxin present. Refinements in processing may further reduce mycotoxin to non-detectable levels in both laboratory- and commercial-scale situations.

Breeding for improved resistance to FHB in oat should be pursued. Based on these trials, it is unlikely that a highly resistant source(s) for use in crosses will be found among domestic oats currently available in Canada. A wider pool of germplasm of cultivated oats, as well as wild relatives, will require screening in inoculated and water irrigated FHB nurseries to identify promising resistance to transfer to adapted oat lines through hybridization.

The technical assistance of Eric Mueller, Marcos Stulzer, Meconnen Beyene and Camille Rhymer is gratefully acknowledged.

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Haplotype diversity at Fusarium Head Blight resistance QTLs in wheat.

C.A. McCartney, D.J. Somers, G. Fedak, and W. Cao. (C.A.M. and D.J.S.) *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB R3T 2M9, Canada; and (G.F. and W.C.) Eastern Cereals and Oilseeds Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON K1A 0C6, Canada.* Fusarium head blight (FHB) reduces grain yield and quality in common and durum wheat. Host FHB resistance is an effective control measure that is achieved by stacking multiple FHB resistance genes. Resistance gene stacking would be facilitated if breeders knew which FHB resistance sources carry different resistance genes. A diverse collection of FHB resistant and susceptible wheat lines was characterized with microsatellite markers linked to known FHB resistance quantitative trait loci (QTLs) on chromosomes 2DL, 3BS (distal to the centromere), 3BSc (proximal to the centromere), 4B, 5AS, and 6BS identified in Maringa, Sumai 3, and Wuhan 1. Putative Sumai 3 QTLs were commonly observed in advanced breeding lines, whereas putative Maringa and Wuhan 1 QTLs were relatively rare. The microsatellite data suggested that the 3BS, 3BSc, and 5AS QTLs in the Brazilian cv. Maringa were derived not from Frontana, as previously thought. Maringa appeared to be closely related to Asian germplasm at the 3BS, 3BSc, and 5AS QTL regions. Other Brazilian wheat lines did not appear closely related to other FHB resistance sources. These Brazilian wheats may have novel FHB resistance that will be useful for stacking with FHB resistance derived from Asian germplasm.

Session 4: Host Resistance Genetics

From QTL mapping to breeding to production. D.J. Somers. *Agriculture and Agri-Food Canada-Cereal Research Centre, 195 Dafoe Rd, Winnipeg, MB R3T-2M9, Canada.*

Fusarium Head Blight (FHB) is the most important wheat disease challenge today and unfortunately, the inheritance of resistance is genetically complex. Genetic resistance to FHB is complex because multiple genes are required, there are likely gene-to-gene interactions, genetic background influences resistance gene expression and there is environmental influence on gene expression. Resistance genes are detected in Asian and Brazilian wheat, but these accessions are very poorly adapted to Canadian growing conditions and are far from Canadian quality standards.

In spite of these complications, current genetic studies and molecular breeding tools facilitate a high degree of optimism that marketable Canadian wheat will be produced with FHB resistance. This presentation will provide an overview of FHB resistance gene mapping in the literature over the last 10 years and how that information is used to consider breeding strategies. Molecular breeding for complex traits is becoming a reality and FHB resistance is a good candidate trait to experiment on. AAFC initiated an aggressive FHB resistance molecular breeding program in April 2002. Lines have been advanced to the BC2F2 level and are ready for testing as homozygous material. An update on this projects progress will provide insight into accelerated breeding and suggest future research.

Transformation to provide new genes for FHB resistance: A summary of current US public research. L.S. Dahleen. *USDA-ARS, Cereal Crops Research Unit, PO Box 5677, SU Station, Fargo, ND 58105, USA.*

Fusarium Head Blight (FHB), caused by *Fusarium graminearum* (Schwabe) has been a serious disease in wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), and durum (*Triticum durum* L.) since the 1990's. The most economical method of control is to develop resistant cultivars. In addition to resistance being bred in through traditional methods, transformation to insert new genes has the potential to increase FHB resistance. This presentation describes progress on public transformation projects funded by the US Wheat and Barley Scab Initiative, including those at several USDA-ARS laboratories, at the Universities of California, Minnesota, and Wisconsin, and at Montana State University. Approaches include insertion of single or combinations of antifungal and antitoxin genes, use of spike-specific promoters, various systems to remove plasmid and marker genes, development of antibodies to test for gene products, and field and greenhouse tests to determine the effectiveness of each gene. Cooperation between laboratories has been essential to avoid duplication of efforts and maximize progress in developing FHB-resistant germplasm with the limited resources available.

Genomics studies to understand the interactions between *Fusarium graminearum* and its cereal hosts. Thérèse Ouellet, Linda Harris and Steve Gleddi, *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa*

Understanding how cereals defend themselves against *Fusarium graminearum* attack and how many strategies *Fusarium* uses to get around the plant defenses is needed to develop durable disease resistance in the cereal crops. Two main molecular approaches are being used to identify and characterize genes from cereals that are associated with the response to *Fusarium graminearum* attack, genetic mapping and study of expressed genes. Progress by many laboratories to understand which expressed genes/pathways in wheat and maize are involved in defense against *Fusarium* will be summarized. Progress in understanding the molecular aspects of the strategies used by the fungal pathogen, *Fusarium*, to attack and invade its hosts will also be presented.

Session 5: Epidemiology

Predicting deoxynivalenol in wheat for Ontario. D.C. Hooker, and A.W. Schaafsma. *Ridgetown College, University of Guelph, Ridgetown, ON, Canada NOP 2C0*

Eight years of data were used to develop DONcast– a model to predict deoxynivalenol (DON) in mature wheat grain for fungicide-spray decisions at heading. A website was launched in 2000 for providing DON predictions to agri-business through the Ontario Weather Network (OWN) (<http://www.ownweb.ca>). The model was adapted for Uruguay, where severe *Fusarium* epidemics have resulted in DON concentrations of up to 5 ppm in baked goods. For predictions in 2004, DONcast will have evolved using an array of weather and agronomic data from over 630 private farms across Ontario and Uruguay. In addition to daily rainfall and temperature data, DONcast for 2004 will include relative humidity (RH) >80% at 11:00 between 3 to 10 d after heading for more accurate decisions of whether or not to apply a fungicide at heading. For the first time, the model will also be extended to include rain and RH between 20 and 36 d after heading (near harvest). Using actual weather and agronomic variables specific to individual farm fields, the overall model explains 75% of the variation of DON using data from 600 farm fields from 1996 to 2003. DON concentrations of less than 1.0 ppm were predicted correctly on 88% of the fields at heading. In other fields where DON concentrations exceeded 1.0 ppm, the model predicted correctly on 72% of the fields at heading.

Progress in forecasting: FHB risk forecasts in Manitoba. David Kaminski, *Plant Pathologist, Manitoba Agriculture and Food*

Manitoba Agriculture & Food and the Agrometeorological Centre of Excellence (ACE) collaborate on several disease risk forecasts one of which focuses on Fusarium Head Blight in wheat. The forecast is based on weather data collected from a network of stations (about 50 in southern Manitoba) and it depicts risk graphically on a regional basis.

In 2002, a new model was employed in the FHB Risk Forecast in an effort to reflect the dynamic nature of infection-conducive environmental conditions and to remove the constraints of an average seeding date. Daily web-posting of the Risk Forecast maps keeps farmers and agronomists aware of risk as crops approach the vulnerable flowering stage. A field study to verify predictability was conducted in commercial fields in a range of agro-ecological zones. That study emphasized the importance of high humidity as a key factor in disease development. The performance of the model in 2003, a year of much lower FHB incidence, will be discussed.

Infection of tolerant and susceptible wheat and barley varieties using GFP-Fusarium. S. Shea Miller, Denise Chabot and Nadia McGoldrick, *Eastern Cereal and Oilseed Research Centre, Agriculture & AgriFood Canada, Ottawa, ON, Canada, K1A 0C6.*

Although the general progression of *Fusarium* infection is known, details concerning the initiation of infection, and spread in specific tissues are unclear. To look at the infection process in wheat and barley, we have been using a strain of *Fusarium graminearum* which was transformed to express the green fluorescent protein (GFP) from jellyfish. Using this GFP-transformed strain, which fluoresces when illuminated with blue light, we are able to use the fluorescence microscope to visualize the fungus *in situ*, on living tissues as well as sectioned material. We have studied both susceptible and tolerant cultivars of wheat (Roblin and Sumai3) and 6-row barley (Chapais and Chevron), in hopes that we will be able to learn more about mechanisms of resistance to the fungus. For both crops, we have used a point inoculation protocol, as this allows us to more easily follow the progression of the fungus from the initial infection site. In wheat and barley, our results indicate that although the spores will germinate on any tissue, the pollen and anthers are major targets in the initial stages of infection for both the resistant and susceptible varieties, with the fungus quickly progressing to the soft tissues of the ovary and then spreading from there. After the fungus moves down the ovary, it infiltrates the node and moves into the rachis. Only after the soft tissues of the floret are consumed does the fungus really start to move beyond the inoculated floret

In wheat, the fungus spread both internally and on the external surface of the spike. Internally, the fungus appears to spread down the rachis from the point of infection, rather than up. The fungus spreads much earlier and more extensively in the parenchyma and vascular tissues in the rachis of the susceptible variety (Roblin) than in the resistant variety (Sumai 3). Although we observed differences in the responses of the two wheat varieties to infection at the macroscopic level, we were unable to determine the cause of the differences microscopically. The fungus spread further, and was more damaging to the adjacent florets in Roblin than in Sumai 3. Both wheats exhibited the well known bleaching of the head above the infection site. Our results suggest that this is due to occlusion of the vascular bundles of the rachis, effectively shutting off the nutrient supply to the spike above the infected floret.

In barley, very little difference was observed in the progression of the disease in the susceptible (Chapais) and tolerant (Chevron) varieties by visual assessment. This is probably due in part to the very small sample size. In addition, the spray inoculation technique has been reported to be more effective in distinguishing differences in FHB resistance among barley cultivars (McCallum and Tekauz, 2002). As is consistent with published observations on *Fusarium* infection in barley, bleaching was generally confined to the inoculated floret, and did not affect the whole head. As in wheat, the fungus spread in the spike both externally and internally. Externally, the fungus would grow out of the top of the inoculated floret, and proceed up or down the rachis along the outside. It tended to accumulate in crevices, and areas with a higher density of trichomes, and often would penetrate into the rachis or node from these sites, as well as growing into uninoculated florets. Internally, the fungus moved from the base of the infected floret into the vascular bundles of the node, and down into the rachis. Typically, the infection travelled farther, faster inside the rachis in Chapais than in Chevron, but in general it was

not sufficient to restrict nutrient supply to the rest of the spike. In the case of the inoculated node itself, Chevron appeared to be more heavily infected, but appeared to resist progression of the hyphae down into the rachis.

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McCallum, B.D., and Tekauz, A. 2002. *Influence of inoculation method and growth stage on fusarium head blight in barley*, Can. J. Plant Pathol., Vol. 24, p. 77-80

Mechanisms of resistance and tolerance to FHB

A. Comeau¹, F. Langevin², F. Eudes³. ¹ Agriculture and Agri-Foods Canada, Ste-Foy, GIW 2B1 ; ² 121 Bon-Air, Ste-Catherine-de-la-Jacques-Cartier, G0A 3M0; ³ Agriculture and Agri-Foods Canada, Lethbridge, T1J 4B1.

Abstract

Correlation between FHB resistance data sets is too often unsatisfactory. However, the diverse types of resistance genes interact with environmental factors according to precise rules, and once those rules are understood, one can improve the methods, and draw more useful conclusions. For gene pyramiding goals it is becoming important to understand the G x E interactions. This can provide clues about which potential parents possess complementary traits. Resistance mechanisms involve a large number of genes that can be classified in many different ways. There is still some mystery about what are the exact underlying mechanisms grouped under the terms: type 1 (resistance to infection, abridged Rtype1), type 2 (resistance to spread, or Rtype2), and other types. Rtype2 is generally correlated with resistance type 3 (destruction of toxins) and 4 (tolerance to toxins). Factors that reduce economic damage without reducing fungal growth are recognized as tolerance factors, and many are morphological in nature. Symptoms above the infection relate more to yield loss and those below that point, to downgrading factors. The role of trichothecene in the infection process is major, but other factors exist for durum wheat. The list of traits, markers and genetic factors that may relate to resistance is a very long one. All of the "end-result" traits like yield, grain class, kernel weight, hectoliter weight, cleaning loss, and toxin content interact with the field conditions, and are less informative about mechanisms. Phenomics, genomics and metabolomics and also test-crossing in search of transgressive segregants are important approaches to replace the more random approaches to gene pyramiding.

Introduction

Our understanding of the mechanisms of resistance and tolerance of plants to pathogens remains minimal for many major diseases. The endeavor of deciphering those mechanisms could be justified by an academic interest. But when dealing with the Fusarium Head Blight (FHB), there are more compelling reasons that justify scientific interest, with practical applications in mind. The goal of geneticists is described as resistance to FHB, but it is understood that there are also many genetic traits that reduce economic damage without reducing fungal growth, and these should properly be identified as tolerance traits. A resistance trait *sensu stricto* must reduce the growth and spread of the pathogen. Many tolerance factors are morphological in nature. Some genes may improve both resistance and tolerance.

Tests of resistance to FHB are notoriously variable among years and sites. Correlation between FHB resistance data sets in Canada and USA ranges from near zero to approximately 0.80 ; the most frequent values are in the 0.40-0.55 range. Correlation between toxins line deoxynivalenol (DON) and symptoms can be weak, often enough below $r = 0.5$. This is enough to allow a slow progress, and yet repeatability is bad enough to cause problems. In fact, a good degree of uncertainty about the value of the FHB resistance or tolerance genes still exists when a line reaches the registration level.

Considerable research funds have been spent on FHB in USA and Canada. And yet, progress in breeding for resistance and tolerance has been slow, while the geographic area where the disease can manifest itself has increased inexorably. Agronomic breeding lines with the same level of resistance as the resistant checks are very difficult to obtain. In Canada, the best available resistant cultivars contain for now only medium resistance. Serious epidemics could still happen in USA and Canada within the next five to ten years.

Thus, it is hoped that in-depth understanding of the diverse existing mechanisms of resistance and tolerance (and of the environmental parameters that can modify the efficiency of those mechanisms) might bring new insight about how to pyramid genes so as to create more efficiently cultivars that have the highest resistance.

Variability factors associated with FHB resistance evaluation

Type 1 resistance mechanisms are easily confused with variations caused by the environment or testing conditions. Field tests of FHB resistance done on the same breeding lines at many sites, or at the same site for many years, show an annoying lack of repeatability. With point inoculations, auxiliary tests done indoors show less variability, but spray inoculations are more unpredictable. In the field, ascospore production can be increased by the use of spawn (often corn inoculum), but this cannot make the ascospore release uniform day after day, and variations in post-anthesis conditions can create major effects. Spraying inoculum in the field is also practiced; for best results this should be done at anthesis, which represents a large task. Even then, the spray equipment cannot mimic the exact deposition patterns caused by the wind, rain and rain splatter, and by insects such as the wheat midge, aphids, mites etc. Rainstorms before flowering increases ascospore release (Fernando et al. 2002), and at flowering, moisture, heat and dew increases spike contamination and spore germination. After a spore has germinated and infected the plant tissue, the climate may still modify fungal growth speed and patterns. All of the above details of test protocol and climate may thus affect: 1) the overall rate of deposition of spores, 2) the pattern of spore deposition on different organs, 3) the rate of survival, germination and successful entry, 4) the infection routes, and 5) the speed of invasion of tissues, and damage levels.

Some of the differences among data sets may thus be due to details in the field test conditions, and in the testing protocols, which are not identical at different sites (Table 1).

<u>Table 1. Sources of variation in FHB testing</u>	-- Notes --
Protocol related	
Indoors tests	
Point inoculation	
Spray	More repeatable
Field tests	Not so easy to master
“Spawn” (corn inoculum) + (with planting dates) (with misting)	More natural More natural Uniformity not easy to obtain
Spray inoculation :	
at specific dates over all plots	
at mid anthesis for each pure line	Time consuming; not ideal for bulks
Spawn plus sprays and misting	Flowering date may still alter results Overkill possible
In any type of test :	
resistance criteria may alter conclusions	
date of observations may alter conclusions	Symptoms not fully reliable Symptoms not fully reliable
Climate, environment related	
Rain before anthesis	
Hot humid climate at anthesis	Increases ascospore release
Hot climate after anthesis	Increases ascospore germination
Hot, humid climate at/after anthesis	Accelerates spread
Strong sunshine with UV, dry heat	Increases routes of spread
Vector insect activity	Kills exposed ascospores
Cool climate	Increased by low wind, less rainy days Reduces infection and spread

All this being said, even within a single test in which none of those factors matters, there is a readability window problem. Indoors, the first symptoms of FHB become visible in 3-7 days, but differences between lines tend to increase till the 14th day or so. The contrast between resistant and sensitive can then be observed easily for one week or one week and a half, till ripening begins. In the field, the early symptoms are more difficult to detect, and due to within-plot variability of flowering and ripening dates, more difficult to separate from ripening. Thus, within a given cultivar, the ideal period to read symptoms may last no more than 7 to 12 days, and under hot conditions this ideal period can become shorter. Since some early cultivars and lines flower about one week before the late ones in practice, the ideal observation date for the late flowering lines is too late for the proper evaluation of the early flowering material.

One example of "readability" problem is shown in the fig 1. The correlation between the two reading dates was 0.598, in a test in which added corn inoculum and natural rainfall led to heavy FHB. Problems were not encountered when comparing extremes, i.e. the resistant and sensitive checks (Alsen as R, and Kohika and Abbey as S). However, with

the lines that have intermediate levels of resistance, there is sometimes a noticeable difference between the two dates. Relative to the group of lines evaluated, AC Voyager and AC Reed seemed much more resistant on Aug. 29 than on Aug 14, and AC Barrie seemed much more resistant on Aug 14 than on Aug 29. In general, a wider spectrum of flowering dates reduces the correlation between the first and last symptom data sets within a given trial. Frequent observations increase the value of conclusions. The calculation of AUDPC (area under disease progress curve) (Buerstmayr et al. 1997) brings another answer to the problem, but is costly.

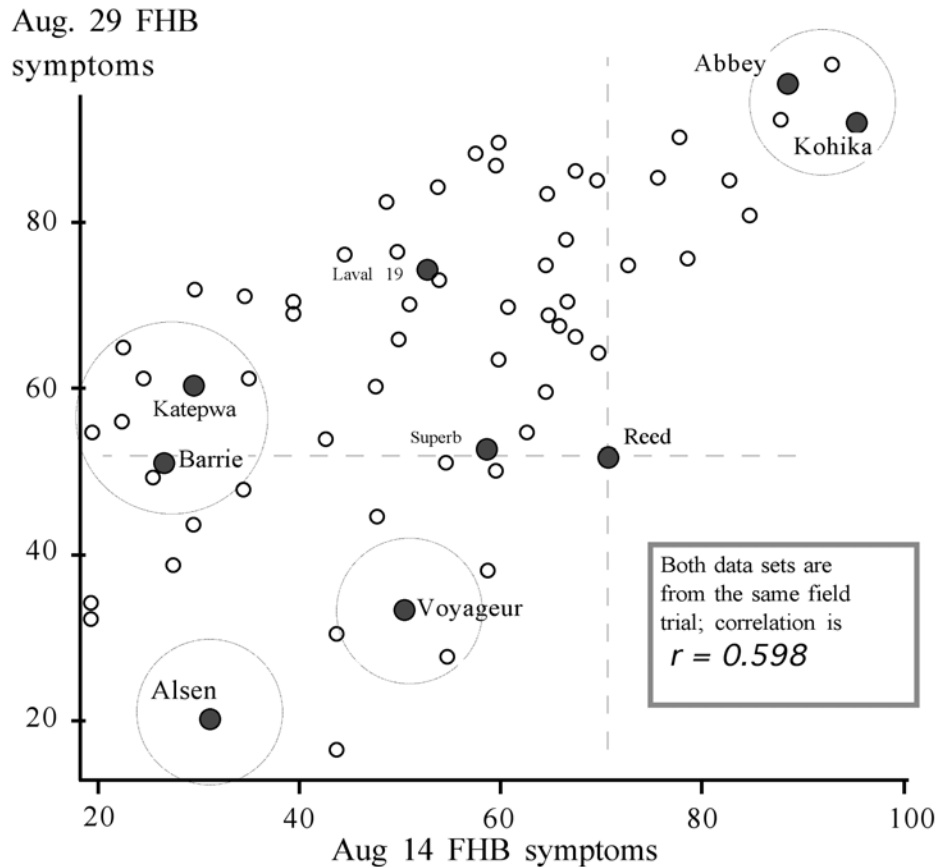


Fig. 1. Visual FHB symptoms taken at two dates, in Quebec, 2003, for candidate lines from Western Canada and local checks (R and S). Kohika and Abbey are used as sensitive checks. The apparent performance of some resistant checks may vary according to reading date. The visual estimate is based on (percent infected spikes) x severity.

The symptomatology approach may be flawed to some extent, because there is tissue-specific resistance, and symptomatology can become biased in favor of glume and rachis resistance unless one takes time to inspect inside the florets, which is seldom done. Thus breeding lines that have good pericarp resistance but bad glume resistance would be downgraded based on field symptoms, and those that have, like triticale, rather good glume resistance but bad pericarp resistance would be judged more resistant than they

truly are. Lower correlation between field symptoms and FDK (Fig. 2) or DON may result from this.

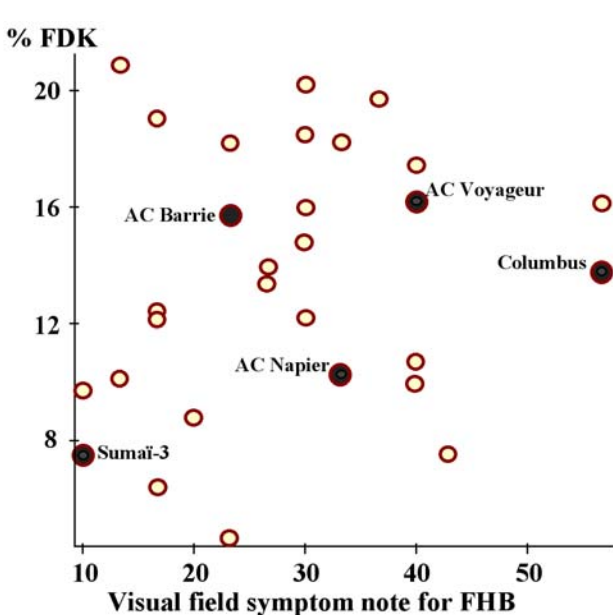


Fig. 2. Data from the CRAAQ provincial trial in Lévis, Qc, 2003, on the field symptoms (based on severity and incidence) vs visual estimate of *Fusarium* diseased kernels (FDK). Checks are identified. This trial includes lines that seem close to Sumai 3, based on both parameters, and yet a low correlation between the parameters (0.263) is observed. The trait ANOVAs were highly significant (FDK, $F=6.88$; symptoms, $F=8.89$). Data by Langevin.

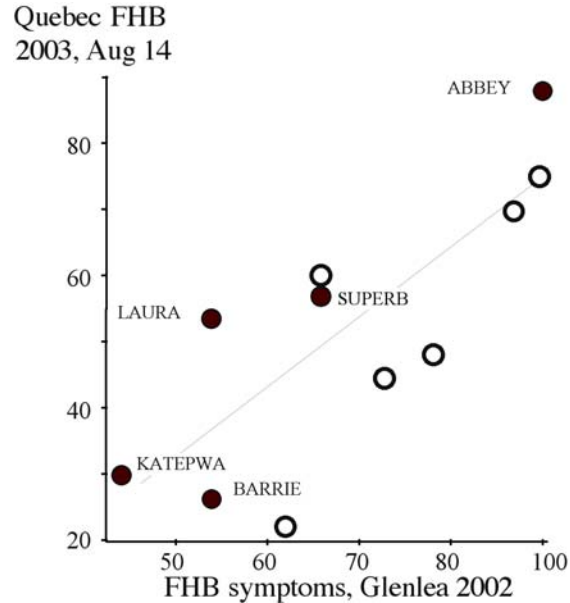


Fig. 3. Comparison of visual symptoms in Glenlea 2002 vs Quebec (Lévis) 2003 ($r = 0.793$), showing that good correlation is possible between locations geographically far apart (Glenlea data by Gilbert ; Lévis data by Langevin). The symptom value is based on percent infected spikes x severity

The geographic location where the test is conducted seems much less important than the climatic variables of temperature and rainfall. Correlation between Western data and Eastern data, for example, can be as good as correlation obtained within a narrow geographic area; in one example shown, it reaches 0.793 (fig 3). This is better than the correlation within one same trial in Quebec for two different reading dates (Fig 1).

Discrepancies between indoors point inoculations and field data can be major because the point inoculation measures the type 2, 3 and 4 resistance complex. In indoors trials related to the lines in Fig 1, the point inoculation of Reed showed this line had good Rtype2, slightly inferior to that of Alsen or Sumai 3. The field-resistant cultivar Barrie, on the contrary, had a speed of *Fusarium* spread almost as rapid as in the sensitive checks Abbey and Kohika. Many sources of Rtype1 share this problem observed in Barrie. Ste-

Foy cultivars and lines like Voyageur, Brio, Drummond, QG 22.24 have a remarkably low level of Rtype2, but good to very good Rtype1. Lines with an incomplete package of resistance factors vary in behavior between years and sites. In the worst epidemics, Rtype1 and Rtype2 are both essential.

Insects and mites play a role in the development of epidemics (Parry et al. 1995). The wheat midge is active in conditions of very low wind intensity and absence of rain, and midge-borne spores can lead to infection even in absence of rain (Mongrain et al. 2000). It is a dangerous extrapolation to assume that dissemination of spores by insects, dry wind, rain, rain splatter leads to spore deposition in the same area and same organs of the spike regardless of agents of dissemination. As to whether a spray with artificial inoculum can perfectly imitate the diverse ways of natural transport of spores into the spike, that also seems unlikely. Attacks initiated on the most sensitive organs (anthers > glumes > rachis) (Pearce et al. 1976) might progress faster and inflict more damage. Attacks in the lower part of the spike causes more yield losses. Attacks in the upper part does not kill as many florets, but is more likely to increase DON. The efficiency of certain Rtype1 genes might thus relate to preferential spore deposition patterns related to the agents of dissemination.

Quantification of genetic differences in resistance and tolerance

Breeders tend to select by “end-result” methods. The end result of FHB is yield reduction, loss of hectoliter weight, 1000 kernel weight loss, infection by *Fusarium* in a certain percentage of the seeds (confirmed by laboratory tests), visual disease symptoms on the seeds, and toxins in the grain after a given cleaning procedure.

Pathologists often use symptomatology at the milk or early dough stages for evaluation and selection. This includes readings of general appearance integrating all visual traits observed, but also readings in which there may be separate notes taken about the percent spikes infected and the severity of the diseases in most spikes. It is also possible to evaluate the relative contributions of Rtype1 and Rtype2 genes through inspecting the fine details of the infection process at many stages within defined testing protocols (Langevin, unpubl.).



Fig. 4. Different responses to point inoculation in the middle of the spike (in.: inoculation).

It was observed that some sensitive lines or cultivars tend to have more symptoms above the infection points than below those points (Fig. 4, spike A) (Bai and Shaner 1996). Savard found that in the sensitive wheat cv. Roblin, there is neither fungus nor toxin above the infection point, although bleaching occurs rapidly. Damage to phloem and xylem causes the whitening and death of tissues (Ribichich et al. 2000). Vessel blocking reduces the yield, since grains die above the infection point. Below the infection point, there is spread of fungus and toxin. In one case, symptoms (Fig 4, spike B, lowest arrow) went further down than the fungal hyphae (Fig 4, spike B, red dot), and this may indicate that toxins paved the way before fungal invasion occurred. *Fusarium*-contaminated grains (likely with toxin) were found mostly below the infection point.

This aspect of the epidemiology brings the loss of quality and the health risks.

Triticale does not bleach so easy, except in severe FHB epidemics. And yet in this species, in a cool year, toxins build up in sensitive lines, despite low symptom levels, as proven by Devaux in 1986-90. In warm humid years, more visible damage to the glumes and rachis can be associated with very severe damage to the grain. Spread of toxins and fungus above an infection point probably exists in triticale. Some fungal growth above an infection point may occur in cereal germplasm that resists phloem blocking.

Schroeder and Christensen (1963) noted that hyphae invade conductive vessels before touching the surrounding tissue. In indoors trials with point inoculation, wheat lines in which the vessel blocking does not cover the full cross section of the rachis were found. Spikelets or flowers on one side of the spike can be sick, and those on the other side, healthy (Langevin, unpubl.). This was repeatable, it represents a form of resistance to radial spread, heritability can be good, the usefulness of the mechanism is worth investigating.

True bleaching is therefore due to photosynthate and mineral starvation, and occurs without presence of fungus in the killed tissue. This is not always easy to identify. The bleaching-like symptoms above the inoculation or infection point being unreliable in triticale, one must raise questions about their reliability in wheat. Some wheat lines could behave somewhat like triticale, at least under certain climatic conditions; in such cases, the casual symptom notation could tend to underestimate the true sensitivity of the line. Some lines with low symptoms in the glumes and rachis can still produce sick looking grain (upper left dot, Fig 2) and contain objectionable toxin. And yet, frequent and thorough inspection of the spikes may often reveal useful details about infection and resistance patterns.

In interspecific hybrids, near-hypersensitive responses can be found, and are characterized by black spots near the infection point (Langevin and Comeau 2001). Very resistant lines like Sumai 3 occasionally have similar symptoms. The fungus is often killed within such black lesions. However, it is not possible to consider black spot symptoms as a sign of high resistance to FHB, since many species of fungi cause black spots.

Mechanisms of resistance : definitions by types

Resistance types are defined based on easily observable effects of resistance, sometimes regardless of precise mechanisms. Schroeder and Christensen (1963) defined resistance to initial infection (Type I) and resistance to the spread of the infection within a plant (Type II). Attention was not paid to the difference between the type of damage above and below the infection point, in any of those definitions. Complex questions arise as to the need of restricting the type 1 definition solely to passive mechanisms, thus excluding any active mechanisms that control infection in its earliest steps.

Other mechanisms were given type numbers by different authors. But their numbering systems involve significant differences in the definitions, which can be confusing. In the current text we adopted the following definitions. The ability to inactivate or degrade trichothecenes like DON is defined as Type III resistance, and a capacity of the cell to tolerate trichothecenes, type IV resistance (Miller *et al.*, 1985; 1986; Wang and Miller,

1988). Type V resistance to kernel infection (Mesterházy, 1995) is measured by threshing infected spikes and observing the damage to the kernels. Type VI, or tolerance to FHB (Mesterházy 1995; Mesterházy *et al.*, 1999), is a concept that deserves more attention.

Mechanisms of resistance and tolerance : morphology and phenology

Observing that a genotype suffers from fewer points of infection does not explain how this Rtype1 is achieved. Observing that a genotype has slower spread of disease also does not explain how Rtype2 is achieved. A search for mechanisms implies an attempt to identify the underlying causes.

Without reference to any of the above six types of resistance/tolerance, the basic reasons for resistance and tolerance may belong to three categories: a) phenology and b) morphology, which could play a role in some mechanisms, and finally c) chemical-biochemical factors. In the study of a wheat line, it is not always possible to separate those categories (a, b,c) in practice, just like it is not possible to separate certain R types like types 2, 3 and 4 which tend to occur together.

The role of morphology is the object of many hypotheses. Long awns, short peduncles, and compact spikes have been observed to favor rapid fungal invasion (Mesterházy 1995). Lax spikes correlated ($r=0.74$) with fewer infected spikelets (Eudes *et al.* 1999). Awns and glume hair can pick up fungal spores and also pollen that serves as food for the fungus. These structures also collect dew or rain droplets, and delay the drying process after dew or rain. It is also possible awns cause abrasion of the wax and cuticle, thus facilitating the entry of hyphae. Short peduncles might put the spike in a too humid microclimate, but could also be a symptom of root problems, which might in turn increase fungal growth. Shorter wheat cultivars are more predisposed to FHB (Couture 1982). This was reconfirmed by others (Buerstmayr 1997, Ban 2000), but the theory that it could be due to higher exposure to spore-carrying rain splash was not supported (Hilton *et al.* 1999). There is a good correlation between peduncle length and plant height, and it would be interesting to see what is important, i.e. it is plant height, or peduncle length, or both. Tall plants tend to lodge. A lodged field has a less ventilated canopy ideal for the growth of *Fusarium*. Spikes are closer to the inoculum source. Moreover, root systems and crowns of lodged plants get damaged, so that the nutrients needed by the plant defense mechanisms may be less available. Thus, lodging resistance genes must be part of a good package of genes of FHB resistance.

Compact spikes allow a larger number of crossing points for the movement of hyphae through different spikelets (Langevin *et al.* 2003). This could explain partly the difference between two row barley and six row barley. Movement of hyphae outside of the rachis seemed difficult to control by trichothecene resistance mechanisms, and that in 6-row barley, this external invasion route was quite important in the field as well as in controlled trials (Langevin *et al.* 2003). Ribichich and Vegetti (2000/2001) observed a long rachilla in Sumai 3, which might perhaps delay the invasion process.

Spike structure differs between species, and this is an important part of the tolerance and avoidance factors. Supposing equivalent and rather good type 2 resistance in all species, in two row barley, one successful spore germination near the base of a grain could infect one grain. In rye, two grains might become diseased; in six row barley, it would be three,

and in wheat, from 4 to 5 grains could be touched by the same path length of fungal growth. Oats offers paths that are one or two orders of magnitude greater between spikelets, making cross contamination between spikelets unlikely (Langevin et al. 2003).

Many FHB resistant wheat have short anthers (ex. Nyu Bay, Fukuho), and considering the nutritive role of pollen for *Fusarium*, it is logical that reducing the amount of excess pollen might help reduce the initial growth rate of the fungus (Miller et al. 2001). Many resistant lines like HY644 were observed to have paler and more translucent glumes, but there was no clear link to resistance. After flowering, some wheat lines close their florets leaving part of the anther exposed ; others open very sparingly and keep the anther inside. There was speculation that some lines with very long anther filaments had anthers that dropped to the ground rapidly, thus reducing the nutriment supply for *Fusarium*. And yet, at this point in time, there is no adequate study of correlation between the anther and glume behavior and the resulting Rtype1 effects.

Root traits were hypothesized to have an effect on *Fusarium* development for quite some time. Maringa for example reacts quite differently from year to year and from site to site. It is a wheat with exceptional roots, able to extract minerals from poor soils, but it lodges easily, which makes matters more complicated since lodging facilitates *Fusarium* growth. It was felt differences in uptake of micronutrients essential to plant defense mechanisms could be involved, since Maringa has shown superior ability to extract nutrients (Comeau, unpubl.). This aspect is still under study; meanwhile, another possible interaction has emerged following the observation of the effects of BYDV on cultivars like Barrie in 2003. The BYDV infected plots had much heavier FHB damage, approximately doubled by the virus at one site where corn inoculum was added. This interaction was visible in a warm midsummer highly favorable to FHB, but had not been observable in previous years that had cooler climate. Previous research at CIMMYT has shown that root disease can increase canopy temperature and spike temperature by 2-3 C in a hot day (Reynolds, pers. comm.). The Ste-Foy research proves that BYDV strongly impairs root growth, and is thus a root disease. It is concluded that BYDV is part of the root problems that increase spike temperature, and by this temperature effect alone, BYDV could increase the rate of spread of the fungus. A warming of 2 C can increase the speed of growth of the fungus by more than 30%, at least between 21 C and 26 C. The root traits discussed above include, besides root disease resistance. a series of physiological and metabolic factors that affect the morphology and functionality of roots, and germplasm selection with BYDV tends to select for better roots.

Phenology traits affecting FHB involves two aspects: earliness, and floral synchrony. In winter wheat, earliness increases the frequency of escape, because ascospore ripening may not be complete by the time winter wheat reaches flowering stage, at least in some parts of Canada. Global warming might decrease the usefulness of this trait. The duration and synchrony of the flowering stages may also be important. Devaux showed circa 1991 that AC Pollet had a sensitivity window only 2 days, whereas other Quebec wheat cultivars evaluated all had a 4 day sensitivity window. In 2003, Barrie had a more synchronous flowering than most cultivars; a relatively synchronous flowering was also observed in Torka (Langevin unpubl.). Barrie and Torka have better FHB resistance than most Canadian wheat cultivars, and both have Rtype1, although the resistance of Barrie

seems more stable across environments. It is thus possible that better floral synchrony and a short duration of flowering explains some of the Rtype1.

The mechanisms discussed above do not have a true repressive effect on the fungus growth or spread. These mechanisms are clever ways of avoiding fungus in the grain or reducing the severity of damage. Such mechanisms should more properly be called mechanisms of tolerance and/or passive resistance to the fungus.

The success rate of seed cleaning may vary quite a lot between cultivars. A cultivar in which many seeds have intermediate levels of FHB damage and carry toxin is not as desirable as a cultivar that tends to have an all-or-nothing response, and in which the seed either remains clean and sound, or else dies and gets easily discarded mechanically. An all-or-nothing response can represent a form of tolerance, at least from the viewpoint of the end users. Many triticale lines probably tend to produce seeds with intermediate to high levels of pericarp infection in years of medium FHB pressure, which would explain the higher DON content that was found in triticale for rather low symptom levels in Quebec.

Biochemical resistance mechanisms

The biochemical FHB resistance mechanisms may be constitutive or induced, and most are tissue- or organ-specific. Most of these may belong to the horizontal resistance category, according to current literature. The abundance of defense metabolites can increase following pathogen inoculation, due to signal-triggered responses, with complex transduction and regulation mechanisms. Hundreds of nonredundant genetic sequences were expressed within 48 h after *Fusarium* infection in Sumai 3 (Kruger et al. 2002). Using the same cultivar, Fellers et al. (2002) observed the induction of ten defense response genes, nine gene expression /regulation genes, 29 genes involved in other cell functions, and 32 genes with unknown function after 24 h. The induced genes having known roles catalyze key steps in the formation of lignin, energy production, and production of phytoalexins.

Dahleen et al. (2001) classified the modes of action of potential biochemical resistance mechanisms ; these may (i) degrade fungal cell wall or membranes, (ii) interfere with pathogen metabolism (protein synthesis, DON), (iii) bolster host defense system, and (iv) interfere with pathogenesis. So far, the research in that potentially valuable area has not brought elements of understanding that could be used in conventional breeding approaches, and yet low cost methods could change that situation. There could be a good use for specific knowledge about active metabolites in gene pyramiding efforts through more conventional means. The current state of the art certainly gives a complex picture relative to the resistance mechanisms. One approach that may have value is to increase resistance metabolites that are not overly phytotoxic; ferulic acid for example is an insecticidal and fungicidal defense molecule, but is also phytotoxic (Abdel-Aal et al. 2001, McKeehen et al. 1999).

One constitutive mechanisms relates to the level of choline (and glycinebetaine), especially in the anthers, but also in the glumes, lemma, and palea (Pearce et al. 1976). Higher levels of choline in anthers correlated to a higher initial growth rate of the fungus. Alondra's had 1300 mg/g choline in anthers, two times more than Wangshuibai (refs Li

and Wu 1994, Chen et al. 2000). Attempts to use this information in a practical selection process were envisioned in China, but this was abandoned due to costs. The stigma and ovaries are also tissues that stimulate the growth of *Fusarium* (Miller et al. 2001). Flavonoids and phenolics which can act as phytoalexins are likely to play a role within the seedcoat of older ovaries. Such compounds are known to be present, and in certain breeding lines, a contrast exists between rather heavy spike symptoms and lower than expected visible seedcoat infection levels or FDK (Fig. 2). This could explain the low correlation between DON and spike symptoms in certain trials.

Some phenolics may have stimulatory effects, and others, or repressive effects. Chlorogenic acid correlated with sensitivity, but total phenolics were not correlated. The activity of phenylalanine ammonia lyase (PAL) was also higher in sensitive wheat lines (Ye et al. 1990, Chen et al. 2000). And yet, phenolics are known as defence compounds, and PAL is involved in resistance mechanisms. One might see this as a clue that perhaps the speed of response of the defense mechanisms in a situation of fungal invasion is a more critical parameter than the amount of antifungal metabolite produced. After trichothecenes are produced, the protein synthesis in the cells is paralyzed, which automatically defeats all the active defense systems.

In another study, FHB resistance was correlated with the activity of SOD enzymes and catalase. Other enzymes and metabolites have also been investigated (Chen et al. 1997, Chen et al. 2000). In trials that are still ongoing and to be reported at the present workshop, Hamzehzarghani and Kushallappa have observed a number of wheat metabolites that may play a role as defense substances, shortly after inoculating spikes with *Fusarium*. Their findings include changes in the levels of phenolics, epoxy ethers, aromatics, furans, phytosterols, heterocyclics, organic acids, ketones, and other substances. Roles can be proposed for some substances known as involved in pathogen recognition, signal transduction, defense induction and antimicrobial or antifungal activity. The resistant cv. Sumai 3 had four times more constitutive phenolics than the susceptible Roblin. Constitutive high level of phenolics before infection were also reported by Siranidou *et al.* (2002). Among the phenolics, ferulic acid, a substance involved in midge resistance (Abdel-Aal et al. 2001), may also share a role in FHB resistance to *Fusarium* (McKeehen et al. 1999).

Insect transport of *Fusarium* is known to occur and may be important (Parry et al. 1995); thus it is also possible that certain metabolites act as insect attractants, or that aphid dew attracts vector insects in the spikes, but this idea has not received much attention. In Quebec, FHB damage tends to correlate with midge infestations in the spikes (Mongrain et al. 1999, 2000). This held true for many years, and the correlation is noteworthy (fig. 5).

Based on visual observations, it was hypothesized that active or vestigial areas of suberization might act as retardant barriers; there seems to be a delay of the fungus colonization when crossing a spikelet internode for example, and such areas seem able to develop more polyphenols. Juvenile tissues have some inherent resistance, and in wheat/*Agropyron* hybrids, many resistant lines had glumes that remained juvenile-looking or «stay-green» for a longer time than those of ordinary wheat (Langevin, unpubl.).

The role of trichothecenes as phytotoxic compounds was proven in the last decade. These are for now the only *Fusarium* metabolites shown to be involved in pathogenesis, (Kruger et al. 2002). It was suggested that ability to decrease trichothecene levels (type 3) (Miller and Arnison 1986, Atanassov et al. 1993) or tolerate those toxins (type 4) (Wang and Miller 1988, Miller 1989) generally correlates with resistance to spread (type 2). In practice, it is indeed difficult to investigate separately the resistance type 3 and 4 components without seeing also type 2 resistance (Desjardins et al. 1996, Eudes et al. 2001, Mesterházy et al. 1999). Thus the type 3 and 4 resistances may be conceived as explaining a lot of the Rtype2 and a lot of the overall resistance/tolerance in general.

And yet, infection of at least one cereal species without trichothecenes has been shown to occur. Durum wheat is highly sensitive to FHB and develops disease even if infected by a trichothecene non-producing strain (Langevin et al. 2003). Durum is thus different from the vast majority of hexaploid wheats, since none of the hexaploids were found sensitive to the trichothecene non-producing strain. This implies a) durum has species-specific biochemical weaknesses in its defense mechanisms, compared to bread wheat, and 2) trichothecenes are perhaps not the only phytotoxic substances produced by *F. graminearum*, although it remains clear that they are the most important ones. Organic acids are produced in vitro and might deserve investigation in this respect (Miller, pers. comm.)

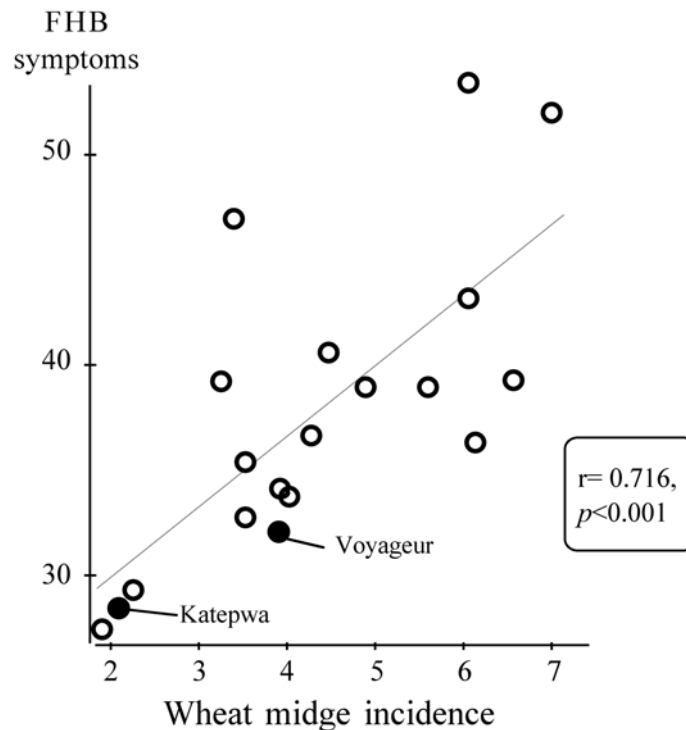


FIG. 5. RELATIONSHIP OF SPIKE SYMPTOMS (MORE THAN 3 YEARS OF ST-HYACINTHE DATA BY DEVAUX, AVERAGED WITH THE LEAST SQUARES METHOD) WITH MIDGE INCIDENCE IN OTHER TRIALS (3 TO 6 YEARS OF QUEBEC DATA BY MONGRAIN AND LANGEVIN, AVERAGED WITH THE SAME METHOD), FOR QUEBEC WHEAT CULTIVARS. THE MORE RESISTANT

CULTIVARS TEND TO HAVE FEWER MIDGES, AND THE FHB CHECKS KATEPWA AND VOYAGEUR FOLLOW THAT TREND.

A rapidly induced response is thus useful, but other mechanisms exist. In Sumai 3, *F. graminearum* invades tissues quite slowly but regularly for up to three weeks. But in one tetraploid experimental line named Blackbird (source : Fedak), the fungus grows rapidly at first but is stopped completely and in a rather sudden manner after 4-6 d. In the field, Blackbird remains more resistant than most durum wheat cultivars but its Rtype1 is quite poor, so the end result is only medium resistance (Langevin and Voldeng, unpublished).

Resistance in complex natural stress situations

Cultivars that look resistant in artificial inoculation trials may perform as expected in the field, but variations occur among years. It was observed that those variations correlated with the environmental factors listed in Table 1. In years with high ascospore inoculum, the climate after flowering may be unfavorable to spread of the disease within a spike, mostly due to lack of heat; in such a case Barrie and other Rtype1 lines can resist quite well. In years with low ascospore release, the climate after flowering may have long periods around 25-29 C, ideal for the spread of the fungus. In such years, a line with good Rtype2 becomes essential, and Alsen or HY644 would be expected to do better than Barrie. In the worst year, a complex combination of Rtype1 and Rtype2 is needed ; such a combination seems nonexistent in commercial cultivars for now.

The role of root health has been mentioned above when discussing morphology. Deficiencies of major and minor nutrients may reduce root length and thus increase spike temperature, which may accelerate fungal growth. Moreover, a number of macro- and micronutrients are essential for the proper functioning of the plant defense mechanisms (Graham 1983, Huber and Graham 1999). Thus, differences in root health and root efficiency may explain part of the variation of the distribution of the disease within a field ; FHB is often somewhat distributed in patches, and thus seems partly related to soil factors.

Resistance in segregating populations

The common understanding about FHB resistance expression is that the genes are mostly additive (Stack et al. 2001), sometimes recessive (Gupta et al. 2001) or epistatic (Jiang and Ward 2002). Field, greenhouse and growth room trials were designed in Ste-Foy based on that hypothesis. In controlled indoors conditions we saw the expected additive behavior when using milder test conditions, at cooler temperatures. However, in severe test conditions outdoors and indoors, we did not observe much additive behavior of the genes. Dominant resistance was not found in any of our hexaploid resistance source so far. In 2003, a year when FHB was severe, more than 6000 F₁ plants containing a source of FHB resistance were thus almost fully destroyed by *Fusarium* inoculation (spray plus corn inoculum). In our indoors trials with F₁s of sensitive x resistant wheat lines, we generally used temperatures that maximize fungal growth rate (24 - 26 C); air moisture may also be important, but is a less controlled factor. The presence of some recessive and

epistatic gene behavior may necessitate special strategies for germplasm development. Due to this, fixation of resistance is not easy, and haploids may serve a good purpose.

A slightly milder FHB stress can help obtain additive behavior, at least within indoors facilities. Yet, previous experience in breeding for BYDV resistance led us to prefer the stronger stresses. With BYDV, the genes that are rather dominant under weak viral stress can be made to behave as additive or recessive by increasing the stress intensity. Also, certain genes identified as useful under medium-weak BYDV epidemic conditions become less efficient under stronger epidemic pressure. Since in Eastern Canada, FHB can often reach very severe levels and also occur together with root-impairing disease like BYDV or *Pythium* and waterlogging stress, there are grounds for using a severe stress, and a complex stress like BYDV plus *Fusarium* inoculation makes sense, since it can intensify FHB in a FHB-prone year. In terms of selection efficiency, it is hoped that the lines thus selected will produce in subsequent generations a stronger proportion of resistant lines, with a more complete resistance package. The observed interactions also support the idea that adding to FHB crosses some genes for root health, like BYDV tolerance, *Pythium* and waterlogging resistance, could perhaps improve the behavior of the FHB resistance genes.

In 2003, at one test site, BYDV and FHB stresses were combined to waterlogging stress, since both BYDV and *Pythium*+waterlogging complex may impair roots enough to reduce general disease resistance. In such conditions, about 1% of the crosses (in F₂-F₄) produced a satisfactory number of multiple-resistant segregants. It was noted that most of those crosses contained in their pedigree certain cultivars known in Brazil for multiple disease and stress resistance, most often EMBRAPA 27 and sometimes BRS 177. On the negative side, these two Brazilian cultivars show weaknesses like Sumai 3 relative to breadmaking potential, and are late flowering. As least, it is proven that FHB resistance is compatible with resistance to BYDV and to the *Pythium*+waterlogging complex.

Conclusions

Many traits play a defined or probable role in resistance. Morphological and phenological traits are often controlled by many genes, and the biochemical clues indicate a number of metabolic pathways are involved, with signaling mechanisms and complex response mechanisms. Creating lines with the higher levels of resistance will necessitate pyramiding complementary genes from very biodiverse sources. A quantitative genetics approach will be needed.

The quality of data sets is important, and this may not be feasible at the lowest cost. A broader spectrum of test conditions and resistance criteria may be needed using more different environments and secondary stresses like those that affect root systems. Symptom readings can be improved by more frequent observations and closer scrutiny of the plant-fungus interaction. End-result criteria may bring useful extra information. Marker assisted selection has been applied in the past based on the existence of a few major genes for type 2 resistance. It could play a role in order to refine the crossing and selection strategies. However, for better results, it may be necessary to combine DNA markers with more precise phenomics and metabolomics. Statistical approaches suitable for simple systems must also be replaced by new approaches that help extract more information from larger data sets, inclusive of phenomics and metabolomics.

In conventional germplasm development, additive or dominant alleles are easier to deal with. Rust, mildew, bunt resistance genes are frequently dominant. But diseases like BYDV and FHB seem to have a frequent pattern of additive behavior of the genes under weak or medium stress, and recessive behavior under severe stress. This brings a new complexity in the germplasm development process. However, previous experience with BYDV is useful in many ways in suggesting crossing/selections strategies that may have value against FHB. New approaches to quantitative genetics are currently being developed. This includes the broadest spectrum of resistance sources, including lines from many countries and a few interspecific hybrids. Crossing blocks including maximum possible biodiversity can help identify complementary gene behavior. Genes that reduce toxins may be found through in vitro selection and use of toxin analyses. Hope for more rapid progress is now permitted based on good results of previous work in Canada and on the new strategies that should accelerate delivery of the results that both the consumer and the industry need. Considering the size the task, collaboration among research groups from many institutions will be a key element in order to achieve further progress.

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Biological control methods to manage fusarium head blight disease of wheat: is it a short or long term solution to the problem? W.G. Dilantha Fernando, *Department of Plant Science, University of Manitoba, Winnipeg, MB R3T 2N2, Canada.*

The broad definition of biological control of plant pathogens is, to use a living organism or organisms or their derivatives to curtail, inhibit, or kill a pathogen or its infective propagules from germinating, and to avoid significant losses to the agricultural industry. In North America, fusarium head blight disease caused mainly by *Fusarium graminearum* reduces yield, grade and quality of wheat and barley. As there is no resistant cultivar available, and fungicide(s) have not produced satisfactory results, searching for alternative methods such as biological control has received prominence in several research programs in the USA and Canada. Biological control using naturally occurring microorganisms such as *Trichoderma* sp., *Bacillus* sp., *Pseudomonas* sp., *Cryptococcus* sp., *Stenotrophomonas* sp., and *Lysobacter* sp., or their biologically active products has shown to significantly reduce FHB disease, and mycotoxin levels in both greenhouse and field experiments. Cover crops such as red clover and medic have been investigated to reduce stubble-borne inoculum from moving to the wheat heads at anthesis. Non-hosts such as flax and canola in crop rotations with wheat, barley and oats are investigated as eco-friendly means of disease management. Examples of these research areas from various labs, their advantages and disadvantages, and complexities of developing integrated disease management strategies will be presented and discussed.

Sources and dispersal of *Gibberella zea*/*Fusarium graminearum* inoculum

J. Gilbert *Cereal Research Centre, AAFC-AAC, 195 Dafoe Road, Winnipeg, MB R3T 2M9*

It has been recognized since the early years of the last century that overwintered stubble provides inoculum for FHB (Atanasoff 1920). Both rain-splash of macroconidia and wind for ascospores were implicated as the means of spore dispersal. More recent studies have attempted to determine the duration of survival of spore-bearing residues in light of an increased move toward tillage practices that leave crop stubble in the field. Khonga and Sutton (1988) examined survival and sporulation of *Gibberella zea* on wheat spikes, grain and stems, and maize stems and ears. Tissues were buried, left at soil surface or suspended from a nylon line 10 cm above the ground. No sporulation occurred on buried tissues. Macroconidia and perithecia developed on tissues placed on the soil or above ground in the first year. In the second year, perithecia, but no macroconidia, formed on maize stems and wheat spikelets and grains, and in the third year, only on wheat spikelets and grain suspended above the ground. A high density of perithecia developed on maize kernels and wheat grains. Similar results were reported for *Fusarium*-infested wheat kernels in Manitoba (Inch and Gilbert 2003).

There is no evidence of spore production on *Fusarium*-infested residues that are buried (Khonga and Sutton 1988, Inch and Gilbert 2003), although these have been shown to have the potential to infect root tissues of both cereal and non-cereal crops seeded in close proximity (Chongo et al. 2000). Rate of decomposition of residues is more rapid in the soil, than above or on the soil surface (Dill-Macky 1999, Khonga and Sutton 1988, Todd et al. 2001) and in conjunction with the lack of spore production means that buried residues contribute little to inoculum load. Wet soil conditions do not favour fungal survival (Dickson, 1923) which, in part, may explain the lack of observed seedling blight in the cool moist clay soils of the Red River Valley, Manitoba where FHB epidemics have been most severe in the last decade.

Several studies have examined the effect of tillage practice on head blight development. Intuitively, tillage systems that leave the most residue on the soil are expected to produce the greatest amount of inoculum and cause the highest severity of FHB. In light of what is known about pathogen survival and sporulation on residues, tillage and crop rotation were recommended as means of reducing inoculum (Khonga and Sutton 1988). However, in Saskatchewan, zero-till did not result in higher levels of FHB than conventional till, while minimum-till fields were more severely diseased (Fernandez et al. 2001). Significant differences were found in disease incidence and severity, and deoxynivalenol (DON) content of grain in a Minnesota study which examined the effects on disease levels of moldboard plow (~10% residue retained), chisel plow (~30% residue retained) and zero-till (~65% residue retained) (Dill-Macky and Jones 2000). However, while statistically significant, differences were small. Disease incidences in moldboard plow, chisel plow, and zero-till plots were 64%, 72% and 71% respectively, disease severities 16%, 20% and 21% respectively, and DON levels 8.1, 10.6 and 11.1 ppm, respectively. The results of a three-year tillage study in Ontario were inconclusive with observations

that *F. graminearum* persists on debris under both till and no-till conditions and that other factors such as rotation and cultivar susceptibility are likely to be more important than tillage practice (Miller et al. 1998).

Conclusions as to the effects of rotations are somewhat similar. Where differences were found, levels of disease on wheat following crops other than cereals or maize were significantly different, but small (Dill-Macky and Jones 2000). For example, FHB incidences in wheat planted into corn, wheat, and soybean residues were 75%, 67% and 64%, respectively, disease severities were 23%, 18%, and 16%, respectively, and DON levels 13.5, 9.2 and 6.9 ppm, respectively. Ahmed et al. (2002) found that FHB was higher in wheat in canola-wheat and pea-wheat rotations than in wheat-wheat and wheat-oat rotations, but disease levels were low and results not conclusive as the experiment had only been conducted for a first year. Other studies have not demonstrated differences in FHB disease levels due to rotations (Fernandez et al. 2001), and even when wheat follows corn as part of the rotation, a report of a three year survey of 230 wheat fields in Kentucky concluded that the principal factor in FHB development is the weather (Hershman 2000).

Guenther and Trail (2002) reported that perithecia form in wheat through stomates above chlorenchyma of the stem internode and from epidermal cells of the stem node region. The light and moisture requirements for *Gibberella zeae* ascospore release have been examined by Trail et al. 2002. Under laboratory conditions, ascospore release was reported to be 8-30% greater in light than in complete darkness, and in constant light, discharge reached maximum rates at relative humidities greater than 92%. This is in contrast to the work of Paulitz (1996) and Inch et al. (2000) who, under natural conditions in Quebec and Manitoba, respectively, found most ascospores were trapped in the evening, reaching a peak before midnight. Schmale et al. (2002) also collected more colonies in sampling periods that spanned sunset to sunrise as opposed to sunrise to sunset. Dufault et al. (2002a, b) identified the conditions under which perithecia develop on corn residues. Under field conditions, an extended period of stalk wetness at temperatures between 15°C and 25°C favoured perithecial development. When temperatures were lower than 15°C, perithecia stopped developing, but resumed when temperatures rose again (Dufault et al. 2002a). Under controlled conditions there were no significant differences in rate of perithecial development at 15°C or 25°C, but none developed at 30°C (Dufault et al. 2002b).

Ascospores appear to be dispersed relatively short distances when measured by level of infection of wheat plants from neighbouring inoculum foci (Fernando et al. 1997). In experiments conducted in Ontario and Quebec, the highest incidence was 0.5 m from the upwind edge of the inoculum area. There was a 50% decline in spikelet infection within 1.6 to 2.0 m upwind and within 2.7 to 4.9 m downwind from disease foci. Seed infection declined to 10% of the maximum within 5-22 m from the focal centre in plots inoculated with corn spawn to promote ascospore infection, and within 5 m in plots inoculated with macroconidia. The suggestion that ascospores might be taken into the planetary boundary layer has recently been given credence by work in New York State. Using remote-controlled model aircraft and boats fitted with spore traps, ascospores have been trapped

at more than 180 m above ground over lakes and regions remote from farm fields (Del Ponte et al. 2003). The relative contributions of external and within-field inoculum sources is unknown, but will vary according to region, crops grown, and tillage practices. The experience in New York, for example, indicates a tendency in fields where wheat follows corn for FHB disease patterns to be aggregated and disease incidence to be high. In fields planted to vegetable crops, or where remnants of old corn residue (2+ years) were small and scattered, disease incidence was random and low (Del Ponte 2003). A combination of within-field inoculum and inoculum from airborne ascospores may explain the uniform and intense level of infection observed across southern Manitoba wheat fields in the epidemic year of 1993.

A number of probably minor sources of inoculum have been examined in an attempt to discover or explain how the disease develops and spreads. Among these, asymptomatic inflorescences of wild grasses have been found to harbour several *Fusarium* spp., including *F. graminearum* (Inch and Gilbert 2003). The fungus, *F. graminearum* survives on kernels of wheat and corn, producing copious perithecia and ascospores on the soil surface for two to three years (Inch and Gilbert 2003; Khonga and Sutton 1988), although sowing *Fusarium*-infested wheat seed does not cause FHB disease (Gilbert et al. 2003). The fungus survives parasitically and saprophytically on wheat leaves throughout the growing season (Ali and Francl 2001, Osborne et al. 2002). The most prevalent species were *F. graminearum* and *F. sporotrichioides*. From 4 to 52% of the non-surface-disinfested diseased leaf tissue was infected with *F. graminearum* (Ali and Francl, 2001). Osborne et al. (2002), recovered up to 1500 spores per leaf. While ascospores were usually most prevalent, in some locations the leaves supported large macroconidial concentrations, suggesting the fungus may grow epiphytically, resulting in higher inoculum levels within the canopy. Soybeans also have been reported as a host for *F. graminearum* (Martinelli et al. 2001), and non-cereal residue, canola and field pea, was found to support high levels of sporulation (Gilbert et al. 2003). Dill-Mackay and Salas (2001) reported that burning of cereal stubble significantly reduced the number of isolations of *F. graminearum* from 26% to 6%, while no isolations were made from charred residues. In areas at low risk from *Fusarium* such as Alberta, efforts have been made to reduce incoming inoculum in the form of infested grain for feed lots. While the fungus does not survive passage through the rumen, wasted feed and spills are cited as potential means of introducing the disease to the province (Calpas 2003, McLaren et al 2003).

Conclusions:

F. graminearum/*G. zeae* sporulates well on residues left on or above the soil surface, especially maize stem and grain, and wheat spikelets and grain.

The fungus does not sporulate on *Fusarium*-infested buried residue, but survives on residues for at least 2 years and can cause root damage to other crops.

Fields in tillage systems that leave large amounts of residue on the soil surface do not appear to suffer consistently higher levels of FHB.

The evidence concerning the effect of rotations is inconclusive, except where cereals follow corn in years that favour FHB development.

Perithecia require temperatures from 15°C to 25°C to develop. In corn stalks more perithecia developed under conditions of high moisture.

Dispersal occurs over short distances at field level, but there is evidence that diffuse concentrations of ascospores are carried in the wind stream and are responsible for long-distance spread.

Different species and plant tissues can be colonized by *F. graminearum*/*G. zeae* and may intensify inoculum buildup during the growing season, and provide an overwintering ground for the fungus.

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Session 6: Disease Management

The Grower's Perspective: Managing Fusarium Head Blight On The Farm

Peter Johnson, Grain Producer and Cereal Specialist, Ontario Ministry of Agriculture and Food, Stratford, Ontario, N5A 5T8

Under Ontario conditions, Fusarium head blight is an ever present threat. While province wide epidemics are still rare, disease incidence has increased and significant losses occur every season in localized areas of the province. Heavy infestations greatly reduce income, with isolated incidences of grain being dumped back in the field due to extreme toxin levels. Awareness and management of the disease has increased dramatically in Ontario, yet many producers are frustrated with inconsistent control. The economics of control is borderline if grade improvement is not achieved. The recent release of several new winter wheat varieties with improved genetic tolerance to Fusarium hold promise for the future. These moderately tolerant varieties, along with fungicide treatments, should increase control consistency and reduce losses to the producer.

Sprayer Technology: How Best to Apply Fungicides. T.M. Wolf and H. Spieser. *Saskatoon Research Centre, Agriculture and Agri-Food Canada, Saskatoon, SK S7N 0X2, Canada; and (H.S.) Ontario Ministry of Agriculture & Food, Ridgetown, ON N0P 2C0.*

The use of fungicides for control of fusarium head blight (FHB) caused by *Fusarium graminearum* (Schwabe) has increased the need to improve spray targeting of vertical structures such as wheat spikes. Experiments were conducted over the last three years to study the interactive effects of travel speed, nozzle orientation and nozzle type on spray deposition and coverage uniformity on vertical artificial targets. The experimental delivery system permitted separate tracing of spray from the front and rear nozzles, and the sampling technique allowed separation of the front and rear side of the target. Results showed that a combination of double nozzles, air-induced sprays, and faster travel speed increased spray retention on vertical targets by more than 100 %. Wider angles separating the nozzles (60° vs 30° from vertical) increased deposition, but more for coarse sprays than fine sprays. Wider angles also increased deposition on the back side of the vertical target. When two spray qualities were combined in a single nozzle, leading with the coarse spray gave a higher deposit than leading with a fine spray. Although these results speak in favour of double nozzles equipped with coarse sprays, the relationship between retention on artificial and biological targets remains to be determined.

Integrated Fusarium head blight management: Employing all the tools
Albert Tenuta, Ontario Ministry of Agriculture And Food, Ridgetown, Ontario, Canada, NOP 2C0.

Fusarium head blight has caused significant economic losses in Manitoba, Ontario, Quebec and the Maritimes over the past decade. Inconsistent control has contributed to the Fusarium head blight problem. At this time it is not realistic to expect a single cultural or management practice to effectively manage this devastating disease alone. The integration of the various cultural or management practices ("tools") into a coordinated FHB management program can minimize producer losses and risk. The components of an integrated FHB management program include: (1) seed quality, (2) seed treatment, (3) variety selection (tolerant/resistant), (4) crop rotation, (5) disease forecasting and fungicide application, (6) residue management, and (7) harvest management.

(1) Seed Quality - Begin by planting good quality seed that is free of Fusarium. Although seed infection is not considered a primary inoculum source for late season head blight, the use of infected seed has been shown to reduce seedling emergence, vigour and tillering. Infected seed increases the survivability of the fungus from one year to the next and is a potential means of spreading or introducing Fusarium into previously non-infected fields or new wheat producing areas.

(2) Seed Treatment - Fungicide seed treatments are effective against many seed-borne and soil-borne seedling blights and seed rots and therefore are a recommended practice. The use of these products on scab affected wheat can increase germination, emergence, and tillering and thereby limiting stand losses due to Fusarium seedling blight. Seed treatments will not prevent late season FHB from developing.

(3) Variety Selection - Although, no resistant varieties are presently available both public and private breeding programs have made significant progress in the development of varieties with partial resistance or "tolerance".

(4) Crop Rotation - Rotation of non-host crops will reduce FHB levels. Avoid planting small cereals following other small cereals or corn. Two years between small grain or corn crops will allow for residue decomposition. Rotation will not eliminate the disease since inoculum from neighbouring fields still pose a risk.

(5) Disease Forecasting and Fungicide Application - Various Fusarium head blight or DON Prediction Models are being evaluated in Canada and the United States. Although, these models provide a new and effective "tool" for managing and timing fungicide applications for FHB, they do not replace good old fashion scouting. The emphasis on weather data requires knowledge of the local (field) weather conditions and crop development. Fungicide efficacy is dependent on delivery systems and variety susceptibility. A number of new fungicides are under development and should provide more consistent control of Fusarium head blight. Understanding the parameters that make

these prediction models work and how the various components interact is necessary for successful implementation and management of Fusarium head blight.

(6) Residue Management - Infested cereal (including corn) residues are a primary source of inoculum. The longer infested residues remain intact on the soil surface, the greater potential for disease development. Removal of residues will reduce inoculum levels but tillage on its own will not eliminate the disease. In areas with a history of the disease, inoculum production from surrounding fields could lead to Fusarium head blight development under favourable environmental conditions.

(7) Harvest and Storage Management - Grain sample improvements are possible at harvest through simple adjustments to the combine. Increasing air blast velocity (speed) will remove many of the smaller, lighter Fusarium infected kernels. Although a small amount of healthy kernels will be removed, the improvement in sample grade will offset these losses. Reducing combine ground speed and adjusting cleaning sieves can further separate out infected kernels. Proper storage and drying will limit further Fusarium development after harvest. Check stored grain frequently to ensure that the grain stays in good condition.

Reducing producer risk and economic losses to FHB requires the integration of available cultural and management "tools" or practices into a sustainable program.

International Perspective on FHB

International Approach to Breeding for Fusarium Head Blight Resistance. Maarten van Ginkel, Lucy Gilchrist, Flavio Capettini, Mujeeb Kazi, Wolfgang Pfeiffer, Manilal William, Tomohiro Ban and Morten Lillemo. *International Maize and Wheat Improvement Center, Mexico*¹. *ICARDA/CIMMYT, Mexico*², *JIRCAS, Japan*³.

CIMMYT (International Maize and Wheat Improvement Center) started breeding for resistance to Fusarium Head Blight (FHB) 20 years ago in Mexico. Breeding for resistance to FHB in barley started in the mid 80's by the ICARDA/CIMMYT program. Adaptation to rainfed environments required resistance to *Septoria tritici* and *Fusarium* spp. A sprinkler-enhanced screening method in a natural high-humidity field site (Toluca) is used. Annually tens of thousands of spikes from thousands of wheat and barley lines bred or obtained from around the world are screened. We measure the five classical Types of resistance. For barley Type I is more important. Our breeding methodology aims to combine resistance mechanisms and accumulate distinct alleles. The international Scab Resistance Screening Nursery (SRSN) is distributed annually. Initial molecular marker studies at CIMMYT and by others indicated that novel resistance genes might be present in CIMMYT wheats, distinct from the full Sumai#3 haplotype. Doubled haploid BSA populations for both wheat and barley are being studied in more detail. Some synthetic wheats show great promise as resistance sources.

Studies for enrichment of wheat germplasm with fusarium head blight (FHB) resistance in Japan. T. Ban, D.H. Xu, Z. Nishio, A. Yanagisawa, and N. Kawada. *JIRCAS, Tsukuba 305-8686, Japan*; (Z. n.) *NARC-Hokkaido, Memuro 082-0071, Japan*; (A. y) *Kitami Agr. Exp. Stn. Kunneppu 099-1496, Japan*; (N. k.) *NARC-Kyushu Okinawa, Chikugo 833-0041, Japan*.

Resistance to fusarium head blight (FHB) varies not only among wheat cultivars but also their wild relatives. No accession yet, however, has been found to be completely immune. Spring wheat cultivars from the Japanese and Chinese gene pools were screened and analyzed for their genetic diversity. It showed the uniqueness of the Japanese gene pool, and its distinction from the Chinese one. Moreover, several wild relatives of *Agropyron* (*Elymus*), indigenous to Japan and with highly effective resistance, were identified. We are tracing the pedigrees of resistant wheat cultivars that were intensively used in Japanese breeding programs since the 1970's. Shinchunaga, in addition to local varieties such as Nobeokabouzu-komugi and Nyubai, is considered to be one of the main donors for moderate resistance to FHB in many Japanese commercial cultivars. Our comparative QTL analysis for different types of resistance in those gene pools revealed several significant regions in the wheat genome. These results showed the potential and indicated a strategy for introgression and pyramiding of genes to enrich FHB resistance in wheat breeding programs.

Fusarium diseases of wheat and barley - an Australian perspective. S. Simpfendorfer. *NSW Agriculture, Tamworth Agricultural Institute, New South Wales 2340, Australia.*

Crown rot caused by the fungus *Fusarium pseudograminearum* is a major constraint to winter cereal production in Australia. Although it is generally more common in the northern cropping belt, it can occur throughout all mainland cereal growing areas and is estimated to cost the Australian grains industry \$56 million per annum. Losses from Fusarium head blight (FHB) caused predominantly by *Fusarium graminearum* have not been estimated in Australia. However, severe FHB on the Liverpool Plains in northern NSW in 1999 and 2000 inflicted yield losses of around 20-100% with associated downgrading in quality. Outbreaks of FHB have occurred sporadically in Australia and have also been associated with the rainsplash of *F. pseudograminearum* macroconidia formed on lower nodes into heads. A strategic initiative on crown rot, common root rot and FHB with funding from the Grains Research and Development Corporation was formed in 2002 to address these disease problems in the Australian grains industry. The initiative encompasses seven projects across four states with the aims of: i) providing an integrated and coordinated approach to the management of these diseases, ii) facilitating communication and collaboration between research groups in Australia and internationally, and iii) extending research outcomes to growers. Research areas include epidemiology and disease management, chemical and biological control, variety development and germplasm introduction and enhancement.

Poster Abstracts

Session 1: Overview and toxins

Prediction of deoxynivalenol (DON) in barley using near-infrared spectroscopy.

Arganosa, G.C.¹, Rossnagel, B.G.¹, Legge, B.G.², Savard, M.³, Zatorski, T.¹ and Tucker, J.². ¹*Crop Development Centre, University of Saskatchewan, Saskatoon, SK, S7N 5A8,* ²*Agriculture and Agri-Food Canada, Brandon Research Station, Brandon, MB, R7A 5Y3,* ³*Agriculture and Agri-Food Canada, Eastern Cereal and Oilseed Centre, Ottawa, ON, K1A 0C6.*

Our objective was to develop a calibration equation to predict the deoxynivalenol (DON) levels in barley using near-infrared spectroscopy. A FOSS NIR Systems 6500 Near Infrared Spectrometer was used to collect the spectra of 577 samples grown at Brandon and Ottawa in 2001 with DON levels ranging from 0 to 161 ppm. The DON levels of these samples were measured using the ELISA test. The computer programs CENTER and SELECT were used to define the population boundaries of these samples and to select those samples to be used for calibrations. Population boundaries were established with a maximum standardized H distance from the average spectrum of 3. Calibration samples were selected with a minimum standardized H distance between samples of 0.6. Calibrations were developed using different math treatments with multiplicative scatter correction (de-trend). This equation was used to predict the DON levels in sample sets grown at Brandon in 2002. The results show that this equation has the potential to be used as a screening tool to eliminate those samples with high levels of DON.

Toxin content in wheat seeds in Quebec in 2002. Y. Dion, S. Rioux and M. Lauzon. (Y.D. and M.L.) *Centre de recherche sur les grains, Saint-Bruno-de-Montarville, QC J3V 4P6 Canada;* (S.R.) *Centre de recherche sur les grains, Sainte-Foy, QC G1P 3W8 Canada.*

Fusarium head blight (FHB) is the most important disease of wheat in Quebec and is a major constraint on the development of spring wheat production. The incidence and the severity of the disease are largely dependent on climatic conditions. Previous surveys conducted in wheat fields in Quebec had shown that *Fusarium graminearum* Schwabe is the most prevalent species associated with the disease and that deoxynivalenol (DON) is the major concern among the toxins present in wheat seeds after an infestation. Surveys conducted in 1999, 2000 and 2001 had also shown that the FHB incidence and toxin content were generally low for most of the regions surveyed. In 2002, the climatic conditions were not favourable to FHB development. DON content in seeds sampled from 81 wheat fields was usually low in all regions. We observed that of the 81 samples tested, 27% had a DON content equal to or higher than 1.0 ppm and only 9% had a DON content of 2.0 ppm or higher. The highest DON levels were found in the Montreal area, the main region for wheat production in Quebec. T-2 toxin was undetected in all samples collected in 2002. However, 23% of the samples had a HT-2 toxin content exceeding 0.025 ppm.

Fusarium head blight in the Atlantic region in 2003. R. A. Martin¹ and M. E. Savard², Agriculture and Agri-Food Canada, ¹Crops and Livestock Research Centre, 440 University Ave., Charlottetown, PEI, C1A 4N6 and, ²Eastern Cereal and Oilseed Research Centre, Ottawa, Ontario, K1A 0C6.

The worst epidemic of fusarium head blight (FHB) in the Atlantic Region (New Brunswick, Nova Scotia and Prince Edward Island) in the last 20 years occurred in 2003. Wheat, oats and barley all harboured some level of Fusarium infection and DON contamination. Based on samples collected at elevators and on farms, the levels of *Fusarium graminearum* and DON ranged from very low to severe. Initial data indicated a wide variability in the severity of FHB across the region in 2003. Seed infection levels as high as 90% were recorded. DON levels were as high as 24 ppm. DON levels were in excess of 2.5 ppm in 22% of barley, 72% of oat, 58% of spring wheat and 17% of winter wheat samples. While crop and cultivar selection affected the level of FHB, field location appeared to have an impact as well.

Fusarium- A Serious Threat to the Australian Wheat Industry. V. Mitter^{1,2}, O.A. Akinsanmi^{1,2}, S. Simpfendorfer³, D. Backhouse⁴, D. Yates⁵ and S.Chakraborty^{1,2} ¹CSIRO Plant Industry, QBP ,306 Carmody Road, St Lucia, Brisbane 4067, ²CRC for Tropical Plant Protection, University of Queensland, Brisbane 4072; ³New South Wales Agriculture, Tamworth ; ⁴University of New England, Armidale, Australia.

Fusarium head blight (FHB) and crown rot (CR) are two of the most serious disease threats affecting production and quality of wheat in Australia. CR is a chronic problem costing the industry over \$56 million annually. Changing cropping practice and above average rainfall have also increased the incidence of FHB in recent years. During the 1999 FHB epidemic the yield of some crops in northern New South Wales was reduced by 20-100%. We have collected over 700 *Fusarium* isolates from crops in three states and identified them using species-specific PCR primers and morphology. A total of 20 *Fusarium* species were identified with *F. pseudograminearum* (59% of isolates) and *F. graminearum* (22% of isolates) being the two most dominant species. Bioassays developed have rapidly and accurately detected small but consistent difference in aggressiveness among 283 isolates from 17 species for FHB and 76 isolates from 10 species for CR. Although isolates between species and isolates within species differed significantly in aggressiveness, the difference between *F. graminearum* and *F. pseudograminearum* was not significant. Aggressiveness of isolates for FHB and CR were not correlated but 20% of isolates caused severe to highly severe infection for both diseases. Amplified fragment length polymorphism shows distinct genetic groups within *F. graminearum* and *F. pseudograminearum* but does not appear related to aggressiveness. The quantitative bioassays are now being used to screen and select germplasm resistant to the two diseases.

Fusarium Head Blight in Northern Quebec. D. Pageau, J. Lajeunesse, M. Savard, C. Coulombe and Y. Lefebvre. *Research farm, Normandin, Agriculture and Agri-Food Canada, QC G8M 4K3, Canada; and (M.S.) Eastern Cereal and Oilseed Research Centre, Ottawa, Agriculture and Agri-Food Canada, ON K1A 0C6 Canada; (C.C. and Y.L.) La Financière Agricole du Québec, Alma, QC G8B 7A6 Canada.*

In the Saguenay-Lac-Saint-Jean area (Northern Québec), barley (*Hordeum vulgare* L.) and oat (*Avena sativa* L.) production is very important whereas wheat production (*Triticum aestivum* L.) is limited because the growing season is very short. Fusarium head blight (FHB) has become the major problem in barley production in this region. In 2001, about 10% of barley acreage (1 987 ha) had a DON content of more than 2 mg kg⁻¹. In 2002, the problem was more severe with 40% of the barley acreage (7 986 ha) infected with FHB. The economic losses associated with FHB in barley were estimated at 2.4 millions dollars during those two years in the Saguenay-Lac-Saint-Jean region. In 2002, seed samples collected from the two locations of the Québec Barley and Oats performance trials in the Saguenay-Lac-Saint-Jean area (Hébertville and Normandin) were analysed for DON content. No inoculum was applied to the performance trials. DON content in oat samples varied from 0.4 to 1.9 mg kg⁻¹. At both locations, there was no difference between naked and covered oat cultivars. A total of 60 barley samples were also analysed at each location (12 samples of two-row barley and 46 samples of six-row barley). DON content in barley samples varied from 0.3 to 3.0 mg kg⁻¹. The results indicate that none of the oat or barley cultivars were immune to FHB.

Deoxynivalenol production by *Fusarium graminearum* isolates in four winter wheat cultivars. L. Tamburic-Ilincic and A. W. Schaafsma. *Ridgetown College, University of Guelph, Ridgetown, Ontario, N0P 2C0, Canada*

A common way to screen wheat (*Triticum aestivum* L.) cultivars for fusarium head blight (FHB) resistance and deoxynivalenol (DON) production is by spray-inoculation with single or mixed isolates of *Fusarium graminearum* (Schwabe) in the field. We wanted to ensure that there was no strain-specific resistance with respect to DON accumulation.

DON production was quantified after two FHB susceptible (Harus, Pioneer 2540), and two FHB moderately resistant (AC Morley, Pioneer 25W60) winter wheat cultivars were spray-inoculated with four *F. graminearum* isolates (1-4) in the field.

The level of DON ranged from 0.5 ppm to 11.1 ppm in the experiment. Pioneer 2540 had the highest mean DON content (9.3 ppm), while AC Morley had the lowest (1.3 ppm). Average DON content after spray-inoculation with isolates 1, 2, 3, and 4 was 3.7, 6.6, 6.5, and 6.4 ppm, respectively across the cultivars. Isolate DAOM178148 (1), used for FHB resistance screening for performance trials in Ontario produced significantly lower DON levels, than the other 3 isolates. Cultivar ranking based on DON accumulation did not vary with isolate.

Session 2: Industry and consumer issues

Interactions of weather factors and fusarium head blight, and its effect on wheat grain quality. W.G. Dilantha Fernando, X.W. Guo, P. Bullock, H. Sapirstein, J. Dexter and T. Nowicki. *Department of Plant Science; (P.B.) Department of Soil Science; and (H.S.) Department of Food Science, University of Manitoba, Winnipeg, Manitoba, R3T 2N2, and (J.D., and T.N.) Canadian Grain Commission, Winnipeg, Manitoba, Canada*

Fusarium head blight (FHB) disease has emerged as a major threat to wheat grain quality. The pathogens involved in FHB disease produce mycotoxins. Successful disease management, especially well-timed fungicide applications based on a disease forecasting system, will improve the net returns. This study aims at understanding quantitative relationships between inoculum levels of *Fusarium* sp. and FHB incidence, FHB index and mycotoxins; the effects of different mycotoxins on grain proteins; and establishment of a weather-based model. This study is conducted in 19 wheat fields growing either AC Barrie or AC Superb, spread throughout different geographic regions in Manitoba, documenting their cropping history, collecting weather data through Agrometeorological Centre of Excellence, pooling information on cultivars and agronomic factors, investigating inoculum levels before seeding and from flowering to early milk stage, assessing wheat head wetness duration, and quantitatively analyzing and integrating data for disease modeling. The experiment was initiated in 2003 and will continue through 2006.

***Fusarium* Toxins in Infant Cereal Foods and Adult Breakfast Cereals from the Canadian Retail Market.** Gary A. Lombaert, Peter Pellaers, Veronica Roscoe, Meena Chettiar, David Kitchen, Susan Kotello, Thomas Krakalovich, Don Lavalley, Greg Sliva, Robert Trelka, Gary Neumann, and Peter M. Scott. *Health Canada, Health Products and Food Branch, 510 Lagimodiere Blvd., Winnipeg, MB R2J 3Y1 Canada; and (P.M.S.) Health Canada, Health Products and Food Branch, Address Locator 2203D, Tunney's Pasture, Ottawa, ON K1A 0L2 Canada.*

Over 300 samples of infant cereal foods and over 150 samples of adult breakfast cereals were collected from the Canadian retail marketplace during the years 1997 - 2000 and 1999 - 2001 respectively. The infant cereal foods included oat-, barley-, soy-, wheat-, and rice-based infant cereals, mixed grain infant cereals and teething biscuits. The adult breakfast cereals included oat-, rice-, wheat-, and corn-based cereals, and mixed grain cereals. The samples were analyzed for targeted *Fusarium* mycotoxins (deoxynivalenol, nivalenol, HT-2 toxin, and zearalenone). Overall, deoxynivalenol was detected in 55% of the infant cereal foods and 40% of the adult breakfast cereals. Nivalenol was detected in one multi-grain adult breakfast cereal, and HT-2 toxin was detected in one oat-based adult breakfast cereal. Zearalenone was detected in 27% of the infant cereal foods and 9% of the adult breakfast cereals. Among the adult breakfast cereals, wheat-based cereals exhibited the highest incidence of deoxynivalenol (72%), while multi-grain cereals exhibited the highest incidence of zearalenone (17%). Among the infant cereal foods, soy-based products (which usually contain corn) exhibited the highest incidences of deoxynivalenol (100%) and zearalenone (71%). Survey results demonstrated the regular occurrence of low levels of *Fusarium* mycotoxins in infant cereal foods and adult breakfast cereals.

Survival of *Fusarium graminearum* through the digestive tract of cattle – fact or fiction? D. L. McLaren, S. L. Scott, T. K. Turkington, Y. Wang and T. McAllister. *Brandon Research Centre, Agriculture and Agri-Food Canada, P.O. Box 1000A, R.R.#3, Brandon, MB R7A 5Y3; (T.K.T.) Lacombe Research Centre, Agriculture and Agri-Food Canada, 6000 C&E Trail, Lacombe, AB T4L 1W1; (Y.W.&T.M.) Lethbridge Research Centre, Agriculture and Agri-Food Canada, P.O. Box 3000, Lethbridge, AB T1J 4B1.*

Fusarium-infested grains are often fed to cattle as opportunity feeds because of their apparent tolerance for higher levels of dietary deoxynivalenol (DON) than swine. The westward movement of feed grain infested with *F. graminearum* Schwabe is a concern, due to the possibility that feeding these opportunity feeds may increase the spread of fusarium head blight (FHB) into Alberta. However, this would require survival of the pathogen through the bovine digestive tract. If *F. graminearum* can survive digestion by livestock, FHB may be introduced into areas where it has not previously been found. Three studies were conducted to evaluate the potential for spread of *F. graminearum* via feed grain after passage through the digestive system of feedlot cattle. Many fungal species were detected from feed bunk barley grain sampled from Alberta and Manitoba feedlots, but they were not detected in whole intact grain screened from manure from these same feedlots. When *F. graminearum*-infested barley kernels were placed directly into autoclaved or non-autoclaved manure, pathogen survival was reduced, with almost complete elimination in non-autoclaved manure. In preliminary feeding trials with infected barley, *F. graminearum* was almost completely eliminated after passage through the digestive tract of beef steers. All three studies indicate that the risk of survival of *F. graminearum* in infected grain after passage through beef animals followed by incubation in manure is very limited.

***Fusarium* spp. from commercial seed samples in Saskatchewan in 2001 and 2002.** R.A.A. Morrall, P.G. Pearse, B. Carriere, D. Schmeling, and C. Pearse. (*R.A.A.M.*) *Dept. of Biology, Univ. Saskatchewan, Saskatoon, S7N 5E2*, (*P.G.P.*) *Saskatchewan Agriculture, Food and Rural Revitalization, Regina, S4S 0B1*, (*B.C.*) *Discovery Seed Labs Ltd., Saskatoon, S7J 4M2*, (*D.S.*) *Lendon Seeds Ltd., Weyburn, S4H 1T1*, (*C.P.*) *Priority Lab Services, Nipawin, S0E 1E0*.

Agar plate test results for *Fusarium* spp. on commercial seed samples of wheat, durum, and barley, plus small quantities of other cereals, were collected from three private laboratories. Common species detected were *F. avenaceum* (Fr.:Fr.) Sacc., *F. poae* (Peck) Wollenw., *F. sporotrichioides* Sherb. and *F. graminearum* Schwabe. Lesser quantities of *F. culmorum* (W.G.Smith) Sacc., *F. equiseti* (Corda) Sacc. and other species were found. Mean percent seed infections with *F. graminearum* and with total *Fusarium* spp. were calculated for each rural municipality for wheat, durum, barley, and all cereals combined and mapped for 2001 and 2002. Seed harvested in 2002 was more heavily infected with *Fusarium* spp. than in 2001 throughout Saskatchewan, but this was not due to fusarium head blight. In most areas conditions during flowering were unfavorable for infection in both years. However, excessive wet weather in August and September, 2002 delayed harvest, leading to saprophytic invasion of seed and poor seed quality. Infection levels were highest on durum, probably because production is concentrated in southern Saskatchewan, where more rainfall was received in both years. *Fusarium graminearum*

was widely distributed in eastern and central areas. However, seed infection levels were very low, except in S.E. Saskatchewan. In this region, higher levels in 2001 than in 2002 probably reflected conditions during flowering more favorable for head blight.

A medium and procedure for identifying *Fusarium graminearum* in cereal seed. S. Pouleur, L. Couture, R. Clear, and A. Comeau. *Agriculture and Agri-Food Canada, Soils and Crops Research and Development Centre, Sainte-Foy, QC, Canada G1V 2J3* ; and (R. c) *Canadian Grain Commission, Grain Research Laboratory, Winnipeg, MB, Canada R3C 3G8*.

A working procedure was developed to detect *Fusarium graminearum* Schwabe in cereal seed. To test it, 10 non-surface disinfested cereal seed samples were analysed using the following procedure. First, seeds were plated onto an agar medium (PCNB) that is semi-selective for *Fusarium* spp. and incubated for seven days. Next, mycelium from each *Fusarium* colony was transferred to a second medium (ID) and incubated for two days. Red colonies on the ID medium were counted as Fg. A total of 225 *Fusarium* colonies were transferred from PCNB onto the ID media. Colonies with and without red pigment on ID media were then transferred to PDA to confirm their identity. Of the 135 red colonies obtained on the ID medium, 114 were confirmed as Fg. Nearly all of the 21 false positives were *F. sporotrichioides* Sherb., while only 6 of the cultures that failed to form a red colour on ID media were Fg. Typically, these false negatives contained other Fusaria with Fg. Seed contamination levels obtained with the new procedure were compared to the ones obtained with the conventional method on PDA. On average, the new approach detected twice as many Fg as did the PDA method. Moreover, the ID medium was also able to segregate *F. pseudograminearum* Aoki and O'Donnell from Fg. The new procedure is very promising for Fg evaluation in seeds. It is more sensitive than the method based on PDA, especially with non-surface sterilized seeds, and it does not require any particular training. We are working on improving this test for large scale use.

***Fusarium* spp. infection of wheat grains in the Czech Republic and its relation to bread-making quality parameters** L. Tvaruzek, *Department of Integrated Plant Protection, Agricultural Research Institute Kromeriz, Ltd., Havlickova 2787, 67 01 Kromeriz, Czech Republic*.

Fusarium spp. incidence on wheat grains was monitored in the period 1999 – 2002 in the territory of the Czech Republic. Samples from farmers' fields were assessed for *Fusarium* infected grains (%) as well as bread-making quality parameters and germination power (%). *Fusarium* species were identified microscopically. The mean infection level in particular years was 7.7 % in 1999 (476 samples), 5.3 % in 2000 (400 samples), 7.8 % in 2001 (318 samples), and 20.8 % in 2002 (222 samples). The regional distribution of infection in the year with a high infection level (2002) was the lowest in South Moravia Region (17.3 %) and the highest in North Moravia Region (28.1 %). *Fusarium graminearum* Schwabe dominated on infected grains in all years of monitoring. The other most frequent species were *Fusarium avenaceum* (Fr.:Fr.) Sacc., *Fusarium poae* (Peck) Wollenw., *Fusarium culmorum* (Wm. G. Sm.) Sacc., and *Fusarium sporotrichioides* Sherb. The increased level of infected grains (%) significantly correlated with lower germination power in all years. Also, falling number was influenced significantly negatively by *Fusarium* infection in all years. Except the year 1999, highly significant reduction of grain protein content was correlated with increasing *Fusarium* infection level.

Session 3: New sources of, and breeding for, FHB resistance

Reaction of intergeneric and synthetic spring wheat lines to *Fusarium graminearum*.

A. Breker, P.J. Hucl, and G. Hughes. *Department of Plant Sciences, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK S7N 5A8 Canada.*

Intergeneric hybrids of wheat (*Triticum aestivum*) and *Agropyron repens* syn. *Elytrigia repens*, and synthetic wheat lines (*Triticum turgidum* ssp. *durum* x *Aegilops tauschii*) produced at CIMMYT, were evaluated under greenhouse conditions for resistance to fusarium head blight caused by *Fusarium graminearum*. Two point inoculation methods, the canaryseed and spore suspension methods, were used to evaluate these lines over two or three screening repetitions. Approximately 18% of the intergeneric lines and 13% of the synthetic lines consistently displayed moderate to high levels of Type II resistance. Pollen staining was conducted to determine the fertility levels of the intergeneric lines. 80% of the F₃ lines and 75% of the F₅ and F₆ lines displayed fertility levels above 75%. Control cultivars displayed fertility levels ranging from 88-94%. Generally, lines that rated as moderately to highly resistant were also highly fertile. These lines could be of significant value in wheat breeding programs aimed at integrating new sources of resistance to fusarium head blight.

Critical control points for large-scale fusarium head blight field screening trials.

A.L. Brûlé-Babel, W.G.D. Fernando, P. Hucl, G. Hughes, S. Fox, R. DePauw, M. Fernandez, J. Clarke, R. Knox, J. Gilbert, G. Humphreys and D. Brown. *Department of Plant Science, University of Manitoba, Winnipeg, Manitoba; (P.H. and G.H) Crop Development Centre, University of Saskatchewan, Saskatoon, Saskatchewan; (R.D., M.F., J.C. and R.K.) Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, Swift Current, Saskatchewan; (J.G., G.H. and D.B.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, Manitoba.*

Fusarium head blight (FHB) continues to be a serious disease of wheat in western Canada and in particular, the eastern prairies. In 2001, breeders and pathologists entered into a collaborative agreement to establish a common FHB screening nursery at Carman, Manitoba. Lines were evaluated in 2001, 2002 and 2003, either in replicated or non-replicated trials. In each year, five checks with known reactions to FHB were included every 50 plots. The objectives of this study were to use the results from the five checks to characterize the variation observed across the nursery and the different environments, and to identify critical control factors that could improve large-scale screening trials. The factors analysed were variation for incidence, severity and FHB Index measurements within and among nurseries, the effect of different evaluators on these measurements, and frequency of change in ranking or classification among the checks. Environmental factors were also assessed. The results from this analysis will assist in the interpretation of data from large field screening nurseries.

Collaborative study of barley Fusarium head blight nurseries. G. Butler, K.M. Ho, M.J. Morrison, A.G. Xue, W.G. Legge, S. Rioux, R.A. Martin, Q. Shen, J. Yang, J.R. Tucker, M.E. Savard, J.L. Gale, M. Kuc and C. Danjou. *Eastern Cereal and Oilseed Research Centre, Ottawa, Agriculture and Agri-Food Canada, ON K1A 0C6, Canada;* (W.G.L., J.R.T) *Brandon Research Centre, Brandon, Agriculture and Agri-Food Canada, MB R7A 5Y3;* (S.R.) *Centre de recherche sur les grains, Sainte-Foy QC G1P 3W8, Canada;* (R.A.M.) *Crops and Livestock Research Centre, Charlottetown, Agriculture and Agri-Food Canada, PE C1A 4N6, Canada;* (Q.S., J.Y.) *Zhejiang Academy of Agricultural Sciences, Hangzhou, Zhejiang 310021, China;* and (C.D.) *Centre de recherche sur les grains, Saint-Hyacinthe, QC J2S 7B8, Canada*

Inoculated nurseries have been established in Canada and China to screen and evaluate barley (*Hordeum vulgare* L.) for resistance to Fusarium head blight (FHB). Results within and between nurseries can be conflicting due to different environments, methods of assessment or other factors. A collaborative trial was designed to investigate deoxynivalenol (DON) content, visual assessments, and their relationships within and across five diverse locations; Brandon MB, Ottawa ON, Saint-Hyacinthe QC, and Charlottetown PEI, in Canada, and Hangzhou in China. Twenty-five cultivars, selected to represent a range of susceptible and resistant cultivars of two-row and six-row barley, were grown in 2002 and 2003 in replicated plots, visually rated for severity of FHB and, where possible, harvested at maturity and sampled for DON. The 2002 results indicate that significant correlations within locations between visual assessments and DON content are possible as were seen in Brandon and Ottawa. Correlations were also significant between these two locations. Environmental conditions, escapes and differences in protocol may have contributed to the lack of correlation within and among some of the locations. Significant correlations were also seen between DON and severity when ranks across locations were combined. The data suggest that multiple environments should be evaluated to obtain reliable results in the assessment of FHB and DON.

Two seeding date system in Fusarium head blight nurseries. G. Butler, H.D. Voldeng, S. Rioux, A.G. Xue, M.E. Savard, J. L. Gale, P.H. Matthew, C. Danjou, F.E. Sabo, Y. Chen, R. Stanley. *Eastern Cereal and Oilseed Research Centre, Ottawa, Agriculture & Agri-Food Canada, ON K1A 0C6, Canada;* (S.R.) *Centre de recherche sur les grains, Sainte-Foy, QC G1P 3W8, Canada;* and (C. D.) *Centre de recherche sur les grains, Saint-Hyacinthe, QC J2S 7B8, Canada.*

Inoculated nurseries have been established in Ottawa, ON and Saint-Hyacinthe, QC to evaluate spring wheat (*Triticum aestivum* L.) genotypes for their susceptibility to Fusarium head blight (FHB). Although misting or irrigation partially dampens the effect of environment there is still considerable variation in the assessment of disease from one environment to another. In an attempt to capture some of this environmental variation within a site year and thus improve and possibly accelerate the evaluation process, the field replicates in a number of trials have been sown on two different dates. In this investigation we present an evaluation of a two seeding date system based on spring wheat trials conducted in Ottawa since 2001 and in Saint-Hyacinthe since 2002. Visual assessments of the percentage of infected heads and the percentage of infected spikelets on infected heads as well as seed levels of deoxynivalenol (DON) were considered. There was significant genotype by planting date interaction for FHB incidence and

severity in most of the trials. This was less often the case for DON. This data indicates that a second planting in spring wheat nurseries is one method of capturing genotype by environment information, which is particularly effective for the visual measures of FHB.

Germplasm enhancement for FHB resistance in spring wheat through alien introgression. George Fedak, Wenguang Cao, Fangpu Han. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-food Canada, 960 Carling Ave. Bldg. 50, Ottawa, Ontario K1A 0C6 Canada.* Fusarium head blight inoculum of cereals seems to have become well established in many regions of Canada such that the occurrence of rainfall during flowering will cause infection in the crop. There is no immunity to the disease in the primary gene pool of wheat. The Chinese cultivar Sumai 3 is the best known source of resistance in wheat and has been used worldwide in breeding and genetics studies. However, under epiphytotic nursery conditions of 2003, for example, Sumai 3 gave incidence, severity and FDK levels of 19%, 7.5% and 10%, respectively. The DON level in the harvested grain was of 7.6 ppm. There is a need therefore to enhance the level of FHB resistance. One possible method is by means of alien introgression. Fairly extensive screening has been carried out of accessions of several species in the secondary and tertiary gene pools of wheat to find additional sources of resistance. In the secondary gene pool of wheat, levels of resistance have been found in *Triticum monococcum* (AA), *Aegilops speltoides* (BB), *Ae squarossa* (DD), *Triticum timopheevii* (AAGG) and *Triticum miguschovae* (AAGGDD). These have all been crossed to hexaploid wheat (a few accessions have been crossed to durum also) and progenies are in various stages of backcrossing and FHB screening. From the tertiary gene pool, chromosome 1 and 2 additions of *Hordeum chilense* to hexaploid wheat have shown fairly good FHB resistance and low DON contents while this variability is being introduced into durum wheat through Tritordeum (AABBHH). The best resistance to FHB that we have identified to date in an alien species is that in the tetraploid form of *Thinopyrum elongatum*. This source of resistance should be transferrable to durum wheat through the existing amphiploids and into hexaploid wheat by means of addition and substitution lines.

Effect of incubation period and plant growth stage on Fusarium Head Blight in barley under greenhouse conditions and screening with single *F. graminearum* isolates. T. S. Grewal, B. G. Rossnagel, and G. J. Scoles. *Department of Plant Sciences/Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK S7N 5A8, Canada.*

Fusarium head blight (FHB) is the most significant disease of barley in Canada. Effective and efficient indoor screening techniques are critical for breeding resistant barley cultivars. Field screening methods are not efficient due to environmental effects and confounding effects of plant architecture, heading date and other field diseases. An indoor screening technique using spray inoculation and a humidity chamber has been evaluated and a screening protocol was developed.

FHB severity increased with the period of incubation in the humidity chamber. A 48 h incubation period was the best. There were some escapes after a 24 h of incubation and incubation of 72 h led to very high disease and deoxynivalenol (DON) levels. When inoculated at ear emergence, early heading lines showed less disease than normal/late

heading lines. All lines had high disease severity when inoculated 14 days after ear emergence. Testing 15 barley cultivars with six isolates of *F. graminearum* Schwabe demonstrated significant cultivar × isolate interaction. Single isolates will now be used to screen segregating populations against FHB. Inoculation will be performed at ear emergence followed by 48 h of incubation in the humidity chamber.

Co-selection for resistance to both wheat streak mosaic virus (WSMV) and *Fusarium graminearum* (cause of fusarium head blight, FHB): a novel approach for the rapid development of elite wheat lines with multiple disease resistances. S. Haber and J. Gilbert. *Cereal Research Centre, Agriculture & Agri-Food Canada, 195 Dafoe Rd., Winnipeg MB, R3T 2M9, Canada.*

Seed from WSMV-infected susceptible wheat lines gave rise in the next generation to plants with unexpected variations in response both to WSMV infection as well as in other, seemingly unrelated, traits. Individual plants exhibited improved resistance to WSMV while others were more susceptible. Some also showed altered pigmentation or number of internodes, traits which were passed in varying proportions to subsequent generations. The unexpected variation was induced in plants stressed in repeated cycles of WSM or Barley Yellow Dwarf (BYD)-plus-WSM disease pressure, and formed the basis of our 'Stress- Directed Selection' (SDS) protocol. Starting in June 2001 with the cross (FHB-resistant) HY644/CO960293-4 we selected F1s and F2s under combined BYD+WSM pressure; 11 WSMV-resistant F3 lines were evaluated in an artificially-inoculated FHB nursery in summer 2002. One line combined FHB and leaf rust resistance from which F4 progeny lines were selected indoors under BYD+WSM pressure followed by spray inoculation with *Fusarium graminearum* Schwabe of heads at anthesis. The best F5s were again selected indoors under severe BYD+ WSM pressure. In summer 2003, 56 SDS-generated F6 lines were tested in the FHB field nursery. Eleven performed better than the resistant checks FHB37 and HY644, and were the source of 49 F7 head-row lines increased in winter 2003/04 in New Zealand.

Metabolic profiling for phenotyping resistance in wheat to fusarium head blight. Hamzehzarghani, H., Kushalappa, A. C., Dion, Y., Comeau, A. and Mather, D. E. *McGill University, Ste- Anne-de-Bellevue, QC, H9X 3V9; (D.Y.) CEROM, St-Bruno-de-Montarville, QC J3V 4P6; (C.A.) AAFC, Ste. Foy, QC G1V 2J3.*

Breeding for quantitative resistance to fusarium head blight (FHB) is hampered by a poor understanding of mechanisms and lack of cost-effective, fast and efficient screening methods. In this study, metabolic profiling has been evaluated as an alternative to disease assessment. Spikes of wheat cultivars Sumai3 (resistant) and Roblin (susceptible) were inoculated at anthesis with a spore suspension of *Fusarium graminearum* and incubated for 24 or 48 h. Metabolites were extracted, derivatized and analyzed using a gas chromatograph and mass spectrometer system. 919 compounds were detected in wheat-FHB system but only 94 more consistently occurred 24 h after inoculation. Among these, 21 and 11 compounds were found only in Sumai3 or Roblin, respectively, while 62 metabolites were common to both. The abundance of phenolics in Sumai3 was four times than that found in Roblin, the most abundant class of compounds being anthraquinones. Following inoculation, aromatic and heterocyclic compounds were up-regulated in Sumai3. Polyphosphoinositides, messenger compounds, were observed in trace amounts

only in Sumai3. Statistical models, using frequency of compounds in a group, and metabolic fingerprints based on mass ion abundance, were used to identify metabolic phenotypes.

Molecular characterization of partial amphiploids from *Triticum durum* x tetraploid *Thinopyrum elongatum* as novel sources of resistance to wheat Fusarium Head Blight. Fangpu Han and George Fedak. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave, Bldg 50. Ottawa, Ontario, K1A 0C6, Canada. Phone: 613-759-1393, Fax: 613-759-6559, Email: fedakga@agr.gc.ca* Three amphiploids ($2n=6x=42$), namely 8801, 8802 and 8803 derived from *Triticum durum* x tetraploid *Thinopyrum elongatum*, were screened for Fusarium Head Blight resistance. The three amphiploids had high levels of resistance to FHB. The disease did not spread beyond the inoculated spikelet in all the tested plants. Genomic in situ hybridization (GISH) using genomic DNA of tetraploid *Th. elongatum* as a probe, indicated that they contained 14 alien chromosomes from tetraploid *Th. elongatum*. There were no gross chromosome structural changes in the three amphiploids. High molecular weight (HMW) glutenin and gliadin analysis showed that 8802 and 8803 had the same protein bands from wheat. The three lines contained a specific HMW band coming from tetraploid *Th. elongatum*. These new amphiploids offer an excellent means of introducing Fusarium head blight resistance into durum wheat.

Fusarium resistance in Western wheat lines tested across three environments. F. Langevin, J Gilbert, H Voldeng, A Comeau. *121 Bon-Air, Ste-Catherine-de-la-Jacques-Cartier, GOA 3M0; (JG) CRC, Agriculture and Agri-Foods Canada, Winnipeg R3T2M9; (HV) ECORC, Agriculture and Agri-Foods Canada, Ottawa K1A0C6; (AC) Agriculture and Agri-Foods Canada, Ste-Foy, G1V2J3.* Western spring wheat lines were grown at Lévis, near Quebec City, and at Ottawa, so that Fusarium Head blight (FHB) reaction could be compared with that observed in Glenlea, Manitoba. Testing methods differed among sites. A high dose of corn inoculum was used in Ste-Foy and Ottawa so as to obtain a better evaluation of type 1 resistance mechanisms. Sprinkler irrigation was also used at Glenlea and Ottawa. At flowering time the weather was warm and rainy for many days in Quebec, leading to an extremely severe test. The results show that most of the current germplasm is susceptible or highly susceptible to FHB, especially in the High Yield and Western Bread Wheat coop tests. Some lines suffered total yield loss from Fusarium infection of the spikes in Lévis. Ranking according to symptoms varied according to the reading date, and was only a moderately good predictor of resistance as measured by other criteria such as FDK count and residual yield under disease pressure. In the Central Bread Wheat test, a few lines were more resistant than the checks. Those may deserve special attention as parental material or for registration purposes. Many resistant lines are late maturing; these may be partly escapes and signify out the need for evaluation over more sites and/or years.

Haplotype diversity at Fusarium Head Blight resistance QTLs in wheat. C.A. McCartney, D.J. Somers, G. Fedak, and W. Cao. (C.A.M. and D.J.S.) *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB R3T 2M9, Canada; and (G.F. and W.C.) Eastern Cereals and Oilseeds Research Centre, Agriculture and Agri-Food Canada, 960 Carling Avenue, Ottawa, ON K1A 0C6, Canada.* Fusarium head blight (FHB) reduces grain yield and quality in common and durum wheat. Host FHB resistance is an effective control measure that is achieved by stacking multiple FHB resistance genes. Resistance gene stacking would be facilitated if breeders knew which FHB resistance sources carry different resistance genes. A diverse collection of FHB resistant and susceptible wheat lines was characterized with microsatellite markers linked to known FHB resistance quantitative trait loci (QTLs) on chromosomes 2DL, 3BS (distal to the centromere), 3BSc (proximal to the centromere), 4B, 5AS, and 6BS identified in Maringa, Sumai 3, and Wuhan 1. Putative Sumai 3 QTLs were commonly observed in advanced breeding lines, whereas putative Maringa and Wuhan 1 QTLs were relatively rare. The microsatellite data suggested that the 3BS, 3BSc, and 5AS QTLs in the Brazilian cv. Maringa were derived not from Frontana, as previously thought. Maringa appeared to be closely related to Asian germplasm at the 3BS, 3BSc, and 5AS QTL regions. Other Brazilian wheat lines did not appear closely related to other FHB resistance sources. These Brazilian wheats may have novel FHB resistance that will be useful for stacking with FHB resistance derived from Asian germplasm.

Fusarium head blight assessments in barley lines after inoculation with *Fusarium graminearum* and *Fusarium sporotrichioides*. S. Rioux. *Centre de recherche sur les grains, Sainte-Foy, QC G1P 3W8 Canada.*

A previous survey conducted in barley (*Hordeum vulgare* L.) fields in Quebec showed the presence of *Fusarium graminearum* Schwabe along with in cool seasons the presence of *F. sporotrichioides* Sherb. known to produce HT-2 and T-2 toxins. *F. sporotrichioides* is a concern as the barley crop in Quebec is more concentrated in cooler climate regions. Two-row and six-row barley lines were tested in separate trials at the Saint-Hyacinthe nursery in 2001 and 2002. Each year, there were for both barley types one trial inoculated with an *F. graminearum* conidial suspension and one trial inoculated with *F. sporotrichioides*. Significant differences were observed between lines for fusarium head blight (FHB) index (% infected spikelets) and for deoxynivalenol (DON) content in all trials inoculated with *F. graminearum*. Significant differences were also observed for the FHB index in all *F. sporotrichioides* trials, whereas only one trial out of four showed significant differences for T-2 toxin content. In this study, correlation analyses showed that the response of barley lines to *F. graminearum* and to *F. sporotrichioides* was similar for symptom ratings, but not for toxin content. Barley cultivar Chevron previously identified as resistant under *F. graminearum* inoculum was also among the least susceptible lines after inoculation with either *F. graminearum* or *F. sporotrichioides*.

Dodging the Exponential Challenge of Breeding Fusarium Head Blight Resistant Cultivars.

J.B. Thomas and R.M. DePauw. *Cereal Research Center, 195 Dafoe Road, Winnipeg, MB R3T 2M9 Canada; and (R.D.) SemiArid Prairie Agricultural Research Centre, POB 1030, Swift Current SK S9H 3X2 Canada.*

As a desirable addition to existing wheat breeding priorities, multigenic Fusarium Head Blight (FHB) resistance will add exponentially to the complexity of developing superior cultivars. To this end, cross complexity and breeding method were investigated in terms of a Desirable Genotype Quotient (DGQ). DGQ is the proportion of lines in which a segregating locus is either fixed for the plus allele or is heterozygous. With no selection, $DGQ=P^N$ in which P varies by generation (3/4 in F2, 5/8 in F3 etc) and N is the number of segregating loci; the reciprocal of DGQ is the minimum population that contains one such desirable genotype. Theoretical and practical considerations show that selection in segregating generations (pedigree method) is a good approach to cope with complex crosses. Doubled haploidy works well with narrow crosses but is inefficient for complex crosses. Disomic reversion was then introduced as a cytogenetic method to reduce cross complexity. The name refers to the tendency of aneuploids such as telosomics and non-reciprocal translocations to revert to the disomic condition and fix the gene content of their hemizygous segments in the process. Based on this idea, it was shown that two FHB QTLs could be fixed by introducing two telocentrics into an elite cross with an FHB resistant parent while four FHB QTLs should be fixable by using two translocations. In terms of the DGQ criterion, the benefit of fixing two or four FHB QTLs represents a 4 to 14 fold increase in the frequency of desirable genotypes in F6.

Evaluation of agronomic performance, disease resistance and malting quality of advanced barley breeding lines selected for fusarium head blight resistance.

J. R. Tucker, W. G. Legge, M. E. Savard and A. Tekauz. *Research Centre, Agriculture and Agri-Food Canada, P.O. Box 1000A, R.R. 3, Brandon, MB R7A 5Y3, Canada; (M. E. S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave., Ottawa, ON K1A 0C6, Canada; and (A. T.) Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Rd., Winnipeg, MB R3T 2M9, Canada.*

In response to Fusarium head blight (FHB) epidemics incited by *Fusarium graminearum* Schwabe in the 1994-95 Manitoba barley (*Hordeum vulgare* L.) crops, several crosses with reputed resistance sources (three eastern Canadian cultivars and six 'exotic' sources) were initiated in 1996 by the two-row, malting barley breeding program at Brandon. After evaluating progeny from these crosses over several years, 25 lines showing improved FHB resistance have entered advanced yield tests. Although 'exotic' germplasm such as Chevron, CI 4196, Seijo II and Zhedar #1 are among the best resistance sources, retrieving recombinants carrying their full potential with adequate agronomic performance has been difficult. Crosses involving Gobernadora have shown moderate yield potential and some of the best FHB resistance, but development as malting cultivars may be problematic due to hull peeling. Several of these lines are also resistant to scald [*Rhynchosporium secalis* (Oud.) Davis]. Lines from the Morrison cross were the highest yielding, with one line having moderate FHB resistance. The Symko cross was less promising. Harbin and AC Sterling crosses have produced lines combining good FHB resistance and agronomic performance, although lines from the AC Sterling cross have inferior malting quality and susceptibility to stem rust (*Puccinia*

graminis Pers.:Pers. f. *sp. tritici* Eriks & E. Henn). Further testing will determine cultivar potential of the FHB resistant lines.

Marker-assisted backcrossing selection of near isogenic lines for 3BS Fusarium head blight resistance QTL in hexaploid wheat. Wenchun Zhou^{*}, Frederic L. Kolb, Guihua Bai. *Department of Crop Science, University of Illinois, 1102 South Goodwin Avenue, Urbana, IL 61801; and (G. B.)USDA-ARS Plant Science and Entomology Research Unit, 4008 Throckmorton Hall, Kansas State University, Manhattan, KS 66506;*

^{*}*Current address: Lethbridge Research Centre, Agriculture and Agri-Food Canada, P. O. Box 3000, 5403 1st Avenue South, Lethbridge, Alberta, Canada.*

Near-isogenic lines (NILs) differing in disease resistance quantitative trait loci (QTL) are valuable materials for the study of the genetic basis of quantitative resistance. A marker-assisted backcrossing selection project was begun in 1999 to develop NILs for the 3BS major FHB resistance QTL. Based on AFLP mapping results and FHB screening tests, a recombinant inbred line, RIL90, was selected from 133 RILs derived from Ning7840×Clark for use as the donor parent of the 3BS QTL. The genome region containing the 3BS QTL was controlled during backcrossing through SSR marker analysis. NILs differing in Type II FHB resistance and carrying marker alleles from Ning7840 and Clark were identified in BC₄F₂ populations. Greenhouse evaluation of FHB resistance and SSR analysis confirmed the identification of NILs differing in the 3BS QTL. Genetic similarity between NILs and Clark was tested based on 121 SSR markers polymorphic between Ning7840 and Clark. Plants obtained after the fourth generation of backcrossing resembled the recurrent susceptible parent based on phenotypic and genotypic evaluation. NILs had a genetic similarity with Clark of more than 98%, but retained a region containing the 3BS FHB resistance QTL from Ning7840. These NILs will be useful for further molecular characterization of the major QTL on 3BS.

Molecular characterization of Fusarium Head Blight resistance in Wangshuibai with SSR and AFLP markers. Wenchun Zhou^{*}, Frederic L Kolb, Jianbin Yu, Guihua Bai, Larry K. Boze, Leslie L Domier. *Department of Crop Science, University of Illinois, 1102 South Goodwin Avenue, Urbana, IL 61801; and (J. Y. and G. B.) Department of Agronomy, USDA-ARS, 4008 Throckmorton Hall, Kansas State University, Manhattan, KS 66506; and (L. L. D.) Department of Crop Sciences, USDA-ARS-MWA, 1102 South Goodwin Avenue, Urbana, IL 61801.*

^{*}*Current address: Lethbridge Research Centre, Agriculture and Agri-Food Canada, P. O. Box 3000, 5403 1st Avenue South, Lethbridge, Alberta, Canada.*

Evaluation of wheat FHB resistance is laborious, costly, and time consuming. Marker-assisted selection of FHB resistant QTL will speed up breeding resistant cultivars by reducing phenotypic evaluation and increasing selection efficiency. However, most reported scab resistance QTL are from Sumai 3 and its derivatives. To broaden the genetic base of scab resistance, it is important to identify QTL from new scab resistant sources. Wangshuibai is a scab resistant landrace originated from China and is not related to Sumai 3. A mapping population of 139 F₅ recombinant inbred lines derived from a cross of Wangshuibai × Wheaton was used to map scab resistant QTL in Wangshuibai. This population was evaluated for Type II scab resistance in the greenhouse at two

locations in 2003. A total of 1196 SSR and AFLP markers were mapped on this population, and four scab resistance QTL were detected. A major QTL near the end of 3BS explained 37.3% of the phenotypic variation. Another QTL on 3BS located close to the centromere, explained 7.4% of the phenotypic variation. Two additional QTL on 7AL and 1BL explained 9.8% and 11.9% of the phenotypic variation, respectively. The SSR and AFLP markers closely linked to these FHB resistance QTL may be useful for stacking QTL to develop transgressive resistant cultivars.

Session 4: Host resistance genetics

Improvement of Fusarium head blight resistance in barley through *in vitro* selection. M. Banik¹, W.G. Legge¹, B. Bizimungu², J. R. Tucker¹, M.C. Therrien¹, A. Tekauz², F. Eudes³, M. Savard⁴ and B. G. Rossnagel⁵. ¹Brandon Research Centre, Agriculture and Agri-Food Canada, Brandon, MB R7A 5Y3; ²Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB R3T 2M9; ³Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB T1J 4B1; ⁴Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON K1A 0C6; and ⁵Crop Development Centre, University of Saskatchewan, Saskatoon, SK S7N 5A8, Canada.

Infection of barley (*Hordeum vulgare* L.) by *Fusarium graminearum* (Schwab) is associated with accumulation of mycotoxins such as deoxynivalenol (DON) which play a significant role in Fusarium head blight (FHB) pathogenesis. A study was conducted to determine the effectiveness of using such mycotoxins in anther culture system for doubled haploid (DH) production to select mycotoxin tolerant barley plants with improved FHB resistance in the field. Twelve crosses varying in FHB resistance were subjected to *in vitro* selection (IVS) using a mixture of 2 or 3 mycotoxins. All fertile IVS and control DH lines from 7 crosses involving “exotic” FHB resistance sources were evaluated for FHB resistance in the Brandon nursery in 2001 and 2002, while 5 standard breeding crosses were evaluated in 2002. DON content was determined by the ELISA technique at Ottawa. Of 7 exotic crosses, only the two-row sub-group of Chevron/CDC Fleet cross showed significantly lower DON content of IVS vs. control group in 2001. Among the 5 standard crosses, only IVS lines from Rivers//Rivers/SB93806 cross had significantly lower DON content than control lines. Several IVS lines from both populations had substantially lower DON content than their parents. In conclusion, *in vitro* selection was effective in improving FHB in only some crosses but further testing is needed.

Physical mapping of a Fusarium head blight QTL on chromosome 3BS of wheat using a bacterial artificial chromosome (BAC) library. A. Brown Hoepfner¹, D.J. Somers¹, S. Cloutier¹, A. Walichnowski¹, S. Liu², J. Anderson². ¹Cereal Research Centre, Agriculture and Agri-food Canada, 195 Dafoe Road, Winnipeg, MB, R3T 2M9, Canada; ²Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN, 55108, U.S.A.

A major quantitative trait locus (QTL) for Fusarium head blight (FHB) resistance was identified on chromosome 3BS of wheat (*Qfhs.ndsu-3BS*). Physical mapping of the targeted region is an important step toward further marker development and map based cloning of a resistance gene. The objective of this study is to physically map the *Qfhs.ndsu-3BS* region. Microsatellite and STS markers previously mapped to *Qfhs.ndsu-3BS* were used to screen a BAC library, and clones were fingerprinted using the SNaPshot labelling kit and capillary electrophoresis. BAC end sequencing was also used to develop new markers and extend the contigs. Currently, 69 BAC clones are identified from the BAC pools of the Glenlea library using 19 markers. FPC was used to assemble 57 of these clones into 14 contigs. Only one contig (ctg01) was comprised of BAC clones identified by different markers. The contigs range in length from 72 to 169 Kbp and have a total length of 1,481 Kbp. More markers need to be identified to complete the physical

map. We will continue to exploit the rice genome sequence, as well as BAC end sequencing to develop more markers in this region and continue with BAC fingerprinting. Concurrently, large populations are being developed and phenotyped to fine map the region between GWM533 and GWM493.

Enhancement of spring wheat FHB resistance through pyramiding of genes from different sources. Wenguang Cao¹, George Fedak¹ and Jeannie² Gilbert. ¹. *Eastern Cereal and Oilseed Research Center, Agriculture and Agri-Food Canada, Ottawa, ON K1A 0C6 Canada*; ². *Cereal Research Center, Agriculture and Agri-Food Canada, Winnipeg, MA R3T 2M9 Canada*. Germplasm development for spring wheat fusarium head blight (FHB) resistance is a critical first step in a breeding program. The objective of this study was to enhance spring wheat FHB resistance through pyramiding genes from two different sources: Sumai 3 and Frontana. Two hundred and ninety four advanced lines (F₇) were derived from two crosses : Frontana/Sumai 3//N 894013/Cimmyt 11 and Frontana / Sumai 3// Ning 894013 / Wuhan 2-37e using pedigree method. In addition, one hundred and seventy four doubled haploid lines were obtained from the cross HC 467/AC Superb. After preliminary FHB screening with two replications in the field (2002), twenty advanced or homozygous lines were selected based on Fusarium symptoms and DON level. These twenty lines were further evaluated in 2003 for incidence, severity, DON content and yield with four replications in two FHB nurseries: Winnipeg (spray inoculation) and Ottawa (corn and barley kernel inoculation). The cultivars or lines: Quantum, Sumai 3, Roblin and HY 644 were included as checks. Correlation analyses showed that DON level has a positive correlation with FDK, severity and incidence, with coefficients of 0.77, 0.61 and 0.53, respectively and that yield had a negative correlation with DON level and FDK, with coefficients of -0.60 and -0.76. The results indicated that 10 lines had better resistance and higher yield than HY644 or Quantum. Lines L662-27-9, L662-43-8, HC 1090, HC 933, HC 1123 and H12637 were close to or better than Sumai 3 in terms of FHB resistance, DON content and yield. These lines could be used as parents for development of FHB-resistant cultivars.

Progress in developing cultivars and germplasm with FHB resistance in Eastern Canada. Comeau A., Dion Y., Rioux S., Butler G., Langevin F., Martin R.A., Nass H., Fedak G., Xue A., Voldeng H., Gilbert J., Dubuc J.P. *Agriculture and Agri-Foods Canada (abridged AAFC), Ste-Foy, GIW 2B1*; (YD, SR) CEROM, St-Bruno-de-Montarville, Qc, J3V 4P6; (GB, HV, GF, AX) ECORC, AAFC, Ottawa, K1A0C6;(FL) 121 Bon-Air, Ste-Catherine-de-la-Jacques-Cartier, G0A 3M0; (RAM, HN) AAFC, Charlottetown, PEI, C1A 7M8; (JG) Cereal Research Center, Agriculture and Agri-Food Canada, Winnipeg, MB, R3T 2M9, Canada; (J.D.) 1499 J.C.Cantin, Cap Rouge, Qc, G1Y 2X7.

Efforts to develop germplasm and cultivars resistant to Fusarium Head Blight (FHB) were initiated in 1979 in Eastern Canada. After a long and difficult struggle with the genetic complexity of the problem, efforts are now paying off. Among the showcase examples of success, there is a line that was supported for registration in 2002, CRGB-O-623.4. This line has resistance not too far from that of Sumai 3, but the yield and other agronomic traits are quite adequate. Another line, AW488, is an intriguing case. It does

not have any source of improved type 2 resistance, and is derived from a cross to Napier. It was observed that Napier has type 1 resistance but shows this resistance with a bit more variability from year to year, whereas AW488 shows good stability of FHB resistance over years. In the germplasm development area, progenies from crosses to Brazilian cultivars show promise, as do progenies from crosses to Chinese and Japanese sources, and interspecific hybrids. Germplasm with medium severity of symptoms and yet low DON were also noted.

A progress report on the incorporation of *Fusarium* head blight resistance into Canadian wheat cultivars using an *in vitro* selection technique. François Eudes¹, Sadash Sadasivaiah¹, Robert Graf¹, Sylvie Rioux², André Comeau³, François Langevin³ and Nathalie Lanoie⁴. ¹*Agriculture and Agri-Food Canada, Lethbridge, Alberta T1J4B1, Canada;* ²*CÉROM, Sainte-Foy, Québec, G1P3W8, Canada;* ³*Agriculture et Agro-Alimentaire Canada, Sainte-Foy, Québec, G1V2J3, Canada;* ⁴*Semico Inc. Ste-Rosalie, Québec, J0H1X0, Canada.*

A total of 70 crosses were made between Canadian cultivars and *Fusarium* head blight (FHB) resistant genotypes. *In vitro* selection of embryos produced from anthers of F1 hybrids cultured on a medium containing trichothecenes was conducted. The number of doubled haploids regenerated was about 10% compared with the trichothecene free treatment. Some of the DH lines were tested for *Fusarium* resistance in the greenhouse (F>32) and under field conditions at two locations (F>6). In general, about 16% of the DH lines had significantly fewer diseased spikelets than their parents, 71% had intermediate susceptibility, and 13% had significantly more diseased spikelets than the parental lines. Populations derived from crosses with CM82036 and Ning894013 failed to show transgressive segregation for FHB resistance. The *in vitro* selection technique failed to identify lines with the main type II resistance QTL *Qfhs.ndsu-3BS* from crosses with CM82036 and Ning894013. The best results were obtained when CIMMYT and Brazilian lines were crossed with Canadian cultivars. These results suggest that parental recombination may have an impact on the efficiency of *in vitro* selection.

The use of an *in vitro* selection technique utilizing trichothecenes has potential to reduce time and resources required to screen for FHB resistance, and is a useful tool in developing FHB resistant cultivars.

Impact of trichothecene on *Fusarium* head blight type II resistance in six cereal species. François Eudes¹, François Langevin² and André Comeau². ¹*Agriculture and Agri-Food Canada, Lethbridge, Alberta T1J4B1, Canada;* ²*Agriculture et Agro-Alimentaire Canada, Sainte-Foy, Québec, G1V2J3, Canada;*

To test the impact of trichothecenes on cereal *Fusarium* head blight, six species were inoculated with two *F. graminearum* strains, the trichothecene non-producing GzT40 strain and the wild parental Gz3639 strain. During three weeks of observation, the fungal strains showed extreme differences in aggressiveness in wheat, durum wheat, barley and triticale. While the GzT40 mutant did not spread into wheat, barley and triticale rachis, the wild-type strain quickly spread in the spike, as previously reported in wheat. In oats and rye, the fungal spread was not significantly different between strains, which we believe to be related to two factors: the inflorescence morphology for oat and the prolonged opening of the flowers for rye. Durum wheat is the only species where the

trichothecene non-producing strain GzT40 could spread in the spike beyond the inoculation step-point. Species responses to inoculation with both strains will be discussed according to genomes (especially D and R), inflorescence morphology, and in the light of resistance types II to IV. A new hypothesis is proposed: the basic level of resistance to fungal spread (type II) is very low in durum wheat, while genotypes tested from common wheat, barley, rye, triticale and oats all possess basic factors of type II resistance, which trichothecenes can inhibit to diverse degrees.

Androgenic ability of FHB resistant barley accessions. G. Fonquerne, I. Clermont, L. Laroche, S. Marchand, and F.J. Belzile. *Département de phytologie, Université Laval, Québec (QC), Canada, G1K 7P4.*

Most Fusarium head blight (FHB) resistant barley (*Hordeum vulgare*) accessions are relatively poor from an agronomic point of view. Due to the complex inheritance of FHB resistance, introgression of this trait into well adapted local germplasm will likely require multiple generations of crossing and selection in order to combine resistance and agronomic performance, even with the use of doubled haploids. Unfortunately, little is known concerning the androgenic ability of genotypes providing FHB resistance and so it is not known which of these could prove interesting in the production of doubled haploid populations. The objective of a first experiment was to compare the androgenic ability of eight barley accessions, known to offer some resistance (Chevron, Gobernadora, Seijo II, Shyri, Svanhals, Zhedar I, F104-250-9 and C97-21-38-3), with three cultivars (ACCA, Léger and Cadette) whose androgenic response was already well characterized. In a second experiment, the androgenic ability of F₁ hybrids, involving some of these genotypes used as parents, was measured and compared to that of the parental genotypes. Very large and significant differences were observed in the number of green plants produced by the different accessions and F₁s. In some cases, the androgenic potential proved so low that only a conventional approach, based on selfing to reach homozygosity, would seem justified.

Tissue specific biochip microarray analysis of genes differentially expressed in *F. graminearum*-challenged wheat heads. Saber Golkari, Suvira Prashar, Jeannie Gilbert and J. Douglas Procnier. *Cereal Research Centre, AAFC, 195 Dafoe Rd., Winnipeg , MB,R3T 2M9.*

Fusarium head blight of wheat caused by *Fusarium graminearum* Schwabe (teleomorph *Gibberella zeae* (Schw.) Petch) remains a significant threat to wheat production throughout the world. Genome profiling of gene expression using microarray technology provides a new opportunity to discover novel genes specifically up or down-regulated during the course of pathogen attack on resistant plants. A cDNA biochip representing 5664 wheat genes derived from *F. graminearum*-challenged wheat heads was used for tissue specific profiling of differentially expressed genes in response to plant infection by *F. graminearum*. A FHB-resistant line carrying three major resistance related QTLs mapped on chromosomes 3BS, 6BS and 5AL was used for this experiment. The wheat head was dissected into its composite tissues including glumes, lemma, palea, ovary, anther and rachis. Dissection was conducted to eliminate the contamination of transcriptomes from non-responsive tissues or cells that may compromise the resulting gene expression data. Reproducibility and accuracy of data were obtained by using five

biological replicas and “chip filtering”. Different tissue specific subsets of responsive genes were detected. These tissue specific expression patterns will enable the generation of microgenomic signatures that can be used to discriminate between the susceptible and resistant plants.

Isolation, characterization and physical mapping of differential clones from a SSH library for Fusarium Head Blight (FHB) resistance. Fangpu Han, George Fedak, Therese Ouellet and Daryl Somers. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, 960 Carling Ave, Bldg 50. Ottawa, Ontario, K1A 0C6, Canada. Phone: 613-759-1393, Fax: 613-759-6559, Email: fedakga@agr.gc.ca.* Isolation, physical and genetic mapping of FHB resistance ESTs in wheat are reported. About 1794 ESTs were sequenced and screened for differential gene expression following infection by *F. graminearum*. Thirty-five ESTs have been confirmed by Northern blot analysis to be either up or down-regulated following infection by *F. graminearum*. Twenty ESTs were selected for mapping. The ESTs were screened against Southern blots of digested genomic DNA from the whole series of nullisomic-tetrasomic and ditelosomic lines of Chinese Spring. Two ESTs were found to be non-specific sequences, as they hybridized to maize, barley, rye, *Elymus* and *Thinopyrum*, so they may be present throughout the grass family. Five ESTs were group specific sequences. Six ESTs belonged to non-specific sequences, but were mapped to several chromosomes of the A, B and D genomes. Six ESTs were repetitive sequences. We determined the more precise location of the differential ESTs by using the series of chromosome deletion stocks. It was shown that the 14 ESTs detected 99 fragments using restriction enzyme EcoRI, and 59 bands were assigned to chromosome bins, while 40 fragments were not assigned to chromosomes. The various clones employed in this study were screened for polymorphism on parents of two mapping populations. Nine ESTs showed polymorphic patterns on the Wuhan x Maringa DH population. One EST related to a QTL for TypeII resistance located on chromosome 2AL with SSR markers.

Quantitative trait loci for fusarium head blight resistance in Chevron x AC Stephen barley. L. Langille¹, K. Armstrong^{1,2}, K.M. Ho¹, G. Fedak^{1,2}, M. Kuc¹, R. Martin³, M. Savard¹, G. Butler¹, M. Burvill⁴. ¹*Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, K1A 0C6*; ²*Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, R3T 2M9*; ³*Crops and Livestock Research Centre, Agriculture and Agri-Food Canada, Charlottetown, PEI, C1A 4N6*; ⁴*Canadian Food Inspection Agency, Ottawa, ON, K1A 0Y9*.

Selection of fusarium head blight (FHB)[*Fusarium graminearum* Schwabe]-resistant barley (*Hordeum vulgare* L.) genotypes has been difficult because screening methods are labour- intensive and disease levels are greatly affected by environment. Molecular markers are now used by some breeders to select barley germplasm with *Fusarium*-resistance genes. The purpose of this study was to map the genome of a Chevron x AC Stephen doubled haploid population using simple sequence repeats (SSRs), and to identify markers linked to FHB-resistance quantitative trait loci (QTL) from Chevron. Data were collected from six tests grown at three locations (Ottawa and Charlottetown, Canada, and Hangzhou, China) between 1999 and 2001. A partial map covering 596 centimorgans was constructed from 83 SSR loci. QTL were identified associated with

deoxynivalenol (DON) concentration, visual symptoms of FHB infection, plant height, days to heading, and plot yield. A total of three DON and four visual symptoms QTL associated with FHB-resistance from Chevron were detected on chromosomes 5H(7), 6H, and 7H(1). Only the DON and visual symptoms QTL on 5H(7) were independent of QTL for increased plant height and days to heading. Although results are promising, more data are needed to determine if markers linked to these QTL will be useful for marker-assisted selection.

Session 5: Epidemiology

Genetic and pathogenic diversity of *Fusarium pseudograminearum* and *F. graminearum* causing head blight of wheat in Australia. O. A. Akinsanmi^{1,4} V. Mitter¹, S. Simpfendorfer², D. Backhouse³, D. Yates⁴ and S. Chakraborty¹ *CSIRO Plant Industry, CRC for Tropical Plant Protection, University of Queensland, 4072 QLD; ²NSW Agriculture, Tamworth; ³University of New England, Armidale; ⁴Department of Botany, University of Queensland, Australia.*

Fusarium head blight (FHB) of small grain cereals has emerged as a significant problem worldwide. Its incidence in Australia is sporadic causing 20-100% loss in some wheat paddocks. *Fusarium* spp. isolated from wheat in Queensland and northern New South Wales were identified using species-specific PCR assays. A total of 199 *F. pseudograminearum* isolates and 118 of *F. graminearum* were evaluated for quantitative differences in aggressiveness and for specialization on wheat cultivars with varying levels of resistance to FHB in plant infection assays. Genotypic diversity of the isolates was evaluated using amplified fragment length polymorphism technique. There were significant ($P < 0.0001$) differences in aggressiveness among the 10 *Fusarium* spp. evaluated, but there were no significant differences among isolates of the two dominant species *F. graminearum* and *F. pseudograminearum*. There was a significant cultivar by isolate interaction in FHB severity at 14 day after inoculation (DAI), but this disappeared when FHB severity data at 3, 5, 7, 10 and 14 DAI were examined as the Area Under the Disease Progress Curve. This indicates a lack of a clear cut pathogenic specialization, however further work is needed to confirm this. Both *F. graminearum* and *F. pseudograminearum* populations displayed high genotypic diversity and random association among loci. Among the 53 isolates of *F. graminearum* and 62 isolates of *F. pseudograminearum* analyzed, 52 and 60 unique AFLP multilocus haplotypes, respectively, were identified. Effective number of alleles, G_{ST} and Nei's gene diversity measures were greater among *F. graminearum* isolates than *F. pseudograminearum*. These indicate that sexual reproduction has a considerably larger influence on the population structure of *F. graminearum* than in *F. pseudograminearum*.

Identification of *Fusarium* species responsible for *Fusarium* head blight of barley in Quebec. J.V. Bourdages¹, S. Marchand¹, S. Rioux², F.J. Belzile¹. ¹*Département de phytologie, Université Laval, Sainte-Foy (Québec), Canada G1K 7P4; ²CEROM, Sainte-Foy (Québec), Canada G1P 3W8*

One of the particular challenges of developing new cultivars with increased resistance to *Fusarium* head blight (FHB) is the large number of *Fusarium* species involved. In the context of a plant breeding program as well as in the development of a better understanding of the epidemiology of the disease, it would be highly desirable to have a good working knowledge of the most common species and how their prevalence may vary in space and over time. In this work, we identified the causal species of FHB in infected barley (*Hordeum vulgare*) seed harvested in various regions of Québec in 2000, 2001 and 2002. Overall infection levels (% of infected seed) were 8.3%, 8.2% and 36.3%, respectively, for the three years. Using conventional identification methods (growth and observation of macroconidia under the microscope), we found that three species were responsible for most of the infections : *F. graminearum* (41.0% of infected

seed), *F. poae* (23.1% of infected seed) and *F. avenaceum* (15.8% of infected seed). A fourth species, *F. sporotrichoides*, although less prevalent (5.6%), was found in each of the three years. In future work, we plan to examine the genetic diversity among this collection of *Fusarium* species. Such data will prove useful in devising the best strategies in the development of effective FHB control.

Identification of crop production factors associated with the development of Fusarium head blight in spring wheat in southeast Saskatchewan. M.R. Fernandez, F. Selles, D. Gehl, R.M. DePauw, and R.P. Zentner. *Agriculture and Agri-Food Canada (MRF, FS, RMD, RPZ) P.O. Box 1030, Swift Current SK, S9H 3X2; and (DG) P.O. Box 760, Indian Head SK, S0G 2K0*

Because of the increasing significance of Fusarium head blight (FHB) in western Canada, identification of crop production factors (CPF) associated with the development of this disease would help to devise a strategy for its control. From 1999 to 2002, 648 wheat fields were sampled in southeastern SK. Environment was the most important factor determining disease development. The effects of the various CPFs on FHB were lower in years with high (2001) and low (1999 and 2002) disease pressure, compared to a year with moderate (2000) disease pressure for this region. The CPFs that affected FHB the most were application of a glyphosate formulation (GF), tillage practice, crop rotation, and cultivar susceptibility. GF application in the previous 18 months was significantly associated with higher FHB levels every year; it was the only CPF in 1999, and one of only two CPFs in 2002, that affected FHB, indicating that its effect was not influenced by environmental conditions as much as other CPFs. The relative effect of the other CPFs on FHB varied from year to year, and were significant in only one or two years. When wheat grown under minimum-till was analysed separately, GF application displayed an even greater effect on FHB. It is not known if similar effects of GF on FHB would occur in environments different from the ones encountered in this study, or more conducive to FHB development. Based on the significant and consistent effect of previous GF application on FHB throughout the four years, further research to elucidate the underlying mechanisms is warranted.

***Fusarium* spp. in residues of cereal and noncereal crops grown in eastern Saskatchewan.** M.R. Fernandez¹, P.G. Pearse², and G. Holzgang². ¹*Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, P.O. Box 1030, Swift Current, SK, S9H 3X2;* ²*Saskatchewan Agriculture, Food and Rural Revitalization, 3085 Albert St., Regina, SK, S4S 0B1.*

In July of 2000 and 2001, residues of wheat (*Triticum aestivum* L.), canola (*Brassica* spp.), flax (*Linum usitatissimum* L.), lentil (*Lens culinaris* Medik.) and pea (*Pisum sativum* L.) crops grown the previous season were sampled from over 300 fields in eastern SK. The noncereal crops had been preceded by a cereal crop. Residues were plated on nutrient agar for fungal identification. Based on total isolations, most *Fusarium* spp. had similar relative frequencies in all residue types. These species ranged from pathogenic to weakly pathogenic on cereals. The most common was *F. avenaceum* (Fr.) Sacc. Among those at lower levels were *F. acuminatum* Ellis & Everh., *F. culmorum* (W.G. Sm.) Sacc., and *F. graminearum* Schwabe. All *Fusarium* spp. found in residues were also previously isolated from wheat and barley heads affected by Fusarium head

blight (FHB), and from discolored roots of cereals and noncereals in SK. One of the most common fungi in heads and roots was *F. avenaceum*. Colonization of canola, flax, lentil and pea residues by fungi commonly isolated from cereals affected by FHB or root rot suggests that rotations with these noncereal crops might not be an effective control strategy against cereal diseases caused by *Fusarium* spp. in SK. This is the first report of isolation of *F. graminearum* from residues of noncereal crops in western Canada.

***Fusarium* populations in underground tissue of pulse, oilseed and cereal crops grown in the Black soil zone of southeastern Saskatchewan.** M. R. Fernandez. *Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, P.O. Box 1030, Swift Current, SK, S9H 3X2.*

Underground plant tissue from 643 cereal, oilseed and pulse fields in southeastern SK was examined for *Fusarium* populations in 2000 and 2001. Many of the *Fusarium* spp. isolated from discolored roots/subcrown internodes had also been isolated from heads affected by *Fusarium* head blight in SK. The most abundant *Fusarium* spp. were *F. avenaceum* (Fr.) Sacc. and *F. equiseti* (Corda) Sacc. *F. avenaceum* was present at the highest levels in pulses, particularly in lentil (*Lens culinaris* Medik.). *F. acuminatum* Ellis & Everh., *F. culmorum* (W.G. Sm.) Sacc., *F. graminearum* Schwabe, and *F. sporotrichioides* Sherb. were isolated from cereal and noncereal tissue at lower levels. When the fungal populations in wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) subcrown internodes were analysed according to crop history, a preceding noncereal crop did not change the incidence of most species, including *F. culmorum* and *F. graminearum*. In addition, in most cases, the isolation frequency of total *Fusarium* spp., and particularly that of *F. avenaceum*, was higher when wheat or barley were preceded by a noncereal crop than when preceded by another cereal crop or summerfallow. This is also the first report of isolation of *F. graminearum* from roots of field-grown pulse and oilseed crops in western Canada.

Inhibition of *Fusarium* sp. by hen egg white lysozyme. Y. Gao, S. Krentz and S. Smith. *INOVATECH BIOPRODUCTS, 31212 Peardonville Road, Abbotsford, BC V2T 6K8*

The objective of this study was to investigate the efficacy of hen egg white lysozyme in inhibiting the growth of *Fusarium* sp. to reduce the production of deoxynivalenol (DON) by the fungus during malting processes. The experiments were carried out under *in vitro* conditions. The *Fusarium* culture was isolated from infected barley grains. The culture was inoculated on potato dextrose agar (PDA). PDA plugs with *Fusarium* sp. were placed into petri dishes containing various concentrations of lysozyme solutions prepared in potato dextrose broth (0 – 250 ppm). The petri dishes with the fungal plugs and lysozyme solutions were incubated at room temperature (25±1°C). The fungal growth was monitored by measuring the area of the petri dishes covered by the fungus. On day 14, the areas in the petri dishes covered by the fungus for the treatments with 100 and 250 ppm lysozyme were only 24.7% and 16.6%, respectively, compared to 71.7% for the control. These results indicate that lysozyme is effective in inhibiting the growth of the *Fusarium* isolate. Future trials are needed to confirm the results under *in vivo* conditions.

The effect of *Trichoderma harzianum* on the production of perithecia of *Gibberella zea* on wheat residue. S. Inch and J. Gilbert. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, Manitoba, Canada, R3T 2M9*

Fusarium head blight (FHB) is currently the most important disease of wheat and other small grains in Canada. In Manitoba, the principal pathogen associated with FHB is *Gibberella zea* (Schw.) Petch. Perithecia and ascospores of *G. zea* develop on residue in the spring and are the primary source of inoculum. Presently, there are no registered resistant wheat varieties, and no reliable chemicals or biological agents to control FHB. The objective of this study was to investigate the effect of *Trichoderma harzianum* (Rifai) on the production of perithecia and ascospores of *G. zea* on wheat residue. Spore suspensions, or cell-free filtrates of *T. harzianum* isolates, were applied to wheat residues at 24 h before, co-inoculated, or 24 h after inoculation with *G. zea*. Petrie dishes containing the treated residues were placed under UV light in a randomized complete block design with 4 replicates per treatment. Development of perithecia and ascospores of *G. zea* were monitored. Compared to controls, perithecia and ascospore development were substantially reduced on residues that were inoculated with either spore suspensions or cell-free filtrates of *T. harzianum* 24 h before *G. zea*. Residues that were co-inoculated showed moderate reduction. No reduction in perithecial development was achieved when the residues were inoculated first with *G. zea*.

A method to observe barrage zone formation in *Fusarium graminearum* (*Gibberella zea*). B.D. McCallum, A. Tekauz, and J. Gilbert *Agriculture and Agri-Food Canada, Cereal Research Centre, Winnipeg, Manitoba, R3T 2M9*.

Vegetative incompatibility within *Fusarium graminearum* Schwabe has previously been determined through the use of auxotrophic mutants. While this method provides direct proof of heterokaryon formation, it is labour intensive. A rapid method for determining vegetative compatibility through the formation of barrage zones was developed. Under the proper cultural conditions, barrage zones of raised mycelia were observed at the junctions of vegetatively incompatible isolates. The distinctiveness of these zones was influenced by the isolates used, the medium, and light intensity. *Fusarium graminearum* isolates in different vegetative compatibility groups formed distinct mycelial interaction zones at their junctions; whereas pairs in the same vegetative compatibility group had no visible reaction, a minimal reaction, or in a few pairs a "line gap" of sparse mycelium at their junctions. Barrage zone formation was used to identify the proportions of *F. graminearum* isolates recovered from barley spikes inoculated with an isolate mixture. This could also be used to investigate the epidemiology and population biology of *F. graminearum*.

Molecular genetic variation, geographical differentiation and hybridisation in populations of *Fusarium graminearum* in Canada. P. K. Mishra, J. P. Tewari, R. M. Clear and T. K. Turkington. (P.K.M., J.P.T.) Univ. of Alberta, Department of Agricultural, Food, and Nutritional Science, 410 H Agric./Forestry Centre, Edmonton, AB, T6G 2P5; (R.M.C.) Canadian Grain Commission, Grain Res. Laboratory, 1404-303 Main Street, Winnipeg, MB, Canada R3C 3G8; and (T.K.T.) Lacombe Res. Centre, Agric. and Agri-Food Canada, 6000 C&E Trail, Lacombe, AB, T4L 1W1.

In this investigation, 309 monoconidial isolates of *Fusarium graminearum* Schw. obtained from fusarium-damaged kernels of wheat collected across the Canadian prairie provinces were analyzed. Population genetic variation was assayed by the restriction digestion of polymerase chain reaction amplified intergenic spacer region of nuclear ribosomal DNA and by inter-simple sequence repeat (ISSR) fingerprinting. Significantly high genetic diversity and frequent gene flow among/between population samples of *F. graminearum* was found. The analysis of molecular variance indicated that most genetic variability was present within populations. The distribution of genetic diversity across western Canada showed a random genetic structure of *F. graminearum*, suggesting the movement of genotypes across the provinces perhaps either by infected seed and/or by wind-borne spores. Analysis of multilocus associations showed that all populations were in linkage equilibrium, indicating that sexual recombination is a frequent phenomenon in *F. graminearum* populations of Canada. The hybridisation between different genotypes of *F. graminearum* was also revealed by ISSR fingerprinting. High gene flow and genetic diversity, and frequent recombination and hybridisation between genotypes observed in the populations of *F. graminearum* have substantial implications for further pathogen adaptation and development of fusarium head blight in Canada. Taken together these results underscore the need for an integrated, coherent and sustainable approach for the effective management of fusarium head blight in Canada.

Determination of *Fusarium graminearum* chemotype based on upstream sequences of the *Tri5* gene. M.K. Tan^A, S. Simpfendorfer^B, D. Backhouse^C and G.M. Murray^D. ^ANSW Agriculture, Menangle, New South Wales 2568, Australia; ^BNSW Agriculture, Tamworth, NSW 2340; ^CUniversity of New England, Armidale, NSW 2351; ^DNSW Agriculture, Wagga Wagga, NSW 2650.

The *Tri5* gene which encodes trichodiene synthase, the first step in the trichothecene biosynthetic pathway, is reported to co-segregate with the locus governing the type of trichothecene produced. Sequence analysis of 26 isolates with known chemotype, representative of the global lineages of *F. graminearum*, revealed that all deoxynivalenol (DON) chemotypes displayed characteristic deletions in a region in the upstream sequences of the *Tri5* gene. The distinct length polymorphisms in this region between the DON and nivalenol (NIV) chemotypes allowed a PCR assay to be developed in this study to distinguish between these chemotypes. Six *F. graminearum* isolates from southern NSW in Australia and twenty overseas isolates were analysed using this technique and compared with published assays utilising polymorphisms in the *Tri7* and *Tri13* genes to distinguish DON and NIV chemotypes. Results demonstrated the potential for reliable use of the molecular tool targeting the upstream sequences of the *Tri5* gene to differentiate NIV and DON chemotypes. Two of the isolates from southern NSW were of the DON chemotype while the other four were of the NIV chemotype. Further research is

required to establish the relative distribution of DON and NIV chemotypes in the NSW and Australian grain-belt.

Moisture retention of cereal spikes and fusarium head blight risk. T. K. Turkington and K. Xi. *Agriculture and Agri-Food Canada, Lacombe Research Centre, 6000 C&E Trail, Lacombe, AB, T4L 1W1*; (K.X.) *Alberta Agriculture, Food and Rural Development, c/o Lacombe Research Centre, 6000 C&E Trail, Lacombe, AB, T4L 1W1*.

In western Canada all commercially available wheat cultivars are vulnerable to fusarium head blight (FHB) caused by *Fusarium graminearum*, but vary in their level of susceptibility. In general, awned wheat cultivars tend to be somewhat more susceptible than awnless types, although exceptions occur. A study was conducted to compare the level of moisture retention by awned (cv. McKenzie) and awnless (cv. AC Barrie) cultivars of CWRS wheat. Spikes with a small portion of stem of each cultivar were sampled and weighed at the early anthesis and early dough stages of development. Spikes were then mounted on styrofoam, sprayed with 2L of water in a research spray cabinet, and then allowed to stand for 10 minutes before weighing. At early anthesis there was significantly more moisture retained by the awned cultivar in three of four experimental trials, while in the remaining trial the awnless cultivar retained more moisture. No consistent pattern was observed at early dough where, in four of six trials no significant cultivar effect occurred, while opposite trends were observed for the remaining two trials. Awned wheat cultivars may have a greater propensity to intercept and retain moisture at early anthesis, which may result in increased disease risk. The awnless characteristic of some wheat cultivars may complement the incorporation of active mechanisms of resistance.

Assessment of the environmental suitability of the western Prairie region of Canada for fusarium head blight caused by *Fusarium graminearum*. T.K. Turkington, O.O.

Olfert, R. Weiss, R.M. Clear, K. Xi, and J.P. Tewari. (T.K.) *Lacombe Res. Centre, Agric. and Agri-Food Canada, Lacombe, AB, T4L 1W1*; (O.O., R.W.) *Saskatoon Research Centre, Saskatoon, SK, S7N 0X2*; (R.M.C.) *Canadian Grain Commission, Winnipeg, MB, Canada R3C 3G8*; (K.X.) *Alberta Agriculture, Food and Rural Development, c/o Lacombe Research Centre, Lacombe, AB, T4L 1W1*; (J.P.T.) *Univ. of Alberta, Department of Agricultural, Food, and Nutritional Science, Edmonton, AB, T6G 2P5*.

Fusarium graminearum (Schwabe) has continued to appear in more westerly regions of the Canadian Prairies after becoming well established in Manitoba and southeastern Saskatchewan. The potential for extensive development of fusarium head blight in the western Prairie region is explored. In central and northern Alberta, long-term June and July mean minimum and maximum temperatures are often 2-4°C lower than in the Red River valley of Manitoba. Temperatures are similar in southern regions of Alberta and the Red River valley of Manitoba. Rainfall, however, follows an opposite trend for central and northern Alberta, while irrigation in southern Alberta may be a factor to consider. Most sites in central and northern Alberta tend to have similar or even higher total rainfall and number of days with rain in June and July than epidemic areas in Manitoba. The CLIMEX™ model was used to predict the potential distribution and abundance of head blight caused by *F. graminearum* in western Canada. Higher ecoclimatic index (EI) values generated by CLIMEX™ indicated areas more favourable

for FHB. EI values suggested areas outside of Manitoba and eastern Saskatchewan could support survival and development of *F. graminearum*. The highest EI values occurred in the Red River valley and in areas around Edmonton. These projections are consistent with reports of *F. graminearum* infecting small grain cereals at temperatures below 15°C, given favourable moisture conditions. Furthermore, temperature variations coupled with favourable moisture conditions in some regions of Alberta may compensate for slightly lower summer temperatures.

***Fusarium* spp. associated with cereal and corn residue in Alberta.** T. K. Turkington, A. Kuzyk, R. Dunn, D.D. Orr, K. Xi., R. Lange, D. Benard, K. Kumar, J. Calpas, S. Ali, and B. Chapman. (T.K.T., D.D.O.) Agriculture and Agri-Food Canada (AAFC), Lacombe Research Centre, Lacombe, AB, T4L 1W1; (A.K.) AAFC Lethbridge Research Centre, Lethbridge, AB, T1J 4B1; (R.D.) Alberta Agriculture, Food and Rural Development (AAFRD), Lethbridge, AB, T1J 4V6; (K.X., K.K.) AAFRD, c/o Lacombe Research Centre, Lacombe, AB, T4L 1W1; (R.L., D.B.) Alberta Research Council, Vegreville, AB, T9C 1T4, (J.C., S.A.) AAFRD, Edmonton, AB, T6H 5T6; (B.C.) AAFRD, Barrhead, AB, T7N 1A4.

Since 2001, *Fusarium* spp. associated with cereal and corn residues in Alberta have been evaluated. *Fusarium avenaceum* (Corda ex Fr.) Sacc., *F. acuminatum* Ell. & Ev., *F. equiseti* (Corda) Sacc., and *F. culmorum* (W.G. Smith) Sacc. were the species most frequently recovered, although *F. graminearum* (Schwabe) was found in some samples, especially from southern Alberta. In 2001, *Fusarium graminearum* was isolated from grass residues at low levels from only 4 of 31 sites in southern Alberta and from 0 out of 15 sites in central Alberta. In central Alberta, 3 cereal fields out of 82 had low levels of *F. graminearum*. In southern Alberta in 2001, 2 of 3 cereal fields and 3 of 7 corn fields had detectable levels of *F. graminearum*. In 2002, this pathogen was detected in 14 of 30 cereal fields in southern Alberta, but only 3 of 41 cereal fields in central Alberta. No *F. graminearum* was detected in cereal residue from the Peace River region (11 fields). While not detected in grass samples from the Peace region (14 locations) or central Alberta (25 locations), *F. graminearum* was found at 4 of 33 locations in southern Alberta. In 2002 it was also detected in 9 of 12 southern Alberta corn fields, but none of 10 corn fields in central Alberta. Isolates from 2001 and 2002 are being checked to confirm that they are *F. graminearum* and not *F. pseudograminearum*. Residue samples from 2003 are currently being processed.

Histological study of stem infection in barley and wheat by *Fusarium graminearum*.

K. Xi⁺ and T.K. Turkington. Alberta Agriculture, Field Crop Development Centre, 6000 C & E Trail, Lacombe, AB, T4L 1W1, (T.K.T.) Agriculture & Agri-Food Canada, Lacombe Research Centre, 6000 C & E Trail, Lacombe, AB, T4L 1W1.

Systemic fungal growth by *Fusarium culmorum* in winter wheat has been demonstrated by isolation in studies from the Netherlands and UK. In Canada, *Fusarium graminearum* isolates from roots and crowns of wheat have been shown to cause fusarium head blight. The current study was undertaken to evaluate the potential for stem infection by artificially inoculating the growth medium in pots grown seedlings of AC Lacombe barley. Furthermore, naturally infected wheat kernels were grown out to assess the potential for systemic infection of *F. graminearum* into the stem. Crown and stem

discoloration was found from the inoculated barley and naturally infected wheat seed. Mycelial infection and sporulation were observed in the crown area of barley using light and electron microscopy. Systemic infection evidenced by the presence of fungal hyphae was observed in the stem tissues of both barley and wheat above the crown up to 15-20 cm at approximately the 3rd internode. *Fusarium graminearum* was identified through isolation. In conclusion, no evidence was found for systemic infection leading to head blight of barley or wheat. Under growth chamber conditions seed and seedling infection by *F. graminearum* and subsequent systemic fungal growth can lead to infection of the lower stem. Research is needed to determine if similar results occur under field conditions.

Effect of harvesting time on incidence of seedborne *Fusarium* spp. in spring wheat in eastern Ontario. A.G. Xue, J. Frégeau-Reid, J. Rowsell, C. Babcock, G.J. Hoekstra, E. Sparry, Y. Chen, and F. Sabo. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, K.W. Neatby Building, 960 Carling Avenue, Ottawa, ON K1A 0C6; (J.R.) New Liskeard Agricultural Research Station, Box 6007, 340 Armstrong St., New Liskeard, ON P0J 1P0; (G.J.H.) 506 Clothier St. W, R.R.#5, Kemptonville, ON K0G 1J0; (E.S.) C&M Seeds, Palmerston, ON N0G 2P0.*

The effect of five harvesting times on incidence of seedborne *Fusarium* spp. was examined using three spring wheat cultivars grown at two locations in Ontario in 1999 and 2000. Twelve *Fusarium* spp. were isolated from 3,831 of the 24,000 seeds which were surface disinfected and plated onto modified potato dextrose agar. *Fusarium sporotrichioides*, *F. graminearum*, *F. poae*, *F. equiseti*, and *F. avenaceum*, were the most frequently detected species and were isolated from 6.8, 3.7, 2.8, 1.8, and 0.6% of the seeds, respectively. The remaining species, *F. acuminatum*, *F. crookwellense*, *F. culmorum*, *F. oxysporum*, *F. sambucinum*, *F. solani*, and *F. tricinctum*, collectively infected only 0.3% of the seeds. The incidence of *F. graminearum*, *F. sporotrichioides*, and total *Fusarium* spp. increased about two fold, from 1.7, 3.9, and 9.5% in seed harvested very early to 5.5, 8.7, and 19.8%, respectively after delayed harvest. Also, *F. poae* had significantly lower incidence at very early and early harvest times compared to normal or later harvest dates. Incidence of the other *Fusarium* spp. were relatively low and not affected by harvesting time. Cultivar, location, and year variation in the incidence of *Fusarium* spp. were observed and likely related to the different levels of varietal resistance to these pathogens, inoculum present, and weather conditions before and during harvesting times.

Pathogenicity of *Fusarium* species causing head blight in barley. A.G. Xue, K.M. Ho, C. Babcock, Y. Chen, F. Sabo, and M. Kuc. *Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, K.W. Neatby Building, 960 Carling Avenue, Ottawa, ON, K1A 0C6, Canada*

The pathogenicity of eight *Fusarium* spp. causing fusarium head blight (FHB) in barley was studied under controlled conditions. Six barley lines varying in resistance to FHB were artificially inoculated with six isolates each of *F. acuminatum*, *F. avenaceum*, *F. crookwellense*, *F. culmorum*, *F. equiseti*, *F. graminearum*, *F. poae*, and *F. sporotrichioides* at the late-flowering stage. Symptoms of FHB were rated as disease severity on a 0-9 scale, 4, 7, 14, 21, and 28 days after inoculation, and as percentage of

infected spikelets (IS) after 21 days. All species caused visible infections in the barley lines, but only *F. crookwellense*, *F. culmorum*, and *F. graminearum* resulted in severe disease development (>60% IS) and were considered highly pathogenic. *F. avenaceum* had IS of 48.3%, which was significantly lower than those of the three highly pathogenic species, being moderately pathogenic; and, the remaining species had <20% IS, being weakly pathogenic. There were significant differences ($P < 0.05$) in aggressiveness among isolates within species and in susceptibility among barley lines, suggesting that screening for resistance to FHB requires the use of aggressive isolates or a mixture of several isolates. This is also the first report showing that *F. crookwellense* is highly pathogenic and *F. avenaceum* is moderately pathogenic in barley.

Session 6: Disease management

Crop rotation as a tool for fusarium head blight disease management. *H. U. Ahmed, J. Gilbert, W. G. D. Fernando, A.L. Brûlé-Babel, and M.H. Entz. Department of Plant Science, University of Manitoba, Winnipeg, R3T 2N2, Canada; and (J.G.) Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, R3T 2M9 Canada.*

Fusarium head blight (FHB) of wheat is a devastating disease reducing yield, grade, and quality. As there is no FHB resistant cultivar available, FHB disease management largely depends on fungicides. Modification of crop sequence with non-host crops might be an eco-friendly alternative. A long-term rotation with oats, canola and peas was initiated in 2001 to determine the influence of crop rotation on FHB of wheat. The experiment was exposed to natural infection. The overall disease incidence and severity was low as the weather was not favorable for disease development. However, in two-crop rotations, the average of two years data showed higher disease incidence, severity and percent damaged kernel (%FDK) on wheat when wheat followed canola or peas compared to wheat-wheat or oat-wheat crop sequences. However, in three-crop rotations, the disease incidence, disease severity and %FDK were higher when wheat was grown for two or three years in a row. Growing wheat in monoculture and leaving the stubble in the field have supported the build up of inocula in the wheat plots contributing to higher disease incidence. These results suggest that crop rotation with non-hosts might be a potential option for FHB management.

Effect of fungicides on fusarium head blight and leaf pathogens in winter wheat. *A.L. Brûlé-Babel and W.G.D. Fernando. Department of Plant Science, University of Manitoba, Winnipeg, Manitoba, Canada R3T 2N2.*

Producers who routinely use fungicides for leaf disease control in winter wheat have questioned whether a single fungicide application could be used to control both fusarium head blight (FHB) and leaf diseases. The objectives of this study were to compare control of FHB and leaf diseases of winter wheat with the application of different fungicides and combinations of fungicides. Trials were conducted at one location in 1999 (Carman, Manitoba), and two locations (Carman and Winnipeg, Manitoba) in 2000, 2001 and 2002. Three fungicides, Tilt, Folicur and Bravo were compared in eight treatment combinations (including un-inoculated and inoculated controls). Yield, leaf disease incidence, and FHB incidence and severity were measured for all plots. Under high disease pressure fungicide treatments reduced both FHB and leaf diseases. Yield differences were primarily associated with differences in leaf disease control. Tilt provided the best control of leaf diseases. Folicur applied at heading provided some level of leaf disease control. Folicur and Bravo provided similar levels of FHB control. Weather conditions during flowering of winter wheat were often not conducive to FHB development. Therefore, disease forecasts may be useful to determine whether fungicide application is warranted in winter wheat.

The impact of rotation and fertility on root rot pathogens of wheat in the Brown soil zone of western Canada. M.R. Fernandez, and R.P. Zentner. *Semiarid Prairie Agricultural Research Centre, Agriculture and Agri-Food Canada, P.O. Box 1030, Swift Current, SK, S9H 3X2.*

Spring wheat (*Triticum aestivum* L.) grown in a replicated trial for one or two years after fallow, lentil (*Lens culinaris* Medik.), flax (*Linum usitatissimum* L.), or continuously (with and without fertilizer N) was examined for subcrown internode discoloration from 2000 to 2002 in southwestern SK. Lesioned tissue was plated on nutrient agar for fungal identification. The most common species were *Cochliobolus sativus* (Ito & Kurib.) Drechs. ex Dast. and *Fusarium* spp. Among the latter, *F. avenaceum* (Fr.) Sacc., *F. equiseti* (Corda) Sacc. and *F. pseudograminearum* O'Donnell & T. Aoki sp. were the most frequent. *F. avenaceum* is the most common *Fusarium* head blight (FHB) pathogen in western SK. Continuous wheat grown with recommended N rates and wheat after fallow had similar root rot severities, but the frequency of fungi differed. The highest root rot level was in wheat after lentil, and the lowest in continuous wheat at low N. The frequency of *Fusarium* spp. was low in the latter but high in wheat after lentil. Among the rotations examined, it appears that the most favourable to the development of root rot in wheat is a one year rotation with lentil. This rotation may also contribute to the build-up of inoculum for the development of FHB, which is an important emerging disease in many areas of SK, including the southwest.

Dry heat treatments to control *Fusarium graminearum* in infested wheat seed. J. Gilbert, A. Tekauz, S.M.Woods, and T.K. Turkington. *Cereal Research Centre, Agriculture and Agri-Food Canada, 195 Dafoe Road, Winnipeg, MB, R3T 2M9, Canada and (T.K.T) Lacombe Research Centre, 6000 C&E Trail, Lacombe, AB, T4L 1W1. Email: jgilbert@agr.gc.ca*

Fusarium head blight (FHB) is one of the most serious diseases of small grains in Canada and one mechanism of spread is through *Fusarium*-infested seed. Dry heat treatment of seed has been proposed as an alternative to chemical seed dressings and several treatments were examined to determine if they could control seed-borne *Fusarium* without detrimental agronomic effects. *Fusarium*-free and *Fusarium*-infested seed samples of CDC Teal were heat-treated at 90°C, 70°C, 50°C, 30°C or kept at room temperature (Control) for 5 days. Additional treatments at 70°C included 10 and 12 days, with and without a pre-heat treatment for 2 days at 38°C. Germination of seed and levels of *F. graminearum* Schwabe were assessed after treatment. Field assessments included emergence, height, yield, thousand kernel weight, and hectoliter weight in 2002 and 2003, and heading and maturity in 2002. Temperatures below 70°C were ineffective in reducing *Fusarium* infection. Treatment at 90°C killed both fungus and seed. After most 70°C treatments, recovery of *F. graminearum* was less than 1%. There were no significant differences for agronomic characteristics within either *Fusarium*-free or -infested seed among controls and treatments at 70°C for 5 days, and 70°C for 10 and 12 days with pretreatment.

Fungicide efficacy for control of FHB in large-scale wheat plots.

A. Tekauz, B. Hellegards and M. Savard. *Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, R3T 2M9; (B.H.) James Richardson International, Kelburn Farm, St. Adolphe, MB, R5A 1A1; and (M.S.) Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada, Ottawa, ON, K1A 0C6.* In most years, fusarium head blight (FHB) reduces the yield and quality of wheat grown in Manitoba and eastern Saskatchewan. Moreover, contamination of grain by deoxynivalenol limits end-uses and jeopardizes export sales. Control of FHB is needed, but is difficult to achieve as no wheat cultivars are resistant, and registered fungicides are not fully effective. To optimize the contribution of fungicides to management of FHB, several registered products were compared either singly, or as split treatments, in 0.5 ha field plots of AC Barrie, AC Snowbird and AC Superb spring wheat. Split fungicide treatments were applied to spikes and foliage at late boot and anthesis; single applications at anthesis only. Visual FHB damage was quantified on spikes prior to maturity and after harvest grain was assessed for levels of *Fusarium* damaged kernels, *Fusarium* spp. and deoxynivalenol. Plots were combine harvested to obtain grain yields. Compared to the untreated check, the best control (65-70%) and most consistent results with single or split treatments were achieved with Folicur, and Tilt + Bravo or Tilt + Folicur, respectively. The greatest yield boost (126% of check) was achieved with a split application of Folicur + Folicur. AC Snowbird and AC Superb developed higher natural levels of FHB than AC Barrie.

Eradication of *Fusarium graminearum* from infested barley seed by heat treatment.

A. Tekauz, T.K. Turkington and J. Gilbert. *Cereal Research Centre, Agriculture and Agri-Food Canada, Winnipeg, MB, R3T 2M9; and (T.K.T.) Research Centre, Agriculture and Agri-Food Canada, Lacombe, AB, T4L 1W1.*

Fusarium head blight of barley results in infestation of seed by the causal fungi, *Fusarium graminearum* Schwabe and other *Fusarium* species. When planted, infested seed may not germinate optimally, and may contribute to the spread of *Fusarium* fungi to new locations. Fungicide seed treatments are registered to mitigate 'seedling blight' in barley, but are not necessarily 100% effective in eradicating the fungi infesting the seed; consequently, an alternative control method, heat treatment, was tested for its effects on *Fusarium* fungi and subsequent seed and crop performance. *Fusarium*-infested and *Fusarium*-free seed lots of CDC Stratus barley were incubated at 30 - 90C temperatures for up to 12 days, and compared with a control treatment (room temperature). Percent seed-borne *Fusarium* and seed germination were assessed, and field trials were planted in 2002 in southern Manitoba to measure emergence, plant height, heading date, grain yield and 1000 kernel and hectoliter weights. *Fusarium graminearum* was eradicated from infested seed incubated at 70C for 5-12 days and 90C for 5 days; germination of infested (34% total *Fusarium*, 28% *F. graminearum*) seed was unaffected compared to the control (83%) except by treatment at 90C (0%). Germination in non-infested (1% *F. graminearum*) seed (86%), likewise was reduced only by the 90C treatment (23%). Only the 90C treatment affected agronomic components in the field, substantially lowering emergence and final grain yield.

Breakout Group Comments

1. Mycotoxins and FDK
2. Milling and Brewing
3. Livestock, Industrial End-use, ethanol production/Seed production and trade
4. Breeding for FHB Resistance- Wheat/Oat
5. Breeding for FHB Resistance- Barley/Corn
6. Molecular Breeding and Biotechnology
7. Disease Management
8. Epidemiology (including the role of infected seed)

Issues and Priorities Recommendations

Dear Industry and Government Sponsors and Workshop Participants:

Once again, we wish to extend our sincere thanks to sponsors for your generous support of the 3rd CWFHB, Winnipeg, Manitoba, December 9-12, 2003, and to all delegates who participated and provided valuable inputs. The meeting was well-attended and held the participants' interest to the very end. After the invited oral presentations to review our current understanding of the FHB situation in Canada, and new research highlighted in 60 posters, we held informed discussions on what is needed to forward the struggle against this devastating disease. Based on these deliberations, participants attending the 3rd CWFHB identified the following as priority needs during the next two years:

Industry/Regulatory

1. In wheat, to re-examine the requirement for kernel visual distinguishability (KVD) among classes, as less stringent, and alternative identification methods would allow for access to, and more rapid development of improved wheat varieties, including the production of 'feed' quality grain for the livestock, ethanol and other value-added industries.
2. For the Canadian Seed Growers Association (CSGA) to consider adding a test for the level of *Fusarium graminearum* and other *Fusarium* spp. as a component of seed certification for the information of the buyer.
3. For representatives of government regulatory agencies to attend future workshops to clarify regulations and interact constructively with other relevant parties to implement such changes to regulatory guidelines as are deemed desirable.
4. That the Canadian Food Inspection Agency's (CFIA) definition of plants with novel traits as 'PNT's be abolished, and that Canada use the same definition for genetically modified organisms (GMOs) used by other countries, i.e. as plants produced by a rDNA transfer, rather than those plants produced by any process, including conventional breeding, but having different or unusual traits.
5. To develop methodologies to mitigate the effects of DON-contaminated by-products from ethanol production, and the milling, malting and brewing processes, and to identify strategies to enhance utilization of FHB-contaminated grains by the livestock sector.
6. To investigate novel uses for FHB-contaminated grain as safe and useful, value-added products.

Mycotoxins

1. To significantly expand laboratory facilities and resources available for testing for deoxynivalenol (DON), particularly to expedite progress in developing resistant varieties in breeding programs and to assist in research related to food safety and quality.
2. To develop and validate alternative, rapid methods for detection of DON in whole grain or grain flour (e.g. near infra-red spectrometry - NIR) and for fusarium-damaged kernels (FDK) (e.g., image analysis), recognizing that different standards are needed for quality control (i.e., high accuracy) vs selection and screening.
3. For economic tests to screen for mycotoxins other than DON (e.g. nivalenol, DAS, T-2 and HT-2) which can be produced by *Fusarium* spp. other than *Fusarium graminearum*, which contribute to FHB in crops such as barley and oat.
4. For CFIA and Health Canada to expand their surveillance for DON and other mycotoxins in grain, foodstuffs, and other sectors of the market place.
5. To investigate sampling protocols for DON and other mycotoxins, as sampling has been identified as the greatest single source of error in determining accurate, meaningful, and reproducible mycotoxin levels.
6. To standardize analytical methods for detection of mycotoxins among laboratories by use of reference materials to obtain consistent, reliable, and comparable results.
7. To develop a better understanding of the implications of mycotoxin contamination for human health, especially for high risk groups, i.e., infants and children.

Conventional/Molecular Breeding

1. To continue investigating FHB in oats, assess its impact on the crop, develop mitigation strategies, and identify sources of resistance for use in breeding programs.
2. To support continued refinement and development of improved phenotyping, i.e., phenomics, to validate the identification of resistant sources, confirm the basis of genetic resistance, and effectively screen breeders' lines, as well as enable the best use of genomics, proteomics, metabolomics, etc.
3. To fully investigate the pathogenic diversity in the principal causal agent, *Fusarium graminearum*, and evaluate for the occurrence of specialization to different crops and different environments (warm vs cool), both to explain its pervasiveness in existing epidemic areas, and the likelihood of *F. graminearum* moving to new regions, or developing resistance to fungicides.

4. For additional research, in particular histological and histochemical studies, on the mechanisms of resistance, to better understand disease progress and identify weak points where attack on the pathogen can be targeted, or host resistance enhanced.
5. Continue other approaches such as molecular breeding and transformation, re. development of FHB-resistant varieties, as alternative routes to achieve the goal of minimizing the impacts of FHB.
6. For a continued research effort in development of *Fusarium*-resistant corn inbreds, that can be crossed to develop *Fusarium*-resistant hybrids.

Epidemiology/Disease Management

1. To clarify the role of corn, both as an additional major host of FHB (pink ear rot and stalk rot) and as a source of abundant and persistent *Fusarium* inoculum for the infection of small grain crops.
2. To study the role of crop residues as sources of primary inoculum, particularly residues of non-host crops (e.g. canola), to assist in the understanding of disease development, and in formulating effective integrated control strategies.
3. To clarify the impact of management practices (e.g., application of herbicides), and better understand the role of crop residues in pathogen survival for overwintering and increasing inoculum potential the following spring.
4. To identify or develop simple, standardized methods to assess and measure *Fusarium* spore concentrations in the atmosphere and to assist in determining the environmental factors responsible for inoculum production and release.
5. To refine and validate current FHB disease forecasting models for effective and economic fungicide application, and to provide for their timely and universal accessibility.
6. To develop more effective fungicides for management of FHB, and to provide Canadian producers with a registered product(s) for use in barley and oat.
7. For producers, agronomists, and industry to be better informed on the various management strategies available to combat FHB. This includes awareness of the various agronomic practices used (e.g., impact of planting seed infected with *Fusarium*, variety selection, crop rotations), as well as the environmental factors that affect disease (e.g., crop staging relative to spore release and the use of disease forecasting models).

Common Issues

1. To develop a committed long-term approach to resolve the FHB problem, one that is not de-railed by complacency arising from one or more years of low FHB.
2. For CFIA and Health Canada to re-examine Canada's FDK and DON standards and bring these into compliance with the recent or imminent changes to such standards both by our individual customers and international conventions, and to communicate with and assist industry in achieving this.
3. To slow or prevent movement of FHB to new areas (e.g., Alberta) and as such to protect our ability to source unaffected high quality grain over a wide region, maintaining Canada's competitive advantage.

We trust recognition of these needs and their implementation will serve to focus future endeavours on FHB in Canada, and lead to timely solutions to minimize the devastating impact of this disease.

Sincerely,

Jeannie Gilbert and Andy Tekauz
Chair and Member, Organizing Committee
3rd CWFHB/CCF

Destinataires : À l'intention des membres de l'industrie céréalière, des partenaires du gouvernement et des participants aux ateliers.

Mesdames, Messieurs,

Encore une fois, nous souhaitons remercier sincèrement nos partenaires de leur généreux appui lors du 3^e Colloque canadien sur la fusariose, qui s'est déroulé à Winnipeg, au Manitoba, du 9 au 12 décembre 2003, et tous les délégués qui y ont participé et offert une contribution significative. La réunion a été un franc succès, et le sujet abordé a retenu l'attention des participants jusqu'à la toute fin. À la suite des présentations orales sur la situation actuelle de la brûlure de l'épi causée par le *Fusarium* au Canada et sur les nouvelles recherches mises en vedette dans les 60 affiches, on a entamé une discussion approfondie sur les éléments essentiels permettant de lutter davantage contre cette maladie dévastatrice. À la suite des discussions, les participants au troisième colloque sur la fusariose ont dressé la liste des priorités pour les deux années à venir.

Industrie céréalière/réglementation

1. Blé : On doit réexaminer l'exigence liée à la distinction visuelle des grains (DVG) relativement au classement du blé, au fur et à mesure que d'autres types d'épreuves rapides et moins exigeantes permettront de mettre au point plus rapidement des variétés améliorées de blé et d'y avoir accès, y compris les variétés fourragères et celles en vue de la production d'éthanol, ainsi que celles créées pour répondre aux besoins de l'industrie de produits à valeur ajoutée.
2. L'Association canadienne des producteurs de semences (ACPS) doit se pencher sur le processus de certification une épreuve pour déterminer le taux de *Fusarium graminearum* et de d'autres espèces de *Fusarium* afin de fournir l'information à l'acheteur.
3. Les représentants des organismes de réglementation gouvernementaux doivent assister à des ateliers dans le futur afin de clarifier la réglementation et d'interagir de façon constructive avec les autres intéressés, pour faire en sorte que les modifications voulues soient apportées aux lignes directrices réglementaires.
4. Que l'Agence canadienne d'inspection des aliments (ACIA) abolisse ce qu'elle a établi comme définition pour les végétaux à caractères nouveaux (VCN), et que le Canada emploie la même définition que d'autres pays pour les organismes génétiquement modifiés (OGM) (c'est-à-dire, des plantes produites par la technique de transfert de l'ADN et ayant des caractères différents ou inhabituels, plutôt que des plantes produites par tout autre moyen que ce soit, y compris la sélection classique des végétaux).
5. Établir des méthodologies qui permettent d'atténuer les effets de la contamination par la vomitoxine DON des produits dérivés de la production d'éthanol, et des procédés de meunerie, de maltage et de brassage, et cerner des stratégies menant à une meilleure utilisation des grains contaminés par la brûlure de l'épi dans le secteur de l'élevage.
6. Se pencher sur de nouvelles façons d'employer le grain fusarié comme produit utile et sûr, à valeur ajoutée.

Mycotoxines

1. Agrandir les installations de laboratoire de façon importante et obtenir plus de ressources permettant d'effectuer l'analyse du désoxynivalénol (DON), particulièrement pour développer plus rapidement des variétés résistantes dans le cadre de programmes de sélection et pour appuyer la recherche sur la qualité et la salubrité des aliments.
2. Concevoir et valider des méthodes de rechange rapides pour déceler la présence de DON dans le grain ou la farine de grain entier (par exemple, la spectrométrie dans le proche infrarouge - NIRS) et pour détecter les grains endommagés par le *Fusarium* (par exemple, l'analyse d'images), reconnaître que différentes normes sont nécessaires pour le contrôle de la qualité (par exemple, la haute précision) en comparaison des normes pour la sélection et le criblage.
3. Développer des tests peu coûteux pour déceler la présence de mycotoxines autres que le DON (par exemple, nivalénol, *DAS*, *T-2* et *HT-2*) pouvant être produites par des espèces autres que celle du *Fusarium graminearum*, lesquelles peuvent favoriser la brûlure de l'épi causée par le *Fusarium* dans des cultures d'orge et d'avoine.
4. L'ACIA et Santé Canada doivent élargir leur surveillance du DON et d'autres mycotoxines dans le grain, les produits agroalimentaires et d'autres secteurs du marché.
5. Enquêter sur des protocoles d'échantillonnage du DON et d'autres mycotoxines, car l'échantillonnage a été cerné comme étant la plus grande source d'erreur lorsqu'on doit établir des niveaux précis, significatifs et pouvant être reproduits.
6. Normaliser les méthodes d'analyse utilisées dans les laboratoires pour déceler la présence de mycotoxines à l'aide d'éléments de documentation permettant d'obtenir des résultats uniformes, fiables et comparables.
7. Développer une meilleure compréhension des effets de la contamination par mycotoxines sur la santé humaine, particulièrement pour les groupes à haut risque (par exemple, les bébés et les enfants).

Sélection classique des végétaux/sélection moléculaire

1. Poursuivre nos études sur l'effet de la brûlure de l'épi causée par le *Fusarium* dans les cultures d'avoine, évaluer son impact sur les récoltes, développer des stratégies pour atténuer la présence du *Fusarium* et identifier la résistance au *Fusarium* afin d'intégrer ces variétés aux programmes de sélection.
2. Appuyer la mise au point continue et l'élaboration de méthodes améliorées du phénotypage, comme la phénomique, permettant de valider l'identification des sources de résistance, de confirmer le fondement de la résistance génétique et de cribler efficacement les lignées sélectionnées, ainsi que la meilleure utilisation de la génomique, de la protéomique, de la métabolomique, etc.

3. Évaluer à fond la diversité pathogénique de l'agent causal principal, *Fusarium graminearum*, et étudier la présence des cas particuliers qui se manifestent dans différentes récoltes et selon différents environnements (froid et chaud) pour expliquer sa présence plus marquée dans des régions d'épidémie et la possibilité que le *F. graminearum* se déplacera vers de nouvelles régions ou qu'il développera une résistance aux fongicides.
4. Poursuivre nos efforts de recherche, et particulièrement les examens d'ordres histologique et histochimique relatifs aux mécanismes de résistance, afin de mieux comprendre la progression de la maladie et d'identifier les points faibles pour mieux cibler l'agent pathogène et le détruire, ou encore, améliorer la résistance de l'hôte.
5. Poursuivre d'autres approches telles que la sélection et la transformation moléculaires, c'est-à-dire, développer des variétés résistantes à la fusariose comme solution de rechange, pour minimiser l'impact de la brûlure de l'épi causée par le *Fusarium*.
6. Poursuivre sans relâche les activités de recherche pour développer des lignées de maïs autofécondées résistantes au *Fusarium* qui pourraient être croisées avec d'autres variétés ou espèces pour développer des hybrides résistants au *Fusarium*.

Épidémiologie/gestion des maladies

1. Clarifier le rôle du maïs comme hôte principal additionnel de la brûlure de l'épi causée par le *Fusarium* (la pourriture rose de l'épi du maïs et la fusariose de la tige) et comme source abondante et persistante d'inoculum de *Fusarium* pour contaminer les céréales à petit grain.
2. Étudier le rôle des résidus de culture comme source d'inoculum primaire, particulièrement les résidus de cultures qui ne sont pas hôtes (par exemple, le canola), pour nous aider à mieux comprendre le développement de la maladie, et à développer efficacement des stratégies de contrôle intégrées.
3. Clarifier les retombées des pratiques de gestion (par exemple, l'application d'herbicides), et mieux comprendre le rôle des résidus de culture par rapport à la survie hiémale des agents pathogènes et le potentiel accru d'inoculum le printemps suivant.
4. Identifier ou développer des méthodes simples et normalisées pour évaluer et mesurer la concentration de spores de *Fusarium* dans l'atmosphère, et participer à l'étude des facteurs environnementaux favorisant la production et le relâchement d'inoculum.
5. Peaufiner et valider les modèles de prévision de la brûlure de l'épi causée par le *Fusarium* pour permettre l'application efficace et économique de fongicides, et en favoriser l'accès à tous et en temps opportun.
6. Développer des fongicides plus efficaces pour contrôler la brûlure de l'épi causée par le *Fusarium* et offrir un ou plusieurs produits homologués aux producteurs canadiens pouvant être appliqués aux cultures d'orge et d'avoine.

7. Mieux informer les producteurs, les agronomes et l'industrie céréalière sur les diverses stratégies de gestion à leur portée pour lutter contre le *Fusarium*. Ceci comprend la sensibilisation aux différentes pratiques agricoles employées (par exemple, les conséquences de l'ensemencement de graines atteintes de fusariose, le choix des variétés, la rotation des cultures) ainsi que les facteurs environnementaux qui influent sur la maladie (par exemple, le stade phénologique relativement à la dissémination des spores et l'utilisation des modèles de prévision de la maladie).

Questions communes

1. Développer une approche à long terme faisant preuve d'engagement pour résoudre le problème du *Fusarium*, soit une approche qui ne serait pas estompée si jamais on enregistrait une faible incidence du *Fusarium* pour une ou deux années.
2. L'ACIA et Santé Canada doivent examiner de nouveau les normes sur le *Fusarium* et la présence de DON, et les rendre conformes aux modifications récentes ou éventuelles apportées à ces normes par nos clients et par les organisations internationales, et communiquer avec les représentants de l'industrie et les aider à réaliser ce but.
3. Ralentir la progression du *Fusarium* ou l'empêcher de se répandre à de nouvelles régions (par exemple, en Alberta) et protéger ainsi notre capacité d'avoir accès à du grain intouché par la maladie sur une grande superficie, afin de maintenir l'avantage concurrentiel que le Canada détient sur le marché.

Nous espérons que ces besoins seront reconnus et que la mise en œuvre des mesures proposées nous permettront de poursuivre avec vigueur nos efforts pour traiter du *Fusarium* au Canada, et de trouver des solutions en temps opportun afin de minimiser les effets dévastateurs de cette maladie.

Veillez agréer, Mesdames, Messieurs, l'expression de mes sentiments les plus respectueux.

Jeannie Gilbert, *présidente du comité organisateur*

Andy Tekauz, *membre du comité organisateur*

3^e Colloque canadien sur la fusariose

List of Registrants

Dave Abramson

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1469
204-983-4604
dabramson@agr.gc.ca

Hafiz Ahmed

Plant Science, University of Manitoba
Winnipeg, MB R3T 2N2
204-474-7888
204-474-7528
ahmedhu@ms.umanitoba.ca

Olufemi Akinsanmi

Botany
CRC for Tropical Plant Protection, University of Queensland
Level 4 John Hines Building, UQ St. Lucia
Brisbane, Queensland, Australia
011-617-3214-2632
011-617-3214-2950
femi.akinsanmi@csiro.au

Shaffeek Ali

Branch Head
Pest Risk Management
Alberta Agriculture, Food & Rural Development
204, 7000 - 113 Street
Edmonton, AB T6H 5T6
780-422-4909
780-427-1057
shaffeek.ali@gov.ab.ca

Erin Armstrong

President & CEO
Brewing and Malting Barley Research Institute
303 - 161 Portage Avenue East
Winnipeg, MB R3B 2L6
204-927-1407
204-947-5960
earmstrong@bmbri.ca

Epifania Austria

Dept of Plant Science, University of Manitoba
Winnipeg, MB R3T 2N2
204-474-6060
204-474-7528
austriae@ms.umanitoba.ca

Rob Bahry

Crop Inputs, Paterson Grain
22nd Floor, 333 Main Street
Winnipeg, MB R3C 4E2
204-926-3456
204-926-9598
rbahry@patersongrain.com

Tomohiro Ban

Biological Resources Division
Japan International Research Center for
Agriculture Sciences
1-1 Ohwashi
Tsukuba Ibaraki 305-8686
011-81-29-838-6364
011-81-29-838-6364
tomohiro@affrc.go.jp

Mitali Banik

AAFC - Brandon
18th St. & Grand Valley Road
Brandon, MB R7A 5Y3
204-726-7650
204-728-3858
mbanik@agr.gc.ca

Francois Belzile

Plant Science, Laval University
Marchand Building #1243
Quebec, PQ G1K 7P4
418-656-2131x5763
418-656-7176
fbelzile@rsvs.ulaval.ca

Benoit Bizimungu

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0466
204-983-4604
bbizimungu@agr.gc.ca

Luc Bourgeois

Bayer CropScience
#3, 35 Crawford Cres.
Campbellville, ON
N0B 1P0
905-854-4474
905-854-4478
luc.bourgeois@bayercropscience.com

Rejean Boyer

Global Sales & Business Development Manager,
Acurum, DuPont Canada Inc.
7070 Mississauga Road
Mississauga, ON L5M 2H3
905-821-5586
905-821-5653
rejean.g.boyer-1@can.dupont.com

Amy Breker

University of Saskatchewan
51 Campus Drive, Agriculture Bldg
Saskatoon, SK S7N 5A8
306-966-8359
alb547@mail.usask.ca

Brad Brinkworth

Editor, Meristem Information Resources Ltd.
12, 3109 Palliser Drive SW
Calgary, AB T2V 4W5
403-543-7425
403-543-7419
brinkworthb@meristem.com

Doug Brown

AAFC, CRC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0413
204-983-4604
dbrown@agr.gc.ca

Allison Brown-Hoepfner

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0936
204-983-4606
ahoepfner@agr.gc.ca

Anita Brule-Babel

Dept of Plant Science
University of Manitoba
Winnipeg, MB R3T 2N2
204-474-6060
204-474-7528
ababel@ms.umanitoba.ca

Jadwiga Budzinski

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-3505
jbudzinski@agr.gc.ca

Patrick Bulman

Bayer Cropscience
#100, 3131-114 Ave SE
Calgary, AB
T2Z 3X2
403-723-7452
403-723-7488
patrick.bulman@bayercropscience.com

Greg Bunkers

Agronomic Traits
Monsanto Co
700 Chesterfield Pkwy N
St. Louis, MO, USA
63198
636-737-7647
636-737-7670
greg.j.bunkers@monsanto.com

Gail Butler

Innovation & Renewal, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1697
613-759-1701
butlergm@agr.gc.ca

Wenguang Cao

ECORC, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1540
613-756-6559
caowen@agr.gc.ca

Gord Carson

Robin Hood Multifoods Inc.
191 Atwell Drive, Unit 4
Rexdale, ON, M9W 5Z1

Valerie Chabot

Cooperative Federee de Quebec
15050 Ch. De la Federee
St-Hyacinthe, PQ J2R 1J2
450-799-2326
450-799-2328
chabotv2@co-op.ca

Sukumar Chakraborty

Plant Industry, CSIRO
Queensland Biosciences Precinct
306 Carmody Road
St. Lucia Queensland, Australia 4067
011-617-3214-2677
011-617-3214-2950
sukumar.chakraborty@csiro.au

Sherman Chan

Technical Director, Rahr Malting (Canada) Ltd.
800 West First Avenue
Shakopee, MN, USA 55379
952-496-7028
952-496-7118
schan@rahr.com

Philip Clarke

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0805
204-983-4604
pjclarke@agr.gc.ca

John Clarke

AAFC
Box 1030
Swift Current, SK S9H 3X2
306-778-7221
306-773-9123
clarkej@agr.gc.ca

Randy Clear

Grain Research Laboratory
Canadian Grain Commission
1404 - 303 Main Street
Winnipeg, MB R3C 3G8
204-983-7797
204-983-0724
rclear@grainscanada.gc.ca

Sylvie Cloutier

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-2340
204-893-4604
scloutier@agr.gc.ca

Andre Comeau

Agriculture Canada
2560 boul Hochelaga
Sainte-Foy, PQ G1V 2J3
418-657-7980 x216
418-648-2402
comeaua@agr.gc.ca

David Curry

Ag-Quest, Inc.
Box 144
Minto, MB R0K 1M0
204-776-5556
204-776-2250
david.currey@agquest.com

Janice Cuthbert

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1142
204-893-4604
jcuthbert@agr.gc.ca

Patricia Cuthbert

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1142
204-983-4604
pcuthbert@agr.gc.ca

Lynn Dahleen

Cereal Crops Research Unit
USDA-ARS
1307 - 18th St. N., PO Box 5677, SU Station
Fargo, ND, USA 58105
701-239-1384
701-239-1369
dahleen1@fargo-ars.usda.gov

Ron DePauw

Cereal Section, AAFC
Box 1030
Swift Current, SK S9H 3X2
306-778-7241
306-773-9123
depauw@agr.gc.ca

Lyle DePauw

Agronomic Services, AgPro
Box 43
Forrest, MB R0K 0W0
204-727-6669
204-725-2690
lyle.depauw@swp.com

Grant Deveson

Bayer CropScience
58 MacKenzie Lake Place
Calgary, AB
T2Z 2T5
403-723-7443

Jim Dexter

Grain Research Laboratory
Canadian Grain Commission
1404 - 303 Main Street
Winnipeg, MB R3C 3G8
204-983-6054
204-983-0724
jdexter@grainscanada.gc.ca

Yves Dion

CEROM
225 Chenier des Vingt-cinq, Est
St.Bruno-de-Montarville, PQ
J3V 4P6
450-653-4413x225
450-441-5694
yves.dion@sympatico.ca

Daryl Domitruk

Manager, Crop Development
Manitoba Agriculture, Food & Rural Revitalization
65 - 3 Avenue NE
Carman, MB R0B 0J0
204-745-5636
204-745-5690
ddomitruk@gov.mb.ca

Patrick Doyle

Syngenta Crop Protection
140 Research Lane, Research Park
N1G 4Z3
519-837-5314
519-823-0504
patrick.doyle@syngenta.com

Karen Dupchak
Manitoba Agriculture & Food
204 - 545 University Cres
Winnipeg, MB R3T 5S6
204-945-7668
204-945-4327
kdupchak@gov.mb.ca

Mark Etienne
Hyland Seeds
RR#1, 11087 Petty St
Ailsa Craig, ON N0M 1A0
519-232-4341
519-232-4345
metienne@hylandseeds.com

Francois Eudes
AAFC
5403 - 1 Avenue South
Lethbridge, AB T1J 2L5
403-317-3338
403-382-3156
eudesf@agr.gc.ca

George Fedak
ECORC, AAFC
960 Carling Ave, Bldg 50
Ottawa, ON K1A 0C6
613-759-1393
613-759-6559(1701)
fedakga@agr.gc.ca

Myriam Fernandez
Cereal Section, AAFC
Box 1030
Swift Current, SK S9H 3X2
306-778-7255
306-773-9123
fernandezm@agr.gc.ca

Dilantha Fernando
Plant Science, University of Manitoba
Winnipeg, MB R3T 2N3
204-474-7889
204-474-7529
d_fernando@umanitoba.ca

Glenn Forster
Technical Sales Specialist, Sales-Seed
Treatments
BASF
#1 - 411 Downey Road
Saskatoon, SK S7S 1J3
306-653-7533
306-652-6745
mitzeln@basf.com

Greg Fox
WestBred LLC
717 - 14 Street South
Fargo, ND, USA
58103
701-293-5146
701-293-5146
gfox@westbred.com

Stephen Fox
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0573
204-983-4604
sfox@agr.gc.ca

Jerome Franckowiak
North Dakota State University
166 Loftsgard Hall, Fargo
ND, USA, 58105
701-231-7540
j.franckowiak@ndsu.nodak.edu

Peter Freeman
Alberta Network for Proteomics Innovation
120 Varsity Estates Mews N.W.
Calgary, AB T3B 3C8
403-288-8157
pfreeman@hordeum.com

Judith Fregeau-Reid
ECORC, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1675
613-759-1701
fregeauja@agr.gc.ca

Don Gaba
Grain Research Laboratory
Canadian Grain Commission
1404 - 303 Main Street
Winnipeg, MB R3C 3G8
204-984-5704
204-983-0724
dgaba@grainscanada.gc.ca

Audrey Gall
Agricultural Fieldman
Northern Sunrise County
Bag 1300,
Peace River, AB T8S 1Y9
780-624-0013
agall@northern sunrise.net

Yuncaï Gao
Canadian Inovatech Inc.
31212 Peardonville Road
Abbotsford, BC V2T 6K8
604-852-2582
604-857-2679
ygao@inovatech.com

Denis Gaudet
AAFC, Lethbridge
Box 3000
Lethbridge, AB T1J 4B1
403-327-4561
403-382-3156
gaudetd@agr.gc.ca

Habib Ghazvini
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1142
204-983-4604
hghazvini@agr.gc.ca

Jeannie Gilbert
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0891
204-983-4604
jjgilbert@agr.gc.ca

Patricia Gilbert
Winnipeg, MB

Mike Giles
Growth Stage
150, 251 Midpark Blvd SE
Calgary, AB T2X 1S3
403-256-9557
mjgiles@growthstage.com

Saber Golkari
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0487
204-983-4604
sgolkari@agr.gc.ca

Robert Graf
AAFC
5403 - 1st Ave South
Lethbridge, AB T1J 4B1
403-317-2258
403-382-3156
rgrafr@agr.gc.ca

Mike Grenier
The Canadian Wheat Board
423 Main Street, Box 816, Stn M
Winnipeg, MB R3C 2P5
204-983-2996
204-984-1699
mike_grenier@cwbc.ca

Tajinder Grewal
University of Saskatchewan
51 Campus Drive
Saskatoon, SK
S7N 5A8
306-966-8476
306-966-5015
tajinder.grewal@usask.ca

Xiaowei Guo
Plant Science, University of Manitoba
222 Agriculture Building
Winnipeg, MB R3T 2N2
204-474-6089
204-474-7528

Jana Hansen
North Dakota State University
Walster Hall, Room 306
Fargo ND, USA
58105
701-231-8868
jana.hansen@ndsu.nodak.edu

Scott Henry
Bayer CropScience
430-B Dovern Court Drive
Winnipeg, MB R3Y 1N4
204-989-5435
204-487-3648
scott.henry@bayercropscience.com

Bernie Hill
AAFC - Lethbridge
5403 - 1 Avenue South
Lethbridge, AB T1J 4B1
403-317-2267
403-382-3156
hillb@agr.gc.ca

David Hooker
Ridgetown College, University of Guelph
Main Street East
Ridgetown, ON N0P 2C0
519-644-2036
519-644-2043
dhooker@skynet.ca

Robert Hornford
BASF
306 - 155 Carlton
Winnipeg, MB, R3J 2G3
204-831-6572
204-885-1688
hornfor@basf.com

Jim House
Animal Science, University of Manitoba
12 Dafoe Road
Winnipeg, MB R3J 2N2
204-474-9523
204-474-7623
j.house@umanitoba.ca

Peter Hucl
Crop Development Centre
University of Saskatchewan
51 Campus Drive
Saskatoon, SK S7N 5A8
306-966-8667
306-966-5015
hucl@sask.usask.ca

Gavin Humphreys
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0123
204-983-4604
ghumphreys@agr.gc.ca

Sharon Inch
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0223
204-983-4604
sinch@agr.gc.ca

Ian John
Manager, Barley Procurement
Westcan Malting Ltd.
Box 113
Alix, AB T0C 0B0
403-747-2777
403-747-2660
ian@westcanmalt.com

Peter Johnson
Grain Producer
London, ON

Mark Jordan
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1453
204-983-4604

mcjordan@agr.gc.ca

Trish Jordan
Monsanto Canada
67 Scurfield Blvd
Winnipeg, MB
R3Y 1G4
204-985-6964
204-488-8064
trish.l.jordan@monsanto.com

Richard Joy
Rahr Malting (Canada) Ltd.
Alix, AB

Pat Juskiw
AFFRD
5030 - 50 Ave
Lacombe, AB
T4L 1W8
403-782-8691
patricia.juskiw@gov.ab.ca

Ron Kaethler
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-0895
204-983-4604
rkaethler@agr.gc.ca

David Kaminski
Manitoba Agriculture, Food, and Rural Initiatives
65 - 3rd Ave NE
Carman, MB R0G 0J0
204-745-5656
dkaminski@gov.mb.ca

Ho Keh Ming
Barley Breeder, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1557
613-759-1926
hokm@agr.gc.ca

Larry Kitz
Alberta Seed Growers'
Box 487
Two Hills, AB, T0B 4K0
780-592-2200
780-592-2335
larry.kitz@lonespruce.com

Jeremy Klassen
DSV Canada Inc.
PO Box 99
St. Norbert Postal Station
Winnipeg, MB R3V 1L5
204-261-6338
204-261-7414

jeremyk@byseeds.com

Ron Knox

AAFC
Box 1030
Swift Current, SK S9H 3X2
306-778-7262
306-773-9123
knoxr@agr.gc.ca

Eric Kokko

AAFC - Lethbridge
5403 - 1 Avenue South
Lethbridge, AB T1J 4B1
403-317-2255
403-382-3156
kokko@agr.gc.ca

Uwe Kromer

AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0895
204-983-4604
ukromer@agr.gc.ca

Krishan Kumar

Field Crop Development Centre
Alberta Agriculture
6000 C&E Trail
Lacombe, AB T4L 1W1
403-782-6120
krishan.kumar@gov.ab.ca

A (Kush) Kushalappa

Plant Science, McGill University
St. Anne de Belleville, PQ H9X 3V9
514-398-7851 x7867
514-398-7897
ajjamada.kushalappa@mcgill.ca

Paul Laflamme

Integrated Crop Management Specialist
Crop Diversification Centre North
Alberta Agriculture, Food & Rural Development
1001, 10320 - 99 Street
Grande Prairie, AB T8V 6J4
780-538-5631
780-538-5288
paul.laflamme@gov.ab.ca

Julie Lajeunesse

Agriculture et Agroalimentaire Canada
1468 St Cyrille
Normandin, PQ G8M 4K3
418-274-3378
418-274-3386
lajeunesseju@agr.gc.ca

Francois Langevin

Agriculture & Agri-Food Canada
121 Bon-Air
Sainte-Catherine-de-la-ji'e, PQ
G0A 3M0
418-875-1691
langevinf@agr.gc.ca

Nathalie Lanoie

Semico Inc.
5175 Boul. Laurier Est
St-Hyacinthe, PQ J2R 2B4
450-799-3225
450-799-3229
nlanoie@semico.ca

Roger Larios

Dept of Plant Science
University of Manitoba
Winnipeg, MB R3T 2N2
204-474-6104
204-474-7528
rlarios@ms.umanitoba.ca

Andre Laroche

Bioproducts & Bioprocesses
AAFC
5403 - 1st Ave South
Lethbridge, AB
T1J 4B1
403-317-2224
403-382-3156
laroche@agr.gc.ca

Eric Lefol

University of Saskatchewan
51 Campus Drive
Saskatoon, SK
S7N 5A8
306-966-5009
306-966-5015
eric.lefol@usask.ca

Bill Legge

AAFC - Brandon
PO Box 1000A, RR#3
Brandon, MB R7A 5Y3
204-726-7650
204-728-3858
blegge@agr.gc.ca

Yueshu Li

Canadian Malting Barley Technical Centre
1365 - 303 Main Street
Winnipeg, MB
R3C 3G7
204-984-0561
204-984-5843
yli@cmbtc.com

Garrett Martin
DuPont Canada Inc.
Box 2200, Streetsville
Mississauga, ON L5M 2H3
613-548-5796 - Kim Dailey
613-548-5041 - Kim Daily

Richard Martin
AAFC
440 University Ave
Charlottetown, PEI C1A 4N6
902-566-6851
902-566-6821
martinra@agr.gc.ca

Ray Mazinke
Grain Producer
Morris, MB

Rob McCaig
Canadian Malting Barley Technical Centre
1365 - 303 Main Street
Winnipeg, MB
R3C 3G7
204-983-1981
204-984-5843
rmccaig@cmbtc.com

Brent McCallum
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0771
204-983-4604
bmccallum@agr.gc.ca

Kevin McCallum
Proven Seed, Agricore United
Box 2549
Morden, MB R6M 1C2
204-435-2063
204-435-2369
kmccallum@agricoreunited.com

Curtis McCartney
Visiting Fellow, CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0936
204-983-4604
mccartneyc@agr.gc.ca

Debra McLaren
AAFC - Brandon
18th St. & Grand Valley Road
PO Box 1000A, RR#3
Brandon, MB R7A 5Y3
204-726-7650
204-728-3858
dmclaren@agr.gc.ca

Rod McLeod
Gustafson
#10, 2712 37 Ave NE
Calgary, AB
T1Y 5L3

Monique McTiernan
Atlantic Grains Council
925 Coverdale Road
Riverview, NB E1B 5E6
506-856-2277
506-856-2092
monique.mctiernan@gnb.ca

Mary Meleshko
Dept of Plant Science
University of Manitoba
Winnipeg, MB R3T 2N2
204-474-6104
204-474-7528
meleshko@ms.umanitoba.ca

Rich Michta
Acurum, DuPont Canada Inc.
7070 Mississauga Road
Mississauga, ON L5M 2H3
204-261-3125
204-261-3124
rmichta@mts.net

David Miller
Carleton University
Ottawa, ON

Shea Miller
ECORC, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1760
613-759-1701
millers@agr.gc.ca

Jennifer Mitchell Fetch
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1460
204-983-4604
jfetch@agr.gc.ca

Rick Mitzel
BASF
#1 - 411 Downey Road
Saskatoon, SK S7S 1J3
306-653-7533
306-652-6745
mitzeln@basf.com

Kevin Morgan
AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0895
kmorgan@agr.gc.ca

Robin Morrall
University of Saskatchewan
112 Science Place
Saskatoon, SK S7N 5E2
306-966-4410
306-966-4461
morrall@sask.usask.ca

Al Morris
Agricore United
Winnipeg, MB

John Morrison
The Ontario Wheat Producer's Marketing Board
190 Nicklin Road
Guelph, ON N1H 6N1
519-767-9713
louise.bignell@ontariowheatboard.com

Eric Mueller
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-0795
204-983-4604
emueller@agr.gc.ca

Hans Nass
Research Branch, AAFC
440 University Ave
Charlottetown, PEI C1A 4N6
902-566-6865
902-566-6821
nassh@agr.gc.ca

Gary Neumann
Chemist, Health Products & Food Branch
Health Canada
510 Lagimodiere Boulevard
Winnipeg, MB R2J 3Y1
204-983-5490
204-983-5547
gary_neumann@hc-sc.gc.ca

Matt Nichols
Technical Services, Neogen Corp
620 Leshner Place
Lansing, MI, USA 48808
517-372-9200
517-372-2006
mnichols@neogen.com

Sasanda Nilmalgoda
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-3505
nilmalgodas@agr.gc.ca

Tom Nowicki
Grain Research Laboratory
Canadian Grain Commission
1404 - 303 Main Street
Winnipeg, MB R3C 3G8
204-983-3345
204-983-3036
tnowicki@grainscanada.gc.ca

Therese Ouellet
ECORC, AAFC
960 Carling Ave
Ottawa, ON
K1A 0C6
613-759-1658
613-759-1701
ouellettr@agr.gc.ca

Helen Page
Plant Products Program Officer
Canadian Food Inspection Agency
613 - 269 Main Street
Winnipeg, MB R4H 1B1
204-983-2158
204-984-6008
hpage@inspection.gc.ca

Denis Pageau
Ferme de recherche, Normandin
Agriculture et Agroalimentaire Canada
1468 St Cyrille
Normandin, PQ G8M 4K3
418-274-3378
418-274-3386
pageaud@agr.gc.ca

Radhey Pandeya
ECORC, AAFC
Central Experimental Farm
Ottawa, ON
K1A 0C6
613-759-1636
613-759-1701
pandeyars@agr.gc.ca

Meryle Pankewich
Canadian Grain Commission
900 - 303 Main Street
Winnipeg, MB R3C 3G8
204-983-2781
mpankewich@grainscanada.gc.ca

Susan Patrick
Grain Research Laboratory
Canadian Grain Commission
1404 - 303 Main Street
Winnipeg, MB R3C 3G8
204-983-7797
204-983-0724
spatrick@grainscanada.gc.ca

Penny Pearse
Crop Development Branch
Saskatchewan Agriculture
#125, 3085 Albert St
Regina, SK S4S 0B1
306-787-4671
306-787-0428
ppearse@agr.gov.sk.ca

Trevor Pizzey
EVP – Operations, Can-Oat Milling
#1 Can-Oat Drive
Portage la Prairie, MB R1N 3W1
204-856-5920
204-857-9500
trevor.pizzey@can-oat.com

Stephan Pouleur
Agriculture et Agroalimentaire Canada
2560 boul Hochelaga
Sainte-Foy, PQ G1V 2W7
418-657-7985 x220
418-648-2402
pouleurs@agr.gc.ca

Curtis Pozniak
Assistant Professor, Crop Development Centre
University of Saskatchewan
51 Campus Drive
Saskatoon, SK S7N 5A8
306-966-2361
306-966-5015
curtis.pozniak@usask.ca

Manika Pradhan
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1142
204-983-4604
mpradhan@agr.gc.ca

Suvira Prashar
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-6927
204-983-4604
sprashar@agr.gc.ca

Doug Procnier
CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-1466
204-983-4604
dprocunier@agr.gc.ca

Darcy Rafoss
Seed Business Unit
Saskatchewan Wheat Board
2625 Victoria Ave
Regina, SK S4T 7T9
306-569-4444
306-569-4897
darcy_rafoss@swp.com

Khalid Rashid
AAFC - Morden
Unit 100 - 101 Rout 100
Morden, MB R6M 1Y5
204-822-7220
204-822-7207
krashid@agr.gc.ca

Curtis Rempel
Monsanto Canada
67 Scurfield Blvd.
Winnipeg, MB R3Y 1G4
204-985-1093
204-488-1577
curtis.b.rempel@monsanto.com

Erica Riedel
CRC, AAFC
195 Dafoe Road
Winnipeg, MB
204-984-3505
eriedel@agr.gc.ca

Bob Robertson
VP, Agriculture and Food
Diagnostix
Ontario L4Z 1N8
800-282-4075
905-890-6024
bob_robertson@diagnostix.ca

Brian Rossnagel
Crop Development Centre
University of Saskatchewan
51 Campus Drive
Saskatoon, SK S7N 5A8
306-996-4976
306-966-5015
brian.rossnagel@usask.ca

R.S. Sadasivaiah

Sustainable Production Systems
Agriculture & Agri-Food Canada
5403 - 1 Avenue South
Lethbridge, AB T1J 4B1
403-317-2273
403-382-3156
sadash@agr.gc.ca

Don Salmon

Field Crop Development Centre, AAFRD
5030 - 50th Street
Lacombe, AB T4L 1W8
403-782-8694
403-782-5514
donald.salmon@gov.ab.ca

Harry Sapirstein

University of Manitoba
250 Ellis Ave
Winnipeg, MB, R3T 2N2
204-474-6481
204-474-7630
harry-sapirstein@umanitoba.ca

Marc Savard

ECORC, AAFC
960 Carling Ave
Ottawa, ON K1A 0C6
613-759-1683
613-759-1701
savardme@agr.gc.ca

Art Schaafsma

Department of Plant Agriculture
University of Guelph
Main Street East
Ridgetown, ON N0P 2C0
519-674-1624
519-674-1555
aschaafs@ridgetownc.uoguelph.ca

Harold Schmaltz

Agricore United
Winnipeg, MB

Paul Schwarz

Associate Professor, Plant Sciences
North Dakota State University
Bolley Drive
 Fargo, ND, USA 58105
701-231-7732
paul.schwarz@ndsu.nodak.edu

Ian Seddon

MB Agriculture and Food
204 - 545 University Cres
Winnipeg, MB, R3T 5S6
204-945-0353
204-945-4327

iseddon@gov.mb.ca

Nandita Selvanathan

Food & Rural Initiatives
Manitoba Agriculture & Food
65 - 3 Avenue NE
Carman, MB R0G 0J0
204-745-5640
204-745-5690
nselvanath@gov.mb.ca

Catherine Shearer

Communications & Consultations, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-0894
204-983-4604
cshearer@agr.gc.ca

Steven Simpfendorfer

Crop Protection, NSW Agriculture
Tamworth Agricultural Institute
RMB 944 Calala Lane
Tamworth NSW, Australia 2340
011-61-2-676-31261
011-61-2-676-31222
steven.simpfendorfer@agric.nsw.gov.au

Linnea Skoglund

Busch Agricultural Resources, Inc
3515 East Country Road 52
Ft Collins, CO,
USA, 80524
970-472-2332
970-472-2334
linnea.skoglund@anheuser-busch.com

Daryl Somers

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-4503
204-983-4604
somersd@agr.gc.ca

Veldon Sorensen

Bayer Cropscience
Ste 100, 3131 - 114th Avenue SE
Calgary, AB T2Z 3X2
403-723-7454
403-723-7496
veldon.sorensen@bayercropscience.com

Ellen Sparry

C&M Seeds
RR#3
Palmerston, ON
N0G 2P0
519-343-2126
519-343-3792
esparry@redwheat.com

Helmut Spieser

Ontario Ministry of Ag and Food
Ridgetown, ON

Goran Srnica

Biology & Logistics
Syngenta Crop Protection
Jealott's Hill Research Centre
Bracknell, Berkshire, UK
RG4Z GEY
01344-416-580
goran.srnica@syngenta.com

Robert Stack

Plant Pathology Department
North Dakota State University
Fargo, ND, USA 58105

Jo-Ann Stebbing

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0827
204-983-4604
jstebbing@agr.gc.ca

Jeff Stewart

Sustainable Production Systems, AAFC
5403 - 1 Avenue South
Lethbridge, AB T1J 4B1
403-317-2208
403-317-2197
stewartj@agr.gc.ca

Marcos Stulzer

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-0795
204-983-4604
mstulzer@agr.gc.ca

Ljiljana Tamburic-Ilicic

Ridgetown College, University of Guelph
Main Street East
Ridgetown, ON N0P 2C0
519-851-6896
519-674-1600
ltamburi@ridgetownc.uoguelph.ca

Marc Tardif

Agent de Commercialisation
Centre de Recherche Sols et Grandes Cultures
Agriculture et Agroalimentaire Canada
2560 boul Hochelaga
Sainte-Foy, PQ G1V 2J3
418-657-7985x243
418-648-2402
tardifm@agr.gc.ca

Andy Tekauz

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-983-0944
204-983-4604
atekauz@agr.gc.ca

Albert Tenuta

Field Crop Plant Pathologist, Crop Technology
Ontario Ministry of Ag and Food
PO Box 400, 120 Main Street East
Ridgetown, ON N0P 2C0
519-674-1617
519-674-1564
albert.tenuta@omaf.gov.on.ca

Chad Thaubergger

Syngenta Crop Protection

Paul Theil

Bayer Cropscience
100, 3131 - 114 Ave SE
Calgary, AB T2Z 3X2

Julian Thomas

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-3505
jthomas@agr.gc.ca

Jill Thomson

Seed Check Technologies Inc.
110 Gymnasium Road
Saskatoon, SK S7N 0W9
306-975-0990
306-975-0991
jill@seedcheck.ca

Lawrence Thomson

Agri Products - Saskatoon Seed Lab
Saskatchewan Wheat Board
102, 407 Downey Road
Saskatoon, SK S7N 4L8
306-933-0541
lawrence.thomson@swp.com

Barry Todd

Management & Regional Agricultural Services
Manitoba Agriculture & Food
Winnipeg, MB

Bill Toews

Western Grain Research Foundation
Box 35
Kane, MB
R0G 1E0
204-343-2002
204-343-2108

bbtoews@cici.mb.ca

James Tucker

AAFC - Brandon
PO Box 1000A, RR#3
Brandon, MB R7A 5Y3
204-726-7650
204-728-3858

jtucker@agr.gc.ca

Kelly Turkington

AAFC - Lacombe
6000 C&E Trail
Lacombe, AB T4L 1W1
403-782-8138
403-782-6120

turkingtonk@agr.gc.ca

Gary Turnbull

Field Research & Development
Dow AgroSciences Canada Inc.
39 Scurfield Blvd
Winnipeg, MB R3Y 1G4
204-488-5757
204-488-5788

gcturnbull@dow.com

Ludvik Tvaruzek

Integrated Plant Protection
Agricultural Research Institute Kromeriz, Ltd.
Havlickova 2787
Kromeriz, Czech Republic 767 01
011-420-573-317-138
011-420-573-339-725

tvaruzek@vukrom.cz

Maarten van Ginkel

Head, Fusarium Research and Gene Bank
Wheat Program, CIMMYT, Int.
Apdo. Postal 6-641 Mexico
D.F., Mexico 6600
011-52-55-5804-2004
011-52-55-5804-7558

m.van-ginkel@cgiar.org

Chris Van Natto

DuPont Canada Inc.
Box 2200, Streetsville
Mississauga, ON L5M 2H3
613-548-5796 - Kim Dailey
613-548-5041 - Kim Daily

William Van Tassel

1st Membre du Comité Exécutif
Fédération des Producteurs de Cultures
Commerciales du Québec
Maison de l'UPA
555, Boul. Roland-Therrien,
Bureau 505
Longueuil, PQ J4H 4G4
450-679-0540
450-679-6372

vanhug@digicom.qc.ca

Harvey Voldeng

ECORC, AAFC
960 Carling Avenue
Ottawa, ON K1A 0C6
613-759-1652
613-759-1701

voldenhd@agr.gc.ca

Andrzej Walichnowski

CRC, AAFC
195 Dafoe Road
Winnipeg, MB R3T 2M9
204-984-0222
204-983-4604

awalichnowski@agr.gc.ca

David Wall

Science Director, Saskatoon Research Centre
107 Science Pl.
Saskatoon, SK S7N 0X2
306-956-7211
306-956-7248

walld@agr.gc.ca

Shannon Warren

Adfarm
250, 333 - 24 Ave SW
Calgary, AB T2P 3E6
403-410-7619
403-410-7601

shannon.warren@adfarmonline.com

John Waterer

Cargill
300 - 240 Graham Ave
Winnipeg, MB R3C 4C5
204-947-6385
204-947-6495

john_waterer@cargill.com

Ron Weik

Manager, Intellectual Property & Regulatory
Affairs
Quality Assured Seeds Inc
418B McDonald Street
Regina, SK S4N 6E1
306-791-0500x29
306-791-0142

rweik@qas-online.com

David Whaley

Director
The Ontario Wheat Producer's Marketing Board
190 Nicklin Road
Guelph, ON N1H 6N1
519-767-9713

dwhaley@wincom.net

Larry White

General Manager, SeCan Association
201, 52 Antares Dr
Ottawa, ON K2E 7Z1
613-225-6891
613-225-6422
seed@secan.com

James Wilkie

General Manager, Agrameteorological Centre of Excellence
182 Main Street
Carman, MB R0G 0J0
204-745-2320
204-745-6299
jim.wilkie@aceweather.ca

Allan Wilson

Gustafson
#10, 2712 37 Ave NE
Calgary, AB
T1Y 5L3

Tom Wolf

AAFC, Saskatoon
107 Science Place
Saskatoon, SK S7N 0X2
306-956-7625
306-956-7247
wolft@agr.gc.ca

Sheila Woods

Agriculture & Agri Food Canada
195 Dafoe Road
Winnipeg, MB
R3T 2M9
204-983-1330
204-983-4604
smwoods@agr.gc.ca

Kequan Xi

Field Crop Development Centre
Alberta Agriculture
6000 C&E Trail
Lacombe, AB T4L 1W1
403-782-8861
403-782-6120
kequan.xi@gov.ab.ca

Allen Xue

ECORC, AAFC
960 Carling Ave
Ottawa, ON K1A 0C6
613-759-1513
613-759-1926
axue@agr.gc.ca

Jennifer Zantinge

Field Crop Development Centre
Alberta Agriculture
6000 C&E Trail
Lacombe, AB T4L 1W1
403-782-8894
403-782-5514
jennifer.zantinge@gov.ab.ca

Tom Zatorski

U of Saskatchewan
51 Campus Drive
Saskatoon, SK
S7N 5A8
306-966-8480
306-966-5015
zatorski@sask.usask.ca

Kevin Zaychuk

20/20 Seed Labs Inc.
#201, 509 - 11 Avenue
Nisku, AB T9E 7N5
780-955-3435
780-955-3428
kevin@2020seedlabs.com

Wenchun Zhou

AAFC
5403 - 1st Ave South
Lethbridge, AB
T1J 4B1
403-317-3430
zhouw@agr.gc.ca

