

1994 GRADUATE STUDENT SYMPOSIUM



Program and Abstracts

January 29, 1994

1994 GRADUATE STUDENT SYMPOSIUM

Participating Universities:

Department of Plant Science, University of Manitoba, Winnipeg, Manitoba (Host)
Crop and Weed Sciences Department, North Dakota State University, Fargo, North Dakota
Department of Crop Science and Plant Ecology, University of Saskatchewan, Saskatoon, Saskatchewan

Friday, January 28, 1994

8:00 p.m. **Reception**
Travelodge Hotel, 2935 Pembina Highway

Saturday, January 29, 1994

8:30 - 8:45 a.m. **Registration (Room 543 University Centre)**

8:45 - 9:00 **Welcome**
Introduction of Judges (Sessions I and II)

9:00 **Seminars begin**

SESSION I: WEED SCIENCE (Chairman, Sessions I & II: GSA Pres. Luc Bourgeois)

9:00 - 9:15 **D. J. Debreuil, University of Manitoba**
"Investigations on Phenoxy Herbicide Resistant Wild Mustard, Sinapis arvensis"

9:15 - 9:30 **J. B. Frie, North Dakota State University**
"Wild Oat (Avena fatua L.) Resistance to Diclofop in the Red River Valley"

9:30 - 9:45 **J. D. Harbour, North Dakota State University**
"Herbicide Retention with Air-Assist and Conventional Sprayers"

9:45 - 10:00 **T. W. Roebke, North Dakota State University**
"The Interaction of Sugarbeet Herbicides and Aphanomyces cochlioides"

10:00 - 10:15 **G. Stilkowski, University of Manitoba**
"Purple Loosestrife - The Beautiful Killer"

10:15 - 10:35

Break

SESSION II: PLANT BREEDING AND CROP PRODUCTION

10:35 - 10:50

W. J. Bullied, University of Manitoba
"Removing Alfalfa Stands From the Crop Rotation With Herbicides"

10:50 - 11:05

S. A. Fitterer, North Dakota State University
"Amaranth (Amaranthus sp.) Stand Establishment and Harvest
Timeliness"

11:05 - 11:20

D. H. Gibson, University of Manitoba
"Selection of Early Maturing, High Yielding, Spring Wheat (Triticum
aestivum)"

11:20 - 11:35

H. A. Roman, North Dakota State University
"Inbreds vs Single Crosses as Topcross Testers in Early Maize (Zea
mays L.)"

11:35 - 11:50

N. Sissons, University of Manitoba
"The Effect of Continuous and Interrupted Leaf Wetness Periods on the
Infection Process of Tan Spot in Wheat"

11:50 - 12:50 p.m.

Lunch (Room E319, Plant Science)

12:50 - 1:10

Description of University of Manitoba
Introduction of Judges (Sessions III and IV)

SESSION III: MOLECULAR GENETICS (Chairman, Sessions III & IV: GSA Vice Pres. Rob Gulden)

1:10 - 1:25

M. A. Chowdhury, University of Saskatchewan
"Inheritance and Linkage of Morphological, isozyme and RAPD markers
in Grasspea (Lathyrus sativus L.)"

1:25 - 1:40

C. A. Goblirsch, North Dakota State University
"Identification of Genes Conferring resistance to Pathotype Pgt-QCC of
Puccinia graminis f. sp. tritici"

1:40 - 1:55

A. Kumar, University of Manitoba
"Cloning and characterization of a debranching enzyme - Limit
dextrinase"

- 1:55 - 2:10 H. Nair, University of Saskatchewan
"Inheritance and Linkage of Isozyme and Morphological Markers in
Fenugreek (Trigonella foenum-graecum)"
- 2:10 - 2:25 W. S. Stock, University of Manitoba
"Developmental Problems of Molecular Markers for a Wheat Breeding
Program"
- 2:25 - 2:40 S. Tewari, University of Manitoba
"What Role Does Differential Induction of Multigene Families Play in
Defense Response?"
- 2:40 - 3:00 Break

SESSION IV: PLANT PHYSIOLOGY AND BIOCHEMISTRY

- 3:00 - 3:15 K. S. Hamed, North Dakota State University
"Effects of Nitrogen Fertilizer on Wheat Flour Quality"
- 3:15 - 3:30 G. W. Massie, University of Saskatchewan
"The Effect of Photoperiod on Cold Acclimation of Three Winter Cereals"
- 3:30 - 3:45 S. Shirtliffe, University of Manitoba
"The Use of Nodulation Mutants of Common Bean (*Phaseolus vulgaris*
L.) in Nitrogen-15 Isotope Dilution Studies"
- 3:45 - 4:00 K. Ward, University of Manitoba
"The Effects of Processing and Storage on the Chlorophyll Derivatives
Present in Commercially Extracted Canola Oil."
- 6:00 Banquet (Travelodge) Social time - 6:00 p.m., Dinner - 6:30.
Speaker: Prof. Tim Ball, University of Winnipeg

Sunday, January 30, 1994

8:00 a.m. Breakfast - Travelodge Hotel

ABSTRACTS

SESSION I: WEED SCIENCE

INVESTIGATIONS ON PHENOXY HERBICIDE RESISTANT WILD MUSTARD, *SINAPIS ARVENSIS*

Debreuil D. J., Morrison I. N. Department of Plant Science. University of Manitoba.

In 1991 Dr. Ian Heap of the University of Manitoba confirmed the existence of a population of wild mustard resistant to phenoxy, as well as other auxin type herbicides. The resistant population originates from the Gilbert Plains region of Manitoba, where there are currently eight confirmed sites.

The objectives of my thesis research are: 1) to confirm the level of resistance of wild mustard to 2,4-D and dicamba under field conditions; 2) to obtain quantitative information on fitness of R (resistant) and S (susceptible) biotypes with the herbicide "off" and the herbicide "on"; 3) to determine the herbicide selection intensity based on seed return; and 4) to verify theoretical models that predict the enrichment of phenoxy herbicide resistance by inserting relative fitness and selection intensity values obtained from the experiment.

Growth analysis data provided quantitative information on fitness and seed return (effective kill) for the wild mustard populations, R and S, treated with increasing rates of 2,4-D and dicamba. Observations from both years of research confirmed moderate resistance to both 2,4-D and dicamba under field conditions. Dicamba treatments affected both mustard populations less than 2,4-D treatments. The selection intensity calculated using relative fitness data generated from plots treated with 2,4-D at 420 g a.i. ha⁻¹ was 1.0. By inserting this figure in predictive models, a rapid rate of enrichment is predicted, no doubt due to the significantly reduced seed return of the susceptible wild mustard plants versus the resistant plants.

WILD OAT (*Avena fatua* L.) RESISTANCE TO DICLOFOP IN THE RED RIVER VALLEY. Jeremy B. Frie, Richard K. Zollinger, Frank A. Manthey, and Beverly R. Durgan, North Dakota State University, Fargo, ND, University of Minnesota, St. Paul, MN.

Isolated sites of wild oat resistant to diclofop have been identified in the Red River Valley (eastern North Dakota and western Minnesota). The objective of this research was to determine the magnitude of diclofop resistant wild oat in the Valley.

Possible sites for wild oat seed collection were determined by a survey sent to growers, a random sampling, and cooperation with local crop consultants. Plants from 11 locations were treated with diclofop at 1120 g/ha at the 3-leaf stage. Known susceptible and resistant wild oat biotypes were also planted. Resistance to diclofop ranged from 0 to 100 percent from all plants tested. Plants from locations that had known to be exposed to diclofop expressed high susceptibility to the herbicide. Plants from nine locations with histories of high diclofop use expressed varying degrees of resistance to diclofop. For example, all wild oat plants from one location were susceptible to diclofop. However, at other locations, 40 to 100% of wild oat plants were resistant. Plants from only one location expressed 100% susceptibility to diclofop.

Most wild oat plants that expressed resistance to diclofop were from fields with a small grain/sugarbeet rotation. Diclofop is used extensively in small grains and sethoxydim is the only postemergence herbicide labeled for grass control in sugarbeet. Both are ACCase inhibiting herbicides. Repeated use of herbicides with the same mechanism of action increases the risk for resistance to develop. The Red River Valley is at high risk for the development of wild oat resistance to diclofop and sethoxydim.

HERBICIDE RETENTION WITH AIR-ASSIST AND CONVENTIONAL SPRAYERS. James D. Harbour and Calvin G. Messersmith, Crop and Weed Sciences Department, North Dakota State University, Fargo, ND 58105.

Air-assist nozzle development has allowed for low-volume application of sprays. The objective of this research was to determine glyphosate retention with adjuvants, including sheared and nonsheared polyvinyl polymer (PVP), using air-assist and conventional sprayer systems.

A modified air-assist sprayer system delivered 5 L/ha using 28 kPa air pressure. The conventional sprayer system, using a 650067 nozzle, delivered 160 L/ha at 280 kPa air pressure. Glyphosate at 0.5 kg ae/ha plus 7.5 g/L Chicago sky blue dye were applied with both sprayer systems.

Glyphosate retention with both sprayer systems was determined with surfactants at 0.5% (v/v) including a check (no surfactant), MON-0818, oxysorbic (20), and allinol (810-60). Treatments were sprayed onto 1- by 10- cm cattail leaf segments mounted on nails angled at 35 degrees from vertical. Leaf segments were washed with water, absorbance by dye was measured at 635 nm, and glyphosate retention was calculated from a standard curve. Glyphosate retention was determined with the same surfactants plus sheared and nonsheared PVP to simulate shearing of PVP caused by a standard pump.

Glyphosate retention was greater with the conventional than air-assist sprayer system both with and without PVP. However, retention was the same with sheared and nonsheared PVP for both sprayer systems. Retention with MON-0818 and oxysorbic (20) surfactants increased compared to the check for both sprayer systems. Glyphosate retention with allinol (810-60) surfactant was substantially greater with conventional compared to air-assist sprayer system and retention with PVP increased with all surfactants.

THE INTERACTION OF SUGARBEET HERBICIDES AND APHANOMYCES COCHLIOIDES. Troy W. Roebke, Alan G. Dexter, and Carol E. Windels, North Dakota State University, Fargo, ND58105, and University of Minnesota, Northwest Experiment Station, Crookston, MN56716.

Aphanomyces cochlioides causes rootrot in sugarbeet in North Dakota and Minnesota. This soil borne fungus can infect sugarbeet seedlings and older plants throughout the growing season causing extensive stand losses, decreased yields, and reduced quality. The following three factors are needed for disease development: 1) presence of A. cochlioides in the soil, 2) presence of sugarbeet plants, and 3) adequate to excess soil moisture and 20 to 37 C soil temperature. The objective of the experiment was to determine the effect of herbicides on A. cochlioides. Herbicides used in the experiment were preplant incorporated diethatyl, EPTC, ethofumesate, and cycloate. Postemergence herbicides were desmedipham, desmedipham + phenmedipham, clopyralid, and triflusulfuron. Plots treated with EPTC, ethofumesate, cycloate and desmedipham had significantly higher root yield and extractable sugar compared to the untreated check. The level of disease was evaluated at harvest by visually rating the scurfiness (root roughness) of the sugarbeet roots from zero = no scurfiness to 4 = more than 75 % of root surface covered by scurfiness. Disease rating for all the herbicide treated sugarbeet was lower than for untreated sugarbeet. However, cycloate was the only treatment that had a disease rating significantly less than the untreated check. Cycloate apparently suppressed and other herbicides may have suppressed A. cochlioides infection resulting in improved sugarbeet yield. Higher yields from herbicide treated sugarbeet compared to the untreated check also may have been partially due to early season weed competition. The untreated check and plots with poor control of red root pigweed had high populations of red root pigweed. Clopyralid does not control red root pigweed. Triflusulfuron and desmedipham plus phenmedipham gave less control than desmedipham.

Purple Loosestrife - The Beautiful Killer

Gayle Stilkowski, University of Manitoba

The biology, distribution, environmental impact, and possible control methods of purple loosestrife will be examined. Purple loosestrife (*Lythrum salicaria* L.) is a herbaceous perennial plant that was introduced into North America from Europe in the early 19th century. Purple loosestrife invades native wetland habitats, forming monospecific stands which eliminate food and shelter for wildlife species. It is also capable of migrating from wetland habitats onto agricultural lands. Purple loosestrife is now found across southern Manitoba and is spreading rapidly. Attempts at controlling purple loosestrife have not been successful. A variety of cultural control methods, including burning and mowing, have been tried. Currently there is no herbicide registered for use over open water in Canada that will control purple loosestrife. Efforts are underway to test the effectiveness of biological control agents. Insects have been imported from Europe with the hope that they will control purple loosestrife in North America. Biological control offers a long-term control method that would reduce population levels to an ecologically acceptable level.

SESSION II: PLANT BREEDING AND CROP PRODUCTION

Removing Alfalfa Stands From the Crop Rotation With Herbicides

W. J. Bullied and M. H. Entz, University of Manitoba

Perennial alfalfa provides many agronomic benefits in a crop rotation. However, many producers prolong the transition of alfalfa to annual crop in the rotation due to the difficulty of terminating the alfalfa stand with traditional tillage methods. Herbicides and tillage were evaluated for control of alfalfa at Portage la Prairie, and Glenlea MB. The time of alfalfa termination (Sept. and May) was also investigated. Roblin wheat and Bedford barley were seeded into chemically suppressed and tilled alfalfa residue. Alfalfa regrowth measured after harvest in both the fall and spring suppression indicated Roundup 5.00 L ha⁻¹ suppressed alfalfa as well as or better than the tillage treatment. There was no difference in the competitive ability of the wheat and barley to suppress regrowth of alfalfa. Wheat and barley yield at Portage la Prairie was increased by the use of herbicides over that of tillage for fall suppression, however the tillage treatment obtained higher yields under spring suppression. Alfalfa regrowth the following year at Portage la Prairie was less with Roundup 5.00 L ha⁻¹ and Lontrel 1.50 L ha⁻¹ than tillage for fall time of alfalfa suppression. However, there was no difference in the regrowth of alfalfa for the spring time of alfalfa suppression by Roundup 5.00 L ha⁻¹, Lontrel 1.50 L ha⁻¹, and tillage treatments. These experiments indicate that herbicides can provide increased control of alfalfa over that of tillage.

AMARANTH (Amaranthus sp.) STAND ESTABLISHMENT AND HARVEST TIMELINESS.

Scott A. Fitterer, Albert A. Schneiter, and Burton L. Johnson, North Dakota State University, Fargo, ND.

Amaranth (Amaranthus sp.) was cultivated by native Americans in Central and South America before their invasion by the Spanish. The crop was all but forgotten until recent years when a renewed interest in its use as a human food resurfaced. Amaranth has many industrial, medicinal and food product uses.

Research has been conducted on many aspects of amaranth but not in the areas of stand establishment and harvest timeliness. The first objective of this research is to identify those agronomic procedures which optimize field stand establishment and seedling survival of amaranth. This will be accomplished by a depth of planting study in the greenhouse and a tillage methods study in the field. Four depths of planting will be tested in the greenhouse for seedling vigor and percent germination. In the field, four tillage methods, each planted with clay coated and uncoated amaranth seed, will be tested for vigor index and percent emergence. The second objective is to determine the proper time for combine harvest to minimize shatter loss. This will be accomplished by a harvest timeliness study in the field. Periodic dates at first shattering will be harvested to determine moisture content, seed yields, percent germination, and percent dockage at harvest. Development of techniques for stand establishment and harvest timeliness will increase the probability of successful amaranth production in North Dakota.

Selection of Early Maturing, High Yielding, Spring Wheat (Triticum aestivum).

Darin H. Gibson, University Of Manitoba

In many areas of the prairies, cool wet growing conditions often lead to the production of high yielding, poor quality spring wheat. Losses in quality are mainly due to: 1) delays in maturity which increase the risk of frost damage, and 2) poor harvesting conditions which lead to weathering. Earlier maturing spring wheat cultivars would significantly reduce the risk associated with delayed crop maturity under cool wet conditions. Unfortunately, attempts to reduce time to maturity have usually resulted in a significant loss in yield potential. A better understanding of the relationship between plant development and yield is essential in order to make significant improvements. The main objectives of current research are to 1) Evaluate response to selection for a number of plant development and yield component parameters, and develop a selection strategy which will assist in the development of early maturing, high yielding cultivars. 2) Refine plant development models. Objective 1 is being met by assessing the relationship between plant development, yield and yield components of 50 F_8 lines from each of six crosses made between genotypes that differ in maturity and yield potential. Initial analysis of 1993 data shows that there is a great deal of variability among the lines within each cross for times to key developmental stages, height, spike number, plant stand, and total plot yield. This provides a very good range of genetic variability from which to assess response to selection, and develop selection strategies. Computer models will be used to compare expected response to selection with actual response in the field. Objective 2 is being met by conducting experiments in the field in order to develop a data base of environmental, phenological and yield data and apply this information to the plant development models.

INBREDS vs SINGLE CROSSES AS TOPCROSS TESTERS IN EARLY MAIZE (*Zea mays* L.). H.Z. Cross and H.A. Roman. Crop and Weed Sciences Department. North Dakota State University, Fargo, ND.

Accurately predicting topcross yields of advanced generation lines based on their early generation topcross performance would improve efficiency in plant breeding programs. A series of S2, S4, and S6 versions of 22 inbred lines were each crossed to 10 testers. The 10 testers were four inbred lines and the diallel set of single crosses among the four inbred testers. The S2, S4, and the S6 sets of topcrosses were each grown in separate experiments at two locations for two years. Correlations were computed between S2-S4, S4-S6, and S2-S6 performances for yield and other agronomic traits for all possible combinations of years and locations for each tester. Testers differed in their efficiency at predicting S6 topcross performance based on S2 or S4 topcross performance. Single cross testers appeared to be more efficient than inbred testers.

The effect of continuous and interrupted leaf wetness periods on the infection process of tan spot of wheat.

Norm Sissons, University of Manitoba

Pyrenophora tritici-repentis, the causal organism of tan spot, requires four to six hours of leaf wetness to infect wheat leaves. Little is known about the effect of interrupted leaf wetness on the infection process of tan spot of wheat. This study dealt with the effect of continuous and interrupted leaf wetness on the infection process of *Pyrenophora tritici-repentis*. Glenlea, a necrotic wheat cultivar, and 6B365, a chlorotic wheat line, were inoculated with isolate ASC1 (nec⁺chl⁺) to runoff. Plants were placed in a humidity chamber for 0, 2, 4, 6, 8, 12, 24, 36, 48 and 72 hours in the continuous leaf wetness experiment. Interrupted leaf wetness treatments consisted of 0, 2, 4, 6, 8, 10 or 12 hours of continuous wetness followed by a 6 hour dry period. All plants in the interrupted leaf wetness experiment were returned to the humidity chamber to complete a 24 hour leaf wetness period. Results showed that longer periods of continuous leaf wetness increased lesion size and number. Interrupting leaf wetness periods disrupted the infection process and reduced disease severity when it occurred during appressorial formation. Our data suggest that under field conditions short periods of leaf wetness (2-6 hours) followed by a dry period may reduce severity of tan spot of wheat.

SESSION III: MOLECULAR GENETICS

Inheritance and linkage of morphological, isozyme and RAPD markers in grasspea (*Lathyrus sativus* L.)

Mahoob Alam Chowdhury, Dept. of Crop Science and Plant Ecology, University of Saskatchewan, Saskatoon, SK, S7N 0W0

Grasspea is a minor pulse crop grown principally in the Indian subcontinent and also in North Africa and other parts of the Mediterranean basin. It has a great potential both as a food and fodder crop, due to its high nutritional value and its ability to fix large amounts of nitrogen and withstand water stress. However, its use as a grain crop has been limited by the presence of a neurotoxic element, ODAP (β -diaminopropionic acid), which causes lathyrism, a neuropathologic disease that results in permanent crippling. Few genetic studies have been conducted on the grasspea. The development of a detailed linkage map for *Lathyrus sativus* will greatly increase the efficiency of genetic and breeding studies in this crop. Marker assisted selection for low or zero ODAP lines would be an efficient approach in developing high yielding grasspea cultivars having low or zero toxicity. Furthermore, information on the chromosome location of a gene provides an alternative route to gene isolation and cloning. In the present experiment six crosses were made among 11 parents involving seven morphological, 20 isozyme and 80 RAPD markers. Data on individual F_2 plants will be analyzed to determine the inheritance and linkage. Inheritance will be determined by chi-squared test and the recombination fractions (r) will be estimated along with standard error (SE) by the maximum likelihood method. A computer software package, Mapmaker, will be used to analyze linkage and construct the linkage map.

Identification of Genes Conferring Resistance to Pathotype Pgt-QCC of *Puccinia graminis* f. sp. tritici. C.A. Goblirsch, and R.D. Horsley. North Dakota State University, Fargo, ND.

Allelism tests with crosses between six partially resistant barley accessions (Diamond, Q21861, Heitpas 5, PC11, PC84, and PI452421) were completed. The F_1 plants from crosses, between the other resistant parents and Q21861 segregated for resistance, which suggests that the resistance of Q21861 is different than the other partially resistant parents and appears to be a complete resistance. The results indicate that there is no relationship between the resistance of Heitpas 5 and the other resistance parents. Also, Diamond, PC84, and PI452421 may have the same gene(s) for resistance but different than PC11. Crosses between the susceptible parent (ND11055) and resistant accessions were completed. Using RAPD's we evaluated 329 primers (OPERON). A total of 67 primers identified putative polymorphisms between resistant and susceptible parents. Of the 67 primers, 19 produced single bands found in all resistant lines, but absent in ND11055. The remaining primers produced a band found in at least one of the resistant lines, but not in either of the susceptible lines. One primer (AG-15) produced a putative polymorphism between Q21861 and the susceptible parents. Two primers identified putative polymorphism between the three resistant parents (Diamond, PC84, and PI452421) and the susceptible parent.

Cloning and characterization of a debranching enzyme - *Limit dextrinase*.

Alok Kumar and R. D. Hill, University of Manitoba, Winnipeg, MB, R3T 2N2, Canada.

Hydrolysis of starch in the cereal endosperm is being carried out by α - and β -amylases. α -amylase randomly cleaves α -(1-4)-linkages from the side chains of amylopectin whereas β -amylase sequentially removes the β -maltose residues from the non reducing ends. This cleavage stops just before the branch point is reached. The remaining structures called limit dextrins can not be hydrolysed by these amylases. *Limit dextrinase*, a debranching enzyme along with other hydrolytic enzymes is required for the complete hydrolysis of starch. This enzyme also complements the degradation of cereal starch to the fermentable sugars in the brewing process.

Our initial experiments suggest very low abundance of limit dextrinase both in the developing as well as in the germinating barley kernels. Considering the economic value of this enzyme in the brewing industry and its importance in the basic studies, efforts to alter the enzyme levels or its activity *in vivo* have been frustrated due to lack of enough information on its molecular properties. Therefore, we are attempting to clone the gene coding for limit dextrinase.

Using western blot analysis, we have observed maximal expression of this protein in endosperm tissue of developing barley kernels. Likewise, we assume the message for this protein to be maximum in the above tissue. Thus a representative cDNA library is being prepared from endosperm tissue. This library will be screened using antibodies raised against limit dextrinase. The isolated cDNAs should enable us to study the expression of limit dextrinase in different physiological contexts. The sequence of this gene will allow us to deduce the amino acid sequence and will be helpful in delineating the molecular properties of the protein with respect to amino acid composition, folding and secondary structure. In future, this data and other can be used to possibly engineer the gene, thereby modulating its abundance.

Inheritance and Linkage of Isozyme and Morphological Markers in Fenugreek (*Trigonella foenum-graecum*).

Harikumar Nair¹ and A.E. Slinkard², 1. Department of Crop Science and Plant Ecology. University of Saskatchewan; 2. Crop Development Centre, University of Saskatchewan.

Fenugreek is a new crop to Canada. It is widely grown in India, Argentina, the Middle East and Mediterranean countries. Fenugreek seeds are used as a condiment and make up about 10% of many spice mixtures. The plant and the seeds reputedly have various medicinal properties and the seeds are a good source of dietary fibre. Forty-eight fenugreek lines were screened for isozyme polymorphisms in 20 enzyme systems using horizontal starch gel electrophoresis. Isozyme polymorphisms were found in seven enzyme systems. Morphological markers (cotyledon colour, pubescence, zero tannin seed coat and single vs. double pods per leaf axil) were also studied. Crosses were made between selected parental lines to obtain 34 F₂ populations segregating for isozyme and morphological markers out of which seven populations were further analyzed. Close genetic linkage was observed between the gene locus controlling cotyledon colour, *Yc*, and the loci coding for two isozymes of esterase, *Est-2* and *Est-3*. The data also suggest weak linkage between the locus coding for triose phosphate isomerase, *Tpi-1*, and both *Yc* and *Est-3*.

Developmental problems of molecular markers for a wheat breeding program.
Wade Stock, University of Manitoba

With the development of molecular genetic technologies, interest has arisen in the development of molecular markers (RAPDs and RFLPs) for facilitating selection procedures in wheat. The use of molecular markers is best suited for traits that are too complex or expensive to score. However, the development of these markers is difficult in wheat for several reasons. Stringent selection for improved quality traits has resulted in reduced genetic variation. Consequently, limited polymorphism exists for the development of molecular markers. Secondly, the homoeologous nature of wheat with its three basic genomes results in the co-migration of DNA fragments. Because wheat is a hexaploid, approximately three-fold more loci must be screened in order to find the same number of polymorphic markers as compared to a diploid relative. Thirdly, wheat DNA contains a nuclease that is stimulated by the presence of EDTA. EDTA is commonly used in plant DNA extraction methodologies to inhibit nucleases. As a result, DNA extraction procedures must be more complex in order to attain stable storage of DNA. Lastly, there is a question of whether or not a molecular marker can be used outside the segregating population from which it was derived. Recent data suggests that only 20 percent of all markers can be used in other populations. Additional research needs to be directed towards molecular marker development and application in wheat before it becomes a standard tool in the selection procedure.

What role does differential induction of multigene families play in defense response? Sandhya Tewari, Stuart M. Brown and Brian Fristensky, University of Manitoba, Winnipeg, MB R3T 2N2. Canada.

Multigene families consist of closely related genes coding for identical or very similar proteins. The *drr49* disease resistance response multigene family of pea is one such example of a defense related family with at least four members cloned so far: *drr49a*, *drr49b*, *drr49c* and *drr49f*. Wild species of pea also contain copies of *drr49* which correspond very closely to those in *P. sativum*.

To test if the defense response of pea is characterized by differential induction of individual members of *drr49* gene family, different disease contexts were selected by using two different pathogens (*Fusarium solani* f.sp. *pisi* and f.sp. *phaseoli*), two different time points during the interaction (8 and 48 h) and six different host species: Garden pea, *Pisum sativum* cv Alaska, which shows a moderate to high level of resistance to both pathogens; Three wild species of pea, *P. humile*, *P. elatius* and *P. fulvum*, which show a lower degree of resistance; Two species of *Lathyrus*, *L. sativus* and *L. tingitatus* which show extremely low levels of resistance.

Northern blot data shows that *drr49c* and *drr49f* accumulate to very high levels in susceptible *L. tingitatus* at the 48 h time point in the f.sp. while *drr49a* is hardly detectable. In the *P. fulvum-phaseoli* interaction, there is no detectable expression of *drr49a* and *drr49c* at the 8h time point while *drr49f* shows considerable expression which increases to very high levels by 48h. Expression of *drr49a*, on the other hand increases only marginally by 48h and that of *drr49c* increases to moderate levels.

SESSION IV: PLANT PHYSIOLOGY AND BIOCHEMISTRY

Effects of Nitrogen Fertilizer on Wheat Flour Quality. Kamal S. Hamed. Crop and Weed Sciences Department. North Dakota State University, Fargo, ND.

It has been known that quality characteristics of wheat flour generally improve as wheat grain protein is increased. There are reports of baking quality declining at very high grain protein levels, particularly when these are the results of late nitrogen fertilizer (N) application. It has been suggested that these discrepancies in the correlation between protein content and baking quality may be caused by an imbalance between N and sulfur (S) in the flour.

The positive relationship between N application rate and grain protein percentage is well documented. But little information has been reported on how N application rate and N application rate X cultivar interactions influence milling and baking qualities of a certain class of wheat. Moreover, responses to increasing N levels were not consistent over cultivars. It is suggested to cross a bread wheat cultivar with superior N accumulation capacity to another wheat cultivar with superior grain yield in an attempt to develop a cultivar with improved N use efficiency.

The Effect of Photoperiod on Cold Acclimation of Three Winter Cereals

Garth W. Massie

Dept. of Crop Science and Plant Ecology

University of Saskatchewan, Saskatoon, SK, S7N 0W0

Two major environmental stimuli which effect the ability of plants to acclimate to a cold stress are cold temperature, and photoperiod. The importance of photoperiod to initiate cold hardening in woody plants has been well established, however, the role of photoperiod on the hardening of herbaceous plants is more dubious. Recently, it has been suggested that a short photoperiod (8h light) was more effective in hardening Puma rye than long photoperiod (16h light), and constant illumination (24h light) was intermediate between the two. In this experiment three winter cereals: Puma fall rye, Norstar winter wheat, and Elmira winter barley were subjected to four photoperiod regimes to determine their effect on cold hardiness. The four treatments used were: (1) 24h light, (2) 16h light, 8h dark, (3) 8h light, 16h dark, (4) 8h light, 8h dark. These treatments were under an irradiance of 400 $\mu\text{Mol photons/m}^2/\text{s}$, at constant temperature. L_{50} tests of freezing tolerance indicate photoperiod does not have a dramatic effect on the freezing tolerance on the crowns of the winter cereals tested.

The Use of Nodulation Mutants of Common Bean (*Phaseolus vulgaris* L.) in Nitrogen-15 Isotope Dilution Studies. S.J. Shirliffe and J.K. Vessey, University of Manitoba.

The ^{15}N isotope dilution method is a common field method of measuring the nitrogen fixation of the legume:rhizobium symbiosis. This method requires a reference crop that is incapable of fixing atmospheric nitrogen. Ideally this reference crop should assimilate fertilizer and soil nitrogen in the same proportion as the nitrogen fixing crop. Barley is commonly used as a reference crop in ^{15}N studies of nitrogen fixation in common bean. Given the differences between the growth rate and root morphologies of barley and common bean it is questionable that barley meets the above criteria. A ^{15}N isotope dilution study was undertaken to determine the suitability of the nodulation mutants of common bean (*Phaseolus vulgaris* L. cv. OAC Rico) R-69 and R-99 for use as reference crops.

OAC Rico, R-69 and R-99, and barley (*Hordeum vulgare* cv. Ellice) were grown at two different levels of fertilizer nitrogen in a field RCBD. One metre square subplots were enriched with ^{15}N . All plants were harvested at physiological maturity and analyzed for atmospheric excess ^{15}N . The nitrogen derived from the atmosphere (Ndfa) of OAC Rico was calculated using the isotope dilution method for each of the three different reference crops. Ndfa of OAC Rico as estimated by the mutants R-69 and R-99 was not significantly different. Barley gave estimates of Ndfa that were significantly lower than those derived from the mutants. The treatment standard deviations for barley was much higher than those of the non-fixing mutants. The results suggest that the nodulation mutants R-69 and R-99 are more suitable than barley as reference crops for OAC Rico in field ^{15}N isotope dilution studies.

The Effects of Processing and Storage on the Chlorophyll Derivatives Present in Commercially Extracted Canola Oil. Kerry Ward, University of Manitoba, Winnipeg.

Green canola seed results in oil that contains a lot of chlorophyll. Chlorophyll pigments can degrade to pheophytins, which have even stronger prooxidant activity, as well as other degradation products, so the pigment composition may have significant effects on the shelf life of the oil. We conducted a study to determine which chlorophyll pigments were present in canola oil at different stages of the extraction process, and in what proportions. The second objective was to examine changes in these pigments during oil storage under various conditions. Samples of pressed, solvent-extracted, crude and degummed canola oils were stored for one month in the freezer, refrigerator, and at room temperature both in the light and in the dark. Each chlorophyll derivative (chlorophylls, pheophytins, pyropheophytins) was measured by HPLC on a weekly basis. The main pigments in the oil prior to degumming were pheophytin a, pyropheophytin a, chlorophyll a and chlorophyll b. Degumming converted chlorophylls to pheophytins and pyropheophytins. The "a" derivatives comprised 81 to 100% of total chlorophyll pigments in fresh canola oil. The proportion of "a" derivatives was dependent on the type of oil and the total chlorophyll content of the oil. During oil storage, chlorophyll a was converted to pheophytin a and pyropheophytin a and chlorophyll b was converted to pheophytin b. Both light and higher temperatures facilitated these reactions.