Detection of quantitative trait loci for ear row number in F_2 populations of maize

C. Yang[1,2], J. Liu[1,2] and T.Z. Rong[1,2]

[1]Maize Research, Sichuan Agricultural University, Wenjiang, Sichuan, China
[2]Key Laboratory of Biology and Genetic Improvement of Maize in Southwest Region, Ministry of Agriculture, Wenjiang, China

Corresponding author: T.Z. Rong
E-mail: rongtz@sicau.edu.cn

Genet. Mol. Res. 14 (4): 14229-14238 (2015)
Received May 20, 2015
Accepted July 14, 2015
Published November 13, 2015
DOI http://dx.doi.org/10.4238/2015.November.13.6

ABSTRACT. Ear row number (ERN) is not only a key trait involved in maize (*Zea mays* L.) evolution but is also an important component that is directly related to grain yield. In this study, quantitative trait loci (QTLs) for ERN were detected across two F_2 populations that were derived from a same cross between B73 with 16 rows (N = 233) and SICAU1212 with four rows (N = 231). As a result, 33 QTLs were associated with 12 agronomic traits: three plant traits, four ear-related traits, and five kernel-related traits. The total phenotypic variation explained by the QTLs for each trait ranged from 8.60 to 72.67%, and four QTLs were identified for ERN in the two populations. Each QTL explained between 6.78 and 36.76% of the ERN variation. Notably, three of the four QTLs (*qERN2-1*, *qERN4-2*, and *qERN8-1*) were associated with ERN, and *qERN8-1* simultaneously influenced grain yield, plant diameter, ear diameter, and kernel length. In addition, only one significant epistatic interaction was detected in all 33 QTLs. This study should provide a foundation for further fine-mapping and map-based cloning of these consistent QTLs, and for controlling maize ERN by marker-assisted breeding.

Key words: Maize; QTL; Ear row number; Grain yield

INTRODUCTION

Maize (*Zea mays* L.) is one of the most important global cereal crops,andis an ideal plant type in terms of its photosynthetic reaction. The growing human population requiresfurther improvementsin grain yield; consequently, the genetics of maize grain yield and yield-related traits have been the focus of many studies (Upadyayula et al., 2006; Yan et al., 2006; Liu et al., 2014). Maize grain yield is a complex quantitative trait, and its genetic basis can be explained by the effects of quantitative trait loci (QTLs) that control its components, such as ear row number (ERN).

ERN isan important agronomic trait, and differs significantly between maize and teosinte. With the rapid development of molecular genetic marker technology and quantitative genetics, segregation populationshave been used to detect QTLs for ERN, and have contributed to maize domestication and diversification (Doebley and Stec, 1991; Lauter and Doebley, 2002; Li et al., 2011; Lu et al., 2011; Lemmon and Doebley, 2014; Li et al., 2014; Yang et al., 2015). The most consistent result obtained from previous studies is that certain regions on the short segment of chromosome 2 have a large effect on ERN during domestication. Few QTLs have been detected thatplay roles in later improvement processes, probably because of the use of different parental materials and different segregation population sizes, marker densities, or environments, which can influence the power of QTL detection, the accurate location of QTLs, and the estimation of QTL effects (Beavis, 1998). In addition, the domestication process (from teosinte to maize) may have been affected by three major mutations. The first mutation was the liberation of the kernel, the second was the lack of abortion of the pedicellatespikelets, and the third was that ERN increased to above four (Iltis, 2000). The missing genetic variation from four to more rows could not be determined due to limited variation in the ERN, although MT-6 (with an ERN of six) was crossed with B73 in order to detect the QTLs responsible for ERN (Cai et al., 2014).

Over the last 10 years, genome-wide association studies (GWAS) have provided a more powerful and complementary tool to connect the genotype-phenotype map than QTL mapping. GWAS may provide insights into trait architecture or candidate loci, and have been widely applied in plants and animals (e.g., *Arabidopsis thaliana*, maize, rice, mice, cattle, and humans) (Korte and Farlow, 2013). Byconducting joint linkage and GWAS, Brown et al. (2011) found 36 QTLs associated with ERN and 261 significant single-nucleotide polymorphisms in anest association-mapping population, which consisted of 5000 recombinant inbred lines from 25 families that represented the global diversity of maize. To date, several genes that influence or control ERN inheritance, namely *zfl2* (Bomblies and Doebley, 2006), *fea2* (Bommert et al., 2013), and *ub2* and *ub3* (Chuck et al., 2014), have been identified. The molecular mechanism underlying ERN variation has only been provisionally determined, although numerous QTLs and genes that control ERN have been identified.

In this study, two F_2 populations derived from a same cross between B73 and SICAU1212 were developed in Yunnan, China in 2009 and Sichuan, China in 2010, respectively, in order toidentify the QTLs responsible for ERN. This study shouldprovide useful insights into the inheritance of ERN, and supply effective molecular tools for improving ERN and consequently grain yield.

MATERIAL AND METHODS

Plant materials

The experimental materials were the maize inbred lines B73 (approximately 16 rows;

F_2, N = 233) as the female parent and SICAU1212 (approximately 4 rows; F_2, N = 231) as the pollen parent. SICAU1212 was derived from *Silunuo*, a four-rowed waxy corn that was planted in Xishuangbanna, Yunnan Province, China and subsequently self-pollinated for seven generations.

Field experiments

F_2 individuals and both parents were planted following a completely random design in two experimental stations in Yunnan (B73) and Sichuan Provinces (SICAU1212), China, in 2009 and 2010, respectively. The abbreviations 09YN and 10SC represent Yunnan in 2009 and Sichuan in 2010, respectively. Each row (approximately 14 plants) was 3.5 m in length with a space of 0.75 m between rows. Standard cultivation management practices were conducted at each location, with a density of 52,500 plants/ha.

Phenotypic identification and statistical analysis

After pollination, three plant-related traits of each individual, namely plant height (PH, cm) from plant base to tasseling tip, ear height (EH, cm) from plant base to the first ear base, and plant diameter (PD, cm) halfway up the plant were calculated. After maturity, ERN in the middle of the ear, ear length (EL, cm) from ear base to ear tip, eardiameter (ED, cm) in the middle of the ear, and kernel number per row (KNR) were investigated. Kernels were bulked for each plant and used to measurefive kernel-related traits. Grain yield (GY, g) was calculated based on the weight of total kernels; 100-kernel weight (HKW, g) was the mean of three repeated measurements of 100 kernels randomly chosen from the bulked kernels; 10-kernel length (KL, cm), 10-kernel width (KW, cm), and 10-kernel thickness (KT, cm) were the means of three repeated measurements of 10 kernels randomly sampled from the bulked kernels. Four of the 12 agronomic traits (PH, EH, PD, and ERN) were repeatedly measured in the two F_2 populations.

SPSS19.0 software was used to calculate Pearson correlation coefficients(*r*) between traits at each location, and to conduct phenotypic data analysis for each trait.

Molecular linkage map construction

Total genomic DNA isolation and purification was performed from young leaves of both parental lines and each of the F_2 plants using the modified CTAB procedure (Saghai-Maroof et al., 1984). Approximately 900 simple-sequence repeat (SSR) primer pairs obtained from Maize GDB (http://www.maizegdb.org) were initially used to examine polymorphisms between the parents, B73 and SICAU1212. Ultimately, 109 and 117 SSR markers were used to genotype233 F_2 individuals in 09YN and 231 F_2 individuals in 10SC, respectively. A total of 93 markers were repeatedly used to genotype the two populations. The polymerase chain reaction (PCR) mixture (15 μL) contain 50 ng genomic DNA, 0.5 μM primer, 10X Taq buffer, 0.2 mM dNTP, 0.5 U Taq DNA polymerase (Tiangen, Beijing). PCR was performed in a thermal cycler (BIO-RAD S1000™) with the following by 30 cycles of 95°C for 30 s (denaturation), a specific temperature depending upon the primer pair for 30 s (annealing) and 72°C for 40 s (extension); followed by a final extension at 72°C for 10 min. All of the PCR products were electrophoresed on 6% denaturing polyacrylamide gels and stained withapproximately 0.33% silver nitrate (Santos et al., 1993).

The linkage maps were developed using MapMaker/EXP version 3.0b (http://www.

softpedia.com/get/Science-CAD/MapMaker.shtml; Lincoln, 1992). The total lengths of the molecular linkage maps for 233 F_2 and 231 F_2 were 1290.4 and 1348.5 cM, respectively, across the maize genome, with average intervals between adjacent markers of 11.53 and 11.84 cM, respectively. The orderof most molecular markers was consistent with that of their physical position in both linkage maps.

QTL identification

QTL identification for each trait at each location was performed using ICIM (QTL IciMapping) version 3.0 (Li et al., 2008). The additive and dominant effects (ICIM-ADD) mapping method was used to identify QTLs by stepwise regression, with 1000 permutations and a walk speed of 2 cM. The confidence intervals (*CI*) of the QTLs were estimated as the following:

$$CI = 530/N \times R^2$$

where, *N* is the population size, and R^2 is the phenotypic variation contributed by the QTL (Darvasi and Soller, 1997). If QTLs for different traits were detected within the same marker interval or their confidence intervals overlapped, the corresponding loci were assumed to be common QTLs or QTLs with pleiotropic effects. Epistatic interactions between QTLs for each trait at each location were identified by the QTL Network version 2.0 (http://ibi.zju.edu.cn/software/qtlnetwork), with a linear mixed model based on the composite interval mapping approach (Yang et al., 2007). The testing window, walk speed, and filtration window of the genome scan were set at 10, 2, and 10 cM, respectively. The logarithm (base 10) of odds (LOD) threshold scores for significant QTLs were obtained with a permutation test of 1000 cycles (Churchill and Doerge, 1994). The mapped QTL effects were estimated according to the following criteria: d/a = dominance effects/additive effect; A, additive (d/a = 0.00-0.20); PD, partial dominance (d/a = 0.21-0.80); D, dominance (d/a = 0.81-1.20); OD, (d/a >1.21) (Stuber et al., 1987).

RESULTS

Phenotypic analysis

Of the two parents, B73 exhibited higher values for the PD, ERN, EL, ED, KNR, GY, and KL, whereas SICAU1212 had higher values of EH, HKW, KW, and KT. There were highly significant differences between B73 and SICAU1212 for all traits (except for PH), regardless of the location (Table 1). Regarding the two F_2 populations, the results of a Shapiro-Wilk test showed that the data distributions for each trait were approximately normal, and there were large variations in twolocations. It was noteworthy that all of the trait sexhibitedbi-directional transgressive segregation at each location, indicating polygenic quantitative genetic control.

Correlation analysis between different traits in the two F_2 populations

We found that 54 of the 72 correlation coefficients derived for the 12 traits in the two locations were significant and positive (Table 2), and interestingly GY was significantly correlated with all of the other traits in 10SC.

Table 1. Phenotypic performance of each traitin parents (B73 and SICAU1212) and intwo F_2 populations.

Trait	Environment	B73	SICAU1212	F_2 population						
				Range	Mean	SE	CV	Skew	Kurt	P value
PH	09YN	167.70 ± 1.2[a]	168.20 ± 4.3	110.8-280.3	201.1	2.1	0.16	-0.54	0.08	<0.001
	10SC	171.75 ± 1.8	166.00 ± 3.5	148.5-297.1	228.1	2.2	0.15	-0.41	-0.50	<0.001
EH	09YN	56.90 ± 2.7	91.30 ± 2.5**	45.4-195.6	97.5	1.2	0.18	0.62	3.50	<0.001
	10SC	55.16 ± 1.9	92.80 ± 2.9**	40.2-173.2	106.9	1.5	0.22	-0.01	0.01	0.96
PD	09YN	2.16 ± 0.05	1.66 ± 0.05**	0.96-2.10	1.53	0.01	0.14	-0.17	-0.18	0.31
	10SC	2.14 ± 0.03	1.69 ± 0.04**	1.07-2.55	1.92	0.02	0.13	-0.28	0.58	<0.001
ERN	09YN	15.67 ± 0.17	4.13 ± 0.09**	4-16	8.59	0.20	0.36	0.05	-1.01	<0.001
	10SC	16.00 ± 0.15	4.29 ± 0.17**	4-16	11.58	0.19	0.25	-0.39	-0.41	<0.001
EL	10SC	14.60 ± 0.59	10.84 ± 0.54**	6.60-20.30	13.61	0.19	0.21	-0.08	-0.53	0.24
ED	10SC	3.95 ± 0.17	2.08 ± 0.04**	1.80-4.14	3.03	0.03	0.14	-0.17	-0.22	0.55
KNR	10SC	38.75 ± 0.53	21.20 ± 1.02**	8-50	32.57	0.47	0.22	-0.26	-0.03	0.05
GY	10SC	76.66 ± 1.53	14.87 ± 0.05**	4.63-100.01	42.83	1.35	0.48	0.61	-0.26	<0.001
HKW	10SC	18.54 ± 0.22	23.36 ± 0.19**	5.44-33.85	17.49	0.39	0.33	0.24	-0.34	<0.001
KL	10SC	10.10 ± 0.02	7.16 ± 0.01**	5.50-10.38	7.69	0.06	0.13	0.40	-0.17	0.01
KW	10SC	6.48 ± 0.01	9.21 ± 0.01**	5.56-10.36	7.52	0.06	0.12	0.23	0.14	0.16
KT	10SC	4.46 ± 0.02	5.13 ± 0.03**	3.19-7.26	4.62	0.05	0.15	0.97	0.97	<0.001

[a]Values are means and standard errors (SE). **Difference between the parents was highly significant at the P < 0.01 level, as determined by a Student *t-test*. 09YN, Yunnan in 2009; 10SC, Sichuan in 2010. P values were derived from a Shapiro-Wilk normality test. PH, plant height; EH, ear height; PD, plant diameter; ERN, ear row number; EL, ear length; ED, ear diameter; KNR, kernel number per row; GY, grain yield; HKW, 100-kernel weight; KL, 10-kernel length; KW, 10-kernel width; KT, 10-kernel thickness. CV, coefficient of variation.

Table 2. Pearson correlation coefficients (r) between traits across two environments.

Trait	Environment	EH	PD	ERN	EL	ED	KNR	GY	HKW	KL	KW	KT
PH	09YN	0.48**	0.44**	0.04								
	10SC	0.44**	0.28**	0.18**	0.41**	0.21**	0.33**	0.36**	0.41**	0.39**	0.33**	0.02
EH	09YN		0.37**	-0.01								
	10SC		0.31**	0.08	0.29**	0.22**	0.33**	0.38**	0.36**	0.37**	0.39**	-0.08
PD	09YN			0.14*								
	10SC			0.05	0.32**	0.23**	0.32**	0.34**	0.28**	0.39**	0.21**	0.07
ERN	09YN											
	10SC				0.05	0.39**	0.19**	0.32**	0.02	0.26**	-0.19**	-0.15*
EL	10SC					0.35**	0.79**	0.52**	0.24**	0.40**	0.21**	-0.16*
ED	10SC						0.42**	0.62**	0.30**	0.62**	0.15*	-0.32**
KNR	10SC							0.50**	0.13*	0.36**	0.07	-0.34**
GY	10SC								0.42**	0.69**	0.28**	-0.29**
HKW	10SC									0.52**	0.55**	0.13
KL	10SC										0.46**	-0.20**
KW	10SC											0.17**

*Significantly different at P < 0.05. **Significantly different at P < 0.01. 09YN, Yunnan in 2009; 10SC, Sichuan in 2010. PH, plant height; EH, ear height; PD, plant diameter; ERN, ear row number; EL, ear length; ED, ear diameter; KNR, kernel number per row; GY, grain yield; HKW, 100-kernel weight; KL, 10-kernel length; KW, 10-kernel width; KT, 10-kernel thickness.

QTL identification foreach trait

These results are summarized in Figures 1 and 2 and Table 3. Nine QTLs were identified in the 233 F_2 in 09YN: one for PH, two for EH, three for PD, and three for ERN. Twenty-four QTLs for 12 traits were detected in the 231 F_2 in 10SC, and were located on all 10 chromosomes, except for chromosome 7. Each QTL accounted for between 6.78 and 36.76% of the phenotypic variation,

with *q10sERN8-1* contributing the highest percentage. Fourteen of 33 QTLs had a major effect (explained more than 10% of the phenotypic variation). The total phenotypic variation explained by all of the QTLs identified for each trait ranged from 8.60 to 72.67%. Approximately 60.61% of the detected QTLs had a positive additive effect, indicating that alleles from B73 contributed to increasing the phenotype. In addition, over 57.57% of the identified QTLs exhibited partially dominant genic interactions. Notably, four QTLs, *qEH1-1*, *qERN2-1*, *qERN4-2*, and *qERN8-1*, were consistently identified at each location; *qERN8-1* was co-located with QTLs for GY (*q10sGY8-1*), PD (*q09yPD8-1*), ED (*q10sED8-1*), and KL (*q10sKL8-1*). As stated previously, 10 significant phenotypic correlations were found between ERN, GY, PD, ED, and KL. It seems that *qERN8-1* playsan important role in determining plant architecture, ear improvement, kernel improvement, and grain yield.

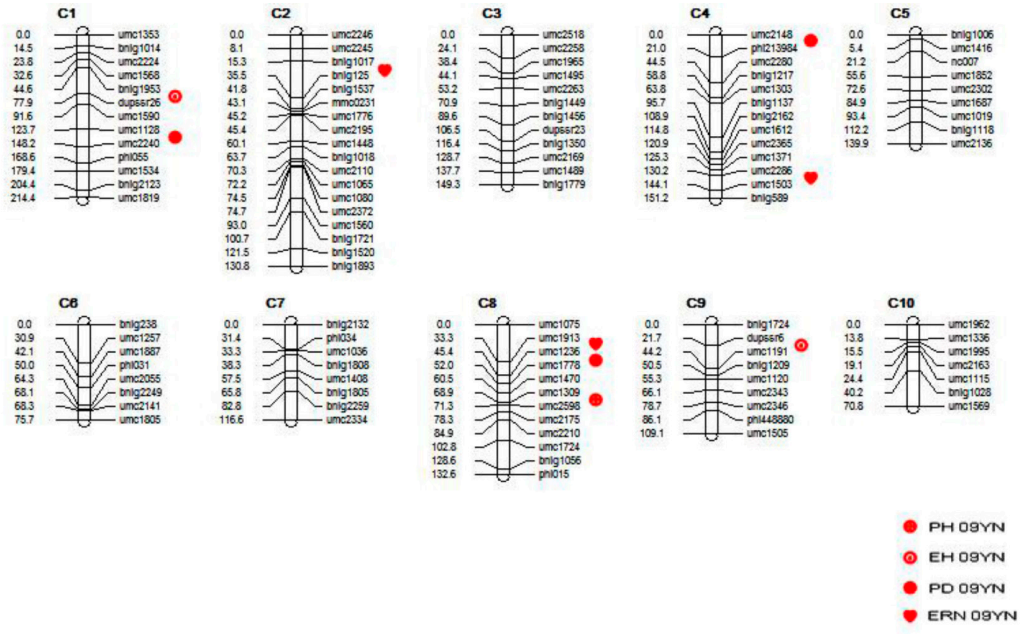

Figure 1. Molecular linkage map of the F_2 population in Yunnan in 2009 (09YN) and locations of quantitative trait loci (QTLs) for four traits. The letter 'C' represents chromosome. QTLs for each trait are differentiated by different red-colored shapes.09YN, Yunnan in 2009. PH, plant height; EH, ear height; PD, plant diameter; ERN, ear row number.

Epistatic interactions between the QTLs

Of the QTLs identified, only one significant epistatic interaction (between *qERN4-2* and *qERN8-1*) was detected for ERN in 10SC (Table 4). However, this effect wasmuch lower than that of corresponding QTLs, indicating that the main effects of significant QTLs may have stronger effectson ERN.

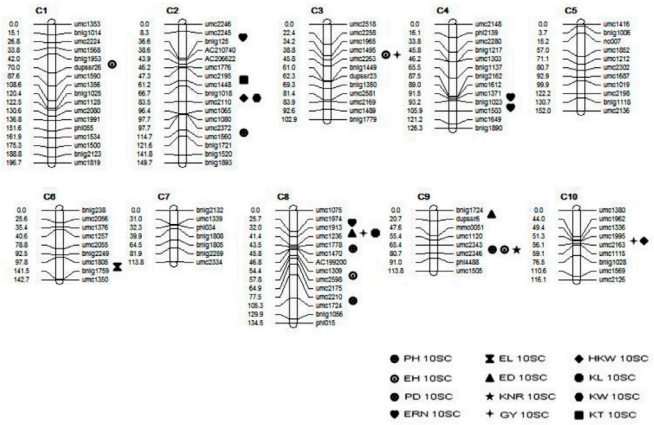

Figure 2. Molecular linkage map of the F_2 population in Sichuan in 2010 (10SC) and locations of quantitative trait loci (QTLs) for 12 traits. The letter 'C' represents chromosome. QTLs for each trait are differentiated by different black-colored shapes.10SC, Sichuan in 2010. PH, plant height; EH, ear height; PD, plant diameter; ERN, ear row number; EL, ear length; ED, ear diameter; KNR, kernel number per row; GY, grain yield; HKW, 100-kernel weight; KL, 10-kernel length; KW, 10-kernel width; KT, 10-kernel thickness.

Table 3. Quantitative trait loci (QTLs) detected for each traitacross two populations.

Trait	Environment	QTL	Chromosome	Position (cM)	Left Marker	Right marker	LOD	A[a]	D[b]	PVE (%)[c]	Gene action[d]
PH	09YN	*q09yPH8-1*	8	70.0	umc1309	umc2598	4.50	13.13	3.83	8.60	PD
	10SC	*q10sPH2-1*	2	108.0	umc2372	umc1560	4.52	-13.68	3.12	8.31	PD
		q10sPH8-1	8	44.0	umc1778	umc1470	5.47	15.00	8.68	9.12	PD
		q10sPH9-1	9	70.0	umc2343	umc2346	5.40	-8.73	5.00	10.06	PD
EH	09YN	*q09yEH1-1*	1	58.0	bnlg1953	dupssr26	4.40	-8.62	1.45	11.13	A
		q09yEH9-1	9	36.0	dupssr6	umc1191	4.64	-8.63	1.26	11.25	A
	10SC	*q10sEH1-1*	1	56.0	bnlg1953	dupssr26	4.57	-8.73	2.64	7.99	PD
		q10sEH3-1	3	42.0	umc1495	umc2263	6.84	-9.78	4.97	10.98	PD
		q10sEH8-1	8	56.0	umc1309	umc2598	5.93	11.02	4.41	9.48	PD
		q10sEH9-1	9	70.0	umc2343	umc2346	4.70	-8.98	4.26	7.95	PD
PD	09YN	*q09yPD1-1*	1	132.0	umc1128	umc2240	7.68	-0.11	0.04	16.03	PD
		q09yPD4-1	4	12.0	umc2148	phi213984	4.73	-0.09	0.01	8.88	A
		q09yPD8-1	8	48.0	umc1236	umc1778	4.53	0.08	0.04	8.00	PD
	10SC	*q10sPH8-1*	8	88.0	umc2210	umc1724	4.37	0.08	0.11	8.86	OD
ERN	09YN	*q09yERN2-1*	2	28.0	bnlg1017	bnlg125	10.67	1.63	0.74	16.12	PD
		q09yERN4-1	4	138.0	umc2286	umc1503	6.41	1.17	0.82	8.73	PD
		q09yERN8-1	8	42.0	umc1913	umc1236	14.07	2.01	0.60	22.24	PD
	10SC	*q10sERN2-1*	2	22.0	umc2245	bnlg125	13.82	1.56	0.65	17.07	PD
		q10sERN4-1	4	92.0	umc1371	bnlg1023	12.95	1.47	0.03	12.06	A
		q10sERN4-2	4	102.0	bnlg1023	umc1503	6.83	0.20	1.50	6.78	OD
		q10sERN8-1	8	28.0	umc1974	umc1913	32.76	2.16	1.79	36.76	D
EL	10SC	*q10sEL6-1*	6	124.0	umc1805	bnlg1759	4.60	0.56	2.50	19.18	OD
ED	10SC	*q10sED8-1*	8	34.0	umc1913	umc1236	4.46	0.17	0.09	8.20	PD
		q10sED9-1	9	10.0	bnlg1724	dupssr6	4.47	0.14	0.19	9.82	OD
KNR	10SC	*q10sKRN9-1*	9	72.0	umc2343	umc2346	4.26	-2.16	3.14	9.29	OD
GY	10SC	*q10sGY3-1*	3	42.0	umc1495	umc2263	5.41	0.39	13.94	11.49	OD
		q10sGY8-1	8	36.0	umc1913	umc1236	4.37	6.82	8.40	7.73	OD
		q10sGY10-1	10	54.0	umc1995	umc2163	4.56	1.30	12.32	9.29	OD
HKW	10SC	*q10sHKW2-1*	2	72.0	bnlg1018	umc2110	7.28	-2.95	-1.08	15.07	PD
		*q10sHKW10-*1	10	54.0	umc1995	umc2163	4.24	0.99	3.09	8.52	OD
KL	10SC	*q10sKL8-1*	8	36.0	umc1913	umc1236	5.66	0.43	0.27	10.21	PD
KW	10SC	*q10sKW2-1*	2	74.0	bnlg1018	umc2110	7.61	-0.40	-0.28	14.80	PD
KT	10SC	*q10sKT2-1*	2	60.0	umc2195	umc1448	4.77	-0.26	-0.21	9.32	PD

[a]A, additive effect of the QTL for each trait(positive values indicate that the alleles from B73 increased the trait score); [b]D, dominant effect of the QTL; [c]*PVE*, percent of the phenotypic variation explained by each QTL; [d]A, PD, D, and OD represent additive, partial dominance, dominance, and over-dominance effects, respectively. 09YN, Yunnan in 2009; 10SC, Sichuan in 2010. PH, plant height; EH, ear height; PD, plant diameter; ERN, ear row number; EL, ear length; ED, ear diameter; KNR, kernel number per row; GY, grain yield; HKW, 100-kernel weight; KL, 10-kernel length; KW, 10-kernel width; KT, 10-kernel thickness. LOD, logarithm (base 10) of odds.

Table 4. Quantitative trait loci (QTLs) with epistatic effects identified in this study.

Trait	Environment	QTL_i	Interval_i	QTL_j	Interval_j	AA[a]	AD[a]	DA[a]	DD[a]	$h^2(aa)^b$	$h^2(ad)^b$	$h^2(da)^b$	$h^2(dd)^b$
ERN	10SC	*q10sERN4-2*	bnlg1023-umc1503	*q10sERN8-1*	umc1974-umc1913	0.7997**	-0.4408	0.3479	-1.6485**	0.0162	0.0022	0.0012	0.0125

[a]AA, AD, DA, and DD represent additive-by-additive, additive-by-dominant, dominant-by-additive, and dominant-by-dominant epistatic interaction effects, respectively. [b]h^2(*aa*), h^2(*ad*), h^2(*da*), and h^2(*dd*) indicate the heritability of additive-by-additive, additive-by-dominant, dominant-by-additive, and dominant-by-dominant epistatic interaction effects, respectively. ERN, ear row number.10SC, Sichuan in 2010. **Significantly different at P< 0.01.

DISCUSSION

The ultimate objective of mapping QTLs for certain traits is to dissect the molecular and genetic basis of their variation. The identification and confirmation of QTLs simultaneously responsible for grain yield and related traits (common QTLs or QTLs with pleiotropic effects) should provide greater opportunities for breeders to improve yield by marker-assisted breeding.

ERN, which is associated with KNR, ED, EL, kernel size, and other features (Lu et al., 2011), was highly correlated with GY. ERN is one of the key traits that distinguish maize and teosinte (Doebley, 2004). QTL mapping has been conducted in many studies, and a large number of QTLs have been identified for ERN across generations and environments in all10 maize chromosomes (Doebley et al., 1990; Doebley and Stec, 1991, 1993; Veldboom and Lee, 1994; Austin and Lee, 1996; Szabó and Burr, 1996; Upadyayula et al., 2006; Yan et al., 2006; Briggs et al., 2007; Li et al., 2007; Guo et al., 2008; Karen Sabadin et al., 2008; Li et al., 2011; Lu et al., 2011; Cai et al., 2014; Lemmon and Doebley, 2014; Li et al., 2014; Tian et al., 2014; Yang et al., 2015). In this study, three QTLs (*qERN2-1*, *qERN4-2*, and *qERN8-1*) were consistently associated with ERN. The first QTL, *qERN2-1* (bin 2.02), was also located onthe short segment of chromosome 2, indicating a potential QTL region near *qERN2-1* that may not only play a novel role in affecting ERN during maize domestication (Doebley and Stec, 1991, 1993), but may also be associated with ERN during later improvement processes. The second QTL (*qERN4-2* at bin 4.08) significantly affects ERN (Austin and Lee, 1996; Li et al., 2011; Lu et al., 2011; Yang et al., 2015). In the genetic region of bin 4.08, one major QTL for ERN has been identified in an $F_{2:3}$ population, and explained the largest percentage (16.4%) of ERN variation (Cai et al., 2014). Although previous reports of QTLs for ERN ingenomic regions near *qERN8-1* in other maize segregations were few, these regions can be mapped in maize x teosinte populations (Doebley et al., 1990; Doebley and Stec, 1991; Briggs et al., 2007), indicating that *qERN8-1* may be a domestication-related QTL that controlled the initial switch from four to eight rowsduring domestication.In addition, this QTL simultaneously controls GY, PD, ED, and KL, suggesting that it could offerfurther opportunities for improving ERN and grain yield.

If one QTL region that is responsible for a certain trait contained, or was adjacent to, a known mutant gene that was the only candidate gene affecting the development of that trait, the mutant gene may be regarded as a logical candidate gene (Upadyayula et al., 2006). In this study, one stable QTL (*qERN8-1*) near 18 Mb (based on the AGI's B73 RefGen_v2 sequence) affected ERN. Twomutant genes (*vt2*and *bif1*) mapped to approximately 17 and 22 Mb, respectively, on chromosome 8are possible candidate genes in this region.*vt2* typically causes barren patches on one or both sides of the ear (Phillips et al., 2011), and this phenotype is similar to that of SICAU1212. *bif1* plays a role in auxin transport, and occasionally causes ears to exhibit four rows (Barazesh and McSteen, 2008). Another QTL (*qERN2-1*) on chromosome *2S* responsible for ERN has *zfl2* as

a flanking marker, which contributed to changes in ERN during maize domestication (Bomblies et al., 2003; Doebley, 2004; Bomblies and Doebley, 2006). The maize *SBP-box* transcription factor gene, *ub3*, which affects the rate of cell differentiation to the lateral domains of meristems and influences yield (Chuck et al., 2014), was located approximately 199 Mb on chromosome 4, near *qERN4-2*. Further studies are needed to determine whether these candidate genes are the causal genetic variants for these QTLs.

Conflicts of interest

All of the authors declare that they have no conflicts of interest.

ACKNOWLEDGMENTS

Research supported by the National Basic Research Program of China (the “973” project, #2014CB138203).

REFERENCES

Austin DF and Lee M (1996). Comparative mapping in F2:3 and F6:7 generations of quantitative trait loci for grain yield and yield components in maize. *Theor. Appl. Genet.* 92: 817-826.

Barazesh S and McSteen P (2008). Barren inflorescence1 function in organogenesis during vegetative and inflorescence development in maize. *Genetics* 179: 389-401.

Beavis WD (1997). QTL analysis: power, precision, and accuracy. In: Molecular dissection of complex traits (Paterson AH, ed.). CRC Press, London, 145-162.

Bomblies K and Doebley JF (2006). Pleiotropic effects of the duplicate maize FLORICAULA/LEAFY genes *zfl1* and *zfl2* on traits under selection during maize domestication. *Genetics* 172: 519-531.

Bomblies K, Wang RL, Ambrose BA, Schmidt RJ, et al. (2003). Duplicate FLORICAULA/LEAFY homologs *zfl1* and *zfl2* control inflorescence architecture and flower patterning in maize. *Development* 130: 2385-2395.

Bommert P, Nagasawa NS and Jackson D (2013). Quantitative variation in maize kernel row number is controlled by FASCIATED EAR2 locus. *Nat. Genet.* 45: 334-337.

Briggs WH, McMullen MD, Gaut GS and Doebley J (2007). Linkage mapping of domestication loci in large maize-teosinte backcross resource. *Genetics* 177: 1915-1928.

Brown PJ, Upadyayula N, Mahone GS, Tian F, et al. (2011). Distinct genetic architectures for male and female inflorescence traits of maize. *PloS Genet.* 7: e1002383.

Cai LC, Li K, Yang XH and Li JS (2014). Identification of large-effect QTL for kernel row number has potential for maize yield improvement. *Mol. Breed.* 34: 1087-1096.

Chuck GS, Brown PJ, Meeley R and Hake S (2014). Maize SBP-box transcription factors unbranched 2 and unbranched3 affect yield traits by regulating the rate of lateral primordial initiation. *Proc. Natl. Acad. Sci.* 111: 18775-18780.

Churchill GA and Doerge RW (1994). Empirical threshold values for quantitative trait mapping. *Genetics* 138:963-971.

Darvasi A and Soller M (1997). A simple method to calculate resolving power and confidence interval of QTL map location. *Behav. Genet.* 27: 125-132.

Doebley J (2004). The genetics of maize evolution. *Annu. Rev. Genet.* 38: 37-59.

Doebley J and Stec A (1991). Genetic analysis of the morphological differences between maize and teosinte. *Genetics* 129: 285-295.

Doebley J and Stec A (1993). Inheritance of the morphological differences between maize and teosinte: comparison of results for two F2 populations. *Genetics* 134: 559-570.

Doebley J, Stec A, Wendel J and Edwards M (1990). Genetic and morphological analysis of a maize-teosinte F2 populaton: implications for the origin of maize. *Proc. Natl. Acad. Sci.* 87: 9888-9892.

Guo JF, Su GQ, Zhang JP and Wang GY (2008).Genetic analysis and QTL mapping of maize yield and associate agronomic traits under semi-arid land condition. *Afr. J. Biotechnol.* 7: 1829-1838.

Iltis HH (2000). Homeotic sexual translocations and the origin of maize (*Zea mays*, Poaceae): A new look at an old problem. *Econ. Bot.* 54: 7-42.

Karen Sabadin P, Lopes de Souza JúniorC, Pereira de Souza A and Augusto Franco Garcia A (2008). QTL mapping for yield components in a tropical maize population using microsatellite markers. *Hereditas* 145: 194-203.

Korte A and Farlow A (2013). The advantages and limitations of trait analysis with GWAS: a review. *Plant Methods* 9: 29.

Lauter N and Doebley J (2002). Genetic variation for phenotypically invariant traits detected in teosinte: implication for the evolution of novel forms. *Genetics* 160: 333-342.

Lemmon ZH and Doebley J (2014). Genetic dissection of a genome region with pleiotropic effects on domestication traits in maize reveals multiple linked QTL. *Genetics* 198: 345-353.

Li F, Jia HT, Liu L, Zhang CX, et al. (2014). Quantitative traits loci mapping for kernel row number using chromosome segment substitution lines in maize. *Genet. Mol. Res.* 13: 1707-1716.

Li YL, Niu SZ, Dong YB, Cui DQ, et al. (2007). Identification of trait-improving quantitative trait loci for grain yield components from a dent corn inbred line in an advanced backcross BC2F2 population and comparison with its F2:3 population in popcorn. *Theor. Appl. Genet.* 115: 129-140.

Li HH, Ribaut JM, Li ZL and Wang JK (2008).Inclusive composite interval mapping (ICIM) for digenic epistasis of quantitative traits in biparental populations. *Theor. Appl. Genet.* 116:243-260.

Li JZ, Zhang ZW, Li YL, Wang QL, et al. (2011). QTL consistence and meta-analysis for grain yield components in three generations in maize. *Theor. Appl. Genet.* 122: 772-782.

Lincoln SE (1992). Mapping gens controlling quantitative traits with MAPMAKER/QTL 1.1. Whitehead Institute Technical Report, Cambridge, MA, USA.

Liu Y, Wang LW, Sun CL, Zhang ZX, et al. (2014). Genetic analysis and major QTL detection for maize kernel size and weight in multi-environments. *Theor. Appl. Genet.* 127: 1019-1037.

Lu M, Xie CX, Li XH, Hao ZF, et al. (2011). Mapping of quantitative trait loci for kernel row number in maize across seven environments. *Mol. Breed.* 28: 143-152.

Phillips KA, Skirpan AL, Liu X, Christensen A, et al. (2011). Vanishing tassel 2 encodes a grass-specific tryptophan aminotransferase required for vegetative and reproductive development in maize. *Plant Cell* 23: 550-566.

Saghai-Maroof MA, Soliman KM, Jorgensen RA and Allard RW (1984). Ribosomal DNA spacer-length polymorphisms in barley: Mendelian inheritance, chromosomal location, and population dynamics. *Proc. Natl. Acad. Sci.* 81: 8014-8018.

Santos FR, Pena SD and Epplen JT (1993). Genetic and population study of a Y-linked tetranucleotide repeat DNA polymorphism with simple non-isotopic technique. *Hum. Genet.* 90: 655-656.

Stuber CW, Edwards MD and Wendel JF (1987). Molecular marker-facilitated investigations of quantitative trait loci in maize. II. Factors influencing yield and its component traits. *Crop Sci.* 27: 639-648.

SzabóVM and Burr B (1996). Simple inheritance of key traits distinguishing maize and teosinte. *Mol. Gen. Genet.* 252: 33-41.

Tian BH, Wang JH and Wang GY (2014).Confirmation of a major QTL on chromosome 10 for maize kernel row number in different environments. *Plant Breed.* 133: 184-188.

Upadyayula N, Da Silva HS, Bohn MO and Rocheford TR (2006). Genetic and QTL analysis of maize tassel and ear inflorescence architecture. *Theor. Appl. Genet.* 112: 592-606.

Veldboom LR and Lee M (1994). Molecular-marker-facilitated studies of morphological traits in maize. II. Determination of QTLs for grain yield and yield components. *Theor. Appl. Genet.* 89: 451-458.

Yang C, Tang DG, Zhang L, Liu J, et al. (2015). Identification of QTL for ear row number and two-ranked versus many-ranked ear across four environments. *Euphytica* 206: 33-47.

Yan JB, Tang JH, Huang YQ, Zheng YL, et al. (2006). Quantitative trait loci mapping and epistatic analysis for grain yield and yield components using molecular markers with an elite maize hybrid. *Euphytica* 149: 121-131.

Yang J, Zhu J and Williams RW (2007). Mapping the genetics architecture of complex traits in experimental populations. *Bioinformatics* 23: 1527-1536.