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Does function follow form? Principal QTLs for *Fusarium* head blight (FHB) resistance are coincident with QTLs for inflorescence traits and plant height in a doubled-haploid population of barley

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Abstract *Fusarium* head blight (FHB), an important disease of barley in many areas of the world, causes losses in grain yield and quality. Deoxynivalenol (DON) mycotoxin residues, produced by the primary pathogen *Fusarium graminearum*, pose potential health risks. Barley producers may not be able to profitably market FHB-infected barley, even though it has a low DON level. Three types of FHB resistance have been described in wheat: Type I (penetration), Type II (spread), and Type III (mycotoxin degradation). We describe putative measures of these three types of resistance in barley. In wheat, the three resistance mechanisms show quantitative inheritance. Accordingly, to study FHB resistance in barley, we used quantitative trait locus (QTL) mapping to determine the number, genome location, and effects of QTLs associated with Type-I and -II resistance and the concentration of DON in the grain. We also mapped QTLs for plant height, heading date, and morphological attributes of the inflorescence (seeds per inflorescence, inflorescence density, and lateral floret size). QTL analyses were based on a mapping population of F_1-derived doubled-haploid (DH) lines from the cross of the two-rowed genotypes Gobernadora and CMB643, a linkage map constructed with RFLP marker loci, and field evaluations of the three types of FHB resistance performed in China, Mexico, and two environments in North Dakota, USA. Resistance QTLs were detected in six of the seven linkage groups. Alternate favorable alleles were found at the same loci when different inoculation techniques were used to measure Type-I resistance. The largest-effect resistance QTL (for Type-II resistance) was mapped in the centromeric region of chromosome 2. All but two of the resistance QTLs coincided with QTLs determining morphological attributes of the inflorescence and/or plant height. Additional experiments are needed to determine if these coincident QTLs are due to linkage or pleiotropy and to more clearly define the biological basis of the FHB resistance QTLs. Plant architecture should be considered in FHB resistance breeding efforts, particularly those directed at resistance QTL introgression and/or pyramiding.

Key words Barley · *Fusarium* head blight (FHB) · QTL mapping · Plant architecture

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Introduction

Fusarium head blight (FHB), caused by a number of *Fusarium* species (principally *Fusarium graminearum)* is an important disease of cereals in environments with prolonged wet climatic conditions from flowering

through the soft-dough stage of kernel development (Parry et al. 1995; McMullen et al. 1997; Miedaner 1997). The disease is of worldwide importance. An example of its impact is the upper Midwest region of the United States where, over the past 6 years, FHB has devastated wheat and barley production (McMullen et al. 1997; Steffenson 1998). Yield losses are due to floret sterility and the formation of shriveled, low test-weight kernels (Barr et al. 1996; Dill-Macky 1996; McMullen et al. 1997; Steffenson 1998). Grain-quality losses are often due to the presence of deoxynivalenol (DON), a trichothecene mycotoxin produced primarily by *F. graminearum*. This family of toxins can be harmful to human and animal health (Desjardins et al. 1996). Even at low DON levels, FHB-infected grain can have negative impacts on malting, brewing, feed, and food quality (Schwarz and Beattie 1995; Vivar et al. 1997). As a consequence, barley producers may not be able to profitably market FHB-infected grain. For example, major buyers of barley in the upper Midwest of the USA established very low tolerances for DON residues. This has contributed to a negative economic climate for barley production in the region. A better understanding of the genetic basis of FHB resistance would be a benefit to researchers throughout the world who are working to develop FHB-resistant varieties adapted to their local conditions.

Breeding for FHB resistance has been difficult, due to the multiple components of resistance, limited understanding of the genetic basis of the various forms of resistance, genotype×environment interaction, and the high cost of phenotyping (Wiersma et al. 1996; McMullen et al. 1997). In wheat, where FHB resistance has been more extensively studied than in barley, FHB resistance is classified as Type I, Type II and Type III (Parry et al. 1995). Type I describes resistance to initial infection (penetration). Type II describes resistance against pathogen spread from the point of infection (Schroeder and Christensen 1963). Different artificial inoculation techniques have been developed to determine Type-I and Type-II resistance in wheat (Bai and Shaner 1994; Miedaner 1997). Type-III resistance was defined as the ability to prevent DON synthesis, or to promote its degradation (Miller and Arnison 1986). The concentration of DON in the grain is of particular importance, due to concerns regarding the effects of the metabolite on human health and the role of the toxin in pathogenesis (Snijders and Krechting 1992; Bai and Shaner 1994; Mesterhazy 1995; Parry et al. 1995; Dill-Macky 1996; Miedaner 1997). FHB resistance is quantitatively inherited (Bai and Shaner 1994; Mesterhazy 1995; Miedaner 1997). Plant architecture and development traits, e.g., inflorescence structure, maturity and plant height, are associated with FHB resistance in wheat (Mesterhazy 1995; Miedaner 1997) and barley (Steffenson 1998; Takeda 1990). Although sources of resistance to FHB are available in barley, most of them are agronomically unacceptable, have poor malting quality, and are two-rowed (Takeda and Heta 1989; Dill-Macky 1996).

"Two-rowed" and "six-rowed" refer to the number of fertile florets per rachis node. The character is determined by the *V* and *I* loci on chromosomes 2 and 4, respectively. Commercial two-rowed and six-rowed varieties are *VVii* and *vvII*, respectively (Hockett and Nilan 1985). While the row-type character is simply inherited, the two-rowed and six-rowed barley types represent two of the principal germplasm groups in cultivated barley (Kjaer and Jensen 1996). A factor complicating the development of FHB-resistant varieties for the upper Midwest of the USA is the preference of the North American brewing industry for six-rowed malting barley.

Quantitative trait locus (QTL) mapping is a tool for investigating traits showing complex inheritance, such as FHB. QTLs determining both qualitative and quantitative disease resistance have been mapped in barley (Chen et al. 1994; Graner 1996; Steffenson et al. 1996). Marker-QTL associations are useful for introgression and the pyramiding of resistance QTL alleles (Tanksley and Nelson 1996; Toojinda et al. 1998). Information regarding the number, genome location, and effects of genes determining the multiple components of resistance to FHB, and the relationships with loci determining morphological and developmental traits, should expedite the development of FHB-resistant varieties. The objective of the present research was to describe QTLs determining FHB resistance and the accumulation of DON in the grain.

Materials and methods

Plant materials and linkage-map construction

A population of 144 F_1-derived doubled-haploid (DH) lines was developed from the cross of Gobernadora×CMB643 using the *Hordeum bulbosum* method (Chen and Hayes 1989). Both parents are two-rowed genotypes developed by the ICARDA/CIMMYT barley breeding program based in Mexico. Gobernadora is grown extensively in the lower Yangtze basin of China under the name "Zhenmai-1". The large-scale production of Gobernadora in this region is due to its FHB resistance (Vivar et al. 1997). CMB643, a germplasm line with the pedigree *Shyri/Gloria//Copal///Shyri/Grit*, has shown different reactions to FHB in different environments. In China, it was classified as "moderately resistant", while in Mexico it showed intermediate levels of FHB resistance (Vivar et al. 1997). The DH population was genotyped with 97 RFLP markers, following standard protocols (Kleinhofs et al. 1993). Locus designations employ the nomenclature used in previous North American Barley Genome Mapping Project (NABGMP) maps (e.g., Kleinhofs et al. 1993). G-Mendel version 3.0 (Holloway and Knapp 1994) was used for linkage-map construction, following the map-construction strategies described by Hayes et al. (1997). The final linkage groups were calculated using a maximum recombination frequency (rmax) of 0.30 and a LOD score of 3.0. The assignment of linkage groups to their corresponding chromosomes was according to Kleinhofs et al. (1993) and Kasha et al. (1995). Recombination values were converted to genetic distances (cM) using the Kosambi function. The χ^2 statistic was used to test for deviations from the expected 1:1 segregation ratio.

Disease evaluations

In this report, we will use "Type I" to refer to resistance assessed under field conditions in environments where there was very little

kernel-to-kernel spread (especially vertical spread) within an inflorescence. As described below, the Type-I resistance phenotype was measured differently in Toluca, Mexico, as compared to the other test environments. We will use the term "Type II" to refer to resistance assessed under field conditions at Toluca, Mexico, following an inoculation protocol described below.

Over a 2-year period (1996 and 1997), the DH population and the parents were grown in a total of four field experiments: Fargo and Langdon, North Dakota, USA; Toluca, Mexico; and Shanghai, China. At Toluca and Shanghai, a randomized complete block design with two replications was employed. DH lines 1–98 were phenotyped. A single replicate was used at both Fargo and Langdon, and DH lines 1–144 were phenotyped at each location. At all locations, artificial inoculation was performed using local isolates of *F. graminearum.*

At Fargo, Langdon and Shanghai, inoculum was prepared using a method modified from Dodman and Wildermuth (1987). The isolates of *F. graminearum* were grown on acidified potato dextrose agar (APDA) at 25°C for 7 days in complete darkness and 2 days under fluorescent light. Agar plugs containing the fungal cultures were added to sterilized barley and maize grains and incubated at 25°C for 7–14 days in complete darkness. At Fargo and Langdon, inoculations were made by scattering infected grain on the soil surface of the nurseries at weekly intervals for 7 consecutive weeks, beginning 10 days before the first inflorescence emerged. Infected kernels were applied once at Shanghai, 10 days before the first inflorescence emerged. Nurseries were irrigated with overhead misters for 20 min during the evening to facilitate spread of the disease. Disease assessments were made on 10–20 randomly selected inflorescences of each line at the mid-dough stage of kernel development. The severity of FHB (%, putative Type I) was determined by dividing the total number of infected kernels by the total number of kernels (Prom et al. 1997). The concentration of DON (ppm) was assessed on two random 3-g grain samples of each line from each plot following the methodology of Tacke and Casper (1996). DON analyses were performed at the Department of Cereal Science, North Dakota State University, on samples from Fargo and Langdon, courtesy of Dr. P. Schwarz. The average of the two sample values was used for QTL analysis. Heading date (days from planting to anthesis) and plant height (cm) were recorded at Fargo and Langdon.

Two planting dates were used at Toluca in order to inoculate inflorescences of each DH line at similar growth stages. Data from the two planting dates were pooled to provide a single data-set for analysis. Inoculations for measuring putative Type-I and Type-II resistance were performed at anthesis. For determining Type-I resistance, a conidial suspension containing 50 000 spores per ml of *F. graminearum* was sprayed on seven flowering heads in each plot. In the absence of natural precipitation, overhead irrigation was applied daily for 30 min to encourage disease development. Fifteen days after inoculation, the number of infected kernels was determined. FHB severity (%, Type I) was calculated as the average percentage of infected kernels per inflorescence. For determining Type-II resistance, a tiny cotton tuft soaked in a suspension of similar spore concentration was placed in a floret of a central spikelet, as described by Van Ginkel et al. (1996). Five heads of each DH line were inoculated per replication. The inoculated heads were then covered with a glassine bag to protect them from allo-infection. Thirty days after inoculation, the number of infected kernels and the total number of spikelets per inflorescence were counted. Type-II disease severity (%) was calculated as the average percentage of infected kernels per inflorescence.

Inflorescence density [(average distance between seeds within the inflorescence (mm)] and lateral floret size [scored as small (1); intermediate (2) or large (3)] were determined on a reference set of dried inflorescences produced under greenhouse conditions at Corvallis, Oregon, USA. The data for QTL analysis were based on an average of three inflorescences per DH line. The inflorescences were obtained from a single grow-out of the population.

Statistical analysis and QTL mapping

Analyses of variance were conducted using SAS software (SAS Institute 1989). Heritability estimates (on an entry mean basis) was calculated using the formula $h^2=\sigma_g^2/(\sigma_g^2+\sigma_e^2/r)$, where σ_g^2 and σ_e^2 are the genotypic and error variance, respectively; and r is the number of replications. Variance components were computed by equating mean squares to their expectations. Pearson correlation coefficients were calculated to determine phenotypic correlations among traits.

QTL analysis was performed with the software package MQTL (Tinker and Mather 1995a, b). The data from each experiment were analyzed separately. Both simple interval mapping (SIM) and simplified composite interval mapping (sCIM) techniques were used for QTL detection. Each data set was analyzed with 1000 permutations, a 5-cM walking speed, and a Type-I error rate of 5%. The significance threshold was used to declare the presence of a QTL. Fifteen background markers were specified for use as cofactors in sCIM, based on their position and a *priori* information from SIM mapping. Interaction among QTL main effects detected via SIM and sCIM was tested by linear regression, using a comparison-wise significance level of 0.01. Single marker loci closest to QTL peaks were considered explanatory variables. Measures of Type-I and Type-II resistance, as well as DON concentration, were considered as response variables.

Results and discussion

Map construction

The 97-marker base map (Fig. 1) comprises a total linkage distance of 1306 cM. The average two-locus interval is 13.5 cM. Marker orders and distances are consistent with published maps (Keinhofs et al. 1993; Kasha et al. 1995; Hayes et al. 1997). Of the 97 marker loci, 25 showed segregation ratios that differed significantly from the expected 1:1 ($P \leq 0.05$). Gobernadora alleles were over-represented at 15 loci and CMB643 alleles were over-represented at ten loci. On the short arm of chromosome 2, there was segregation distortion in favor of Gobernadora alleles at eight contiguous marker loci, beginning with *MWG844A* and ending with *Pox*. CMB643 alleles were favored at three contiguous loci on chromosome 3 (*PSR156–MWG571A*) and at three loci on the short arm of chromosome 4 (*Ole1–CDO542*). Otherwise, loci showing segregation distortion occurred individually or were bracketed by loci showing normal segregation. Segregation distortion is not a common phenomenon in DH populations derived by the *H. bulbosum* technique (Hayes et al. 1997). It is more common in barley DH mapping populations of androgenetic origin (Graner et al. 1991; Zivy et al. 1992; Devaux et al. 1995). Segregation distortion can be caused by a variety of physiological or genetic factors that lend selective advantage to particular gametes, or the selective fertilization of particular genotypes (Xu et al. 1997). The consequences of segregation distortion on linkage-map construction and QTL detection have not been thoroughly investigated (Holloway and Knapp 1994). The *CDO542* locus on chromosome 4 was the only marker showing segregation distortion that coincided with a QTL peak. Loci showing significant distortion fell within QTL significance intervals on chromosomes 3 and 5. There was

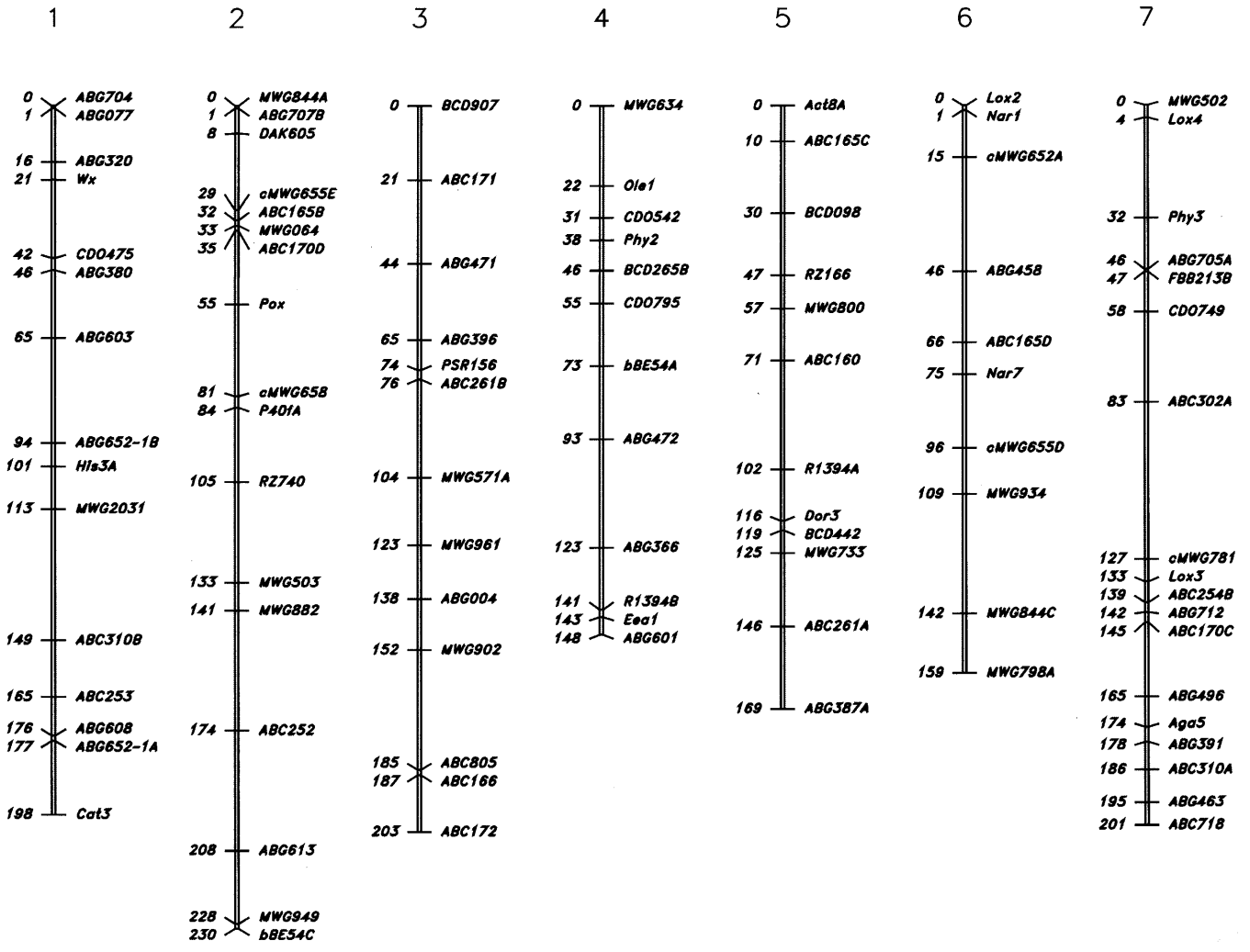


Fig. 1 Linkage map with 97 RFLP markers constructed from 144 F_1-derived DH lines from the cross of Gobernadora×CMB643. Distances are in Kosambi cM units

no pattern of QTL-allele phase and segregation distortion.

Phenotypes

Phenotypic frequency distributions (Fig. 2) support the quantitative inheritance of all FHB resistance, plant architecture, and developmental characters. There were large differences in disease severity between environments. For example, maximum Type-I severities ranged from 15% at Fargo to 48% at Langdon. Maximum DON concentrations were 25 and 80 ppm at Fargo and Langdon, respectively. Using the parental values as standards, there were more susceptible transgressive segregants than resistant transgressive segregants for the three types of FHB resistance. This suggests that unique configurations of alleles at multiple loci are required for resistance. In some cases, however, the distributions were skewed to the lower disease level. A similar phenomenon was reported in the progeny of two FHB-resistant wheat genotypes (Van Ginkel et al. 1996). These authors attributed the phenomenon to the fact that the phenotypic values of the resistant genotypes could not be distinguished as easily as those of the susceptible genotypes. In the Gobernadora×CMB643 population, there are DH lines that combine low Type-I severity, low Type-II severity and a low DON concentration. There were both high and low transgressive segregants for plant height, heading-date, and the inflorescence characters. Heritability estimates, on an entry mean basis, for FHB resistance traits ranged from 0.50 to 0.81 depending on environment, type of resistance, and inoculation procedure (Fig. 2). These heritability values are higher than those reported by Takeda (1990), who used a different germplasm and the "cut-spike" method of assessment.

All significant correlations among Types-I and -II resistance and DON concentration were positive (Table 1). All non-significant correlations were related to Type-I resistance at Toluca, where a conidial spore suspension was applied directly to the inflorescence. When Type-I resistance was measured based on inoculum applied to the soil surface, there were significant positive correla-

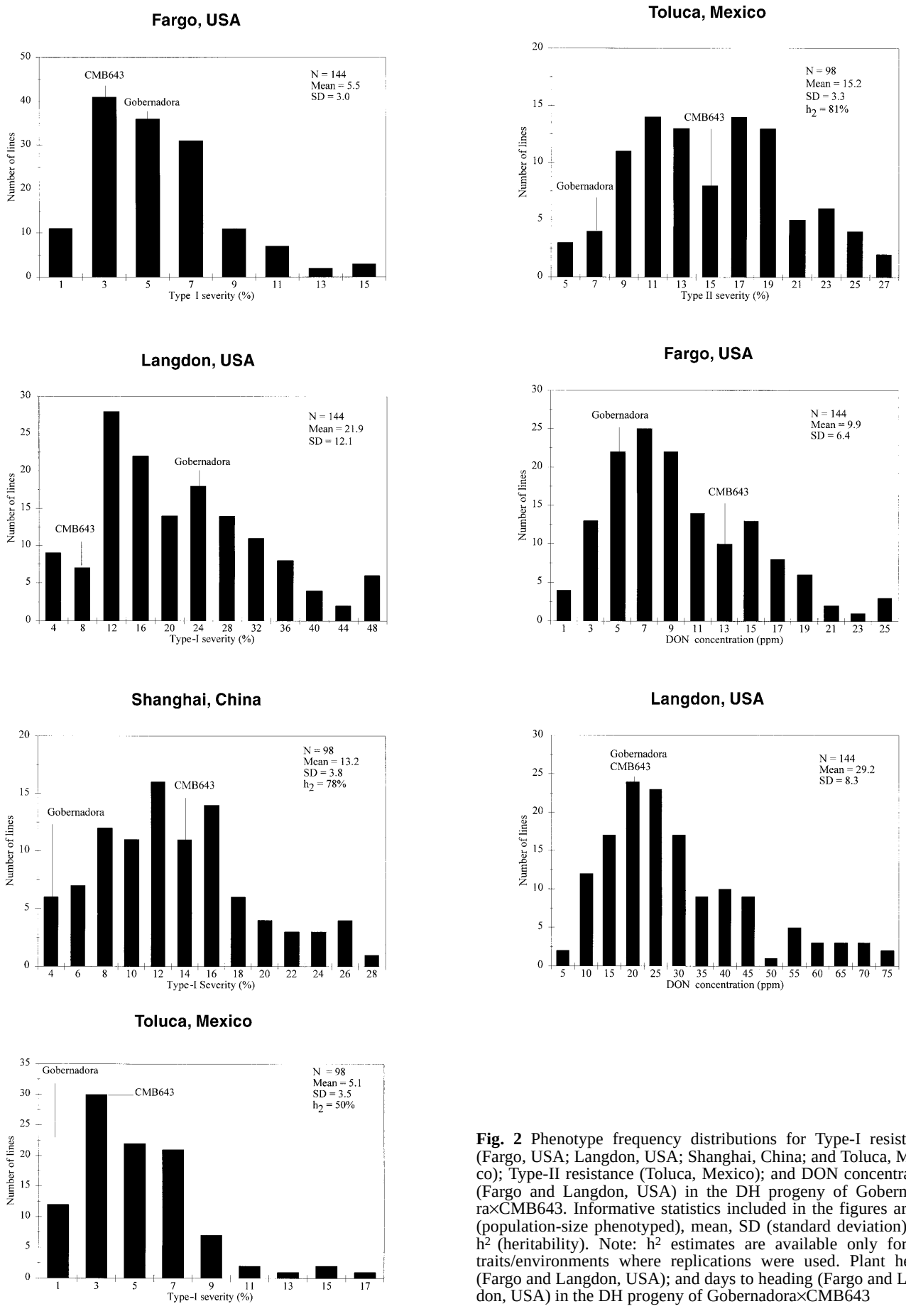


Fig. 2 Phenotype frequency distributions for Type-I resistance (Fargo, USA; Langdon, USA; Shanghai, China; and Toluca, Mexico); Type-II resistance (Toluca, Mexico); and DON concentration (Fargo and Langdon, USA) in the DH progeny of Gobernadora×CMB643. Informative statistics included in the figures are: N (population-size phenotyped), mean, SD (standard deviation) and h^2 (heritability). Note: h^2 estimates are available only for the traits/environments where replications were used. Plant height (Fargo and Langdon, USA); and days to heading (Fargo and Langdon, USA) in the DH progeny of Gobernadora×CMB643

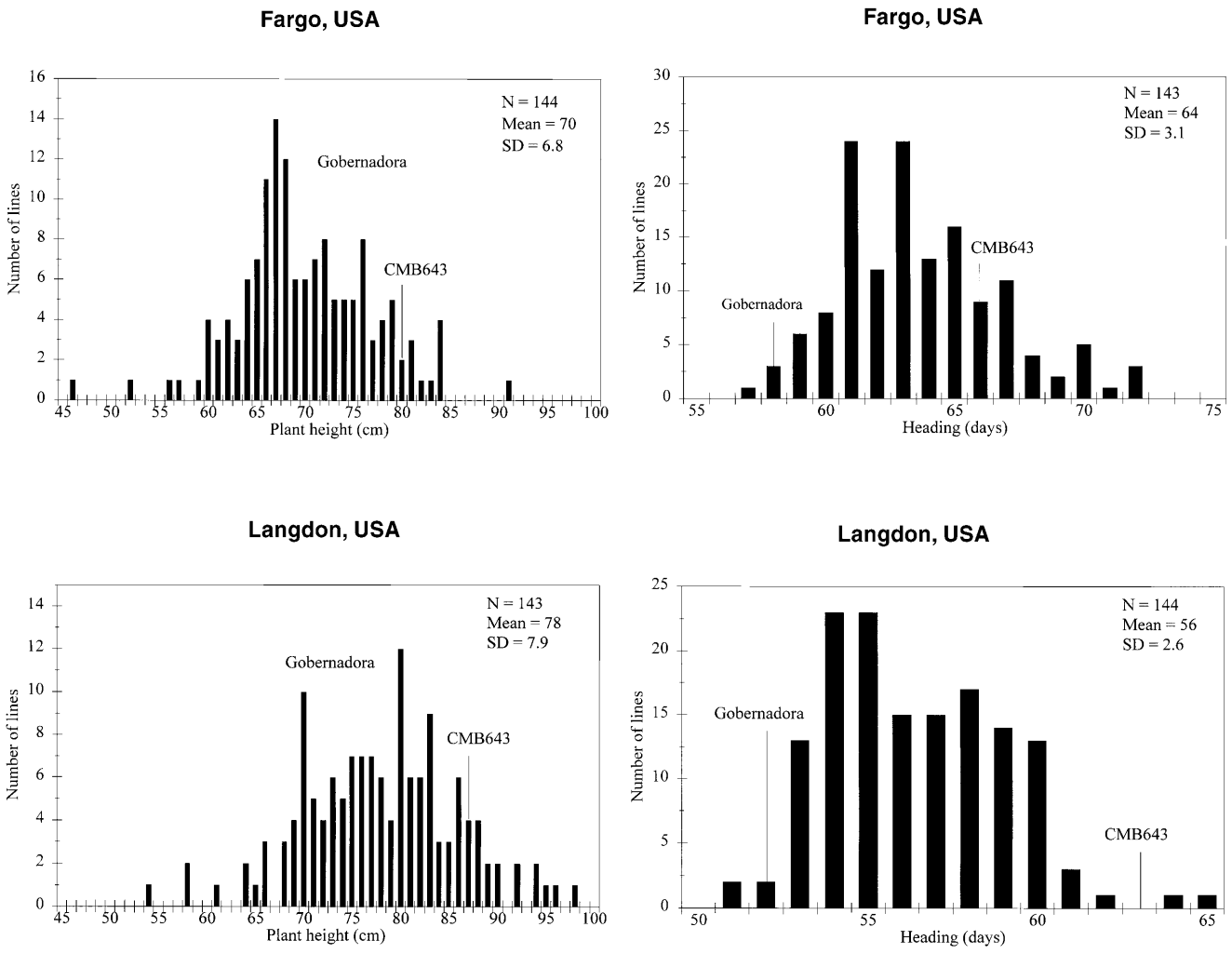


Fig. 2 Legend to Fig. 2 see page 1225

tions with Type-II resistance and DON concentration. Inoculation technique is clearly important in measuring Type-I resistance and the choice of inoculation technique will have an impact on measures of association among resistance phenotypes. The uniqueness of the Type-I (Toluca) resistance phenotype is apparent in the pattern of correlations between FHB resistance traits, heading-date, plant height, and inflorescence morphology characters (Table 2). Correlations between Type-I resistance (Toluca) and other traits were non-significant. The various measures of FHB resistance, with the exception of Type I (as measured at Toluca), were associated with taller plants, more seeds per inflorescence, low-density inflorescences, and small lateral florets. Heading-date was not associated with resistance. These correlations support the argument that plant architecture attributes contribute to lower disease levels (Miedaner 1997).

QTLs

All QTLs were detected with both SIM and sCIM. Significant QTLs, and coincident QTLs that approached, but did not reach, the significance threshold are shown in Fig. 3. Single-locus effects of all QTLs significant in the MQTL analysis, and QTLs that were non-significant but showed a coincident trend with a significant QTL, are described in terms of position, effect, and favorable allele phase in Table 3. In all cases, except for small-effect QTLs on chromosomes 5 and 6, FHB-resistance QTLs were coincident with QTLs for plant height, seeds per inflorescence, inflorescence density, and/or lateral floret size.

The largest-effect resistance QTL detected in this experiment was for Type-II resistance (Toluca) on chromosome 2. There were significant coincident QTLs for DON concentration (Langdon), seeds per inflorescence (Shanghai), and lateral floret size. There were coincident, but non-significant QTL peaks for Type-I resistance (Shanghai) and DON concentration (Fargo). In all cases, CMB643 contributed resistance alleles and Gobernadora had the larger-value allele for lateral floret size. On chromosome 1, the only QTL significant in the MQTL analysis was for Type-I resistance (Fargo). This QTL had a small main effect, accounting for 4% of the variation in phenotypic expression. At this same QTL there were coincident, but non-significant, QTLs peaks for Type-II resistance (Toluca) and inflorescence density.

Table 1 Pearson phenotypic correlation coefficients for Type-I and Type-II resistance and DON concentration across environments and/or types of resistance in the Gobernadora×CMB643 DH population

Resistance type	Type I Toluca, Mexico	Type I Shanghai, China	Type I Fargo, USA	Type I Langdon, USA	Type II Toluca, Mexico	DON conc. Fargo, USA
Type I Shanghai, China	0.22					
Type I Fargo, USA	0.32	0.35				
Type I Langdon, USA	ns[a]	0.30	0.60			
Type II Toluca, Mexico	ns	0.33	0.33	0.45		
DON Fargo, USA	ns	0.44	0.40	0.40	0.30	
DON conc. Langdon, USA	ns	0.37	0.39	0.67	0.49	0.53

[a] ns=not significantly different from zero at the 0.01 probability level

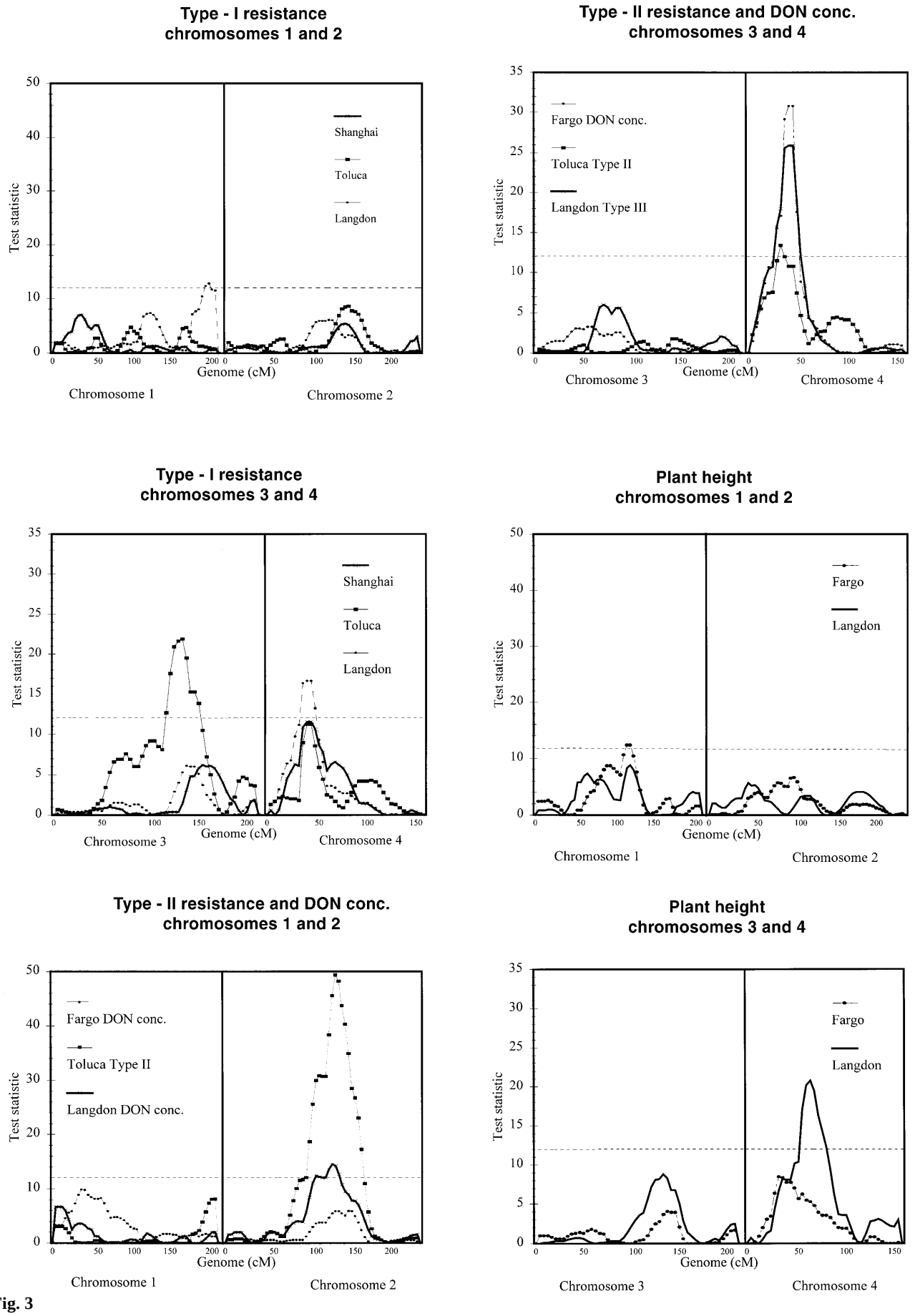
Table 2 Pearson phenotypic correlation coefficients among Type-I and Type-II resistance, DON concentration, inflorescence architecture traits, and plant height across environments in the Gobernadora×CMB643 DH population. **Note**: all phenotypic correlations of FHB resistance traits with heading-date at Fargo and Langdon were non-significant and are not included in this table

Item	Seeds/ inflorescence Toluca, Mexico	Seeds/ inflorescence Shanghai, China	Seeds/ inflorescence Fargo, USA	Seeds/ inflorescence Langdon, USA	Lateral floret size, Oregon, USA	Inflorescence density Oregon, USA	Plant Height Fargo, USA	Plant Height Langdon, USA
Type I Toluca, Mexico	ns[a]	ns	ns	ns	ns	ns	ns	ns
Type I Shanghai, China	–0.38	–0.41	–0.33	–0.25	0.34	0.25	–0.29	ns
Type I Fargo, USA	ns	ns	–0.39	ns	ns	0.28	–0.50	–0.56
Type I Langdon, USA	–0.27	–0.38	–0.52	–0.50	0.33	ns	–0.67	–0.66
Type II Toluca, USA	–0.44	–0.51	–0.44	–0.48	0.63	ns	–0.33	ns
DON Fargo, USA	–0.29	–0.37	–0.41	–0.31	0.41	ns	–0.31	ns
DON Langdon, USA	–0.44	–0.53	–0.51	–0.39	0.54	ns	–0.55	–0.34

[a] ns=not significantly different from zero at the 0.01 probability level

Individual marker loci on the short arm of chromosome 1 – *Wx*, *CDO475*, *ABG380*, and *ABG603* – were significant in single-locus regressions for multiple measures of FHB resistance, heading-date, and plant height. On chromosome 3, a significant QTL for Type-I resistance (Toluca) coincided with non-significant QTL peaks for plant height at Langdon and Fargo and poorly defined peaks for inflorescence density and lateral floret size. In single-locus regression analysis, the *ABG396* and *ABC261b* loci, which are 11-cM apart, accounted for the largest proportion of phenotypic variation in Type-I resistance (Toluca), DON concentration (Langdon), seeds per inflorescence (Shanghai) and inflorescence density. On chromosome 4, significant QTLs for all measures of FHB resistance coincided with significant QTLs for plant height, seeds per inflorescence, and lateral floret size.

Coincident QTLs support the observed pattern of phenotypic correlations, although the latter may also represent the net effects of multiple loci. For example, favorable allele phases for Type-I (Toluca) resistance QTLs are opposite to those for coincident resistance QTLs on chromosomes 1 and 4. All non-significant correlations involved Type-I resistance (Toluca). Measures of FHB resistance other than Type I (Toluca) were positively correlated: at all QTLs except those on chromosomes 5 and 6, coincident QTLs had consistent allele phases. Coincident QTLs for Type-II resistance (Toluca) and DON con-


Type - I resistance
chromosomes 1 and 2
Shanghai
Toluca
Langdon
Test statistic
Genome (cM)
Chromosome 1
Chromosome 2
Type - II resistance and DON conc.
chromosomes 3 and 4
Fargo DON conc.
Toluca Type II
Langdon Type III
Test statistic
Genome (cM)
Chromosome 3
Chromosome 4
Type - I resistance
chromosomes 3 and 4
Shanghai
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Langdon
Test statistic
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Chromosome 3
Chromosome 4
Plant height
chromosomes 1 and 2
Fargo
Langdon
Test statistic
Genome (cM)
Chromosome 1
Chromosome 2
Type - II resistance and DON conc.
chromosomes 1 and 2
Fargo DON conc.
Toluca Type II
Langdon DON conc.
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Fig. 3

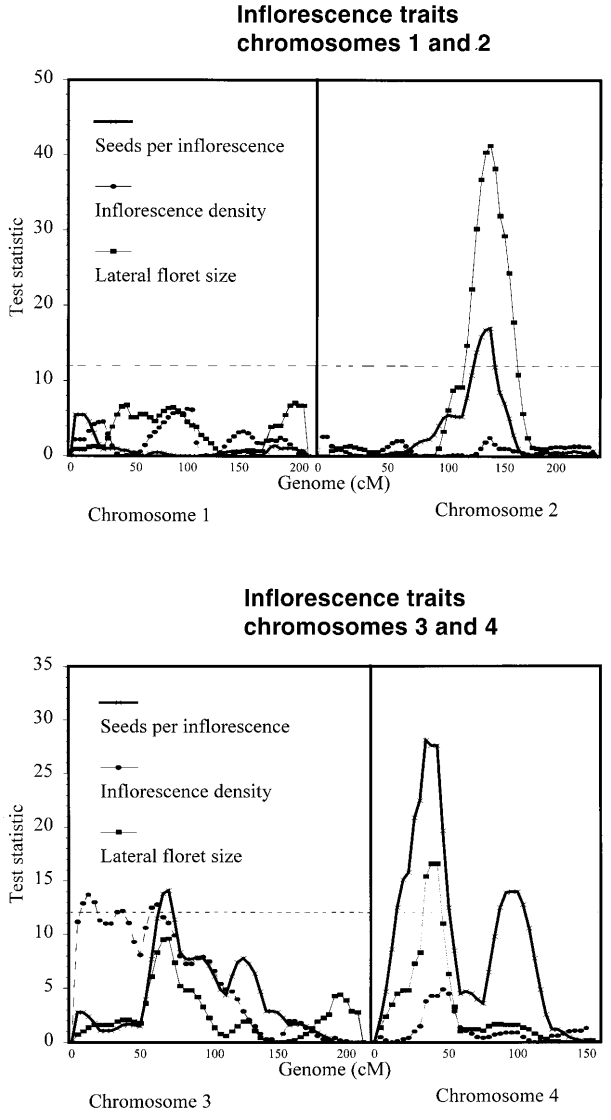

Fig. 3 Test statistics from the simple interval mapping (SIM) analysis of FHB resistance and inflorescence traits in the DH progeny of Gobernadora×CMB643. Test statistics above the horizontal bar indicate significance at an experiment-wise error rate of 0.05. The SIM plots are shown in a plus to minus chromosome arm orientation. The x-axis units correspond to the cM distances shown on the genome map in Fig. 1

centration (Fargo and Langdon) on chromosome 2 may be a contributing factor to the positive correlations between these traits. The negative correlations between lateral floret size and these same measures of resistance may be attributable, in part, to the larger-value allele for lateral floret size contributed by Gobernadora.

Coincident QTLs may be due to linkage or pleiotropy. At the level of resolution afforded by this mapping population, the two cannot be distinguished. Coincidence of resistance QTLs and QTLs for developmental and plant architecture traits could be due to the latter determining the former. It is intuitively appealing that taller plants with longer, more open, inflorescences and insignificant lateral florets would provide an environment less favorable for fungal growth and development. Alternatively, coupling relationships between resistance genes and genes determining components of plant architecture that limit disease development could be an evolutionarily advantageous strategy. Additional experiments with alternative genetic stocks will be necessary to distinguish between linkage and pleiotropy. These experiments will be necessary before attempting to pyramid and/or introgress resistance QTL alleles. An immediate implication is that plant architectural traits need to be considered in FHB resistance breeding.

With the exception of the chromosome-2 QTL for Type-II resistance, individual FHB resistance QTL main effects accounted for, on average, less than 10% of the variation in trait expression. However, heritability values were high, ranging from 50 to 81%. Lande and Thompson (1990) argued, on theoretical grounds, that the proportion of additive genetic variance explained by QTLs is inversely related to the product Nh^2, where N is the population size. In this population, Type-I (Shanghai) and Type-II resistance had similar heritabilities. A single QTL on chromosome 2 accounted for 33% of the variation in trait expression in Type-II resistance, while of five QTLs detected for Type-I resistance at Shanghai, main effects averaged less than 10% of the phenotypic variance explained. Multi-locus R^2 values for FHB resistance traits ranged from 8 to 60%. The genetic variance that remains unaccounted for may be due to QTL×QTL interactions. Two-way interactions between loci with significant main-effects were significant for the FHB resistance phenotypes, except for Type-I resistance in the North Dakota environments, and DON concentration at Fargo (Table 3). The R^2 values for these interactions were nearly half the value of the multi-locus R^2 values, except for Type-II resistance, where the main-effect multi-locus R^2 was high (60%). In all cases, the sign of the epistatic interaction agreed with the sign of the main-effect QTLs.

Higher-order QTL×QTL interactions are probably important in FHB resistance in this population. Estimation of these interactions would require a substantially larger population size, which would also increase the power of QTL main-effect detection (Melchinger et al. 1998). Furthermore, as these authors point out, QTL effects are often overestimated when they are estimated from the same data used for QTL detection and mapping. However, due to the high cost of phenotyping (Steffenson 1998), increasing population size for FHB resistance QTL mapping remains a challenge.

This study represents an initial step in understanding some of the possible factors contributing to lower levels of FHB severity and DON accumulation in barley. Additional finer-structure analysis will be required to determine if coincident plant architecture/development traits and FHB resistance QTLs are due to linkage or pleiotropy. Controlled environment FHB resistance phenotyping could be of assistance in separating factors – such as plant architecture, heading-date, and effects of other diseases – that can confound FHB development in field ex-

Table 3 Summary of quantitative trait loci (QTLs) detected in the Gobernadora×CMB643 DH population. "C" and "G" in the "Effects" column refer to the parent contributing the resistance allele (in the case of the FHB-related traits) and the larger-value allele (in the case of the plant architecture traits). The markers shown are those that were closest to the QTL peak and had the highest R^2 value

Environment	Phenotype	Chromosome 1			Chromosome 2			Chromosome 3			Chromosome 4		
		Closest marker	Effect	R^2 (%)	Closest marker	Effect	R^2 (%)	Closest marker	Effect	R^2 (%)	Closest marker	Effect	R^2 (%)
Shanghai, China	Type-I resistance	*ABG380*	2.31 (G)	4	*MWG503*	2.86 (C)	4	*ABG004*	1.91 (C)	4	*Phy2*	3.46 (G)	10
Toluca, Mexico	Type-I resistance	*CDO475*	1.13 (C)	6				*ABG004*	2.27 (C)	16	*Phy2*	1.62 (C)	7
								ABG396	1.26 (C)	9			
Fargo, USA	Type-I resistance	*Wx*	1.02 (G)	4				*MWG902*	1.20 (C)	4	*CDO542*	1.38 (G)	5
Langdon, USA	Type-I resistance							*ABG004*	4.99 (C)	3	*CDO542*	5.57 (G)	4
Toluca, Mexico	Type-II resistance				*MWG503*	4.36 (C)	33				*CDO542*	4.18 (G)	12
Fargo, USA	DON concentration	*Wx*	1.19 (G)	4							*Phy2*	4.68 (G)	13
Langdon, USA	DON concentration				*RZ470*	10.94 (C)	8	*ABC261B*	7.28 (G)	5	*Phy2*	10.42 (G)	7
Toluca, Mexico	Seeds/inflorescence							*MWG961*	1.68 (G)	8	*Phy2*	2.13 (G)	18
Shanghai, China	Seeds/inflorescence				*MWG503*	2.52 (C)	22	*ABG004*	1.12 (G)	3	*Phy2*	1.90 (G)	14
								ABG396	1.04 (G)	11			
Langdon, USA	Seeds/inflorescence										*CDO542*	1.27 (G)	11
Fargo, USA	Seeds/inflorescence										*Phy2*	1.26 (G)	10
Fargo, USA	Days to heading	*CDO475*	3.25 (C)	26									
Langdon, USA	Days to heading	*CDO475*	2.32 (C)	19									
Fargo, USA	Plant height	*MWG2031*	3.38 (C)	4				*ABG004*	2.29 (C)	3	*CDO542*	3.10 (G)	4
Langdon, USA	Plant height	*ABG603*	2.97 (C)	2				*ABG004*	2.67 (C)	2	*CDO795*	4.69 (G)	9
Corvallis, USA	Lateral floret size				*MWG503*	0.94 (G)	34				*Phy2*	0.62 (C)	14
Corvallis, USA	Inflorescence density							*BCD907*	0.25 (C)	10			

Table 3 Continued

Environment	Phenotype	Chromosome 5			Chromosome 6					
		Closest marker	Effect	R^2 (%)	Closest marker	Effect	R^2 (%)	Multi-locus R^2 (%)	Two-locus interaction	R^2 (%)
Shanghai, China	Type-I resistance				*ABC165D*	3.37 (G)	4	26	*Phy2×ABG380*	13
Toluca, Mexico	Type-I resistance							35	*CDO475×ABG396*	14
Fargo, USA	Type-I resistance							11		
Langdon, USA	Type-I resistance							8		
Toluca, Mexico	Type-II resistance	*MWG733*	1.16 (G)	7				60	*MWG503×CDO542*	4
Fargo, USA	DON concentration							14		
Langdon, USA	DON concentration	*MWG733*	6.54 (G)	5				29	*ABG261B×MWG733*	13
Toluca, Mexico	Seeds/inflorescence							24		
Shanghai, China	Seeds/inflorescence							41		
Langdon, USA	Seeds/inflorescence							11		
Fargo, USA	Seeds/inflorescence									
Fargo, USA	Days to heading							26		
Langdon, USA	Days to heading							19		
Fargo, USA	Plant height				*MWG884c*	2.67 (C)	3	14		
Langdon, USA	Plant height							13		
Corvallis, USA	Lateral floret size							47		
Corvallis,USA	Inflorescence density							20		

periments. Such experiments will also be useful in determining if the terms “Type I” and “Type II”, as defined for wheat, are appropriate for barley. The inoculation technique was an important factor in assessing Type-I resistance. Many of the same loci are involved in determining Type-I resistance as measured by the two different inoculation procedures. However, there were favorable allele phase changes, depending on the mode of inoculation. The biological basis of this change in allele phase needs to be determined. Type-II resistance was the most highly heritable FHB resistance phenotype and the one for which the largest effect QTLs were detected. However Type-II resistance is not of epidemiological importance in all environments. For example, in the upper Midwest USA, high levels of Type-I resistance are required in malting barley cultivars in order to prevent DON levels from exceeding the stringent thresholds established by the malting and brewing industries (B. Steffenson, unpublished data). An understanding of the biology underlying coincident plant architecture and FHB resistance QTLs will contribute to the efficient development of FHB-resistant barley varieties.

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