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Genetic analysis of maize kernel thickness by quantitative trait locus identification

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ABSTRACT. Kernel thickness is one of the most important traits in kernel structure, and is related to yield. To ascertain its genetic information more clearly, an immortal recombinant inbred line segregation population was used to map the quantitative trait loci (QTLs) for kernel thickness. As a result, two QTLs were identified on chromosome 9; both of them had negative additive effects, and could decrease kernel thickness to some extent. The QTLs explained 25.8% of the total phenotypic variation. These results advance our understanding of the genetic basis of kernel thickness in maize-breeding programs.

Key words: Maize (*Zea mays* L.); Quantitative trait locus; Recombinant inbred line; Kernel thickness

INTRODUCTION

Maize (*Zea mays* L.) is one of the most important global cereal crops, is widely consumed, and plays a crucial role in maintaining food security. In addition, maize is used as a raw material in industry. Its high demand makes grain yield a major target of maize breeding (Liu et al., 2014). Maize grain yield is a complex, quantitative genetic trait, and can be divided into smaller components, such as plant number per hectare, kernel number per ear, 100-kernel weight, etc. Kernel thickness is an important agronomic trait, which is related to 100-kernel weight and kernel appearance.

Quantitative trait locus (QTL) mapping is an effective means of understanding the genetic mechanisms of agronomic traits. Numerous studies on QTL identification in crops have been conducted, including in maize (Riedelsheimer et al., 2012; Almeida et al., 2014; Herzog et al., 2014), rice (Chen et al., 2014; Xu et al., 2014; Zhou et al., 2014), wheat (Bennett et al., 2012; Ishikawa et al., 2014; Katkout et al., 2014), sorghum (Rajkumar et al., 2013; Alam et al., 2014), and soybean (Liang et al., 2014; Mamidi et al., 2014). Regarding maize kernel-related traits, most studies that have used QTL mapping have focused on kernel number per ear (Liu et al., 2011; Zhang et al., 2013; Li et al., 2014) or kernel weight (Tang et al., 2010; Ding et al., 2011; Prado et al., 2014). There have only been a few studies conducted on kernel thickness. Li et al. (2009) identified four QTLs on chromosomes 2, 3, 6, and 10, with one on every chromosome, and Peng et al. (2011) mapped three QTLs (using a population derived from Qi319 and Huangzaosi) on chromosomes 2, 3, and 10. Peng et al. (2011) used another segregation population bred from Ye478 and Huangzaosi, and mapped three QTLs on chromosomes 1, 3, and 10. Recently, Zhang et al. (2014) and Liu et al. (2014) found 11 and 18 QTLs, respectively, for kernel thickness. These different results were probably caused by the use of different parental materials, segregation populations, genetic maps, or ecological environments. Therefore, it is significant to select a new population to study the genetic mechanism of kernel thickness by QTL mapping in maize-breeding program.

In the present study, an immortal recombinant inbred line (RIL) population derived from the parental inbred lines Mo17 and Huangzaosi was used in QTL detection for kernel thickness; its aims were to understand the genetic basis of kernel thickness more clearly, and to locate the quantitative loci that are associated with kernel thickness in maize.

MATERIAL AND METHODS

Experimental materials

We used the two parental inbred lines Mo17 and Huangzaosi and an RIL segregation population consisting of 239 RILs derived from the two parents. Mo17 and Huangzaosi are representative inbred lines of the Lancaster (United States) and Tangsipingtou (China) heterosis groups, respectively.

Field design and phenotypic observations

The 241 maize materials were sown at the Nanchong Agricultural Academy, Nanchong City, Sichuan Province, China. Twenty plants of each of three replicates were designed

for every maize line. After harvesting, the thickness of 20 randomly selected kernels from each line was measured with an electronic digital caliper, and Microsoft Excel 2010 was used to compute the mean thickness of the 20 maize kernels.

Descriptive statistics

The mean kernel thicknesses of the parents were compared, and descriptive statistics of the RILs were obtained using SPSS version 11.5, including ranges, minima, maxima, means, standard deviations (SD), skewness, kurtosis, and coefficients of variation (COV); frequency distributions of kernel thickness were also generated.

QTL detection

An established marker linkage map (Liu et al., 2009) that included 100 simple-sequence repeat (SSR) markers and covered 1421.5 cM was used for QTL mapping for kennel thickness, based upon the descriptive statistics of the RILs. The QTLs for kernel thickness were detected by the interval mapping method, using the QTL mapping software Windows QTL Cartographer version 2.5 (Wang et al., 2010), with 1.0 cM of walk speed. The log10 of the odds ratio (LOD) threshold value used to detect QTLs was determined by a 1000-permutation test ($\alpha = 0.05$) (Doerge and Churchill, 1996). The chromosomal positions, additive genetic effects, and percentages of phenotypic variance of the QTLs that were identified were estimated using the same QTL mapping software. The QTLs were then mapped onto the marker linkage map using the MapChart 2.1 software (Voorrips, 2002).

RESULTS

Descriptive statistics of parents and RILs

On average, Huangzaosi had thicker kernels than Mo17 (Table 1). Kernel thickness in the RILs ranged between 4.08 and 8.53 mm (Table 2) and followed a normal distribution (Figure 1), indicating that it is a quantitative trait that is controlled by several genes.

Table 1. Kernel thickness in maize parental inbred lines.

Parental inbred line	Kernel thickness (mm)
Mo17	4.77
Huangzaosi	5.43

Table 2. Descriptive statistics for kernel thickness in maize recombinant inbred lines.

Trait	Range (mm)	Minimum (mm)	Maximum (mm)	Mean (mm)	SD	Skewness	Kurtosis	COV (%)
Kernel thickness	4.45	4.08	8.53	5.28	0.59	0.875	3.309	11.17

SD, standard deviation; CV, coefficient of variation.

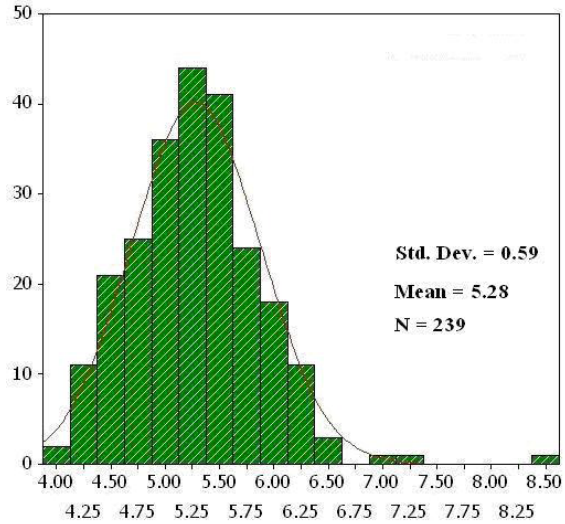

Figure 1. Frequency distribution of kernel thickness in maize recombinant inbred lines.

QTL detection

The results of the QTL scanning are shown in Figure 2. According to the LOD threshold value of 2.20, only two QTLs (*qKT9-1* and *qKT9-2*) were detected, which were on chromosome 9 (Figure 3). The two QTLs had the same adjacent marker (Umc1357), but *qKT9-1* was closer to it (2.1 cM) than *qKT9-2* (10.0 cM). *qKT9-1* was between Phi016 and Umc1357, and *qKT9-2* was between Umc1357 and Bnlg1375 (Table 3). Both QTLs exhibited negative additive effects, suggesting that they decrease kernel thickness to some extent. *qKT9-1* accounted for 10.5% of the total phenotypic variation, and *9KT9-2* accounted for up to 15.3%. Therefore, the two QTLs identified in this study are different chromosomal loci, despite having the same adjacent marker and being in close proximity to each other.

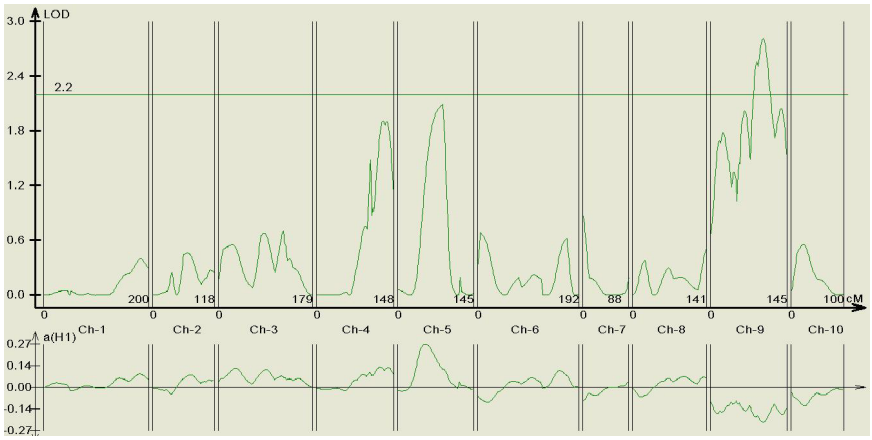

Figure 2. Quantitative trait locus (QTL) analysis result by interval mapping for maize kernel thickness. Only two QTLs were detected on chromosome 9 according to the log10 of the odds ratio (LOD) curves.

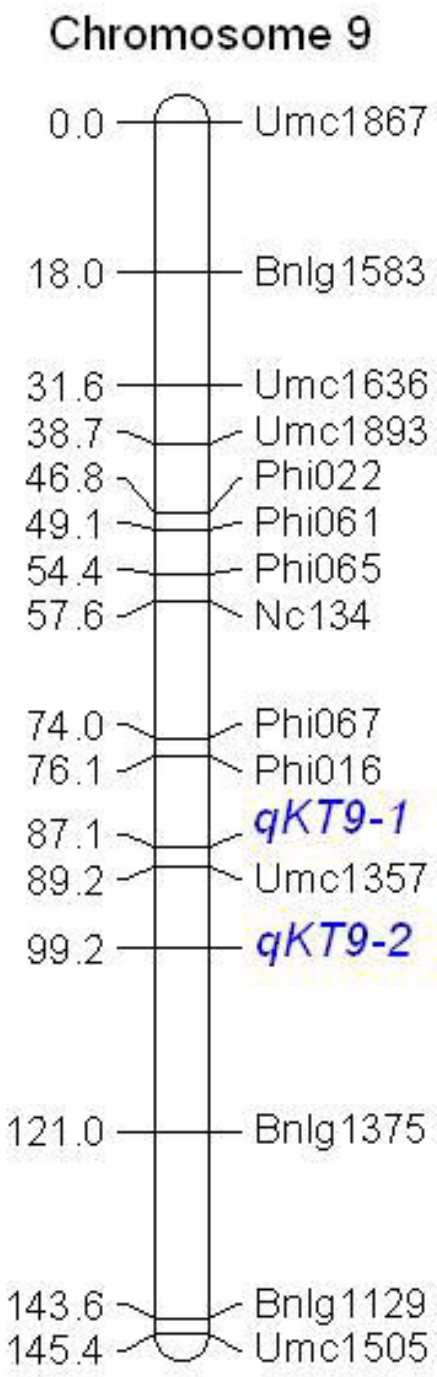

Figure 3. Chromosomal distribution of quantitative trait loci (QTLs). Two QTLs were mapped on chromosome 9, one between Phi016 and Umc1357 (*qKT9-1*) and the other between Umc1357 and Bnlg1375 (*qKT9-2*).

Table 3. Quantitative trait loci associated with maize kernel thickness and their genetic parameters.

QTL	Chr.	Chromosomal position (cM)	Genetic distance to Umc1357 (cM)	LOD	Additive effect	R^2 (%)
qKT9-1	9	87.1	2.1	2.6	-0.18	10.5
qKT9-2	9	99.2	10.0	2.8	-0.22	15.3
Total						25.8

QTL, quantitative trait locus; Chr., chromosome number; LOD, log10 of the odds ratio; R^2, percentage of phenotypic variance explained by QTL.

DISCUSSION

We found two QTLs, both of them on chromosome 9. They both exhibited negative additive effects, and could decrease kernel thickness to some extent.

Previous studies have found several QTLs for kernel thickness, with at least one on each of the 10 maize chromosomes (Table 4). Liu et al. (2014) identified up to 18 QTLs on seven chromosomes, and Zhang et al. (2014) found 11 on six chromosomes. These results suggest that kernel thickness is a quantitative trait and is controlled by several genes. Our results confirm the findings of Liu et al. (2014), who also found two QTLs on chromosome 9. However, the two results differ: the two QTLs identified by Liu et al. (2014) exhibited positive additive effects, whereas the two mapped in our study exhibited negative additive effects.

Table 4. Quantitative trait loci associated with maize kernel thickness and their chromosomal distribution.

Reference	Parental material	No. of mapped QTLs	Chr. No.									
			1	2	3	4	5	6	7	8	9	10
Li et al. (2009)	Qi319 and Huangzaosi	4		1	1			1				1
Peng et al. (2011)	Qi319 and Huangzaosi	3		1	1							1
Peng et al. (2011)	Ye478 and Huangzaosi	3	1		1							1
Zhang et al. (2014)	Xu178 and HuangC	11	1	3	1			2	1	3		
Liu et al. (2014)	V671 and Mc	18	5	1		2	3			2	2	3
This study	Mo17 and Huangzaosi	2									2	

QTL, quantitative trait locus; Chr. No., chromosome number.

Although we detected two QTLs in maize that were associated with kernel thickness, they were relatively far from their adjacent SSR marker (Umc1357) in genetic distance, so more molecular markers need to be added to this chromosomal region in order to more finely map them, and this study is currently ongoing.

Conflicts of interest

The authors declare no conflict of interest.

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