# Potential of Wide Crosses to Improve the Resistance to Vomitoxin Accumulation in Wheat Following Infection by Fusarium Head Blight

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**Abstract**

Deoxynivalenol (DON) levels were determined in landraces of rye from Brazil, in a collection of triticales and a series of triticale amphiploids. Two of three rye landraces showed a resistant reaction to DON. Seven triticale accessions of the 371 score showed lower levels of incidence, severity and DON content. A total of eight Tritordeum (*Triticum durum* × *Hordeum chilense amphiploids*) were scored and showed lower DON levels. Stable lines with lower Fusarium head blight (FHB) and DON levels were selected in progenies from crosses of wheat to preselected accessions of *Triticum monococcum and Aegilops speltoides*. Both selections compared favourably to the check cultivars in term of agronomic traits indicating minimal linkage drag. One stable resistant line with lower DON levels was isolated in the F7 generation of progenies from crosses to *Tritium timopheevii*. Lower DON levels were observed in field trials of advanced generation progeny from crosses of wheat to *Aegilops cylindrica* and *Triticum miguschovae*. The findings indicate that the alien species accessions or segregating populations from the inter-specific or inter-generic hybridization can provide material with variability for DON content.

**Keywords:** deoxynivalenol (DON), interspecific/intergeneric, hybrids, segregating, populations

## 1. Introduction

Fusarium head blight (FHB) is a ubiquitous fungal disease of wheat, barley, oats, rye and ear rot of maize. Deoxynivalenol (DON) is a secondary metabolite of Fusarium head blight. DON content renders the harvested grain unsuitable for food or feed. It can cause malfunction of respiratory, immune and even reproduction systems. It has been estimated that for

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each 1 ppm increase in DON content in harvested grain, feed consumption in swine decreases by 7.5% [1, 2]. Additional costs are incurred in lowering the DON level of threshed grain. In North America, the US-FDA has set tolerance limits for DON of 1 ppm in processed grain [1, 2], whereas Health Canada has set regulations of 2 ppm in uncleaned soft wheat for use in non-staple foods and 1 ppm in uncleaned soft wheat for use in baby foods [3, 4].

In an epidemic year such as 1966 in Southwest Ontario in Canada, samples of winter wheat taken directly from farmer's combine showed a range in DON content of 1.1–13.9 ppm [5]. These findings point to the fact that genetic resistance must be put into the wheat crop to reduce the DON content.

In terms of breeding for resistance to FHB, earlier efforts were focussed on accumulating genes that reduced the symptoms of Type I and Type II resistance. Bai [5] was among the first to consider the inheritance of two other traits, Fusarium damaged kernels (FDK) and DON content. These two factors are receiving additional attention lately.

The correlation between FDK and DON is in the order of 0.81 [6] but they are much lower for incidence/severity and DON content, indicating that FDK and DON deserve additional attention as measures of FHB resistance.

Somers et al. [7, 33] were the first to suggest that DON accumulation was controlled by independent quantitative trait loci (QTL). These QTL were located on chromosome 5A, on 2D (coincident with a plant height QTL) and on chromosome 3BS (coincident with a QTL for Type I resistance). In addition, a number of minor QTL that were not specifically mapped were revealed in that study and shown in **Figure 1** of that publication.



**Figure 1.** Resistance to Fusarium head blight in an accession of *Triticum monococcum*. Disease symptoms developed at 21 days after artificial inoculation.

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This was followed by similar reports [8, 9, 10]. In the latter study, a major QTL for DON content was mapped on chromosome 2AS that was independent of FHB severity. The cultivar CJ9306 was the source of several QTL for resistance to DON accumulation [11]. Two new QTLs were reported, in that study QFhs.nau-2DL and QFhs.nau-1AS, whereas two others, QFhs.ndsu-3BS and QFhs.nau-SAS, were validated in that study.

This overview will discuss the variability for DON content in alien relatives of wheat and in progenies obtained from wide crosses with wheat.

## 2. Materials and methods

At the start of the project, large numbers of accession of alien wheat relatives were acquired from numerous gene banks. In addition, cytogenetic stocks and inter-specific and inter-generic hybrids were screened for resistance. Some of the numbers of accessions acquired for screening included 200 accessions of *Triticum monococcum* and 370 accessions of triticales plus lower numbers of other species and hybrids.

The initial screening invariably consisted of inoculation with point inoculation in greenhouses or corn spawn in field plots, followed by scoring of symptoms. In the initial screen, the obvious susceptible lines were discarded. Screening on promising lines was repeated. Evaluation of DON content on ground seeds harvested from inoculated plots was carried out on lines that showed minimal scab symptoms.

The lines showing lowest scab symptoms and lower DON content were then crossed to wheat. In most cases, this involved the application of growth hormones following pollination, then rescuing of hybrid embryos and culturing on artificial media. In most cases, backcrossing to a recurrent parent was necessary to restore full fertility. Screening of progenies from wide crosses was carried out by selecting resistant segregates with minimal symptoms following inoculation. DON contents were determined on lines with minimal symptoms. In some cases, DON contents were determined directly on alien species or cytogenetic stocks following inoculation.

For point inoculation—Type I resistance—plants were grown in controlled environments at day/night temperatures of 20/15°C and 16 hours photoperiods supplied by a combination of florescent and incandescent lamps. Spikes at the 50% flowering stage were point inoculated by injecting 10 μl of a 50,000 spores/ml suspension into a central floret on the spikes. Inoculated plants were retained in a unit maintained at 25°C for 48 hours and 95% RH, then moved to a normal growth cabinet. Symptoms were read at 21 days after inoculation. Symptom scores were expressed as % infected florets. Other symptoms such as blackened rachis were also recorded [12].

Type II resistance was usually evaluated in field plots in the epiphytotic nursery. Where seed quantities were adequate the plots consisted of two 1-m rows spaced at 6 inches apart and ideally replicated three times. At the boot leaf stages, corn spawn consisting of inoculated corn and barley seed was spread between the rows at the rate of 80 g/m$^2$. Applications

were repeated 1 week later. An irrigation system was activated twice a day to maintain a high relative humidity to enhance sporulation of the inoculum. Flowering dates of each plot were recorded, defined as the stage of 50% anthesis. At 21 days after the flowering date, disease incidence and severity was estimated visually for each plot and recorded. FHB indices were calculated from these readings. The plots were hand harvested at physiological maturity.

Threshing was done with a small plot thresher adjusted to retain the shrunken Fusarium damaged kernels (FDK). Two 1-g aliquots were removed from each sample and ground in a Wiley mill. To ensure homogeneity of the aliquots, the seed was put through a seed divider.

DON contents were estimated by an ELISA test using established methods [13]. Don contents of plots were expressed as parts per million. The check cultivars in field plots were Roblin as the susceptible check and Sumai3 as resistant. Other checks were selected as those that were parents of the various populations.

## 3. Results

### 3.1. Triticum monococcum

Excellent reviews have been written listing the variability for resistance to FHB in alien species [14, 15–18, 34]. *T. monococcum* was not listed in those reviews. *T. monococcum* was one of the species screened for FHB resistance in our studies. We started by screening 200 accessions of *T. monococcum* that were obtained from M. Trottet of INRA. After repeated screening, line 10-1 was identified as having a fair level of FHB resistance (**Figure 1**) [19, 20, 21]. Line 10-1 was crossed to the spring wheat cultivar AC Domain. After repeated backcrossing and screening, line M321 was selected. The values for percent infected florets following point inoculation were 8% compared to 4% for the resistant check Sumai3 and 32% for Roblin the susceptible check. The DON content of M321 was 5.5 ppm compared to Sumai3 at 2.1 and Roblin at 17.2 (**Table 1**). M321 was crossed to AC Domain and a doubled haploid mapping population of 80 lines was produced by the maize pollination method [22]. A QTL for FHB resistance was located in chromosome 5A, linked to the marker Xwme705 [18].

The agronomic characteristics of line M321 are shown in **Table 1**. Line M321 compares favourably with check cultivars in terms of agronomic traits such as plant height, yield, thousand kernel weight (TKW), protein content and even flour yield. The grain yield of this line is reasonable compared to AC Barrie, a check cultivar. The data in **Table 1** indicate that there is minimal linking drag in M321. The lowered DON content relative to the checks could be a useful attribute for improvement of disease resistance of wheat.

### 3.2. Aegilops speltoides

FHB resistance was also sought in *Aegilops speltoides*. In this case, 50 accessions were screened and line S184 selected [19, 23]. It has previously been shown that different accessions of *Ae. speltoides* can lead to different levels of meiotic chromosome pairing in F1 hybrids with wheat. The hybrid

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| Genotypes | Yield (kg/ha) | TSTWT (kg/hl) | HT (cm) | Protein (%) | Flour yield (%) | DON (ppm) |
|---|---|---|---|---|---|---|
| Sumai3 | 2895 | – | 88.5 | 13 | – | 2.1 |
| M 321 | 3272 | 79.3 | 76 | 13.9 | 57.5 | 5.5 |
| S 184 | 3246 | 80.3 | 86 | 13.3 | 67.2 | 3.4 |
| AC Barrie | 3304 | 80.5 | 79 | 13.7 | 66.8 | 6.5 |
| Roblin | – | – | – | – | – | 17.2 |

**Table 1.** Agronomic characteristics and DON content of FHB resistant lines introgressed into wheat from *T. monococcum* (M321) and *Ae. speltoides* (S184).

between AC Domain and the resistant speltoides accession showed an average of 3–4 bivalents at meiosis, i.e. the accession that we chose produced a high level of chromosome pairing in the F1 hybrid. The level of recombination between wheat chromosomes and those of *Ae. speltoides* would then be relatively high. Despite this, three backcrosses were required to restore fertility in the progeny. The agronomic characteristics of line S184 are shown in **Table 1**.

For most agronomic traits, such as plot yield, plant height protein content and even flour yield, the values for S184 compared favourably with the check cultivars (**Table 1**) [35]. Perhaps the most important attribute of this line is the lowered DON content. The DON content as shown in **Table 1** is 3.4 ppm compared to Sumai3 at 2.1, AC Barrie at 6.5 and Roblin at 17.2.

### 3.3. Triticum timopheevii

A resistant accession of *T. timopheevii* (AAGG genome) was crossed to the wheat cultivar Crocus which has all three crossability genes. The F1 was backcrossed to Crocus [24]. A population of 1500 $BC_1F_2$ plants was established and 535 BC1F7 lines were developed in the greenhouse using single seed descent. One hundred lines were selected based on full plant fertility and good agronomic traits and evaluated for their FHB reaction in the field. The line TC67 was selected based on its enhanced FHB resistance (**Figure 2**) and good agronomic traits. To map the resistance trait, a mapping population was established by crossing TC67 to the moderately susceptible cultivar AC Brio. An F7 population of 230 RIL was established by SSD and evaluated for a number of FHB-selected traits in field and greenhouse plantings.

As shown in **Table 2**, the DON content of TC67 and Brio was 1.3 and 3.0 ppm, respectively. The population mean for DON content was 2.2 with a range of 1.0–5.1 ppm. The QTL for this trait was mapped to chromosome 5A [25].

### 3.4. Aegilops cylindrica

*Aegilops cylindrica* is a tetraploid with the CCDD genome constitution. An accession collected in the wild by Alexander Rybalka of the Plant Breeding and Genetics Research Institute at Odessa Ukraine showed resistance to FHB. It was crossed to a local cultivar and a FHB resistant, stable derivative Cyl-1 was selected in the progeny. In our tests that line gave DON ratings intermediate between Sumai3 and Roblin. The DON content of Cyl-1 was 4.5 ppm




**Figure 2.** Resistance to Fusarium head blight expressed in TC67 an introgression from *T. timopheevii*. Disease symptoms expressed at 21 days after inoculation. Roblin is the susceptible check.

| Trait | Parents | | Population mean | Population range | Heritability |
|---|---|---|---|---|---|
| | TC 67 | Brio | | | |
| Disease spread within the spike (%) | 5.1 | 35.1 | 35.2 | 5.1–99.2 | 0.89 |
| Disease incidence (%) | 18.0 | 42.6 | 36.4 | 12.4–65.4 | 0.60 |
| Disease severity (%) | 41.3 | 41.8 | 50.8 | 25.5–76.7 | 0.47 |
| FDK (%) | 2.4 | 6.3 | 7.4 | 1.7–22.3 | 0.67 |
| DON content (ppm) | 1.3 | 3.0 | 2.2 | 1.0–5.1 | 0.69 |

**Table 2.** FHB scores and DON content, means, ranges and heritability in a mapping population derived from TC67, a derivative from *Triticum timopheevii* and wheat cultivar Brio.

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compared to Sumai3 and Roblin at 3.0 and 10.0, respectively. Cyl-1 was crossed to North America cultivars AC Superb, AC Barrie and Alsen as shown in **Table 3**.

The populations were advanced to F4, F6 and F7. Progenies were grown in field plots and DON contents determined. The distribution of DON levels was similar for the three populations. Although the DON levels in the checks Sumai3 and Roblin were at expected levels, the levels in the populations were unusually low and will need to be repeated.

Continued selection for a combination of improved agronomic traits and lower DON content resulted in line Odessa129-2 with a DON content of 9.6 ppm compared to Sumai3 and AC Superb at 3.9 and 47.2, respectively.

### 3.5. Triticum miguschovae

*Triticum miguschovae* is an amphiploid between *T. timopheevii* (AAGG genome) and *T. tauschii* (DD genome) [26, 27].

The spikes of the amphiploid display many alien species traits as shown in **Figure 3**. Following point inoculation with a 50,000 spores/ml, suspension of *Fusarium graminearum* spores, the symptoms did not spread beyond the inoculated floret (**Figure 3**). A similar display of symptoms was observed in $BC_2$ progeny following backcrossing to AC Superb (**Figure 4**). AC Superb has no FHB resistance so the observed resistance must be contributed by the alien parent.

The progenies of $BC_2$ plants were advanced to F5 with selections made on point inoculation symptoms at each generation.

A total of 35 F5 lines were grown in the epiphytotic nursery in single row plots and only one replicate. The DON content of the 35 lines ranged from 0.6 to 11.3 ppm (**Table 4**). Ten of the best F7 lines grown in the field gave a range of DON values of 3.5–8.2 ppm. The mean DON

| Derivatives | Generation | DON levels (ppm) | | | | No. of lines |
|---|---|---|---|---|---|---|
| | | <1 ppm | 1–2 ppm | 2–5 ppm | > 5 ppm | |
| *Cyl-1/AC Superb | F7 | 11 | 8 | 10 | 4 | 33 |
| Cyl-1/AC Barrie | F6 | – | 4 | 14 | 9 | 27 |
| Cyl-1/Alsen | F4 | 7 | 6 | 6 | 4 | 23 |
| Checks | | | | | | |
| Sumai3 | | | 1.2 | | | |
| Roblin | | | | | 11 | |
| Strongfield | | | | | 17.6 | |

*Note*: *Cyl-1 FHB-resistant accession of *Aegilops cylindrical*.

**Table 3.** DON content of FHB-resistant lines derived from progenies of *Aegilops cylindrical* crossed to wheat.



**Figure 3.** Resistance to Fusarium head blight expressed on spike of *Triticum miguschoae* (AGD) (R) at 21 days after inoculation. Roblin (L) is the susceptible check.



**Figure 4.** Symptoms on $BC_2$ spike of hybrid between Superb and *T. miguschovae* at 21 days after point inoculation.

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| Generation | No. of lines | DON content (range) |
|---|---|---|
| F5 | 35 | 0.6–11.3 |
| F7 | 10 | 3.5–8.2 |
| **Checks** | | |
| Sumai3 | | 2.7 |
| Fukuhokomuji | | 13.5 |
| AC Superb | | 17.8 |

**Table 4.** FHB symptoms and DON content (ppm) in progenies from intercrosses of bread wheat cultivar AC Domain with *Triticum miguschovae*.

levels of the checks for the field tests were Sumai3 at 2.7 ppm, Fukuho at 13.5 ppm and AC Superb at 17.8 ppm.

Continued selection for a combination of improved agronomic traits and lower DON content resulted in the line MSB55 that had a DON content of 10.8 ppm compared to Sumai3 at 3.9 and AC Superb at 47.2.

### 3.6. Tritordeum

Resistance to FHB in durum wheat is very poor and variability for this trait in the tetraploid gene pool is very limited [28]. After screening some accessions of *Hordeum chilense* and detecting some variability for reaction to FHB, we crossed the better accessions to the durum cultivar Ma (which in our experience had better crossability than other durum cultivars). A chromosome preparation of the amphiploid is shown in **Figure 5**. Seven of the tritordeum amphiploids were evaluated for DON content and the results are shown in **Table 5**. Compared to Medora, a susceptible check, all seven amphiploids showed improved levels of DON content. Some of the values shown in **Table 5** are unrealistically low and should be

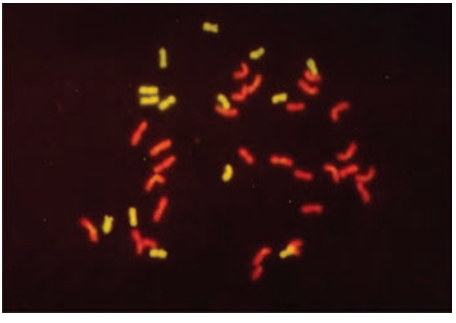

**Figure 5.** GISH pattern on a chromosome preparation from a Tritordeum (*Hordeum chilense* × *Triticum durum* amphiploid) showing 14 *Hordeum chilense* chromosomes (light color) and 28 durum chromosomes.

| Strain | DON content | |
|---|---|---|
| | Mean + SD | Range |
| HT-8 | 0.32 + 0.05 | 0.27–0.37 |
| HT-10 | 1.24 + 1.52 | 0–2.76 |
| HT-18 | 2.52 + 3.36 | 0–5.88 |
| HT-31 | 2.31 + 0.71 | 1.60–3.02 |
| HT-47 | 1.62 + 1.92 | 0.26–3.54 |
| HT-166 | 6.83 + 6.01 | 6.82–12.84 |
| HG-174 | 1.83 + 2.01 | 0.54–3.75 |
| Medora | 8.83 + 2.01 | 6.66–10.54 |

**Table 5.** DON content (ppm) of amphiploids between *Triticum turgidum* (AABB) and *Hordeum chilense* (HH).

re-evaluated; however, this appears to be a potential source of lower DON levels. A variety of FHB responses are shown in **Figure 6**, following the point inoculation of four tritordeum amphiploids plus a Roblin check.

The amphiploids show normal meiotic behaviour, are stable and perfectly fertile with perfect transmission of all chromosomes. There is no meiotic pairing between *Hordeum chilense* chromosomes and those of wheat. In order to induce pairing between homoeologous chromosomes, the amphiploid was crossed to the Capelli *Ph* mutant then backcrossed once to place the mutant in a homozygous recessive condition. The progeny resulting from the *Ph* mutant treatment were further backcrossed and advanced to the $BC_3F_4$ generation. Selection for reduced DON content and desirable agronomic traits was practiced during this procedure. Seventeen $BC_3F_4$



**Figure 6.** FHB symptoms on spikes of Tritordeum lines at 21 days after inoculation.

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lines were evaluated in the epiphytotic nursery, and results are shown in **Table 6**. The DON content in these lines ranged from 3.3 to 27.7 ppm compared to 19.1 for Strongfield the recurrent parent. Derivatives from this process appeared to have lower symptoms.

### 3.7. Triticale

Triticale, a wheat-rye amphiploid is used primarily as a feed grain worldwide, but has never reached its true potential. For a feed grain, DON content is a highly significant component. It has been shown that for each ppm of DON, feed consumption by monogastric animals decreases by 7.5%. Triticale was considered to be a major carbohydrate for bio-fuel production because of its high yields of biomass and tolerance to poor soils. Some consideration has been given for triticale to be used for ethanol production. However, it has been shown that the DON content in distiller's grains can be three lines as high as in the original grain. Therefore to fully realize the potential of triticale, its DON content must be reduced.

In general, triticale strains are notorious for poor FHB resistance. To put a wider perspective on this problem, we started by acquiring 371 strains of triticale from Plant Gene Resources of Canada (PGRC) to begin FHB testing. The testing was done in an epiphytotic nursery to evaluate Type II resistance. Visual rating of incidence and severity was done on the field plots and aliquots of seed ground for DON analysis. For the majority of the strains tested, the incidence and severity values exceeded 50%. Seven of the best strains were selected and shown in **Table 7**.

As shown in **Table 7**, the DON values of the seven strains ranged from 2.1 to 7.7 ppm, compared to Sumai3 at 1.2. The DON values in 2007 were low overall in that year. They were somewhat higher in 2008, ranging from 3.2 to 9.0.

AC Ultima was used a check triticale cultivar. It was a recently licensed cultivar in Canada and superior for most agronomic traits. Its DON content was 17.5 in 2007 and 16.0 in 2008.

The triticale strain TMP16315 was selected for further study. It was tested at numerous locations across Canada and proved to be stable in its reaction to FHB. Its pedigree is undefined, but believed to originate from a Polish gene pool.

A study was initiated to identify the QTL combining the FHB resistance/lower DON levels. Line TMP16315 was crossed to AC Ultima. A mapping population of 150 DH lines using microspore culture (Francois Eudes pc) method was produced from the F1 hybrid. The

| Generation | No. of lines | DON content (range) |
|---|---|---|
| $BC_3F_4$ | 17 | 3.3–27.7 |
| **Checks** | | |
| Strongfield | | 14.1 |
| AC Superb | | 16.3 |
| Roblin | | 17.2 |

**Table 6.** DON content (ppm) in progenies of intercrosses between *Tritordeum* (ABH) and *Capelli Ph* mutant followed by backcrosses to durum cultivar AC Strongfield.

| Line | 2007 | | | 2008 | | |
|---|---|---|---|---|---|---|
| | Incidence (%) | Severity (%) | DON (ppm) | Incidence (%) | Severity (%) | DON (ppm) |
| PI 355949 | 10.0 | 10.0 | 3.6 | 17.5 | 10.0 | 5.2 |
| PI 428748 | 10.0 | 10.0 | 3.2 | 10.0 | 5.0 | 4.4 |
| PI 428754 | 10.0 | 10.0 | 2.1 | 7.5 | 7.5 | 5.4 |
| PI 428814 | 30.0 | 20.0 | 7.7 | 20.0 | 15.0 | 9.0 |
| PI 428846 | 15.0 | 10.0 | 2.4 | 15.0 | 15.0 | 3.8 |
| CN 42948 | 20.0 | 20.0 | 5.0 | 10.0 | 10.0 | 4.5 |
| TMP 16315 | 15.0 | 15.0 | 4.1 | 20.0 | 10.0 | 5.2 |
| Sumai3 | 5.0 | 5.0 | 1.2 | 5.0 | 5.0 | 3.2 |
| AC Ultima | 85.0 | 50.0 | 17.5 | 45.0 | 45.0 | 16.0 |

**Table 7.** FHB symptoms and DON content of seven best resistant accessions of triticale and the checks Sumai3 and AC Ultima in the field nursery in 2007 and 2008.

mapping population in three replicates was grown at three locations in eastern Canada and data collected on incidence severity, FDK and DON content. The QTL for the various FHB related traits will be determined from these data.

### 3.8. Rye

In screening of numerous accessions of rye from numerous sources, we were not able to find any lines with even minor improvements in FHB resistance. There were reports of Brazilian land races of rye with improved levels of FHB resistance [29]. The lines were evaluated for resistance by plating on media containing from $10^{-3}$ to $10^{-6}$ M levels of DON [32] to evaluate their levels of tolerance to DON. Lines that showed no variable effects on a medium containing $10^{4}$–$10^{-3}$ M DON were considered to be resistant, whereas lines showing retarded growth on media containing $10^{-5}$–$10^{-6}$ M levels of DON were considered to be susceptible. As shown in **Table 8**, the landraces from Poula Frontin were susceptible to DON, whereas landraces from Lagoon Vermellia and Sao Paulo gave a resistant reaction. A number of the resistant lines were used as pollen parents on wheat cultivars Encruzilhada, Maringa, Max and NyuBay to produce octoploid amphiploids (as shown in **Table 9**).

| Accession | DON* | Reaction to DON |
|---|---|---|
| Rye from Poula Frontin, Parana | $10^{-5}$ | S |
| Rye from Lagoon Vermellia, Rio di Sul | $10^{-4}$ | R |
| White rye, Sao Paulo | $10^{-3}$ | R |

*Note*: *DON levels in culture media (ppm).

**Table 8.** Tolerance of Brazilian rye landraces to deoxynivalenol (DON) following plating on DON-containing media.

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| Hybrid combination | No. tested | DON* | Reaction to DON |
|---|---|---|---|
| Encruzilhada X 14A | 1 | $10^{-5}$ | S |
| Maringa X 26A | 3 | $10^{-5}$ | S |
| Max X 2C | 1 | $10^{-4}$ | R |
| | | $10^{-6}$–$10^{-5}$ | $10^{-3}$–$10^{-4}$ |
| | | S | R |
| NyuBay X Rye lines | 13 | 8 | 5 |

*Note*: *DON levels in culture media (ppm).

**Table 9.** Reaction of octoploid triticale strains to deoxynivalenol (DON) following plating on DON-containing media.

One amphiploid combination with wheat cultivar Max gave a resistant reaction and five amphiploids with NyuBay were also resistant by growing in a medium with a $10^{-3}$ level of DON [12].

## 4. Discussion

Reviews have been written showing the variability for FHB resistance in alien species [14, 20]. Less information is available on variation for DON content in alien species [18].

This review has shown that there are ranges for DON values in progenies obtained from several combinations of inter-specific/inter-generic hybrids. Although some of the data represent analyses from single years, there are indications of the potential of lowering the DON content by means of wide crosses.

In all cases, the screening of alien species parents was initially conducted by point or spray inoculation. The progenies in most cases were screened by several methods. Perhaps a more concerted effort needs to be employed to initially screen wild species for DON content.

Of various inoculation methods evaluated and methods of disease evaluation scored, including incidence, severity and FDK, it was found that DON evaluation gave the most reliable estimates of FHB resistance [14]. Considering that reducing DON content is the most important aspect of FHB resistance, DON evaluations should receive higher priority in future studies. Transgressive segregation for DON content was obtained in breeding populations of wheat and rye [30]. It was suggested that selection for lower DON content could be initiated as early as F3.

Transgressive segregation for DON content was observed in populations described in this paper, especially in progenies of crosses to *T. timopheevii* derivatives.

It has been shown by numerous studies beginning with Somers et al. [7] that DON content in wheat is controlled by distinctive QTL. That study also showed that minor QTL for DON content were present in the mapping population derived from Wuhan and NyuBay. These observations indicate that the potential exists for employing a combination of marker-assisted

selection plus a high selection pressure in an epiphytotic nursery to increase the overall resistance to DON accumulations as has been done for visual symptoms QTL [31].

Detailed screening of alien species collections for DON content should be done to the same extent as screening for visual symptoms of FHB resistance. Preliminary results shown in this paper indicated that such an approach would be warranted, to be followed by mapping of additional QTL. Such QTL would very likely be unique and would add to the toolbox of resources available for breeding for reduced DON content.

In order to effectively transfer FHB resistance from alien species to wheat, sufficiently large populations need to be grown. It has been showed in numerous studies that sufficient number of major and minor QTL need to be transferred to obtain effective resistance.

In conclusion, these studies have shown considerable variability for DON content can be obtained from species relatives alien to wheat. A focussed approach would be required to tag the various QTL and systematically integrate them into bread wheat. This is anticipated to be an incremental process. The end products would be crop cultivars that would be resistant to the head scab phase of FHB with the added benefit of lower DON accumulation, making them more suitable for feed and food.

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