Identification of QTL for resistance to head smut in maize (Zea mays. L)

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Abstract Head smut (HS) is one of the most devastating diseases of maize in spring production regions in China. Quantitative trait loci (QTL) for HS resistance were identified in this study to provide theoretical and applied tools for breeding HS resistance in maize. QTL associated with HS resistance were identified in a F_2:3 population derived from a T32 (highly resistant genotype) × HC (highly susceptible genotype) cross. Analysis in each of three environments and a collective analysis across all three environments were used to identify QTL in the F_2:3 population. A significant difference in HS resistance was found between the inbred lines, ‘T32’ and ‘HC’. Large genetic variation and transgressive segregation in the F_2:3 population were observed between the three different sites, Guian (GA), Huaxi (HX), and Pingba (PB). Two stable and novel QTL for resistance to HS were detected in the different environments that were located within the bnlg1014 to umc2224 (qHS1) interval on chromosome 1 and in the umc1006 to umc1857 (qHS6) interval on chromosome 6. Both QTL can be used for further fine mapping, marker-assisted selection breeding, and theoretical studies on HS resistance in maize.

Keywords Maize · Resistance · Head smut · Quantitative trait locus

Abbreviations
HS Head smut
QTL Quantitative trait loci
MAS Marker-assisted selection
GA Guian
HX Huaxi
PB Pingba
JA Joint analysis
cM Centimorgans
CIM Compound intervals mapping
HS1 An antidisease position on chromosome 1 in maize
NILs Near-isogenic lines

Introduction

Maize head smut (HS) is a fungal disease caused by Sporisorium reilianum (Sánchez et al. 2011), a systemic that infects maize seedlings. The pathogenic spores remain in the soil and on harvested seeds and infect newly germinated plants the following year (Zhang et al. 2013). The pathogen mainly infects...
individual seeds at the seedling stage and does not infect other established seedlings in the field (Potter 1914). HS primarily destroys the panicles of maize. Symptoms of HS in infected seedlings include dwarf shoots, clumped shoots, yellow striped stems, stem deformity, leaf abnormality, and leaf curling (Little et al. 2012). The panicle of infected plants produces powders of black spores, and infected panicles eventually become a 'black bag' of spores. HS, in recent years, has become more widespread and outbreaks have been more intense due to climate change and various management practices (Ren et al. 2014). HS has become one of the important diseases in the main maize production area of Heilongjiang province and even in the spring maize production areas of China (Wang et al. 2002; Zhao et al. 2012). The most effective way to control HS in maize is to breed and promote the use of disease-resistant varieties (Jin et al. 2003). The selection of resistant germplasm resources and the analysis of HS resistance are essential to maize breeding. In total, more than 7000 genotypes of maize have been identified as resistant to HS in studies conducted worldwide (Song et al. 2000; Gao et al. 2006; Guo et al. 2007). Results of germplasm screening have indicated that the most commonly used inbred lines are highly susceptible to HS and that the use of resistant germplasm resources is seriously deficient (Wang et al. 2001; Wang et al. 2004a, b; Meng et al. 2015).

Once a source of resistance has been identified, genetic analyses can be conducted to identify genetic markers that are closely linked with disease resistance, which can then be used for marker-assisted selection (MAS) (Qian et al. 2007). Many studies have utilized quantitative trait loci (QTL) mapping to identify markers for HS in maize. Gao (2005) used the F2:3 families from Mo17×HZ4 and detected disease-resistant QTL on chromosomes 1.02, 2.08 to 2.09, 2.09, 3.04, and 8.02. Chen et al. (2008) developed an SSR marker (SSR 148,152) using the BC2 population of HZS×J1037. Shi et al. (2009), utilizing two BC3 populations of HZS×Mo17 and HZS×Q319, developed a SCAR marker 5130 associated with HS resistance in maize, and demonstrated that the SSR markers bn1g1893, umc1525, and UIII1C2077 could effectively account for the main effect of the QTL. Yong et al. (2015) identified two resistant QTL on chromosomes 2 (q2.09 HR) and 5 (q5.03 HR) using a RIL population constructed from HZS×Q319. Lübbestedt et al. (1999) used 220 F3 families produced by the hybridization of two European maize inbred lines (D32×D145) identified 11 QTL using the compound interval mapping (CIM) method. Ten of the 11 QTL exhibited significant additive effects, while only the QTL on chromosome 3 showed a significant dominant effect. Liu et al. (2009) used 100 recombinant inbred lines of Hi34×Tzi17 as a mapping population. In that study, CIM detected only one QTL, while four QTL were located on chromosomes 1, 2, 9, and 10. The results of their analysis were not consistent with prior studies, however, most likely due to the use of different genotypes, environmental conditions, and research methods.

In the present study, maize inbred lines of T32 (a highly resistant genotype) and HC (a highly susceptible genotype), lines of germplasm that are commonly used in maize breeding research in the southwest area of China, were used to construct an F2 mapping and a F2:3 population for the evaluation of HS resistance. A genetic linkage map of F2 plants was constructed using 152 SSR markers across ten chromosomes. The map was combined with two years of field evaluation data on the resistance of artificially-inoculated plants grown at three different sites to identify loci associated with HS resistance. This effort provides a foundation for using MAS to develop maize varieties with resistance to HS.

Materials and methods

Plant material

The mapping population included 184 F2:3 families derived from randomly selected F2 plants of the cross between inbred lines T32 (a highly resistant genotype) and HC (a highly susceptible genotype), T32 and HC were selected from the inbred lines based on an analysis of resistance to HS (Tan et al. 2019). T32 is a line of tropical descent representing Thai germplasm Suwan1 C9, which is highly resistant to HS and possesses other excellent traits. T32 is well adapted to the southwest mountainous area of China and has been widely used in the development of maize hybrids used for the development of new varieties. HC is derived from Reid germplasm and is susceptible to HS, as well as other diseases of maize,
however, it is well-adapted to central and southwest regions of China. The F\textsubscript{2} individuals were produced between September, 2017 and January, 2018 in Hainan, the southernmost province of China. The F\textsubscript{2:3} families and their parents were planted in the spring of 2018 in Guian (GA) and in the spring of 2019 in Huaxi (HX) and Pingba (PB), Guizhou Province, located in the southwest of China (Fig. 1).

Field assessment of HS resistance

F\textsubscript{2:3} families and their parents were planted in three environments: GA (29.13°N, 106.25°E; 1356 m altitude) in 2018, and HX (26.43°N, 106.67°E; 1055 m altitude) and PB (26.22°N, 106.13°E; 1255 m altitude) in 2019. Phenotypic resistance to HS was assessed at the milk-ripe stage after artificial inoculation of plants with the HS fungus under conditions of low temperature and low humidity. A completely randomized block design with two replications and 20 plants in each plot was utilized at all three sites. All plants were naturally, cross-pollinated by the wind, and 13 plants from the middle of each plot were used to evaluate HS resistance.

Artificial inoculation and disease evaluation

Sori containing teliospores of *Sphacelotheca reiliana*, the causal agent of head smut in maize, were collected from the field in 2017 and stored in a dry, well-ventilated environment after drying in the shade. Spores were removed from the sori, filtered, and then mixed with soil at a ratio of 1:1000 prior to planting. Artificial inoculation was conducted by applying a mixture of soil and teliospores to cover each maize kernel planted in a seedling tray. The plants were transplanted into the field when 4–5 leaves were visible (Ali et al. 1990). Disease incidence was scored by examining the presence of sori either in ears or tassels as an indicator of susceptibility/resistance to HS in mature plants. A statistical analysis of the phenotypic data was performed using IBM SPSS Statistical 20.0 software. The variances for genotype, environment, and the interaction between genotype and environment were estimated using Microsoft Office Excel-based on the random model. The broad-sense heritability ($H^2$) was calculated as follows: $H^2 = \frac{\delta^2_G}{\delta^2_G + \frac{\delta^2_{GE}}{n} + \delta^2_E/nr}$, where $\delta^2_G$ represents the genetic variance, $\delta^2_{GE}$ represents the genotype and environment interaction variance, $\delta^2_E$ represents the error variance, and $n$ and $r$ are the numbers of environments and replications, respectively (Knapp et al. 1985).

**Linkage map construction for the F\textsubscript{2} population**

Genomic DNA was extracted from a young leaf samples of each plant using cationic detergent cetyltrimethyl ammonium bromide (CTAB) (Chen and Ronald 1999). The genotype of each individual was identified using 10% non-denaturing polyacrylamide gel electrophoresis and select genetic markers. A total of 177 polymorphic markers for T32 and HC were used to develop a genetic map using JoinMap version 4 (Jacobs et al. 1995). The markers were based on publicly available SSR markers retrieved from the Maize Genetics and Genomics Database (http://www.maizegdb.org) (Portwood et al. 2019). The linkage map was constructed from 152 SSR markers across ten chromosomes. The recombination frequency between linked loci was transformed into a genetic distance (centimorgans, cM) using the Kosambi’s function.

**QTL analysis**

A chi-square ($X^2$) test was used to determine if the SSR marker genotype separation ratio conformed to 1:2:1 and if the population gene frequency conformed to 1:1. Inclusive composite interval mapping was performed using QTL IciMapping software to identify the QTL and estimate their effects (Jian, 2009; Li et al. 2008, 2007). Parameters for the regression
analysis were set with a window size and walking speed of 10 and 1 cM, respectively. The significance threshold for the QTL detection was 1000 random permutations of the phenotypic data at the 5% level. The gene action mode of each significant QTL was estimated by the rate (D/A) of additive (A) and dominant (D) effects and classified into additive (A=0–0.20), partial dominant (PD=0.21–0.80), dominant (D=0.81–1.20), and overdominant (OD>1.20) (Edwards et al. 1987; Stuber et al. 1987; Tuberosa et al. 1998). QTL were named as follows: In GAHS1a, ‘GA’ represents the environment in which the QTL was identified (GA, HX, PB, and JA are abbreviated from Guian, Huaxi, Pingba, and Joint analysis, respectively); ‘HS’ indicates the trait (Head Smut); the number ‘1’ is the serial number of the chromosome; and ‘a’ presents the serial alpha code of the detected QTL. Other QTL were named in the same manner.

Results

Phenotypic variation of HS in F2:3 families and their parents

The t test for both parents within a site produced highly significant differences in HS resistance between the two parents (Table 1) (P<0.01). HS resistance in T32 was significantly higher than it was in HC. The incidence of HS in F2:3 families exhibited a continuous and approximately normal distribution at all three sites with low skewness and kurtosis, indicating that HS resistance was a typical quantitative trait controlled by multiple genes (Table 1, Fig. 2). A transgressive segregation of resistance values was observed in the population across the three sites. ANOVA indicated that the effect of genotype (F2:3 families), environment, and the interaction between genotype and environment on HS resistance were highly significant (P<0.01) (Table 2), and heritability was higher than 50.75%.

Linkage map

A total of 152 polymorphic markers across ten chromosomes were used to construct a linkage map for QTL mapping (Fig. 3, Table 3). The total map length was 1147.30 cM, and the average distance between markers was 7.55 cM. The average number of markers per chromosome was 15.2, ranging from 9 on chromosome 5–26 on chromosome 1. A few alterations in the marker order on a chromosome were observed compared to the position derived by the IBM 2008 Neighbors Frame 6. These slight alterations may be due to the different material used in the population.

QTL analysis for resistance to HS at individual sites (environments)

Twelve QTL for HS resistance were detected in F2:3 families, comprising three QTL at GA, five QTL at HX, and three QTL at PB (Table 4). Two of the loci were detected in more than one environment. QTL-GAHS1a, QTL-HXHS1a, and QTL-PBHS1a were located in the same genetic region in the interval between l bnlgl1014 and umc2224 (bins 1.01) (qHS1) on chromosome 1. This QTL explained 11.96%, 12.67%, and 13.03 of the phenotypic variation in the GA, HX, and PB populations, respectively. Negative additive effects of the three QTL indicated that the

Table 1 Phenotype evaluation of HS resistance in F2:3 families and their parents grown in three environments (sites)

| Site | Parents | F2:3 |
|------|---------|------|
|      | T32 (%) | HC (%) | Mean (%) | Range (%) | SDa | CVb (%) | Skewness | Kurtosis | Wc |
| GA   | 0**     | 35    | 13.38   | 0–40     | 8.15 | 60.91   | 0.73     | 0.61     | 0.959 |
| HX   | 0**     | 39    | 14.03   | 0–42     | 8.50 | 53.53   | 0.60     | 0.61     | 0.959 |
| PB   | 0**     | 38    | 14.16   | 0–43     | 8.20 | 57.93   | 0.78     | 1.03     | 0.957 |

a Standard deviation of phenotypic data, bCoefficient of variation of phenotypic data, cShapiro–Wilk statistic for the W test of normality, **significance with P<0.01
increasing trait values were contributed by T32, whose effect values were −0.23, −0.46, and −0.01 respectively. QTL-GAHS6a, QTL-HXHS6a, and QTL-PBHS6a were also located in a common genetic region in the interval between umc1006 and umc1857 (bin6.02–6.04) (qHS6) on chromosome 6. This QTL explained 16.11%, 12.19%, and 18.05% of the phenotypic variation in the GA, HX, and PB populations, respectively. Negative additive effects of the three QTL indicated that the increasing trait values were also contributed by T32, whose effect values were −4.65, −4.21, and −4.73, respectively. Among the twelve QTL, three QTL had additive effects, three QTL had partially dominant effects, and the others had an overdominant effect. The increasing effect of the alleles came from both parents. The mode of gene action was additive, partial dominant, and over dominant. QTL detection in the joint analysis across all three environments

Eighteen QTL associated with HS resistance were identified in the joint (combined) analysis across all three sites (Table 5). The two common genetic regions detected in the single environment analysis were also detected in the joint analysis at the same marker intervals with the relatively higher LOD values of 8.45 and 10.97. The one QTL found only in the GA population, two QTL found only in the HX population, and the one QTL found only in the PB in the single site analysis were also detected in the joint analysis in the intervals between the same markers. QTL-HXHS1b detected in the single site analysis and QTL-JAHS1b detected in the joint analysis were located near the genetic region with the same marker (umc1306). Some QTL detected in the joint analysis across all three environments (sites) were not detected in the single environment analyses.
Therefore, they probably have only a minor effect, and can only be detected in the larger dataset combining data from all three environments.

Discussion

In the present study, 184 F_{2:3} families were evaluated for HS resistance in three different environments (sites); GA, HX, and PB. The level of disease incidence in the three environments fell mostly between the levels present in the two parents, which conforms to the normal distribution rule. Heritability was also higher than 50.75%. This indicates that the HS resistance is inherited in a quantitative manner and has high heritability. This finding is consistent with the results reported by Zhang et al. (2002) and Bai (2009). Environmental and genetic factors have a

\[\text{Fig. 3 Distribution of QTL associated with head smut (HS) resistance in a F}_{2:3}\text{ population of maize derived from a T32 x HC cross plotted on a maize linkage map. The line segment indicates the marker intervals for the QTL, and the node on the line segment indicates the position of the QTL on the linkage map. QTL were named as follows: For GA/HS1a, 'GA' indicates the environment in which the QTL was identified (GA, HX, PB, and JA are abbreviations of Guian, Huaxi, Pingba, and Joint analysis of all three sites, respectively); 'HS' indicates the name of the trait; the number '1' is the serial number of the chromosome; 'a' presents the serial alpha code of the detected QTL.}\]
large effect on QTL detection. Different environments often result in a change in the magnitude of significant QTL effects or the direction of additive effects (Boer et al. 2007; Peng et al. 2011).

Three different environments were assessed in the present study to increase the reliability and consistency of QTL that were detected for HS resistance. HS resistance was a quantitative trait controlled by several QTL in all three of the environments when analyzed singly or jointly. Some of the QTL were identified in the analysis of individual sites and other only in the joint analysis of all three environments. Two genetic regions were commonly detected in the three environments, while other QTL were not stable and were not consistently identified across all sites. Due to the differences in soil and climate in the three environments, some QTL accounted for a relatively large amount of variance at a single site but were not detected at all in the other sites. QTL that are extremely stable in different sites are essential if they are to be used in MAS programs, for fine mapping, and map-based cloning in maize.

Comparisons of the two QTL identified in the present study, located on chromosome 1 (1.01) and chromosome 6 (6.02–6.04), with QTL detected by other groups, resulted in the identification of one similar region in the maize genome. The QTL in bin 1.01 in our study was previously reported by Shi et al. (2005), Lu and Brewbaker (1999), and Chen et al. (2008). Another QTL detected in bin 6.02/4 in our

Table 3 Information on the linkage map constructed for an F_2 population of maize

| Chromosome | Markers | Cover the chromosome length (cM) | Average map distance (cM) |
|------------|---------|---------------------------------|--------------------------|
| 1          | 23      | 199.449                         | 8.672                    |
| 2          | 17      | 119.008                         | 7.000                    |
| 3          | 12      | 86.085                          | 7.171                    |
| 4          | 18      | 127.224                         | 7.068                    |
| 5          | 9       | 81.611                          | 9.068                    |
| 6          | 9       | 66.999                          | 7.444                    |
| 7          | 15      | 147.152                         | 9.810                    |
| 8          | 12      | 70.272                          | 5.856                    |
| 9          | 12      | 102.797                         | 8.566                    |
| 10         | 18      | 102.758                         | 5.708                    |

Table 4 QTL associated with HS resistance as determined in the analysis of an F_{2:3} population of maize grown in single environments (sites)

| Site | Chr | QTL name | Bins | Interval markers | Position | LOD score | PVE | Est A^a | Est D | Gene action |
|------|-----|----------|------|------------------|----------|-----------|-----|---------|-------|-------------|
| GA   | 1   | GAHS1a   | 1.01 | bnlg1014-umc2224| 20.00    | 2.48      | 11.96| −0.23   | 4.32  | OD          |
|      | 6   | GAHS6a   | 6.02–6.04 | umc1006-umc1857| 45.00    | 4.32      | 16.11| −4.65   | −4.68 | D           |
|      | 9   | GAHS9a   | 9.01 | phi033-umc1867  | 102.00   | 2.15      | 6.29 | 2.13    | −0.77 | PD          |
| HX   | 1   | HXHS1a   | 1.01 | bnlg1014-umc2224| 20.00    | 3.82      | 12.67| −0.46   | 5.62  | OD          |
|      | 1   | HXHS1b   | 1.09–1.10 | umc1306-umc2189| 155.00   | 2.63      | 5.94 | 2.37    | −1.80 | PD          |
|      | 3   | HXHS3a   | 3.02–3.03 | bnlg1325-bnlg1447| 17.00    | 2.87      | 9.07 | 0.43    | 4.48  | OD          |
|      | 4   | HXHS4a   | 4.01–4.03 | umc2281-umc1757| 110.00   | 2.23      | 5.62 | 2.14    | 1.77  | D           |
|      | 6   | HXHS6a   | 6.02–6.04 | umc1006-umc1857| 45.00    | 5.63      | 12.19| −4.21   | −4.50 | D           |
| PB   | 1   | PBHS1a   | 1.01 | bnlg1014-umc2224| 20.00    | 2.99      | 13.03| −0.01   | 5.02  | OD          |
|      | 3   | PBHS3a   | 3.02–3.03 | bnlg1325-bnlg1447| 18.00    | 2.31      | 7.42 | 0.45    | 3.72  | OD          |
|      | 6   | PBHS6a   | 6.02–6.04 | umc1006-umc1857| 45.00    | 5.34      | 18.05| −4.73   | −4.89 | D           |

^a A, additive effect of the QTL, negative values indicate that the alleles for increasing the HS resistance are contributed by T32, positive values indicate that the alleles for increasing HS resistance are contributed by the HC parent
study is different from the QTL identified in bin 6.07 of the maize genome by Chen et al. (2008). Notably, the intervals of the two QTL identified in the present study do not overlap the map position for HS resistance reported in other studies, potentially indicating the discovery of novel loci for HS resistance in our study. The two major QTL in bin 1.01 and 6.02/4 represent good choices for resistance gene cloning and for use in marker-assisted selection in breeding programs focusing on HS resistance.

At present, HS resistance in maize is considered a quantitative trait and controlled by additive, dominant, and epistatic effects. Additive effects play a dominant role while non-additive effects serve little function. Lübberstedt et al. (1999) demonstrated that the inheritance of HS resistance is mainly additive, but Ali et al. (1990) believed that the inheritance of HS resistance was related to the incidence of HS, i.e., resistance was dominant when incidence was high, otherwise, it was additive. Bernardo et al. (1992) found that additive effects play a decisive role in the inheritance of resistance, while the impact of non-additive effects are small. The QTL associated with HS resistance detected in the present study were mainly characteristic of dominant inheritance, however, four QTL showed additive inheritance in the joint analysis of all three environments, while the rest of the QTL in the joint analysis were characteristic of dominant inheritance. Therefore, the inheritance of HS resistance in maize can be either an additive or dominant due to the different genetic background of the material, however, whether the inheritance of HS resistance changes with the level of incidence requires further study.

Head smut is a global maize disease causing moderate-to-severe losses in both quality and quantity of annual yield (Shrestha et al. 2019). Considering both economic and ecological elements, cultivation of resistant varieties represents an effective way to control epidemics of head smut.

### Table 5

| Chr | QTL name | Bins | Interval markers | Position | LOD score(A) | LOD score (A by E) | PVE | Est A | Est D | Gene action |
|-----|----------|------|-----------------|----------|--------------|--------------------|-----|-------|-------|-------------|
| 1   | JAHS1a   | 1.01 | bnlg1014-umc2224 | 22.00    | 8.45         | 0.04               | 7.21 | −0.32 | 3.72  | OD          |
| 1   | JAHS1b   | 1.07 | umc1278-umc1147 | 121.00   | 2.57         | 0.22               | 2.65 | 1.44  | −1.11 | D           |
| 1   | JAHS1c   | 1.09 | bnlg1331-umc1306| 153.00   | 3.32         | 0.37               | 3.21 | 1.67  | −0.52 | PD          |
| 1   | JAHS1d   | 1.10–1.11 | bnlg1347a-phi064 | 168.00  | 2.55         | 0.38               | 2.63 | 1.32  | −1.37 | D           |
| 2   | JAHS2a   | 2.02–2.03 | bnlg125-bnlg1537 | 32.00   | 3.61         | 0.17               | 2.92 | −0.03 | 2.37  | OD          |
| 2   | JAHS2b   | 2.06–2.07 | umc1108-umc1536 | 115.00  | 2.36         | 0.08               | 1.97 | −1.29 | 0.81  | PD          |
| 2   | JAHS2c   | 2.08 | umc1464-phi090  | 118.00   | 2.87         | 0.07               | 2.38 | −1.50 | 0.64  | PD          |
| 3   | JAHS3a   | 3.02–3.03 | bnlg1325-bnlg1447| 19.00   | 6.13         | 0.07               | 4.94 | 0.19  | 3.00  | OD          |
| 4   | JAHS4a   | 4.07 | umc2038-bnlg1784| 52.00   | 4.40         | 0.14               | 3.75 | 1.82  | −0.22 | A           |
| 4   | JAHS4b   | 4.01–4.03 | umc2281-umc1757 | 85.00   | 5.12         | 0.08               | 4.27 | 1.93  | 1.04  | PD          |
| 5   | JAHS5a   | 5.03 | umc2294-umc1557a| 1.00    | 3.66         | 0.12               | 3.06 | −1.90 | 0.14  | A           |
| 5   | JAHS5b   | 5.07–5.08 | umc1072-umc1225 | 74.00   | 3.34         | 0.11               | 2.54 | 0.02  | 2.18  | OD          |
| 6   | JAHS6a   | 6.02–6.04 | umc1006-umc1857 | 42.00   | 10.97        | 0.12               | 7.64 | −2.39 | −1.79 | PD          |
| 7   | JAHS7a   | 7.02 | umc1409-umc1585 | 52.00   | 2.81         | 0.28               | 2.23 | −1.52 | −0.42 | PD          |
| 7   | JAHS7b   | 7.05 | umc1671-umc1154 | 134.00  | 2.60         | 0.16               | 2.18 | −0.68 | −1.93 | OD          |
| 9   | JAHS9a   | 9.03–9.04 | bnlg1270-umc2338 | 49.00   | 2.06         | 0.15               | 1.63 | −1.32 | −0.31 | PD          |
| 9   | JAHS9b   | 9.01 | phi033-umc1867  | 102.00  | 4.49         | 0.28               | 3.68 | 1.75  | −0.90 | PD          |
| 10  | JAHS10a  | 10.07 | umc2351-umc1196 | 19.00   | 2.69         | 0.12               | 2.30 | 1.49  | 0.27  | A           |
| 10  | JAHS10b  | 10.01–10.02 | umc1432-umc1291 | 102.00  | 2.60         | 0.01               | 2.21 | −1.49 | −0.14 | A           |

*a LOD (A), LOD score for additive and dominance effects, *b LOD (A by E), LOD score for additive and dominance by environment effects, *c LOD (A by E), LOD score for additive and dominance by environment effects.
Pyramiding of resistant genes/quantitative trait loci (QTL) against HS into elite varieties is a promising approach for improving HS resistance in maize (Chen et al. 2008). The maize inbred line T32 was confirmed to be highly resistant to HS in the present study. Several major QTL associated with HS resistance were identified and will be the subject of fine mapping in near-isogenic lines (NILs) in our further research. The stable QTL associated with HS resistance in maize in the present study can potentially be used to develop molecular marker-assisted selection in maize breeding programs. The economic value of maize production will greatly increase as new varieties with strong HS resistance are developed and released for use by growers.

Conclusions

We successfully identified two stable QTL for HS resistance in maize growing in three different environments. Both QTL can be used in the development of marker-assisted selection (MAS) and in theoretical studies of HS resistance in maize.

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Data availability The authors described the relevant data sources in the manuscript. The data generated or analyzed during this study are included in this manuscript and its supplementary material.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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