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Quantitative and qualitative trait loci affecting host-plant response to *Exserohilum turcicum* in maize (*Zea mays* L.)

P. J. Freymark[1], M. Lee[1], W. L. Woodman[1], C. A. Martinson[2]

[1] Department of Agronomy, Iowa State University, Ames, IA 50011, USA
[2] Department of Plant Pathology, Iowa State University, Ames, IA 50011, USA

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Abstract. Molecular markers at 103 loci were used to identify the location of quantitative sources of resistance to *Exserohilum turcicum* in 150 $F_{2:3}$ lines of a B52/Mo17 maize population. Host-plant response was measured in terms of the average number of lesions per leaf, the average percent leaf tissue diseased (severity), and the average size of lesions. The location of quantitative trait loci were compared with three loci having known qualitative effects, namely *Ht1*, *Ht2* and *bx1*. Chromosomal regions containing the *Ht1* and *Ht2* loci showed a small contribution in determining lesion size, even though alleles with dominant, qualitative effects at these loci have never been reported in either inbred parent. Similar effects were not observed for the number of lesions or for disease severity. Likewise, some contribution was observed for chromosomal regions encompassing the *bx1* locus in determining lesion size but not the number of lesions or disease severity. Overall the contribution of loci in the vicinity of *Ht1*, *Ht2* and *bx1* was small relative to variation attributable to loci with quantitative effects identified in this study. Molecular-marker-facilitated mapping concurred with previous reciprocal translocation mapping stu[illegible] importance of chromosomes 3, 5 and 7, despite the fact that these studies utilized diverse sources of resistant germplasm.

Key words: Genetics – Disease – Mapping – Breeding

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Correspondence to: M. Lee

Introduction

Quantitative and qualitative trait loci affecting disease resistance in plants

The terms horizontal and vertical resistance were introduced by Van der Plank in the 1960s (Van der Plank 1963, 1968). His terminology generated much controversy; the concepts qualitative and quantitative were already widely used and considered adequate (Roane 1973). The term, horizontal resistance soon became considered synonymous with polygenic, minor gene, or quantitative resistance despite the fact that horizontal resistance, as defined by Van der Plank, could have monogenic or qualitative inheritance patterns (Van der Plank 1963, 1978). Perhaps the most onerous of Van der Plank's points was the concept of horizontal resistance being nonspecific, that is, evenly spread against all races of the pathogen (Van der Plank 1984).

One of the first studies to respond to Van der Plank's concepts involved an investigation of differences in pathogenicity and virulence among isolates of *Exserohilum turcicum* to maize (Nelson et al. 1970). The authors concluded that host genes for vertical resistance and horizontal resistance are the same; however, those genes conferring horizontal resistance may not function by themselves vertically. In subsequent studies with wheat isogenic lines and cultures of *Erysiphe graminis* it was argued that a single resistance gene could cause characteristics more commonly associated with several genes, namely a reduction in the apparent infection rate 'r' (Ellingboe 1975; Martin and Ellingboe 1976). In particular, a reduction in the numbers of lesion as well as slower mildew development were noted.

New terminology and concepts continued to evolve and an attempt was made to redefine Van der Plank's

terms (Nelson 1978). Some authors contended that horizontal resistance and vertical resistance, as originally defined, could not co-exist in a single variety (Nelson 1978). This contrasted with Van der Plank's ideas which assumed that vertical resistance probably never occurs independently of horizontal resistance (Van der Plank 1968). In a series of papers Nelson argued that resistance genes, once having been overcome or defeated by a pathogen strain, may retain some residual effect (Nelson 1975, 1978, 1979). Clifford (1975) summarized similar suggestions from other workers, namely quantitative resistance may be an accumulation of residual effects of defeated genes with qualitative effects. Further studies on the reaction of isogenic lines of wheat to *E. graminis* were seen as opposing Van der Plank's concepts in that a reduction in 'r' was demonstrated by residual effects of defeated major genes (Nass et al. 1981). However a reduction in 'r' can be brought about by minor genes irrespective of whether horizontal or vertical resistance is involved (Anderson 1982). Nelson proposed that defeated genes retained some value and suggested pyramiding these into a cultivar to capitalize on the residual effects (Nelson 1975, 1978, 1979). He also highlighted the idea that host background may alter a gene's expression, consequently these differences may be incorrectly attributed to different genes (Nelson 1979). In essence, Nelson proposed that the numbers of genes conditioning disease resistance was not a definitive issue of black and white, but rather a continuum of shades of gray (Nelson 1981).

The debate on the genetic relationship of quantitative and qualitative effects of genes has not been confined to disease resistance. Recently Robertson (1989) has suggested that genes with quantitative effects should be allelic to genes with qualitative effects. The information gained about the location of quantitative trait loci (QTLs) can be compared with that reported for qualitative loci. Such an approach has already been demonstrated for plant height in maize (Beavis et al. 1991), and for traits of significance to the domestication of maize (Doebley et al. 1990). We are proposing an analogous approach for studying host-plant response to *E. turcicum*.

Northern corn leaf blight

E. turcicum Pass. (Syn. *Helminthosporium turcicum* Pass.) is the causal agent of Northern corn leaf blight (NCLB). NCLB occurs sporadically in most temperate, humid areas of the world where maize is grown, and heavy infestations on susceptible germplasm have been known to result in grain yield losses of 30–68% (Lim et al. 1974; Ullstrup 1977; Raymundo and Hooker 1981; Shurtleff 1986; Perkins and Pedersen 1987).

Resistance to the pathogen has been available for some time. The first sources of resistant germplasm exhibited quantitative inheritance patterns controlling the number of lesions (Elliot and Jenkins 1946; Jenkins and Robert 1952; Jenkins et al. 1954; Ullstrup 1977). There are conflicting reports about the effect of quantitative resistance on the size of lesions (Ullstrup 1977; Smith and White 1988). The second type of resistance discovered in maize had qualitative inheritance patterns. Genes with qualitative effects have been identified at several loci, namely *Ht1*, *Ht2*, *Ht3* and *HtN* (Hooker 1961; Hilu and Hooker 1964; Gevers 1975). *Ht1*, *Ht2*, and *Ht3* are characterized by chlorotic lesions or yellow chlorotic margins, reduced fungal sporulation and, subsequently, less inoculum for secondary infections (Hilu and Hooker 1964; Hooker and Kim 1973; Ullstrup 1977). *Ht1*, the first qualitative locus to be mapped, was placed to the central region of the long arm of chromosome 2 (Patterson et al. 1965). More recently restriction fragment length polymorphisms (RFLPs) have been used to gain more precise information on the location of *Ht1* (33, 34) and *Ht2* (Zaitlin et al. 1992). *HtN* is an exception in that it is not characterized by chlorotic lesions but rather by a delay in disease development until after flowering (Gevers 1975). The locations of *Ht3* and *HtN* remain unknown.

The *bx* locus has been implicated in conditioning resistance in seedlings to NCLB through the production of cyclic hydroxamates. Plants of the *bx/bx* genotype are deficient with regard to hydroxamate production (Couture et al. 1971). The deficient genotype results in an increase in both NCLB lesion number and lesion size (Couture et al. 1971).

The objectives of this study were to use RFLP markers to obtain more precise estimates of the chromosomal location of QTLs conferring resistance to *E. turcicum* in maize; in particular to make comparisons with the locations, where known, of loci with qualitative effects on host-plant response.

Materials and methods

Parental inbreds and experimental design

The population studied was created by crossing two inbred maize lines, Mo17 and B52. Mo17 has been an important commercial inbred and shows partial resistance to NCLB. B52 has not been widely used commercially and is very susceptible to NCLB (M. Lee, unpublished). Alleles at *Ht1*, *Ht2*, *Ht3* and *HtN* with qualitative effects have never been reported in either inbred. In our tests chlorotic lesions were not observed. Both inbreds have the normal or *Bx/Bx* genotype. The F_1 hybrid was self-pollinated and 169 F_2 plants were randomly retained. These were self-pollinated to generate the 150 unselected $F_{2:3}$ lines used in the field evaluation. A sets within replications design was used with 27 entries nested within each of six sets. Two replications (12 sets) were grown at Ames, Iowa, and two replications at Urbana, Illinois, in the summer of 1991. Check-inbreds (both parents)

were included in each set. Border rows consisted of bulked F_3 seed from the same population. Row length was 5.5 m with 0.76 m spacing between rows. Plots were thinned to 20 plants per row, approximately 48,000 plants ha^{-1}. Standard management practices regarding fertilization and cultivation were followed. At the Ames location agronomic data including plant height, maturity, and grain yield were recorded. Broad-sense heritabilities were calculated on a progeny mean basis (Hallauer and Miranda 1988). Phenotypic correlations were calculated using Pearson product-moments (SAS Institute Inc. 1988).

Disease inoculation and data collection

At Urbana, Illinois, F_3 plants, check-inbreds, and border plants were inoculated with ground leaf tissue harvested from diseased leaves the previous season. Disease development was negligible and the experiment was abandoned at this location. At Ames, Iowa, F_3 plants, check-inbreds, and border plants were inoculated in the whorl with approximately 10 ml per plant of spore suspension of race 0 on the 24th June and 9th July, 1991 (45 and 60 days after planting respectively). Spore suspension concentrations were 2600 per ml and 2300 per ml, respectively. For spore production, isolate HE62 of *E. turcicum* race 0 was cultured on agar under continuous fluorescent lighting for 12 h daily. Inoculum was applied in the evening when temperatures were cooler and dew formation likely. Disease development was rated twice commencing on the 7th August and 4th September, 1991 (89 and 117 days after planting respectively). Data were collected from the 12 innermost plants per row on four leaves per plant for a total of 48 leaves per entry per replication. The leaves measured on each plant included the leaf attached to the primary ear node, and those attached to the next three internodes below. For each leaf, the number, length and width of all lesions was recorded. The area of lesions, in cm^2, was calculated using the formula: (length × width × 0.7854) (Leath and Pedersen 1986). The percent leaf tissue diseased (severity) was calculated for each leaf by dividing the total area of lesions by the area of the ear leaf and multiplying by 100. The area of the ear leaf was determined using the formula: (length × width × 0.75) (Pearce et al. 1975). Lesion data were then calculated on a per plant basis by averaging over the four leaves, and entry means for each replication were calculated by averaging over the 12 plants sampled. An objective appraisal of disease development was made on 15,552 leaves in each of the two ratings. The large number of leaves to be rated, and the use of multiple inoculations, precluded other factors like infection efficiency, incubation period, or latent period, from being considered. The first assessment took 1 day per replication, the second assessment took 4 days per replication to complete. Trait values in the second replication of the second assessment tended to be a bit higher than those of the first replication. $F_{2:3}$ trait means for QTL mapping were produced by averaging over both replications as this represents the most precise estimate. For QTL mapping, three main aspects of disease development were considered on an entry mean basis: average number of lesions per leaf (number), average percent leaf tissue diseased (severity), and average size of lesions in cm^2 (size). Collectively these will be referred to as traits.

RFLP assays and data analysis

From each of the 150 $F_{2:3}$ lines, ten plants were grown in the greenhouse. An equal quantity of leaf tissue was taken from each plant and bulked to represent the genotype of the F_2 plant from which those lines were derived. Genomic DNA was digested separately with *Eco*RI, *Eco*RV and *Hin*dIII, electrophoresed, and blotted according to Lee et al. (1989). One hundred and three probes that showed polymorphisms between the parental inbreds were chosen from the collections of mapped maize genomic and cDNA clones of the Brookhaven National Laboratories (BNL) (Burr et al. 1988), the University of Missouri-Columbia (UMC) (Coe et al. 1990), Native Plants Inc. (NPI) (Helentjaris et al. 1986), Iowa State University (ISU), and Pioneer Hi-Bred International (PIO). Some probes that are clones of genes with known functions were also included: *Agp2*, ADP glucose pyrophosphorylase (C. Hannah, University of Florida); *C1*, colored aleurone and *P11*, purple plant (K. Cone, University of Missouri); and *dek326* (renamed *ren2*), reduced endosperm (James et al. 1991). Probes were selected such that they represented all ten maize chromosomes and ensured as uniform a coverage of the entire genome as possible.

Linkage maps and interval mapping

The computer program MAPMAKER/QTL (Lander and Botstein 1989) was used to construct the genetic linkage map and for quantitative trait mapping (Lincoln and Lander 1990). To include a locus in a linkage group a maximum LOD score of at least 3.0, and a recombination fraction of at most 0.30, was used. With 94 intervals in this study, a LOD score of 2.83 was calculated as the LOD significance threshold for $P < 0.01$ and 2.31 as the LOD significance threshold for $P < 0.05$. A LOD score of 2.05 corresponds to $P < 0.10$ and, consequently, the highest chance of reporting false positives (Lander and Botstein 1989). The data were analyzed as an F_2 intercross. Scanning the entire genome for putative QTLs was accomplished using the free genetics or unconstrained model. The $F_{2:3}$ means for all disease data tended to be skewed, especially that for the first rating (89 days after planting). This is not entirely unexpected and it should not seriously bias estimates of effects and positions (Knott and Haley 1992). Disease severity, for example, seldom exceeded 25% by virtue of when the rating was taken. If disease development were allowed to proceed too much beyond this point individual lesions would no longer be easily discernable. Consequently most of the disease severity scores lie in the range of from 0 to 25%, rather than from 0 to 100%. Because a normal distribution of phenotypes is an inherent assumption for interval mapping, square root, arc sin, and $\log_{10}$ transformations were attempted for all disease data from both the first and second ratings. The first-assessment data were not normally distributed even after transformation, consequently these data were not used for QTL mapping. Data from the second assessment (117 days after planting) responded more favorably to transformation, in particular when using $\log_{10}$ values. With this transformation, lesion number, percent severity and size of lesions in cm^2 achieved normality as described by the Shapiro-Wilk W statistic ($W = 0.97$, 0.97 and 0.99 respectively, all non-significant at the 0.05 probability level) (Shapiro and Wilk 1965). Consequently $\log_{10}$ transformations of the $F_{2:3}$ line means were used for QTL mapping.

The estimation of QTLs was initially calculated where only one QTL at a time was allowed to explain the variation in the trait. Once these were identified, a multiple QTL model was employed. This was achieved by considering all QTLs for a trait simultaneously in a single model using the 'sequence' and 'map' commands of MAPMAKER QTL. In addition, using each QTL already identified, the genome was re-scanned searching for QTLs, other than those already recorded, which provided a better explanation of the data. This method altered the peak values of QTLs already reported but did not appear to identify any additional QTLs.

Log-likelihood plots were constructed for the entire length of chromosomes 2, 4 and 8 by calculating LOD scores at 2.0 centiMorgan (cM) intervals and then plotting these against map position for each chromosome.

In addition, single factor analysis of variance (SFAOV) (Edwards et al. 1987) was employed to detect significant variation in trait expression associated with the three genotypic classes at each RFLP locus on chromosomes 2, 4 and 8.

Results

Segregation of markers and map size

Of the 103 probes used in this study, one deviated with $P < 0.001$, three deviated with $P < 0.01$, and 22 deviated with $P < 0.05$ from the 1:2:1 ratio expected in the F_2. This has been observed in other maize mapping studies (Edwards et al. 1987); consequently probes that deviated from the expected ratio were not excluded. The 103 probes were used to construct a map of 1454 cM with an average interval length of 15 cM.

Disease data and QTLs affecting host-plant response to E. turcicum

Border plots that were not inoculated were virtually free of NCLB indicating that there was very little natural inoculum and that the disease development that did occur was a direct consequence of the artificial inoculation. Based on the inbred checks, trait values for the 12 sets were not significantly different ($P < 0.05$) indicating that the inoculum procedures and subsequent disease development were relatively uniform across the experiment. On average, B52 had twice as many lesions and over four times as much leaf tissue diseased than did Mo17. In addition, lesions on B52 were over twice the size of those on Mo17 (Table 1). Heritability estimates were 69% for the number of lesions, 62% for disease severity and 31% for the size of lesions.

For declaration of a QTL, a LOD threshold of 2.31, representing a potential false positive rate of 5%, was chosen as an appropriate level of significance for reporting results in this study (Table 2). Three unlinked regions (1S, 3L and 5S) were found to have significant effects on lesion number, each explaining from 6.8% to 13.2% of the phenotypic variation for this trait. Severity was associated with the same regions and, in addition, with QTLs on 7L and 8L; individual loci explained from 7.5% to 13.4% of the phenotypic variation. Two unlinked regions (5L and 7L) were found to have significant effects on lesion size, accounting for 18.1% and 12.3% of the phenotypic variation respectively (Table 2). For each trait the log-likelihood of the multiple QTL model was substantially higher than the individual QTL model. Multiple QTL LOD scores effectively equalled the arithmetic summation of the individual QTL map LOD scores (Table 2). Considering each trait individually, this suggests that while QTLs may explain autonomous portions of the variation, their action is cumulative (Lincoln and Lander 1990). With the exception of chromosome 1, alleles contributing to reduced disease development were derived from Mo17.

Table 1. Trait means and associated standard errors for $F_{2:3}$ lines and inbred checks

Entry	N[a]	Number	Severity	Size (cm^2)
Mo17	6	$1.08^a \pm 0.07$	$0.99^a \pm 0.03$	$4.07^a \pm 0.26$
B52	6	$2.09^a \pm 0.21$	$4.38^b \pm 0.54$	$10.83^b \pm 0.74$
$F_{2:3}$ lines	150	$1.65^a \pm 0.10$	$2.33^a \pm 0.15$	$8.34^b \pm 0.25$
range		(0–10.58)	(0–17.52)	(1.91–31.02)

[a] N, number of observations per replication
Entry means with common superscripts for each trait are not significantly different ($P < 0.05$)

Table 2. Location of QTLs affecting host-plant response to *E. turcicum* in $F_{2:3}$ lines of a B52/Mo17 population

Interval	Chromosome	Variation explained	LOD
Average number of lesions per leaf (number)			
UMC157-UMC67	1S	6.8%	2.31
UMC16-NPI457	3L	13.2%	4.60
BNL6.25-UMC90	5S	11.8%	3.85
Multiple QTL model		29.5%	11.10
Average percent leaf tissue diseased (severity)			
UMC157-UMC67	1S	8.0%	2.71
UMC16-NPI457	3L	9.4%	3.22
UMC90-UMC166	5S	13.4%	4.14
BNL15.21-UMC110	7L	13.2%	3.84
BNL9.08-BNL7.08A	8L	7.5%	2.32
Multiple QTL model		44.6%	17.30
Average size of lesions in cm^2 (size)			
BNL5.71-UMC51	5L	18.1%	4.32
UMC116-BNL15.21	7L	12.3%	3.70
Multiple QTL model		28.9%	7.98

Chromosomes 2, 4 and 8 are of particular interest since they contain the *Ht1*, *bx1* and *Ht2* loci, respectively. Consequently QTL log-likelihood plots are shown for the entire length of each of these chromosomes. Chromosome 2 is depicted in Fig. 1. The *Ht1* locus is flanked by several probes on the long arm of chromosome 2. The extent of the contribution of this region to the size of lesions varies from explaining between 3.2% and 6.5% of the phenotypic variation. The range in the contribution depends on which of the different maps are integrated to determine the position of the *Ht1* locus. The lowest estimate is obtained if the data of Bentolila et al. (1991) (A in Fig. 1) are superimposed on the map for chromosome 2 obtained in the present study (C in Fig. 1). Superimposing the data from Hoisington and Coe (1989) (B in Fig. 1) produces the higher estimate. The contribution of these intervals to the number of lesions and the percent severity ranges from 0.4% to 0.7% and from 0% to 0.4% respectively. SFAOV indicated that loci UMC98 and UMC88 were significantly associated with lesion size ($P < 0.05$, Fig. 1). Additional significant associations were not detected with SFAOV on chromosome 2.

Chromosome 4 is depicted in Fig. 2. Probe MPIK5, representing a putative clone of the *bx1* locus (M. Frey, personal communication), has been assigned to the short arm of chromosome 4 (B. Burr, personal communication), suggesting that UMC123 is 5 cM distal to *bx1*. This information was superimposed on the map for chromosome 4 obtained in the present study (B in

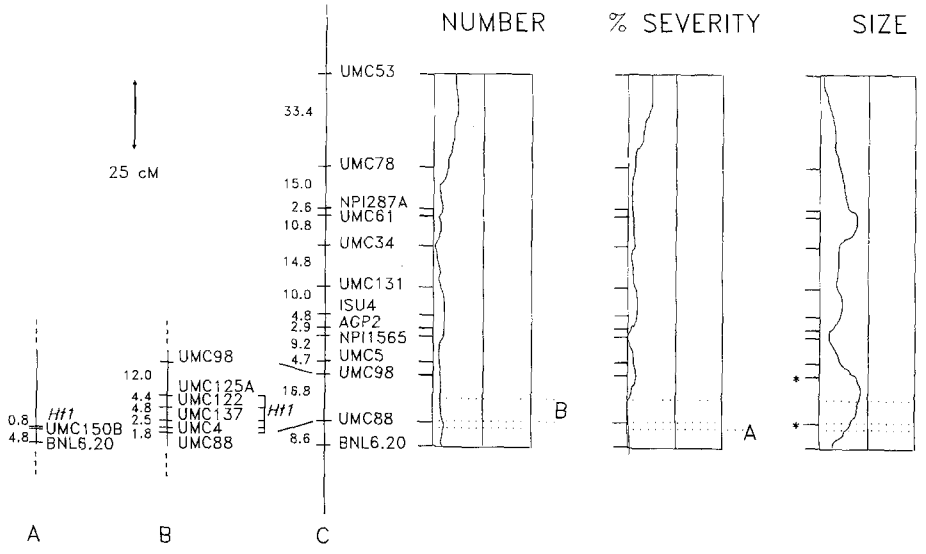

Fig. 1A–C. Linkage maps and log-likelihood plot of chromosome 2. **A** Position of *Ht1* (34). **B** Position of *Ht1* (33). **C** Linkage map and LOD scores from current study. *Dotted lines* indicate estimated position of the *Ht1* locus obtained by superimposing data from **A** on **C** using BNL6.20 for alignment, and **B** on **C** using UMC98 and UMC88 for alignment. Distances between probes are shown in cM. *Vertical lines* in the log-likelihood plots represent LOD scores of 0, 2 and 4. SFAOV results: *, ** and *** indicate significance at 0.05, 0.01 and 0.001 levels respectively

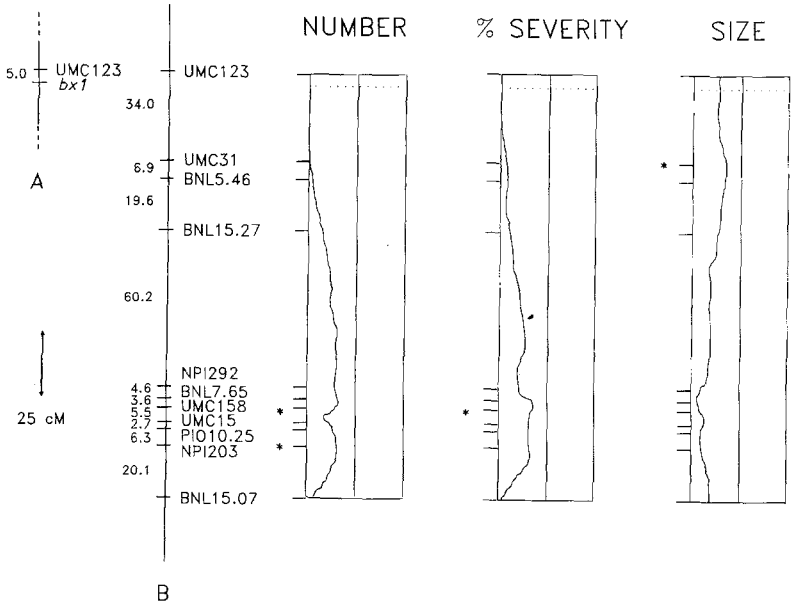

Fig. 2A, B. Linkage maps and log-likelihood plot of chromosome 4. **A** Putative position of *bx1* (B. Burr, personal communication and M. Frey, personal communication). **B** Linkage map and LOD scores from current study. *Dotted lines* indicate estimated position of the *bx1* locus obtained by superimposing data from **A** on **B** using UM123 for alignment. Distances between probes are shown in cM. *Vertical lines* in the log-likelihood plots represent LOD scores of 0, 2 and 4. SFAOV results: *, ** and *** indicate significance at 0.05, 0.01 and 0.001 levels respectively

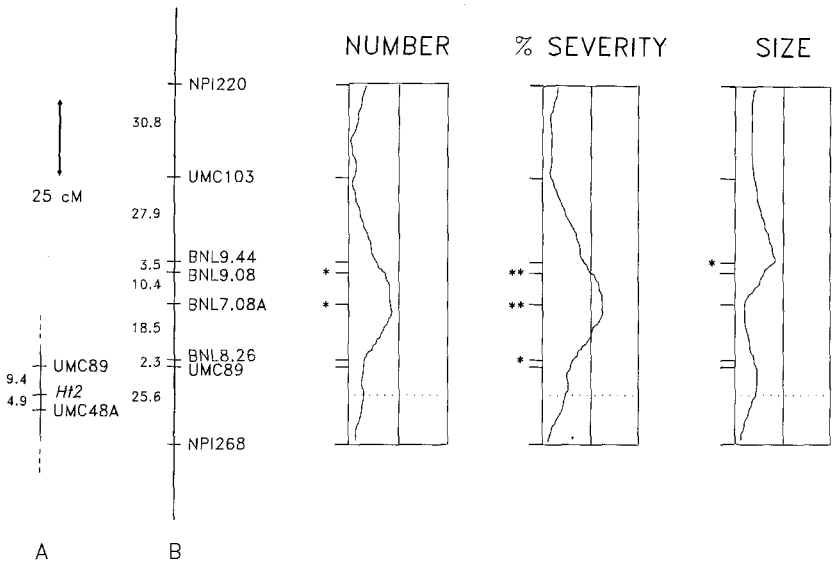

Fig. 3A, B. Linkage maps and log-likelihood plot of chromosome 8. **A** Position of *Ht2* (Zaitlin et al. 1992). **B** Linkage map and LOD scores from current study. *Dotted lines* indicate estimated position of the *Ht2* locus obtained by superimposing data from **A** on **B** using UMC89 for alignment. Distances between probes are shown in cM. *Vertical lines* in the log-likelihood plots represent LOD scores of 0, 2 and 4. SFAOV results: *, ** and *** indicate significance at 0.05, 0.01 and 0.001 levels respectively

Fig. 2) for comparative purposes. The chromosomal region containing the *bx1* locus appears to contribute 5.0% of the phenotypic variation for lesion size. The contribution towards lesion number and severity is negligible. From Fig. 2 it is apparent that regions of the short arm of chromosome 4 play a small role in determining lesion size. SFAOV indicated some significant associations ($P < 0.05$) but the RFLP loci are not in close proximity to the *bx1* locus (Fig. 2).

Chromosome 8 is depicted in Fig. 3. The data of Zaitlin et al. (1992) (A in Fig. 3) suggests that *Ht2* is 9.4 cM distal to UMC89. This information was superimposed on the map for chromosome 8 obtained in the current study (B in Fig. 3). The chromosomal region containing the *Ht2* locus appears to contribute 3.0% of the phenotypic variation for lesion size. The contribution towards the number of lesions and severity is 2.3% and 3.7%, respectively. SFAOV indicates several loci with significant associations ($P < 0.01$ or $P < 0.05$).

Phenotypic correlations between attributes of disease development

The number of lesions per leaf showed a very strong positive correlation with disease severity (0.95, $P < 0.01$ level). The average size of lesions was not correlated with severity (-0.09, not significant, $P > 0.05$). The correlation between the size and number of lesions was -0.23 ($P < 0.01$).

Discussion

QTL and physical mapping

Most of the information currently available on the location of quantitative resistance to NCLB in maize was obtained in a comprehensive series of studies with reciprocal translocations, generally using Mo21A as a source of resistance (Jenkins et al. 1957; Jenkins and Robert 1961). More recently, a translocation study using Mo17 as a source of resistance indicated that chromosomes 3, 4S (short arm), and 6L (long arm) were of importance in reducing disease severity (Brewster et al. 1992). However, the translocation stocks used were not complete; several chromosome arms were not represented (2L, 7S, 8S, 10L and 10S) or else only represented by a single breakpoint (1S, 2S, 4S, 5S, 5L, 6S and 6L). In the latter case, any factors important in determining host-plant response and lying more than 50 cM away from the breakpoint would not be detected. In contrast, the map used in the present study and the translocation stocks used in the early studies provide more complete coverage of the genome. The RFLP mapping results obtained in this study concur with the early translocation mapping experiments (Jenkins et al. 1957; Jenkins and Robert 1961) on the importance of chromosomes 3, 5 and 7, despite the fact that these studies have utilized diverse germplasms and arguably even different fungal isolates given the 30-year time span. Prior to 1974 only one physiological race of *E. turcicum* was known to exist (Smith and White 1988). The only exceptions regarding chromosomal regions involve 5L and 7S. Whereas Jenkins et al. (1957) and Jenkins and Robert (1961) indicate that 5L is important for disease severity (based on visual appraisals), the present study indicates that 5S is important in determining severity while 5L appears to be associated with lesion size. Furthermore, Jenkins et al. (1957) and Jenkins and Robert (1961) indicate 7S is important in determining disease severity, whereas the present study implicates 7L for both disease severity and lesion size. In the latter case, the log-likelihood

peaks appear close to the centromere, based on the RFLP loci positions relative to other published maps (Hoisington and Coe 1989). Since centromere probes were not included in the current study the designation of short or long arm are approximate only, particularly in the case of chromosome 7. This is often also true for the translocation studies and could be a reason for the discrepancy.

Qualitative alleles at quantitative loci

Dominant alleles with qualitative effects have never been reported in either inbred parent used in this study. Chlorotic lesions were not observed in the $F_{2:3}$ lines or parental inbreds, further suggesting the absence of this type of qualitative resistance. The normal or *Bx* genotype has been implicated in imparting resistance to NCLB; however, this is most likely to be expressed in seedlings (Couture et al. 1971). In contrast, the disease appraisals in the current study where conducted on adult plants. The present study suggests that loci in the vicinity of *bx1* appear to have some minor contribution in determining lesion size.

The location of the genes with qualitative effects are approximate and their putative effects modest; however, it seems unlikely that the small log-likelihood peaks, particularly in regard to lesion size on chromosomes 2 and 8, arise purely by chance. The results of this study suggest that loci in the vicinity of the *Ht1* and *Ht2* loci have a small effect in the absence of the marked qualitative effects of the *Ht1* or *Ht2* genes. In this one end of a spectrum of isoalleles as Robertson (1989) suggested? Are the marked qualitative effects of the *Ht1* and *Ht2* genes the opposite end of this spectrum? Genes with qualitative effects, like *Ht1* and *Ht2*, may be, as Robertson's (1989) hypothesis suggests, mutants or alternate alleles at quantitative loci. The small contributions of genes with quantitative effects, such as those observed in the vicinity of the *Ht1* or *Ht2* loci, may be masked by environmental factors and by the use of subjective and imprecise assessment methods in contrast to objective measurements on lesion number and size. Additionally, the extent of these effects may differ according to pathogen race and host background.

Our current knowledge of chromosomal regions contributing to *E. turcicum* resistance is largely limited to alleles fortuitously identified with qualitative effects. Obviously, regions other than *Ht1*, *Ht2*, *HtN* and *bx1* are of importance. (The *HtN* locus has been mapped to the long arm of chromosome eight, distal to *Ht2*; see Simcox and Bennetzen 1994.) The regions identified in this study may include *Ht3* since that has not been mapped; however, the number of QTLs identified in this study exceeds the number of loci with known qualitative effects. Perhaps such QTL mapping initiatives provide insights into genomic regions that merit further study.

The usefulness of graphical presentation of the data is obvious when one compares the possible erroneous conclusion that may be arrived at by solely comparing tables listing QTLs exceeding threshold values. Table 2 indicates that while only three QTLs affect the number of lesions, five affect disease severity. However, a graphical presentation of log-likelihood plots for all chromosomes (data not shown) reveals almost identi cal patterns for both traits and confirm their high phenotypic correlation (0.95). Non-significance in statistical terms may not denote insignificance in biological terms. There are several alternative 'levels of significance' and by solely relying on tables with truncation thresholds we may diminish the power of molecular-marker-facilitated mapping techniques. This concept, along with that of components of resistance, gene action, and correlations with other agronomic traits, will be discussed elsewhere.

References

Anderson MG (1982) Interpreting residual effects of "defeated" resistance genes. Phytopathology 72:1383–1384

Beavis WD, Grant D, Albertson M, Fincher R (1991) Quantitative trait loci for plant height in four maize populations and their associations with qualitative genetic loci. Theor Appl Genet 83:141–145

Bentolila S, Guitton C, Bouvet N, Sailland A, Nykaza S, Freyssinet G (1991) Identification of an RFLP marker tightly linked to the *Ht1* gene in maize. Theor Appl Genet 82:393–398

Brewster VA, Carson ML, Wicks ZW III (1992) Mapping components of partial resistance to Northern leaf blight of maize using reciprocal translocations. Phytopathology 82:225–229

Burr B, Burr FA, Thompson KH, Albertson MC, Stuber CW (1988) Gene mapping with recombinant inbreds in maize. Genetics 118:519–526

Clifford BC (1975) Stable resistance to cereal disease: problems and progress. 1974 Rep Welsh Plant Breed Stn (Aberystwyth), pp 107–113

Coe EH, Hoisington DA, Neuffer MG (1990) Linkage map of corn (maize) (*Zea mays* L.) (2N = 20). In: O'Brien SJ (ed) Genetic maps, 5th edn. Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, pp 6.39–6.67

Couture RM, Routley DG, Dunn GM (1971) Role of cyclic hydoxamic acids in monogenic resistance of maize to *Helminthosporium turcicum*. Physiol Pl Path 1:515–521

Doebley J, Stec A, Wendel J, Edwards M (1990) Genetic and morphological analysis of a maize-teosinte F_2 population: implications for the origin of maize. Proc Natl Acad Sci USA 87:9888–9892

Edwards MD, Stuber CW, Wendel JF (1987) Molecular-marker-facilitated investigations of quantitative-trait loci in maize. I. Numbers, genomic distribution and types of gene action. Genetics 116:113–125

Ellingboe AH (1975) Horizontal resistance: an artifact of experimental procedure? Aust Pl Pathol Soc Newslett 4:44–46

Elliot C, Jenkins MT (1946) *Helminthosporium turcicum* leaf blight of corn. Phytopathology 36:660–666

Gevers HO (1975) A new major gene for resistance to *Helminthosporium turcicum* leaf blight of maize. Plant Dis Rep 59:296–299

Hallauer AR, Miranda JB (1988) Quantitative genetics in maize breeding, 2nd edn. Iowa State University Press, Ames, Iowa, USA

Helentjaris T, Slocum M, Wright S, Schaefer A, Nienhuis J (1986) Construction of genetic linkage maps in maize and tomato using restriction fragment length polymorphisms. Theor Appl Genet 72:761–769

Hilu HM, Hooker AL (1964) Host-pathogen relationship of *Helminthosporium turcicum* in resistant and susceptible corn seedlings. Phytopathology 54:570–575

Hoisington DA, Coe EH (1989) Mapping in maize using RFLPs. In: Gustafson JP (ed) Stadler genetics symposium–Gene manipulation in plant Improvement II. Plenum Press, New York, London, pp 331–352

Hooker AL (1961) A new type of resistance in corn to *Helminthosporium turcicum*. Plant Dis Rep 45:780–781

Hooker AL, Kim SK (1973) Monogenic and multigenic resistance to *Helminthosporium turcicum* in corn. Plant Dis Rep 57:586–589

James MG, Scanlon MS, Robertson DS, Myers AM (1991) Cloning of three putative defective kernel loci by transposon tagging. Maize Newslett 65:10

Jenkins MT, Robert AL (1952) Inheritance of resistance to the leaf blight of corn caused by *Helminthosporium turcicum*. Agron J 44:136–140

Jenkins MT, Robert AL (1961) Further genetic studies of resistance to *Helminthosporium turcicum* Pass. by means of chromosomal translocations. Crop Sci 1:450–455

Jenkins MT, Robert AL, Findley WR Jr (1954) Recurrent selection as a method for concentrating genes for resistance to *Helminthosporium turcicum* leaf blight in corn. Agron J 46:89–94

Jenkins MT, Robert AL, Findley WR Jr (1957) Genetic studies of resistance to *Helminthosporium turcicum* in maize by means of chromosomal translocations. Agron J 49:197–201

Knott SA, Haley CS (1992) Aspects of maximum likelihood methods for the mapping of quantitative trait loci in line crosses. Genet Res 60:139–151

Lander ES, Botstein D (1989) Mapping Mendelian factors underlying quantitative traits using RFLP linkage maps. Genetics 121:185–199

Leath S, Pedersen WL (1986) Differences in resistance between maize hybrids with or without the Ht_1 gene when infected with *Exserohilum turcicum* Race 2. Phytopathology 76:257–260

Lee M, Godshalk EB, Lamkey KR, Woodman WL (1989) Association of restriction fragment length polymorphisms among maize inbreds with agronomic performance of their crosses. Crop Sci 29:1067–1071

Lim SM, Kinsey JG, Hooker AL (1974) Inheritance of virulence in *Helminthosporium turcicum* to monogenic resistant corn. Phytopathology 64:1150–1151

Lincoln SE, Lander ES (1990) Mapping genes controlling quantitative traits using MAPMAKER/QTL. Whitehead Institute for Biomedical Research, Cambridge, Massachusetts, USA

Martin TJ, Ellingboe AH (1976) Differences between compatible parasite/host genotypes involving the *Pm4* locus of wheat and the corresponding genes in *Erysiphe graminis* f. sp. *tritici*. Phytopathology 66:1435–1438

Nass HA, Pedersen WL, MacKenzie DR, Nelson RR (1981) The residual effects of some "defeated" powdery mildew resistance genes in isolines of winter wheat. Phytopathology 71:1315–1318

Nelson RR (1975) Horizontal resistance in plants: concepts, controversies and applications. In: Galvez GE (ed) Proc Sem on Horizontal Resistance to the Blast Disease of Rice. CIAT publication, Series CE-9. Cali, Columbia, pp 1–20

Nelson RR (1978) Genetics of horizontal resistance to plant diseases. Annu Rev Phytopathol 16:359–378

Nelson RR (1979) The evolution of parasitic fitness. In: Horsfall GJ, Cowling EB (eds) Plant disease, an advanced treatise, vol IV. Academic Press, New York, pp 23–46

Nelson RR (1981) Disease resistance breakthrough – resistance not black and white, but various shades of gray. Crops and Soils Mag, Am Soc Agron, Madison, Wisconsin, 34:7–9

Nelson RR, MacKenzie DR, Scheifele GI (1970) Interaction of genes for pathogenicity and virulence in *Trichometasphaeria turcica* with different numbers of genes for vertical resistance in *Zea mays*. Phytopathology 60:1250–1254

Patterson EB, Hooker AL, Yates DE (1965) Location of *Ht* in the long arm of chromosome 2. Maize Newslett 39:86–87

Pearce RB, Mock JJ, Bailey TB (1975) Rapid method for estimating leaf area per plant in maize. Crop Sci 15:691–694

Perkins JM, Pedersen WL (1987) Disease development and yield losses associated with Northern leaf blight on corn. Pl Dis 71:940–943

Raymundo AD, Hooker AL (1981) Measuring the relationship between Northern corn leaf blight and yield losses. Pl Dis 65:325–327

Roane CW (1973) Trends in breeding for disease resistance in crops. Annu Rev Phytopathol 11:463–486

Robertson DS (1989) Understanding the relationship between qualitative and quantitative genetics. In: Helentjaris T, Burr B (eds) Development and application of molecular markers to problems in plant genetics. Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, pp 81–87

SAS (1984) Procedures guide, guide 6.03 edn. SAS Institute Inc., Cary, North Carolina, USA

Shapiro SS, Wilk MB (1965) An analysis of variance for normality (complete samples). Biometrika 52:591–611

Shurtleff MC (1986) *Helminthosporium* leaf spots and blights. In: Compendium of corn diseases, 2nd edn. Am Phytopathol Soc, St. Paul, Minnessota, pp 15–19

Simcox KD and Bennetzen JL (1994) The use of molecular markers to study *Setosphaeria turcica* resistance in maize. Phytopathology (in press)

Smith DR, White DG (1988) Diseases of corn. In: Sprague GF, Dudley JW (eds) Corn and corn improvement, 3rd edn. Am Soc Agron, Madison, Wisconsin, pp 700–766

Ullstrup AJ (1977) Diseases of corn. In: Sprague GF (ed) Corn and corn improvement, 2nd edn. Am Soc Agron, Madison, Wisconsin pp 391–500

Van der Plank JE (1963) Plant diseases: epidemics and control. Academic Press, New York

Van der Plank JE (1968) Disease resistance in plants. Academic Press, New York

Van der Plank JE (1978) Genetic and molecular basis of plant pathogenesis. Springer-Verlag, Berlin New York

Van der Plank JE (1984) Disease resistance in plants, 2nd edn. Academic Press, Orlando, Florida, USA

Zaitlin D, DeMars SJ, Gupta M (1992) Linkage of a second gene for NCLB resistance to molecular markers in maize. Maize Newslett 66:69–70