SESSION 2:

FOOD SAFETY AND TOXICOLOGY

Co-Chairpersons: Jim Pestka and
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IMPACT OF BRAN PROPERTIES ON *FUSARIUM* MYCOTOXIN LEVELS IN WINTER WHEAT (*TRITICUM AESTIVUM* L.) KERNELS

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ABSTRACT

Wheat bran is an important source of dietary fiber, and according to consumers, they prefer the appearance and taste of whole-wheat products made from white wheat compared to those made from red wheat. However, since the predominant FHB mycotoxin, deoxynivalenol (DON) accumulates primarily in the bran layer of kernels, the bran may be a significant source of DON, especially in white-wheat products made using white wheat. Moreover, differences in bran between white and red wheat kernels may impact the DON in such products, presenting a potential food contamination risk. Near-isogenic lines (NILs) of red and white winter wheat were developed for use in this study to examine potential bran differences and identify the impact of any genetic differences on the accumulation of DON in bran. In two greenhouse trials we artificially infected parents, NILs, and control varieties with *Fusarium graminearum* (teleomorph *Gibberella zeae*) to induce FHB, and samples were collected from both infected and uninfected plants. To compare accumulation of DON in the bran layer, the samples were pearl-milled to produce bran and non-bran fractions. DON accumulation in bran fractions was significantly higher than in non-bran fractions. Though the differences were not statistically significant, generally, DON accumulation was higher in the bran fractions of red kernel genotypes compared to accumulation in the bran fractions of white kernel genotypes. To determine the nature of these differences, we are also analyzing possible morphological and chemical bran differences between the white and red NILs. Using the NILs, we will measure bran layer thickness from kernel cross-sections and determine the impact of bran extractions on fungal growth in vitro. Data from these analyses will be compared and presented.

A BARLEY UDP-GLUCOSYLTRANSFERASE FORMING A NOVEL ZEARALENONE-GLUCOSIDE

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ABSTRACT

During work aiming to identify deoxynivalenol (DON) inactivating UDP-glucosyltransferases, several barley genes were DON inducible, but did not confer DON resistance upon heterologous expression in yeast [1]. We found that one of the candidate genes, designated *HvZOG2*, encodes an enzyme capable of glycosylating the second prominent *Fusarium* mycotoxin, zearalenone. Besides the previously described [2] product zearalenone-4-O-glucoside (Z4G) also a second compound consistent with the mass of a glucoside was formed. The two products, Z4G and, ZON-2-O-glucoside were produced in about 1:1 ratio. According to a new nomenclature and numbering system for zearalenone derivatives [3] the novel product should be named and abbreviated ZEN-16-glucoside. Yeast expressing *HvZOG2* was grown in a fermenter and treated with zearalenone. The zearalenone-glucosides released into the medium were purified by solid phase extraction. The ZEN-16-Glc was eluted with 60% methanol and concentrated before further purification by preparative HPLC (Waters Sun Fire C18 column, 40-80% methanol gradient). The structure of the purified compound was confirmed by NMR. The occurrence of ZEN-16-Glc in crop plants is currently under investigation. We will also determine, whether ZEN-16-Glc, like the previously described ZON-4-Glc/ZEN-14-Glc, is easily hydrolyzed back to the parental Fusarium mycotoxin and should therefore be considered as an additional masked mycotoxin [4].

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DON – PAST, PRESENT AND FUTURE

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ABSTRACT

Deoxynivalenol (DON) was discovered by the Japanese from grain that caused 'red mold disease'. However, Fusarium head blight was long recognized in Europe as a substantial problem in both the old and new world. Spanish cultivars of wheat were imported to the new world (by Columbus) followed by English and French cultivars by fisherman and emigrants in the 16th century. What we know as FHB was reported in the 17th century in New England and Quebec & Acadia (New Brunswick). Quebec priests wrote that the grain was in some way toxic. Ergot epidemics were by then known to be caused by a fungus. Modern awareness that that FHB resulted in grain that was toxic began in the 1920s during a series of bad epidemics. American wheat exported to Germany was reported to cause emesis in swine by German researchers in 1929. Reporting in a 1926 Phytopathology, Russian workers discussed scabby wheat producing "inebriant bread" associated with the now recognizable symptoms of acute DON toxicosis. Large FHB screening programs began in the US during that time. Many of these older cultivars have reasonable tolerance to FHB but have poor yield by today's standards.

When FHB devastated farmers around the Great Lakes in the 1980s and later in the mid-1990s, much research was again stimulated. Programs to understand agronomic factors and the nature of the resistance have produced thousands of papers in the past 30 years. None-the-less, after a century of research, cultivars that have adequate resistance to FHB and other diseases and have appropriate quality characteristics remain elusive. We remain in a period where it is necessary to manage the problem to ensure that there is an adequate supply of soft and hard wheat in the affected areas. The strategies to do this involve resistance screening programs, forecasting systems to ensure appropriate use of fungicides, and regulations.

As witnessed by the differing views on appropriate regulation of aflatoxin and ochratoxin A, over-regulation affects all partners in the chain from farm to fork. A determination of reasonable certain of no harm of a particular toxin requires a sophisticated understanding of the mode of action of the compound. Increased understanding allows the uncertainties to be better defined. In 2004, an ILSI-EU workshop on DON identified a number of priorities for action. One of these was that the mechanism of action of the mechanism of food refusal in non-human primates. Although these data have so far not indicated that the PMTDI is insecure, the widespread occurrence of the DON-glycoside in European cultivars forces a re-examination of the question.

QUALITY ASSURANCE ISSUES IN DON TESTING

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ABSTRACT

Mycotoxin testing is an essential component to the USWBSI mission of developing effective control measures that minimize the threat of Fusarium head blight (scab), including the reduction of mycotoxins, to the producers, processors, and consumers of wheat and barley. The four diagnostic labs that are supported through USWBSI funding analyze in excess of 50,000 samples of wheat and barley each year, and support all research areas of the Initiative. As such, an understanding of factors that impact accuracy and precession of deoxynivalenol (DON) data, as well as laboratory throughput, are very important. The intent of this presentation is to review the basics of DON testing methodology, sources of error, and then the limitations on throughput. Discussion of check samples, limits of detection (LOD), limits of quantitation (LOQ), and DON recovery will address the interpretation of data, in terms of quality assurance. Sampling issues, which were previously discussed as the major source of error, will be reiterated.