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QTL Mapping in Tropical Maize: II. Comparison of Genomic Regions for Resistance to *Diatraea* spp.

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ABSTRACT

Southwestern corn borer (SWCB), *Diatraea grandiosella* (Dyar), and the sugar cane borer (SCB), *Diatraea saccharalis* (Fabricius), are serious insect pests in maize (*Zea mays* L.) production areas of Central America and the southern USA. We mapped and characterized quantitative trait loci (QTL) affecting resistance to the leaf feeding generation of SWCB (1SWCB), compared these QTL with those for resistance to the leaf feeding generation of SCB (1SCB) identified in the same mapping population, and assessed the consistency of QTL for 1SWCB across two populations. One hundred seventy-one F_2 genotypes from cross CML131 (susceptible) × CML67 (resistant) and 100 RFLP marker loci were used for the QTL analyses. 1SWCB and 1SCB resistance were assessed in $F_{2:3}$ lines by leaf damage ratings (LDR) after artificial infestation in field experiments with two replications at one subtropical environment in 2 yr. The method of composite interval mapping (CIM) was used for QTL detection. Estimates of genotypic (σ^2_g) and genotype × year interaction variance (σ^2_{gy}) were highly significant for 1SWCB LDR and 1SCB LDR. Phenotypic and genotypic correlations between both traits were 0.62 and 1.02, respectively. For 1SWCB LDR, six QTL were detected explaining 53.3% of $\hat{\sigma}^2_g$, with two QTL displaying significant QTL × year interactions. Ten QTL were detected for 1SCB LDR, accounting for 98.2% of $\hat{\sigma}^2_g$. The QTL showed predominantly additive or partially dominant gene action. Seven out of 10 QTL were pleiotropic to both *Diatraea* spp. Three genomic regions, on Chromosomes 5 and 9, were consistent with a second mapping population derived from cross Ki3 (susceptible) × CML139 (resistant), for which seven QTL for 1SWCB LDR were found. Marker-assisted 'gene stacking' is recommended for transferring pleiotropic QTL into susceptible germplasm and for pyramiding QTL from different sources of insect resistance.

THE SOUTHWESTERN CORN BORER and the sugar cane borer are important insect pests in tropical and subtropical areas of maize production in Central America and the southern USA. Both insect species are closely related, as apparent from their similar life cycles and feeding behavior on maize. Larvae of both insect species can cause extensive damage due to leaf feeding and stem tunneling. However, SWCB larvae are generally larger and more aggressive feeders than corresponding developmental stages of SCB (Hinderliter, 1983).

The regional distributions of SWCB and SCB overlap in Central America and the southern USA. The development of maize germplasm resistant to the leaf feeding and stem tunneling generations of both insect species is important because resistance to only one of these insect species would be insufficient. Therefore, Smith et al. (1989) developed a breeding scheme at the International Maize and Wheat Improvement Center (CIMMYT) to combine different germplasm sources conferring resistance to a number of maize stem borer species, including both *Diatraea* spp., for establishing a multiple-borer-resistant (MBR) population. The level of resistance to these insect species was significantly improved by S_1 recurrent selection, suggesting a preponderance of additive gene action (Thome et al., 1992).

Although host-plant resistance to insects comprises nonpreference, antibiosis, and tolerance, breeding procedures have focused mainly on antibiosis to improve the level of SWCB and SCB resistance (Mihm, 1989). The level of antibiosis can be determined either directly by evaluating parameters of insect development or indirectly by assessing the degree of feeding damage on plants caused by the insect larvae. The biochemical compound DIMBOA provides protection against leaf feeding by larvae of the European corn borer (ECB), *Ostrinia nubilalis* (Hübner), in temperate maize germplasm (Klun et al., 1967). However, in tropical maize germplasm, DIMBOA was not active against leaf feeding damage by SWCB and SCB larvae, indicating a different mode of antibiosis (Hedin et al., 1984).

Two studies have reported mapping the QTL underlying antibiosis type of leaf feeding resistance in maize. Schön et al. (1991) identified four QTL conferring resistance to the first generation ECB (1ECB) damage, one of them being located on Chromosome 4 in the region of the benzoxazinless1 (*bx1*) gene, which contributes to a high DIMBOA content. Bohn et al. (1996) evaluated a population of F_3 lines from the cross of two CIMMYT lines, CML131 × CML67, for resistance to the first generation SCB (1SCB) in three environments. By

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Abbreviations: 1ECB, first generation of European corn borer; 1SCB, first generation of sugar cane borer; 1SWCB, first generation of southwestern corn borer; 2ECB, second generation of European corn borer; cM, centimorgan; CIM, composite interval mapping; CIMMYT, International Maize and Wheat Improvement Center; DR, dominance ratio; ECB, European corn borer; LDR, leaf damage ratings; LOD, $\log_{10}$ odds ratio; LR, likelihood ratio; MAS, marker-assisted selection; MBR, multiple-borer-resistant; QTL, quantitative trait locus (or loci, depending on the context); *RE*, relative efficiency; RFLP, restriction fragment length polymorphism; SCB, sugar cane borer; SWCB, southwestern corn borer.

applying the method of composite interval mapping (CIM) on the means of F_3 lines across environments, they identified 10 QTL including four regions known to harbor genes involved in cell-wall biochemistry. These results supported a hypothesis of Bergvinson et al. (1996), who proposed that mechanisms of SWCB and SCB resistance in maize include protein, fibre, and cell wall phenolic acid contents.

QTL mapping is the first step in marker-assisted selection (MAS) procedures. The efficiency of MAS depends on the consistency of the estimated QTL positions and effects across populations. No reports are hitherto available concerning QTL mapping results for 1SWCB or 1SCB resistance in different populations. For resistance against the second generation ECB (2ECB), Lee (1993) identified 16 QTL in three populations of F_3 lines derived from crosses between two susceptible and two resistant maize inbred lines. Most of these QTL were found in more than one population, indicating a common genetic basis for resistance to 2ECB feeding.

In this study, we investigated the genetic basis of resistance in maize to the leaf feeding generation of SWCB (1SWCB) by means of QTL analyses using the same population of F_3 lines as Bohn et al. (1996) in their study on 1SCB resistance. Our objectives were to (i) estimate the number, chromosomal positions, and genetic effects of QTL involved in antibiosis against 1SWCB and 1SCB in one mapping population, using multiple-trait analysis of QTL developed by Jiang and Zeng (1995), (ii) compare QTL for resistance against 1SWCB with those for 1SCB resistance identified in the same mapping population tested in the same environments, and (iii) ascertain the consistency of QTL for 1SWCB resistance found in this population with those mapped recently in a different population (Khairallah et al., 1997).

MATERIALS AND METHODS

Plant Materials

The materials and part of the methods used in this study have been previously described in detail by Bohn et al. (1996). Briefly, two inbred lines, CML131, a subtropical, intermediate maturity, white dent line and highly susceptible to leaf feeding by SWCB and SCB, and CML67, a tropical, late maturity, yellow semi-dent line selected out of 'Antigua Group 2' with known high resistance to 1SWCB and 1SCB, were used as parents. F_2 plants originating from two randomly chosen F_1 plants were selfed to produce 215 F_3 lines. Leaf samples were taken from a random subset of 190 F_2 plants for subsequent RFLP assays, of which 171 were in common with the 215 F_3 lines.

A second population, described in detail elsewhere (Khairallah et al., 1997), was used in this paper only for comparison of the QTL mapping results on 1SWCB resistance. This population originated from the cross between inbreds Ki3, a tropical, late maturity, yellow flint line, susceptible to *Diatraea* spp., and CML139, a subtropical, intermediate maturity, yellow semi-flint line, with a high level of resistance to 1SWCB and 1SCB. RFLP assays of the 475 F_2 plants and field testing of the corresponding F_3 lines from this population for 1SWCB resistance in three environments (Tlaltizapán, summer season 1990 and winter seasons 1990 and 1992) were performed according to the following described procedures.

RFLP Assays

The procedure for the RFLP assays has been reported in the companion paper (Bohn et al., 1996). The RFLP data for cross CML131 × CML67 given by these authors were also used in the present QTL analyses, because the F_3 lines employed in both studies were identical. A total of 100 RFLP marker loci, well distributed over the maize genome, were used to genotype the parental F_2 plants of 190 F_3 lines.

Agronomic Trials

Separate experiments with (i) infestations of SWCB larvae and (ii) infestations of SCB larvae were conducted at CIMMYT's experimental station at Tlaltizapán, Mexico, (subtropical environment, 940 m elevation, 18° N) during the winter seasons (November through May) of 1992 and 1993. Each experiment included 240 entries: 215 F_3 lines and the parental inbred lines as multiple entries. The experimental design was a 24-by-10 alpha design with two replications and single-row plots 0.75 m apart and a length of 5.0 m in 1992, and 2.5 m in 1993.

For evaluating the level of antibiosis against 1SWCB, every plant was artificially infested with about 30 to 45 neonate SWCB larvae at the six- to eight-leaf stage (mid-whorl) by mixing freshly hatched larvae with maize-cob grits and applying the mixture into the plant whorl with a mechanical dispenser ('bazooka'; Mihm, 1983). Leaf feeding damage by each insect was assessed 2 to 3 wk after infestation with a rating scale from 1 (no visible leaf damage) to 10 (dead growing point, all leaves with long lesions) as described by Thome et al. (1992).

In a companion paper, Bohn et al. (1996) presented the results of a QTL analysis of 1SCB leaf damage ratings (LDR) based on three environments, using the same experimental procedures as described. In their study, QTL analysis was performed on LDR means across environments by applying the method of CIM. However, to avoid confounding with other factors, we compared QTL for 1SWCB and 1SCB resistance only for those two environments (Tlaltizapán, winter seasons 1992 and 1993), for which we had data on both insect species. The reduced 1SCB data set was reanalyzed by the joint CIM approach for multiple environments as suggested by Jiang and Zeng (1995).

Statistical Analyses

Phenotypic Data

Analyses of variance were performed on field data from each experiment within each year. Adjusted entry means and effective error mean squares were used to compute the combined analyses of variance and covariance across years. Orthogonal contrasts among the means of F_3 lines ($\bar{F}_3$) vs. the midparent value ($\bar{P}$ = (P1 + P2)/2) and P1 vs. P2, as well as estimates of the genotypic variance (σ^2_g), the genotype × year interaction variance (σ^2_{gy}), the error variance (σ^2), the phenotypic variance (σ^2_p) and heritability (h^2) for F_3 lines together with 90% confidence intervals as well as phenotypic (r_p) and genotypic correlation coefficients (r_g) were calculated as described in detail by Bohn et al. (1996).

Genotypic Data

The segregation and linkage analyses of the RFLP marker data for F_2 individuals from the cross CML131 × CML67

have been reported by Bohn et al. (1996). To relate the QTL mapping results for 1SWCB and 1SCB leaf feeding resistance in the cross CML131 × CML67 with QTL for 1SWCB resistance in cross Ki3 × CML139 found in a separate QTL analysis, a combined RFLP linkage map was constructed. The genotypic data from both mapping populations (i.e., 190 F_2 individuals and 100 RFLP loci for the CML131 × CML67 population and 475 F_2 individuals and 128 RFLP loci for the Ki3 × CML139 population) were merged into a single data set. MAPMAKER (Lander et al., 1987) was employed to construct the combined linkage map obtained from both populations. RFLP markers only available for one population were treated as missing data in the other population in the subsequent analysis of the combined data set.

QTL Analyses

QTL analyses were performed for the CML131 × CML67 population on the subset of 171 F_2 individuals for which both molecular and phenotypic data on F_3 lines were available. The method of CIM proposed by Jansen and Stam (1994) and Zeng (1994) and recently extended by Jiang and Zeng (1995) to perform a joint analysis of multiple environments and multiple traits was employed to map QTL and estimate their genetic effects.

The Model. First, adjusted entry means of 1SWCB LDR and 1SCB LDR were analyzed separately in a joint CIM across both years in order to identify putative QTL for each trait and to determine the significance of QTL × year interactions. Second, a joint analysis was performed on both traits and both years to test the hypotheses of pleiotropy, pleiotropy vs. linkage, and QTL × trait interactions for putative QTL. Following Jiang and Zeng (1995), the underlying mixture model for QTL detection was:

$$y_{jkl} = b_{0kl} + a_{kl}x_j + d_{kl}z_j + \sum_s^t (a^*_{skl}x^*_{js} + d^*_{skl}z^*_{js}) + e_{jkl},$$

with $j = 1, ..., 171$; $k = 1, 2$; $l = 1, 2$; $s = 1,...,t$. Here, y_{jkl} denotes the phenotypic value for trait k in year l of the jth F_3 line; b_{0kl} is the mean effect of the model for trait k and year l; a_{kl} and d_{kl} are the additive and dominance effects, respectively, of the putative QTL for trait k and year l in the marker interval $(i, i + 1)$ under consideration; x_j counts the number of alleles from the resistant parent at the putative QTL and takes values 2, 1, and 0 if the genotype at the putative QTL is QQ, Qq, or qq, respectively, with probabilities depending on the observed genotype at the flanking marker loci and the recombination frequencies between the QTL and the markers; similarly, z_j is a random indicator variable taking values 1 and 0 for heterozygote and homozygote QTL genotypes, respectively; x^*_{js} and z^*_{js} are corresponding variables for marker s, assuming t markers are selected as cofactors for controlling the residual genetic variation; a^*_{skl} and d^*_{skl} are the partial regression coefficient of phenotype y_{jkl} on x^*_{js} and z^*_{js}; and e_{jkl} is the residual variable for the jth F_3 line for trait k in year l.

For QTL detection, two variants of this model were used: (i) Model A employed selected markers as cofactors not located on the chromosome scanned for the presence of a QTL, and (ii) Model B used all selected cofactors plus markers flanking the target interval with a minimum map distance (referred to as window size) of 30 cM.

Cofactor Selection. Cofactors were selected for each experiment by a stepwise regression procedure (Draper and Smith, 1981, p. 307ff). Final selection was for the model that minimized Akaike's information criterion with penalty = 3.0 (for details, see Jansen, 1993). In order to perform joint CIM for a single trait across both years, or both traits and years, the respective cofactor sets were combined.

QTL Detection. A two-step procedure was employed using both model variants in the QTL detection process. At first, Model A was used to identify putative QTL regions by taking advantage of its high power for QTL detection. In a second step, Model B was employed to resolve QTL linked in coupling or repulsion phases. The decision, whether a QTL was active at a given chromosomal position, π_1, or not was based on the following rules: (i) A QTL was declared as present, whenever a significant likelihood ratio (LR) peak was detected by Model A and Model B also yielded a LR peak at the same or an adjacent position. In this case, it was not required that the LR peak found by Model B exceed the critical threshold. (ii) If the LR peak identified by Model A at π_1 was not confirmed by Model B, we rejected the hypothesis of the presence of a putative QTL at this position. This result was taken as indicative for the presence of multiple, linked QTL in adjacent regions. (iii) A QTL solely detected with Model B was regarded as significant without further confirmation.

Hypotheses Testing. By adopting the joint CIM approach developed by Jiang and Zeng (1995), we were able to test the following genetic hypotheses: (i) pleiotropic effects of QTL on both 1SWCB LDR and 1SCB LDR, (ii) pleiotropy of one QTL vs. two linked QTL each with an effect on one trait only, (iii) QTL × trait interactions, and (iv) QTL × year interactions.

We performed a LR test for pleiotropy in all those cases, where a genomic position showed significant QTL activity in joint CIM of multiple traits, and this region had a significant QTL in single trait analysis for only one trait. If the null hypothesis (H_{10}: $a_{1l} = 0$, $d_{1l} = 0$, and H_{20}: $a_{2l} = 0$, $d_{2l} = 0$) was rejected, we inferred the presence of pleiotropy. When QTL activity was indicated by joint CIM of both traits and this position was significant for both 1SWCB LDR and 1SCB LDR, LR tests were performed for testing the hypotheses H_0: $p(1) = p(2)$ vs. H_1: $p(1) \neq p(2)$ in order to distinguish between pleiotropy and linkage. Here, parameters $p(1)$ and $p(2)$ denote the position of the QTL having an effect on 1SWCB LDR and 1SCB LDR, respectively. The significance of QTL × trait interactions was tested by using trait means across all experiments at chromosomal regions, where pleiotropic QTL had been identified by joint CIM of both traits (H_0: $a_{1\cdot} = a_{2\cdot}$, $d_{1\cdot} = d_{2\cdot}$, where the dot notation indicates averaging over the respective index). LR tests for QTL × year interactions (H^*_{10}: $a_{11} = a_{12}$, $d_{11} = d_{12}$, and H^*_{20}: $a_{21} = a_{22}$, $d_{21} = d_{22}$) were performed for chromosomal regions, where QTL had been detected by joint CIM on single traits. All necessary computations for the above LR tests described in detail by Jiang and Zeng (1995) were performed using special software developed by C. Jiang and Z.B. Zeng (1996, personal communication).

The proportion of the total genome represented by all QTL regions for a specific trait was determined by dividing the sum of the lengths of the 1LOD support interval of each QTL by the total length of the RFLP linkage map. QTL identified separately in the CML131 × CML67 and Ki3 × CML139 populations were regarded as indistinguishable, if the RFLP markers nearest to the LR peak were located in the same chromosomal bin on the 1995 UMC maize linkage map, accessible through the USDA maize genome database, MaizeDB (http://www.agron.missouri.edu).

Critical Thresholds. For testing the presence of a QTL, a genome-wise significance level of $\alpha = 0.30$ was used, corresponding to a LOD threshold of 3.0. Because 89 marker intervals were scanned for QTL activity, the error rate of the test per interval (α') was calculated by a Bonferroni approximation as $\alpha' = 0.30/89 = 0.0034$. To ensure the comparison-wise significance level of $\alpha' = 0.0034$, we used as thresholds for

the joint CIM of a single trait across both years $\chi^2_{0.0034,5} = 17.8$ and for the joint CIM of both traits and years $\chi^2_{0.0034,9} = 24.9$. An experimentwise significance level of $\alpha = 0.05$ was employed in order to test the various genetic hypotheses. The significance level for each individual test was calculated as $\alpha' = 0.05/n$, where n equals the number of LR tests performed. Appropriate $\chi^2_{\alpha/n,df}$ values were chosen as thresholds for testing pleiotropy vs. linkage with $df = 1$ and pleiotropy, QTL × trait interactions, and QTL × year interactions with df = 2. LOD values corresponding to the LR thresholds can be obtained

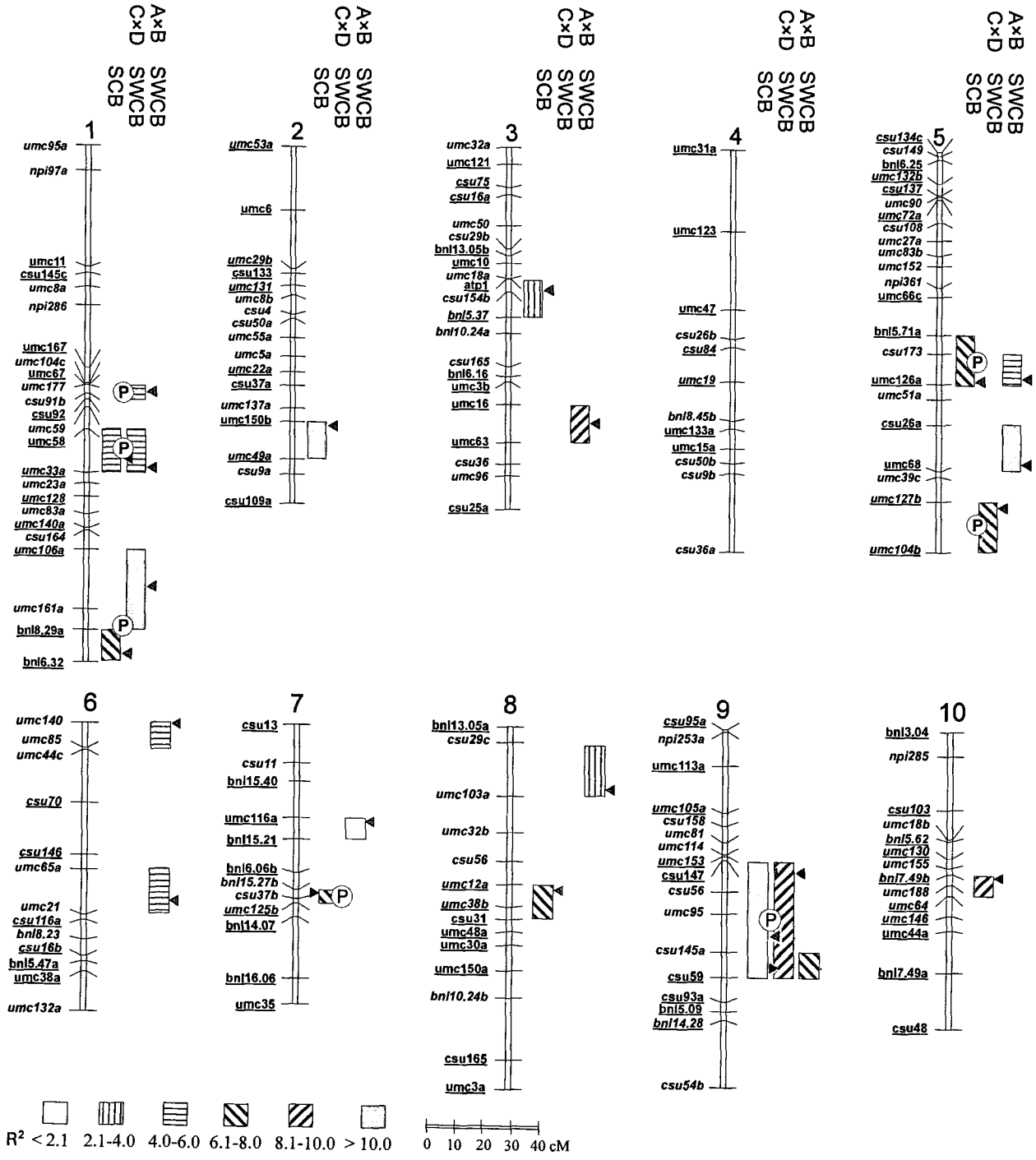

Fig. 1. Combined RFLP linkage map of maize based on F_2 individuals from crosses Ki3 × CML139 (A×B) and CML131 × CML67 (C×D) as well as QTL detected for leaf feeding resistance against 1SWCB larvae in F_3 lines of population Ki3 × CML139 and against 1SWCB and 1SCB larvae in F_3 lines of population CML131 × CML67. The marker interval with the maximum likelihood ratio (LR) peak is indicated by boxes. The box pattern is associated with the phenotypic variation explained by the respective QTL and triangles mark LR peak position. QTL with pleiotropic effects on both traits within population CML131 × CML67 are indicated by a circled P. Underlined RFLP loci in normal letters are common to both mapping populations, whereas underlined RFLP loci in italics are unique for population CML131 × CML67. All other markers are unique for population Ki3 × CML139.

by multiplying the latter with the transformation module $c = 1/(2ln10) \approx 0.217$.

Variance Explained. Considering that $(\hat{a}_k^2/2 + \hat{d}_k^2/4)$ refers to the variance among F_3 lines (Falconer, 1989) contributed by the kth QTL, we estimated the phenotypic variance explained by the kth detected QTL as $R_k^2 = (\hat{a}_k^2/2 + \hat{d}_k^2/4)/\hat{\sigma}_p^2$, where the additive $(\hat{a}_k^2)$ and dominance $(\hat{d}_k^2)$ effects of the kth QTL were determined by applying Model B. The genotypic variance explained by all QTL was estimated by fitting a model to the adjusted entry means from each environment, which included all QTL detected by joint CIM separately for 1SWCB LDR and 1SCB LDR. The sum of squares for genotypes obtained from the combined ANOVA was partitioned into the variation due to regression on the detected QTL and the residual variation. A similar partitioning was performed for the genotype × year interaction sum of squares. The total genetic variance explained by all QTL in the model $(\hat{\sigma}_q^2)$ was estimated by equating the mean squares to the expected mean squares as described by Bohn et al. (1996). Unlike the R_k^2 values, $\hat{\sigma}_q^2$ is not biased by QTL × year interactions. Hence, the proportion of $\hat{\sigma}_g^2$ explained by all QTL in the model (Q^2) was estimated as $Q^2 = \hat{\sigma}_q^2/\hat{\sigma}_g^2$. The multiple regression and the combined ANOVA were performed by using software PLABQTL (Utz and Melchinger, 1996).

The type of gene action at each QTL was characterized by calculating the dominance ratio $DR = |d_k/a_k|$ ratio: additive for $DR < 0.2$; partial dominance for $0.2 \leq DR < 0.8$; dominance for $0.8 \leq DR < 1.2$; overdominance for $DR \geq 1.2$.

RESULTS

Segregation and Linkage of RFLPs

The results of the RFLP analysis, including the linkage map for the F_2 population of the cross CML131 × CML67, based on 100 RFLP marker loci, have been presented in our companion paper (Bohn et al., 1996). Likewise, the RFLP linkage map obtained with the 475 F_3 lines from the Ki3 × CML139 population based on 128 RFLP marker loci were presented by Khairallah et al. (1997). In the combined RFLP linkage map, 58 markers were in common between the crosses CML131 × CML67 and Ki3 × CML139 (Fig. 1). The linear order of the common markers on the linkage map was consistent across both populations and in agreement with published maize maps (Gardiner et al., 1993).

Agronomic Trait Analysis

The distribution of phenotypic means of F_3 lines for 1SWCB LDR and 1SCB LDR followed approximately a Gaussian distribution, with the parent lines representing the extremes. The overall mean of the 215 F_3 lines $(\bar{F}_3)$ for 1SWCB LDR was significantly $(P < 0.05)$ greater than the mean performance of the two parent lines $(\bar{P})$ for 1SWCB LDR and was significantly greater than the $\bar{F}_3$ for 1SCB LDR (Table 1). Variance components $\hat{\sigma}_g^2$ and $\hat{\sigma}_{gy}^2$ of the F_3 lines were highly significant $(P < 0.01)$ for both traits and h^2 estimates were intermediate. Correlations of 1SWCB LDR with 1SCB LDR in the F_3 lines were highly significant $(\hat{r}_p = 0.62, \hat{r}_g = 1.02)$. Relative homozygosity of F_2 plants (determined from RFLP data) was not significantly correlated with 1SWCB LDR and 1SCB LDR in the descending F_3 lines. However, the estimated percentage of P1 genome in the F_2 plants showed highly significant $(P < 0.01)$ correlations with 1SWCB LDR $(\hat{r}_p = 0.55)$ and 1SCB LDR $(\hat{r}_p = 0.59)$.

Table 1. Means of parents CML131 and CML67, and 215 F_3 lines derived from their cross; and estimates of variance components and heritabilities among F_3 lines for 1SCB and 1SWCB leaf damage ratings (LDR) evaluated at one subtropical location in 2 yr.

Parameters	Entries	1SWCB LDR	1SCB LDR
	no.	1–10 scale†	
Means‡			
CML131	12	8.9 ± 0.2	8.4 ± 0.2
CML67	13	4.6 ± 0.2	4.3 ± 0.2
$\bar{P}$§	25	6.6 ± 0.2	6.4 ± 0.1
$\bar{F}_3$	215	6.9 ± 0.1	6.3 ± 0.1
Variance components (F_3 lines)			
$\hat{\sigma}_g^2$		0.33 ± 0.05**	0.29 ± 0.05**
$\hat{\sigma}_{gy}^2$		0.20 ± 0.04**	0.22 ± 0.04**
Heritability (F_3 lines)			
$\hat{h}^2$		0.64	0.59
90% C.I. on $\hat{h}^2$		(0.55–0.71)	(0.49–0.68)

** Variance component was significant at the 0.01 probability level.
† LDR was assessed using a rating scale from 1 (no visible leaf damage) to 10 (dead growing point, all leaves with long leasions).
‡ Standard errors are attached.
§ $\bar{P}$ = mean of CML131 and CML67, $\bar{F}_3$ = mean of F_3 lines.

QTL Analyses

1SWCB Leaf Damage Ratings. Seven markers were used as cofactors in the joint CIM for 1SWCB LDR across both years (Table 2). Six putative QTL affecting 1SWCB LDR were detected on Chromosomes 1 (three QTL), 5, 7, and 9, explaining between 1.6% and 14.9% of $\hat{\sigma}_p^2$ with LR values ranging from 20.2 to 38.2. All alleles increasing the level of resistance (i.e., decreasing 1SWCB LDR) were contributed by the resistant parent CML67. Two QTL showed additive gene action, one partial dominance, one dominance, and two QTL displayed overdominance. A simultaneous fit with all six putative QTL explained a total of 32.4% of $\hat{\sigma}_p^2$ and 53.3% of $\hat{\sigma}_g^2$. QTL × year interactions were significant $(P < 0.05)$ for the first QTL on Chromosome 1 and the QTL on Chromosome 7.

1SCB Leaf Damage Ratings. For 1SCB LDR, 12 RFLP markers were selected as cofactors (Table 2). Nine putative QTL located on Chromosomes 1 (two QTL), 2, 3, 5, 7, 8, 9, and 10 were found to affect 1SCB LDR. The LR values ranged from 18.3 on Chromosome 10 to 95.6 on Chromosome 9. The latter QTL explained 30.8% of $\hat{\sigma}_p^2$, whereas the other QTL explained between 3.8% and 20.2% of $\hat{\sigma}_p^2$. All alleles reducing 1SCB LDR (i.e., improving resistance) were contributed by the resistant parent CML67, except for the QTL on Chromosome 2. One QTL showed additive gene action, five partial dominance, and three displayed dominance or overdominance. All putative QTL showed no interaction with years. A simultaneous fit of all 10 putative QTL accounted for a total of 60.2% of $\hat{\sigma}_p^2$ and 98.2% of $\hat{\sigma}_g^2$.

Joint Analysis of 1SWCB and 1SCB Leaf Damage Ratings. In the joint CIM for 1SWCB and 1SCB LDR, we detected 10 putative QTL on Chromosomes 1 (three QTL), 3, 5 (two QTL), 7 (two QTL), 9, and 10 by using 14 markers as cofactors (Table 3). The LR values ranged

Table 2. Parameters associated with QTL for 1SWCB and 1SCB leaf damage ratings (LDR). Biometric parameters were estimated from phenotypic data of 171 F_3 lines from cross CML131 × CML67 evaluated at one subtropical location in 2 yr.

Bin§	Marker interval	Pos.¶	LR† for QTL position	R_k^2	Genetic effect‡		Gene action#	QTL × year interaction
					Add.	Dom.		
		cM	LR‡‡	%	— 1–10 scale§§ —			LR¶¶
1SWCB LDR††								
1.06	*umc67-csu91b*	58	24.6*	4.4	−0.02	−0.30	OD	13.7*
1.07	*umc58-umc33*	108	36.1*	5.2	−0.23	−0.04	A	6.3
1.10	*umc106-bnl8.29a*	154	38.2*	14.9	−0.31	0.34	D	7.1
5.07	*umc127b-umc104b*	141	21.6*	6.3	−0.19	0.24	OD	4.3
7.02	*umc116a-bnl15.21*	32	20.2*	1.6	−0.13	0.01	A	12.5*
9.05	*csu147-csu59*	68	30.0*	8.1	−0.26	0.18	PD	3.3
1SCB LDR##								
1.07	*umc58-umc33*	103	20.1*	5.7	−0.18	0.32	OD	5.2
1.12	*bnl8.29a-bnl6.32*	182	19.2*	6.4	−0.26	−0.10	PD	1.2
2.07	*[umc131-umc22a]*†††	114	15.4	7.3	−0.26	0.09	PD	0.2
2.08	*[umc150b-umc49a]*	141	25.4*	20.2	0.28	−0.49	OD	1.3
3.05	*atp1-bnl5.37*	48	25.8*	3.8	−0.19	0.04	PD	4.7
5.06	*bnl5.71a-umc126a*	85	27.3*	7.6	−0.22	−0.23	D	2.8
7.04	*bnl15.27b-umc125b*	66	21.5*	7.0	−0.23	−0.18	PD	2.4
8.05	*umc12a-csu31*	66	19.9*	7.5	−0.24	−0.18	PD	0.2
9.05	*csu147-csu59*	84	95.6*	30.8	−0.55	−0.02	A	3.9
10.03/4	*[bnl7.49b-umc64]*	84	18.3*	8.4	−0.27	0.14	PD	4.4

* Test was significant at the respective threshold defined below.
† LR = Likelihood ratio.
‡ Genetic effects were estimated by Model B; QTL alleles with negative effects were contributed by the resistant parent CML67 and QTL allele with positive effects were contributed by the susceptible parent CML131.
§ Bin locations are designated by an X.Y code, where X is the linkage group containing the bin and Y is the location of the bin within the linkage group (Gardiner et al., 1993).
¶ Position of likelihood ratio peak (maximum LR) in cM relative to the first marker on the chromosome according to the RFLP linkage map presented by Bohn et al. (1996).
# A = additive gene action ($|d_k/a_k| < 0.2$), PD = partial dominance ($0.2 < |d_k/a_k| < 0.8$), D = dominance ($0.8 < |d_k/a_k| < 1.2$), OD = overdominance ($|d_k/a_k| > 1.2$).
†† Marker cofactors: *umc33a, bnl8.29a, bnl5.71, cdc48, bnl15.27b, umc48a, csu147.*
‡‡ Threshold of LR-test for QTL detection: $\chi^2_{0.30/89.5} = 17.8$, corresponding to LOD = 3.0.
§§ LDR was assessed using a rating scale from 1 (no visible leaf damage) to 10 (dead growing point, all leaves with long leasions).
¶¶ Thresholds of the LR-test used for the test of QTL × year interactions are $\chi^2_{0.05/6.2} = 9.6$ for 1SWCB ratings, $\chi^2_{0.05/10.2} = 10.6$ for 1SCB ratings.
Marker cofactors: *umc167, bnl6.32, csu46, atp1, umc31a, bnl5.71, umc127b, bnl15.27b, umc12, umc153, csu59, bnl7.49b.*
††† Brackets indicate QTL identified with Model B.

from 25.1 on Chromosome 7 to 102.4 on Chromosome 9. Most putative QTL detected in joint CIM of single traits (Table 2) were also found in the joint CIM of multiple traits. The only exceptions were the QTL detected for 1SCB LDR on Chromosomes 2 and 8, which were not found by joint CIM with multiple traits. In all those cases, where a QTL was confirmed in the joint CIM of 1SWCB and 1SCB LDR, the LR value was substantially increased in comparison to the LR value for the joint CIM of the single traits.

Seven out of 10 QTL identified by joint CIM for both traits showed significant ($P < 0.05$) pleiotropic effects on 1SWCB LDR and 1SCB LDR (Table 3). All but one of them showed no significant QTL × trait interaction between 1SWCB LDR and 1SCB LDR. The only exception was the QTL on Chromosome 9, which showed a large effect for 1SCB LDR, but a smaller effect for 1SWCB LDR.

DISCUSSION

At CIMMYT, diverse sources of resistance to maize borer species were combined to form the MBR population (CIMMYT Population 590). Smith et al. (1989) demonstrated that full-sib families derived from the most resistant S_1 progenies of the MBR population showed resistance to various stem borer species. In a diallel study of 45 F_1 crosses between resistant CIMMYT lines (originating from the MBR population and Antigua germplasm) and susceptible inbreds, Thome et al. (1992) reported extremely high correlations between LDR for 1SWCB, 1SCB, and 1ECB ($r_{SWCB \times SCB} = 0.89$, $r_{ECB \times SWCB} = 0.76$, $r_{ECB \times SCB} = 0.89$). These studies showed that by means of conventional breeding methods, maize genotypes with multiple maize stem borer resistance can be developed. QTL studies can help to answer the question of whether the resistance to multiple insect species is the sum of diverse resistance mechanisms, which are different for each single insect species (Painter, 1951), or partly due to resistance genes with pleiotropic effects.

Genetic Foundation of Resistance to 1SWCB and 1SCB Leaf Feeding

We identified six putative QTL with significant effects on 1SWCB resistance in a cross between a tropical and a subtropical inbred. The parental inbreds were selected on the assumption that CML67 contained all resistance alleles, whereas CML131 had no resistance genes. In agreement with this hypothesis, no transgression of the parental LDR means was observed in the population of 215 F_3 lines derived from their cross. In agreement with the conclusions drawn from phenotypic data, all QTL alleles conferring resistance to 1SWCB and 1SCB leaf feeding were contributed by the resistant parent CML67, except the QTL detected on Chromosome 2 for 1SCB LDR.

Table 3. Joint QTL mapping for 1SWCB and 1SCB leaf damage ratings (LDR) and tests of genetic hypothesis for QTL effects. Biometric parameters of QTL effects were estimated from phenotypic means of 171 F_3 lines from cross CML131 × CML67 evaluated in separate trials at one subtropical location in 2 yr.

				Genetic hypotheses tested							
				Genetic effects†		Pleiotropy		QTL × trait		Pleiotropy vs. linkage	
Bin‡	Marker interval	Pos.§	LR for QTL position¶	Add.	Dom.	Hypothesis#	LR††	Hypothesis‡‡	LR§§	Hypothesis¶¶	LR##
		cM	LR	1–10 scale‡‡‡							
Maize stem borer resistance†††											
1.06	*umc167-umc67*	52	33.8*	−0.20	−0.42	$a_{21} = a_{22} = 0$ $d_{21} = d_{22} = 0$	14.6*	$a_1 = a_2$ $d_1 = d_2$	2.2		
1.07	*umc58-umc33a*	102	44.2*	−0.10	−0.25			$a_1 = a_2$ $d_1 = d_2$	7.7	$p(1) = p(2)$	2.2
1.11	*umc106a-bnl8.29a*	156	44.8*	−0.20	0.26			$a_1 = a_2$ $d_1 = d_2$	9.4	$p(1) = p(2)$	2.8
3.05	*atp1-bnl5.37*	48	27.3*	−0.15	−0.02	$a_{11} = a_{12} = 0$ $d_{11} = d_{12} = 0$	11.8				
5.06	*umc126-csu26*	88	30.7*	−0.11	−0.10	$a_{11} = a_{12} = 0$ $d_{11} = d_{12} = 0$	14.5*	$a_1 = a_2$ $d_1 = d_2$	1.4		
5.07	*umc127b-umc104b*	140	26.7*	−0.09	−0.06	$a_{21} = a_{22} = 0$ $d_{21} = d_{22} = 0$	16.5*	$a_1 = a_2$ $d_1 = d_2$	8.3		
7.02	*umc116a-bnl15.21*	30	25.1*	−0.07	0.06	$a_{21} = a_{22} = 0$ $d_{21} = d_{22} = 0$	10.3				
7.04	*bnl15.27b-umc125b*	62	30.6*	−0.18	0.04	$a_{11} = a_{12} = 0$ $d_{11} = d_{12} = 0$	14.3*	$a_1 = a_2$ $d_1 = d_2$	4.3		
9.05	*csu147-csu59*	82	102.4*	−0.37	0.10			$a_1 = a_2$ $d_1 = d_2$	29.8*	$p(1) = p(2)$	1.3
10.03/4	[*bnl7.49b-umc64*]§§§	84	26.2*	−0.25	0.23	$a_{11} = a_{12} = 0$ $d_{11} = d_{12} = 0$	10.1	$a_{1.} = a_{2.}$ $d_{1.} = d_{2.}$			

* Test was significant at the respective threshold defined below.
† Genetic effects were estimated by Model B; QTL alleles with negative effects were contributed by the resistant parent CML67 and QTL alleles with positive effects were contributed by the susceptible parent CML131.
‡ Bin location is designated by an X.Y code, where X is the linkage group containing the bin and Y is the location of the bin within the linkage group (Gardiner et al., 1993).
§ Position of likelihood peak peak (maximum LR) in cM relative to the first RFLP marker on the chromosome according to RFLP linkage map presented by Bohn et al. (1996).
¶ Threshold of LR-test used for QTL detection: $\chi^2_{0.30/89,9} = 24.9$, corresponding to LOD = 3.0.
# a_{1l}, d_{1l}, and a_{2l}, d_{2l} represent the additive and dominance effects of QTL for SWCB and SCB ratings in year l, respectively.
†† Threshold of LR-test used for testing pleiotropy of QTL: $\chi^2_{0.05/7,4} = 14.0$.
‡‡ $a_{1.}$, $d_{1.}$, and $a_{2.}$, $d_{2.}$ represent the average gene effects of QTL for SWCB and SCB ratings, respectively.
§§ Threshold of LR-test used for testing QTL × trait interaction: $\chi^2_{0.05/7,2} = 9.9$.
¶¶ $p(1)$ and $p(2)$ indicate the QTL positions for both traits.
Threshold of LR-test used for testing pleiotropy vs. close linkage: $\chi^2_{0.05/3,1} = 5.7$.
††† Marker cofactors: *umc167*, *umc33a*, *bnl6.32*, *umc22a*, *atp1*, *umc31a*, *bnl5.71*, *umc127b*, *cdc48*, *bnl15.27b*, *umc48*, *umc153*, *csu59*, *bnl7.49b*.
‡‡‡ LDR was assessed using a rating scale from 1 (no visible leaf damage) to 10 (dead growing point, all leaves with long leasions).
§§§ Brackets indicate QTL identified only with Model B.

Additive genetic effects were the major source of genetic variation for most QTL affecting 1SWCB LDR. A preponderance of additive gene action was also reported in previous studies on resistance to 1SWCB and 1SCB leaf feeding damage (Bohn et al., 1996; Hinderliter, 1983; Thome et al., 1992). These findings for individual QTL were corroborated by the association of 1SWCB LDR and 1SCB LDR with the percentage of CML131 genome in F_2 plants ($0.55 < r_p < 0.59$). However, a substantial proportion of the genetic variation was attributable to dominance ($d \sim 0.38a$). The sum of dominance effects of individual QTL was significantly different from zero, indicating heterosis for 1SWCB LDR (i.e., susceptibility), even though the generation means $\bar{P}$ and $\bar{F}_3$ did not differ significantly.

We performed a joint CIM on both traits to answer the question of whether resistance to 1SWCB and 1SCB leaf feeding was affected by the same chromosomal regions. Seven out of 10 genomic regions were identified with significant ($P < 0.05$) pleiotropic effects on both insect species. By fitting all seven QTL with pleiotropic effects in one model, 55 and 64% of $\hat{\sigma}^2_g$ could be explained for 1SWCB LDR and 1SCB LDR, respectively. This result in combination with the high genotypic correlation between 1SWCB LDR and 1SCB LDR suggests that the antibiotic type of resistance against 1SCB and 1SWCB found in CML67 has largely the same genetic foundation.

QTL positions on Chromosomes 3, 7, and 10 with non-pleiotropic effects could be insect-species specific. However, it cannot be ruled out that this result is merely attributable to the limited power of QTL detection due to the population size available for our study. Even if 1SWCB and 1SCB resistance were governed by the same set of QTL, this would not guarantee the simultaneous detection of every QTL for both traits.

Consistency of QTL across Populations

The prospects of MAS in breeding programs depend heavily on the extent to which QTL results can be extrapolated from one population to another population, because sizeable experiments are required to gain reliable information on QTL number, location, and genetic effects. Information about the consistency of QTL for 1SWCB resistance across different populations of F_3

lines can be gained by comparison with results obtained from a second cross, Ki3 × CML139 (Khairallah et al., 1997), for which the same QTL detection and mapping procedure was applied. In this study, seven QTL for 1SWCB leaf feeding resistance were identified on Chromosomes 3, 5 (two QTL), 6 (two QTL), 8, and 9 (Fig. 1). Most alleles increasing the 1SWCB resistance (i.e., decreasing the 1SWCB LDR) were contributed by the resistant parent CML139.

Three genomic regions on Chromosomes 5 (bins 5.06 and 5.07) and 9 (bin 9.05) were common across both mapping populations. This small number of common QTL between both populations is consistent with previous reports in the literature. Beavis et al. (1991) found no common QTL for plant height across four $F_{2:4}$ mapping populations in maize. Similarly, Bubeck et al. (1993) detected unique sets of QTL for grey leaf spot (caused by *Cercospora zeae-maydis* Theon and Daniels) resistance in three populations of F_3 families with only one QTL in common across all three populations.

Several reasons may explain the observed lack of consistency between QTL identified in different populations. First, the power of QTL detection is a function of the population size. The QTL for 1SWCB resistance in crosses of CML131 × CML67 and Ki3 × CML139 explained only about half of $\hat{\sigma}^2_g$, indicating that due to sampling effects, several QTL remained undetected in each population. The power of QTL detection can be improved by increasing the population size. However, even with the unprecedented high number of F_3 lines used in the cross Ki3 × CML139 (n = 475), a large proportion of $\hat{\sigma}^2_g$ remained unaccounted for by the detected QTL.

Second, inconsistencies between QTL results may reflect the fact that different sets of QTL for LDR segregate in the two crosses. Both resistant inbreds CML67 and CML139 have landrace 'Antigua Group 2' as one common ancestor (CIMMYT, 1991). However, as indicated by their pedigrees, different additional sources of insect resistance were combined to form the base populations from which the inbreds were developed. Answering the question of whether the same resistance mechanisms are active in both populations may help to clarify the reasons for the lack of congruency between both populations.

Third, in a segregating population, a QTL can be detected only if both parental inbred lines contributed different alleles at the QTL. The susceptible inbred Ki3 was developed from Suwan-1, which originated from the Thai Composite #1. In this composite, 36 germplasm sources were combined, including landrace Antigua Group 2 and an insect resistant synthetic [Antigua Group 2-'Veracruz 181'] (Sriwatanapongse et al., 1993). Based on their pedigree, it is possible that Ki3 and CML139 may carry resistance alleles identical by descent at some QTL. By contrast, no resistant progenitor is known in the pedigree of the susceptible inbred CML131. Therefore, a putative common QTL may be polymorphic in the CML131 × CML67 population, but monomorphic in Ki3 × CML139. In addition, different alleles with varying effects on resistance to leaf feeding damage may segregate in each of the populations. As a consequence, the QTL allele with a large effect will be detected, whereas the QTL allele with a smaller effect will remain undetected unless the population size is very large or more precise methods of evaluation of insect damage are employed.

Epistatic interactions between QTL in each of the mapping populations may also account for the observed lack of consistency, because in this case the difference between QTL genotype classes depends on other QTL segregating in the genetic background (Stuber, 1995). However, we found no significant digenic epistatic interactions among the detected QTL using the same procedure as described by Lübberstedt et al. (1997).

Method of QTL Detection

Interval mapping has become the standard method for QTL analyses since its proposal by Lander and Botstein (1989). In this study, we used the new method of joint CIM developed by Jiang and Zeng (1995) for QTL mapping, because it offers several advantages over simple interval mapping. First, the power of QTL detection was increased by using selected markers as cofactors in the regression model. This facilitates the detection of QTL linked in repulsion phase and helps to avoid the erroneous detection of "ghost QTL" (Martinez and Curnow, 1992). As our study shows, multiple linked QTL cannot be regarded as rare exceptions (Table 2). The use of cofactors enabled us to detect the QTL for 1SCB LDR on Chromosome 2 (cM = 141, a = −0.26), which was linked in repulsion phase with a second putative QTL (cM = 114, a = 0.28) yielding a non-significant LR peak.

Second, the test statistic for the joint analysis of correlated traits is generally higher than for the single trait QTL analyses (Jiang and Zeng, 1995). In all cases, where a QTL was detected in the joint CIM for 1SWCB LDR and 1SCB LDR, the LR value was substantially increased in comparison to the LR value of the separate analyses (increments varied between 6–240%). Because of the high phenotypic and genotypic correlations between both insect LDR, the LR values were increased, even if the QTL position showed a significant effect for only one trait. However, Jiang and Zeng (1995) pointed out that the increased test statistic will not necessarily increase the power of QTL detection, because more parameters have to be fitted in the model, which leads to higher thresholds for the test. In our study, one QTL for 1SCB LDR on Chromosome 8 remained undetected in joint CIM with both traits, because the increase in the test statistic did not compensate for the higher threshold.

Third, the joint CIM provides the statistical basis for testing the hypotheses of pleiotropy, pleiotropy vs. linkage, QTL × trait, and QTL × environment interactions using the test procedures developed by Jiang and Zeng (1995). Hitherto, no other method of QTL detection allowed testing these important genetic hypotheses. However, when QTL are tightly linked (<20 cM), the power of the test to distinguish between pleiotropy and

close linkage is low, unless the population size is very large. Improved resolution can be expected from QTL mapping with populations of recombinant inbred lines currently underway at CIMMYT with materials derived from the two populations compared in this study. The ultimate verification of pleiotropy vs. close linkage can only be achieved by identifying and cloning the gene or genes involved in those QTL.

The R_k^2 values for individual QTL were calculated based on their additive and dominance genetic effects. The genetic effects were estimated using linked markers as cofactors (Model B) to reduce the influence of putative linked QTL in adjacent regions. With regard to the population size used in this study, three QTL explained unexpectedly small proportions of $\hat{\sigma}_p^2$ ($R_k^2 < 5\%$). At least in two instances, the observed discrepancy between the high LR values and the small R_k^2 values at these QTL positions could be explained by large QTL × year interactions.

It should be pointed out in this context that the control of Type I error in a genome-wide search for QTL remains to be a problem because the distribution of the test statistic under the null hypothesis is often not clear and multiple tests are performed. In this study, the Type I error rate per marker interval was determined by applying the Bonferroni correction assuming independency of tests in different intervals. In practice, the total number of independent tests is lower than supposed in the Bonferroni correction due to (i) correlation between tests caused by small marker intervals with rare recombination and the use of markers as cofactors and (ii) the inability to separate linked QTL in neighboring intervals. Therefore, the actual genome-wise significance level in the present study can be expected to be lower than $\alpha = 0.30$. In contrast to this parametric approach, which assumes normality for the residuals, a permutation-based method for estimating empirical thresholds for a given set of experimental data was recently proposed (Churchill and Doerge, 1994; Doerge and Churchill, 1996). Application of this method to CIM requires further research and development of appropriate software.

In a companion study, Bohn et al. (1996) identified 10 QTL for 1SCB LDR. Seven of these QTL were also found in the present study by reanalyzing a subset of two environments from the original 1SCB LDR data. In addition, one QTL on Chromosome 5 was consistent with a pleiotropic QTL for 1SWCB LDR detected in the current study. Only two QTL on Chromosomes 2 and 9 were not consistent across both studies reflecting the influence of the third environment and/or different implementations of CIM.

Table 4. Chromosomal bin location of QTL involved in the antibiotic type of resistance against 1SWCB and 1SCB leaf feeding damage in this study and insect resistance and disease resistance genes and QTL found in the same chromosomal regions in other studies (data from McMullen and Simcox, 1995, modified).

Bin location†	Insect resistance (population)‡	Disease resistauce
1.07	2ECB§: QTL (B73 × B52)	*Cochliobolus carbonum* Nelson (Carbonum leaf spot): *hm1*; *Gibberella zeae* (Schwein.) (Fusarium stalk rot): QTL
1.11	2ECB: QTL (B73 × B52)	
3.04	2ECB: QTL (B73 × B52, B73 × DE811)	Fusarium stalk rot: QTL; *Puccinia sorghi* (Schwein.) (Common rust): *rp3*; Maize dwarf mosaic virus; Wheat streak mosaic virus
5.06	2ECB: QBL (B73 × DE811)	*Setosphaeria turcica* (Luttrell) (Northern corn leaf blight): QTL
7.04	2ECB: QTL (B73 × B52, B73 × DE811)	
9.05	2ECB: QTL (Mo17 × B52)	Carbonum leaf spot: *hm2*
10.04	2ECB: QTL (B73 × B52, Mo17 × B52)	

† Chromosomal bin locations refer to the 1995 UMC maize RFLP linkage map. Bin locations are designated by an X.Y code, where X is the linkage group containing the bin and Y is the location of the bin within the linkage group.
‡ QTL detected in F_3 population derived from crosses B73 × B52 (Schön et al., 1993), B73 × DE811 (Lee, 1993), and Mo17 × B52 (Lee, 1993).
§ 2ECB, second generation of European corn borer (*Ostrinia nubilalis* Hübner).

Clusters of Resistance Genes

Based on a review of recent literature, McMullen and Simcox (1995) reported that the majority of disease and insect resistance genes or QTL occur on all maize chromosomes in clusters with the exception of Chromosomes 7 and 9. They used the chromosomal bin location (taken from the 1995 UMC maize RFLP linkage map) in order to group genes and QTL identified in a wide range of maize germplasm. QTL with significant effects on leaf feeding resistance for 1SWCB and 1SCB detected in the joint CIM of both *Diatraea* spp. on Chromosomes 1 (two QTL), 3, 5, 7, 9, and 10 were located in chromosomal bins containing genes or QTL for 2ECB resistance and QTL for resistance to fungal or virus diseases (Table 4). The former were identified in three populations of F_3 lines derived from crosses between susceptible (B73, Mo17) and resistant (B52, DE811) maize inbreds (Lee, 1993; Schön et al., 1993). Three QTL regions with effects on all three maize stem borer species were in common across three mapping populations and four QTL positions were identified simultaneously in two populations. This demonstrates a close relationship between 1SWCB, 1SCB, and 2ECB resistance at the level of the genome.

In addition, the two QTL for 1SCB leaf feeding resistance, which were not detected by joint CIM of both *Diatraea* species (Table 2), were located in bins 2.08 and 8.05, clustering with QTL for 2ECB, northern corn leaf blight [*Setosphaeria turcica* (Luttrell) K.J. Leonard E.G. Suggs], and grey leaf spot resistance. However, no information is yet available on the functional relationship between genes and QTL located in the same bin for different maize diseases and pests.

Marker-Assisted Selection

Based on their theoretical investigations, Lande and Thompson (1990) suggested the use of molecular

marker information to increase the efficiency of selection in those cases, where the heritability of the trait is low and the ratio $Q^2 = \hat{\sigma}_q^2/\hat{\sigma}_g^2$ is high. The relative efficiency (RE) of a single cycle of marker-based selection in comparison with conventional selection can be estimated as $RE = \sqrt{\hat{\sigma}_q^2/\hat{\sigma}_g^2\,\hat{h}^2}$, given the same selection intensity for both selection schemes. For population CML131 × CML67, the predicted RE for using MAS to improve 1SWCB resistance is 0.85, demonstrating that due to the high heritability ($\hat{h}^2 = 0.64$) and the moderate proportion of $\hat{\sigma}_g^2$ explained by the putative QTL, conventional selection methods may work more effectively. However, for improving 1SCB resistance, the estimated RE of MAS was 1.27 mainly because of the high proportion of $\hat{\sigma}_g^2$ explained by the 10 putative QTL. Due to the high genotypic correlation between 1SWCB LDR and 1SCB LDR and the pleiotropic QTL found between both traits, MAS selection for 1SCB resistance will also indirectly increase the level of 1SWCB resistance. In addition, the absence of QTL × year interactions for most QTL for 1SWCB LDR and 1SCB LDR suggests that a limited number of years will be sufficient to identify a suitable set of QTL for MAS. By applying cost efficient PCR based marker systems, MAS may be competitive over conventional selection even with a low RE, because conventional selection for insect resistance requires resource-demanding artificial infestation of maize plants with insect larvae.

In contrast to index selection based on markers, which was characterized by Stam (1994) as 'blind to the (putative) Mendelian factors underlying it,' he proposed a marker-assisted 'gene stacking' procedure. With this approach, QTL will be mapped in an initial step and subsequently favorable QTL alleles are accumulated by crossing those individuals most likely to produce the ideal genotype. The efficiency of this method depends on the accuracy of QTL detection, the number of putative QTL, the distance between the markers flanking the QTL, and how much of the genome they represent. In our study, the seven genomic regions with pleiotropic effects on both insect species represented 9.0% of the total genome and the length of the marker intervals carrying the QTL varied between 8 and 29 cM. These figures suggest that a transfer of all QTL with pleiotropic effects on leaf feeding resistance to both insect species into susceptible inbreds can be accomplished effectively by stacking marked chromosome segments harboring QTL through successive cycles of selection and crossing. Marker information can be employed in selecting for the desired genomic regions as well as against the undesirable remainder genome of donor line CML67.

Although the pleiotropic QTL for 1SWCB and 1SCB resistance explained only about 60% of $\hat{\sigma}_g^2$, it may be useful to transfer these QTL into susceptible germplasm as a meaningful contribution to an integrated pest management system. In addition, MAS may help to utilize different sources of quantitative resistances to SWCB and SCB by pyramiding the underlying QTL in a subsequent breeding process. Even a moderate improvement of host-plant resistance should increase yield stability and prevent substantial yield losses with minimal use of insecticides or other biological control measures.

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