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R. Babu · S. K. Nair · A. Kumar · H. S. Rao
P. Verma · A. Gahalain · I. S. Singh
H. S. Gupta

Mapping QTLs for popping ability in a popcorn × flint corn cross

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Abstract Popping expansion volume (PEV) in popcorn (*Zea mays L.*) is a distinct heritable character and defined as the ratio of the volume after popping to the volume before popping. PEV is quantitatively inherited and 3–4 genes/quantitative trait loci (QTLs) have been implicated. In the present study, we have dissected the quantitative PEV into two component traits, viz., flake volume (FV) and percent unpopped kernels (UPK), and mapped QTLs using SSR markers for all three traits with 194 F_3 families derived from a popcorn (A-1-6) × flint corn (V273) cross. Heritability (broad sense) estimates for PEV, FV and UPK based on F_3 mean bases were 0.72, 0.54 and 0.68, respectively. The QTL analyses for the three traits based on combined environment data were performed by composite interval mapping using QTL cartographer. Four QTLs were identified for PEV on chromosomes 1, 3, 8 and 10, which together explained 62% of the phenotypic variance (σ^2p). Four QTLs were found on chromosomes 1, 5, 9 and 10 for FV (explaining 44% of σ^2p) and five QTLs for UPK on chromosomes 1, 3, 4, 5 and 9 (explaining 57% of σ^2p). The relative efficiency estimates of marker-based selection in comparison to phenotypic selection for PEV (1.10), FV (1.22) and UPK (1.11) indicated that marker-based selection could be relatively more efficient. The QTL on chromosome 1S for PEV was found to be most significant, where QTLs for hard endosperm starch concentration had been detected earlier.

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A. Kumar and H.S. Rao contributed equally to this research work.

R. Babu (✉) · S. K. Nair · A. Kumar · H. S. Rao · P. Verma
A. Gahalain · H. S. Gupta
Vivekananda Institute of Hill Agriculture (ICAR), 263601,
Almora, Uttaranchal, India
E-mail: rbabu_icar@rediffmail.com
Tel.: +91-5962-230060
Fax: +91-5962-231539

I. S. Singh
Department of Plant Breeding, College of Agriculture,
G.B. Pant University of Agriculture & Technology,
Pantnagar, India

Introduction

Popcorn is considered the most primitive maize thus far known (Mangelsdorf and Smit 1949) and its discovery dates back to 2500 B.C. Presently, popcorn is an important commercial crop throughout the world and its production has been steadily increasing for decades (Zeigler 2001). Popcorn is tropical in appearance, with weak stalks, high ear position, prolificacy, day length sensitivity, many husks with no flag leaves, small ears and large tassels. Attempts to develop a tropical popcorn industry have been limited because of the absence of well-adapted, highly productive hybrids (Larish and Brewbaker 1999). Temperate popcorn cultivars and hybrids have poor stalk strength and lodging tolerance relative to field corns and are generally susceptible to tropical diseases and pests with exception to maize mosaic virus in Hawaii (Brewbaker 1981).

The foremost trait that distinguishes popcorn from all other types of corn is the ability to form large flakes after the kernels explode in response to heating, which is referred to as 'popping expansion' (Zeigler 2001). There are flint corns and some rare dent corns that will produce a flake when heated, but these flakes are very small as compared to the standard popcorn. Both pericarp thickness and endosperm starch type have been attributed to the popping ability in popcorn. The most tenable hypothesis advanced so far suggests that a more crystalline arrangement of cellulose and a higher degree of fibrillar packing in popcorn pericarp along with the ratio of hard to soft endosperm is primarily responsible for the explosion and significant flake formation in popcorn (da Silva et al. 1993).

Excellent popping ability is of utmost importance to the popcorn trade and all the available experimental evidences indicate that popping expansion is a heritable

character. The variability of individual ears for popping ability within an open-pollinated variety is similar to other well-known cases of quantitative inheritance controlled by many genes (Brunson 1955). Though detailed information on the inheritance of popping expansion in popcorn × dent/flint corn crosses is lacking, investigations carried out by Dofing et al. (1991) on inheritance of expansion volume in popcorn × dent corn crosses and Robbins and Ashman (1984) on parent–offspring popping expansion correlation in the progeny of dent corn × popcorn and flint corn × popcorn crosses indicate that the trait is quantitatively inherited and mostly additive in nature. Lu et al. (2003) reported three putative QTLs for popping expansion based on a preliminary study with $60BC_1$ families evaluated in one location by Kantety. In a more detailed study, Lu et al. (2003) identified four QTLs on chromosomes 1S, 3S, 5S and 5L, which together explained 45% of the phenotypic variation for popping expansion in a popcorn × dent corn cross. While a number of studies have involved dent corn as one of the parent in the genetic analyses, efforts to unravel information on the ability of normal flint corn to contribute towards superior popping expansion volume (PEV) have been very limited.

Popcorn germplasm, in general, is inferior to normal dent/flint corn in yield and other agronomic traits (Robbins and Ashman 1984; Dofing et al. 1991). It is a frequent practice to introduce dent corn germplasm into popcorn to improve traits such as yield, resistance to disease and stalk and root lodging in popcorn. Improved popping expansion and stalk strength in backcross-derived progenies from popcorn × dent corn crosses (Johnson and Eldredge) and popcorn × flint corn crosses (Robbins and Ashman 1984) have been reported. Nevertheless, complete recovery of PEVs in popcorn × dent/flint corn crosses practically requires several backcrosses with sufficient selection pressure for popping expansion in each breeding generation.

Molecular markers have emerged as powerful tools not only for mapping genes/QTLs governing economically important traits in crops (Dudley 1993), but also for unlocking the useful genetic diversity from unadapted/wild/unrelated germplasm (Tanksley and Nelson 1996). With the availability of robust SSR linkage maps in maize (Smith et al. 1997; Senior et al. 1998), marker-based selection could be useful to rapidly recover popping expansion during recurrent backcrossing programs of popcorn × dent/flint corn (Lu et al. 2003). There are two significant events that occur when popcorn pops—it explodes and produces a flake. The explosion part of the phenomenon is largely governed by the pericarp thickness and moisture in the kernel, while production of large flakes is associated with the ratio of hard (translucent) to soft (opaque) endosperm (Zeigler 2001). In the present study, we split PEV into two component traits, viz., percent unpopped kernels and flake volume, which seemingly correspond to the two events described by Zeigler (2001). The objectives of the present investigation were to study the genomic positions of QTLs for PEV and component traits in a popcorn × flint corn cross, to detect the magnitude and type of their genetic effects and to explore the efficiency of marker-based selection for faster recovery of PEV in the breeding programs.

Materials and methods

Population development

The cross of A-1-6 (popcorn) × V273 (normal flint) was made during the *Kharif* (monsoon) season of 2002 in the Hawalbagh farm of VPKAS, Almora. During the winter of 2002–2003, F_1 plants were selfed in the winter nursery of the Directorate of Maize Research, Amberpet, Hyderabad, India. In the *Kharif* (monsoon) season of 2003, F_2 plants from a single F_1 ear were selfed to develop the 194 F_3 families.

Experimental design

During the monsoon season of 2004, the 194 F_3 families were evaluated for popping expression in two environments, viz., Hawalbagh farm of VPKAS (ICAR), Almora, and Crop Research Centre of G.B. Pant University of Agricultural Science and Technology, Pantnagar, India. The two environments were diverse with respect to altitude (Almora: 1,250 amsl, Pantnagar: 244 amsl), temperature (maximum ranged from 25.5 to 30.3°C at Almora and 30.5 to 34.5°C at Pantnagar; minimum ranged from 10.2 to 20.4°C at Almora and 16.6 to 25.6°C at Pantnagar) and relative humidity (ranged from 72.4 to 84.25% at Almora and 77.5 to 93% at Pantnagar). The experiments at both the environments were conducted in an alpha lattice design (0, 1) along with parents in two replications of single row plots (5×0.75 m^2). Each plot was planted with one seed per hill and adjusted to a total stand of about 66,666 plants/ha. Standard cultural practices were followed at both the environments.

Trait evaluation

In the present study, three traits with respect to popping expansion, viz., PEV, flake volume (FV) and percent unpopped kernels (UPK) have been considered. PEV is defined as the ratio of popped corn volume to original unpopped corn volume. The popping expansion tests were conducted as described by Robbins and Ashman (1984) with slight modifications. The ears were gradually dried until the kernels reached the optimum moisture for popping ($13.5 \pm 1\%$). The middle portion of dried ears of each plot were shelled manually and bulked for popping expansion tests. A sample of 200 kernels from each plot was individually popped in a microwave oven

(LG-MS 304A) with 75% power for constant time. The original volume of unpopped corn was measured in graduated 100 ml glass cylinder while the popped corn volume was measured in 2 l graduated measuring cylinder. Flake volume (FV) refers to the absolute volume of 100 popped kernels and is measured using the 2 l graduated cylinder. The percentage of UPK is calculated based on the number of unpopped kernels in 200 kernels after popping.

Phenotypic data analysis

The initial analyses of variance for the 194 F_3 families were done by alpha program developed by CIMMYT (The CIMMYT Maize Program 1999). The efficiency of the alpha lattice analysis compared with a randomized complete block analysis was less than 1.0 for all the three traits based on combined data and hence unadjusted means were used as input for subsequent analysis. Variance components of the 194 F_3 families were estimated from linear functions of the mean squares. Standard errors of the estimated variances were calculated as the square root of the variance of the estimated variance component according to Anderson and Bancroft (1952). Broad sense heritability for the F_3 families on an entry-mean basis was calculated by dividing the genotypic variance (σ^2g) by the phenotypic variance (σ^2p). (Hallauer and Mirando 1981). Confidence intervals on heritability estimates were calculated according to Knapp et al. (1985). Coefficient of genotypic and phenotypic correlation among the three traits, viz., PEV, FV and UPK based on unadjusted entry means of the 194 F_3 families were calculated using SPAR1.

Molecular marker assay and linkage mapping

Molecular marker assay

DNA was extracted from 194 F_2 individuals of the mapping population employing the procedure described in Babu et al. 2005. Approximately 20 ng of DNA was used as template for PCR in a 15 µl reaction volume in MJ Thermal Cycler, USA (Model PTC-200). PCR cycling consisted of initial denaturation at 94°C for 2 min, followed by 30–35 cycles of amplification, at 94°C for 1 min, 58–68°C (based on T_m values of different SSR primers) for 2 min and 72°C for 2 min. A final extension step at 72°C for 7 min was followed by termination of the cycle at 4°C. Amplification products were separated in 3.5% Super fine resolution (SFR) agarose gel as per the procedure suggested by Senior et al. 1998. A total of 495 SSR loci uniformly spaced in the genome (maize GDB; http://www.maizegdb.org) were assessed for SSR polymorphism in the parental lines of the mapping population. Of the 495 loci, 101 were found to be polymorphic and producing discriminatory banding pattern in the mapping population. These were used for genotyping the 194 F_2 individuals for which phenotypic data were available.

Linkage mapping

Standard χ^2 test was used to test the segregation pattern at each marker locus for the deviations from expected Mendelian segregation ratio of 1:2:1 (for an F_2 mapping population). The overall mean frequencies for the alleles from both the parental lines were calculated, and the deviation, if any, from the expected allele frequencies (0.50:0.50) were tested. Linkage analysis was done with the software package MAPMAKER/EXP version 3.0 (Lincoln et al. 1992). A LOD score of 3.0 and a maximum recombination frequency of 0.40 were used to declare linkage between two markers. After determination of linkage groups and the correct linear arrangement of marker loci along the chromosomes, recombination frequencies between marker loci were estimated by multi-point analysis.

QTL mapping

Quantitative trait loci analysis was carried out on the set of 194 F_2 individuals with phenotypic data for PEV, FV and UPK. The genotypic data consisted of 101 SSR marker loci, and the phenotypic data comprised of untransformed PEV, FV and UPK from pooled data across environments. The method of composite interval mapping (Zeng 1994) was used to map QTLs and estimate their genetic effects. We employed CIM as implemented in QTL Cartographer version 2.5 (Wang et al. 2005). We used Model 6 of Zmapqtl procedure stipulating an F_2 population, scanning the genome every 2 cM, and specifying cofactors identified by forward and backward regressions that explained most of the variation for a given trait. Empirical threshold levels for declaring QTL significance at a Type I error rate of 0.05 were obtained by performing 1,000 permutations of the data according to the method of Churchill and Doerge (1994). The best estimate of QTL location was assumed to correspond to the position having the peak significance level and QTL support intervals were drawn as the entire region of the chromosome that exceeded the threshold value. Additive and dominance effects of the detected QTLs were also estimated by the Zmapqtl procedure. Because the dominance effects calculated for F_3 families are expected to be reduced by half compared with F_2 plants, we doubled the dominance effect estimates. The R^2 value from this analysis was accepted as the percent phenotypic variance explained by the locus.

Gene action was determined by the ratio of the absolute value of the estimated dominance effect divided by the absolute value of the estimated additive effect following Stuber et al. (1987) (additive 0–0.20, partial dominance 0.21–0.80, dominance 0.81–1.20, over dominance >1.20).

Results

Trait analysis

With respect to all the three traits, majority of the F_3 lines exhibited less than mid-parent values. No significant transgressive segregation was observed for any of the three traits. Mean squares and variance components for genotypes and genotype × environment interaction were significant at $P = 0.01$ for all the three traits. Heritability estimates for all the three traits were intermediate/high (Table 1).

Genotypic and phenotypic correlations among the three traits were highly significant. Both genotypically and phenotypically, FV was positively correlated (0.84 and 0.75) with PEV, while as expected, UPK was negatively correlated (−0.92 and −0.63) to popping expansion. Relatively weak correlations (−0.68 and −0.21) were observed between the component traits, viz., FV and UPK.

SSR data analysis and linkage mapping

Missing data in the genotyping of mapping population across 101 SSR loci were negligible (6.29%). χ^2 tests showed that eight markers deviated from the expected ratio of 1:2:1. Allele frequencies of parent V273 (flint) were significantly larger for *umc1155*, *umc1705, umc1822, umc2115, umc1456* and *umc1524*, whereas allele frequencies of the popcorn parent were significantly higher than 0.5 for the other two markers (*umc1357* and *umc2163*). Overall, the contribution of SSR alleles from the flint corn parent (V273) in the individuals of the mapping population was found to be 52.4%, ranging from 44.1 to 63.2%, while the contribution of SSR alleles from the popcorn parent (A-1-6) was 47.6%, ranging from 35.2 to 52.7%.

All the ten maize chromosomes were represented in the linkage map constructed with ten linkage groups (corresponding to ten chromosomes), spanning a total length of 2,022.2 cM at an average marker interval of 20.02 cM (Fig. 1). Almost all bin locations in the maize genome were represented in this linkage map except for some, particularly at the terminal regions of the chromosomes due to lack of polymorphic loci obtained from these regions. In the linkage map, there were some discrepancies regarding placements of markers as specified in the maize GDB. In some linkage groups, minor order changes were observed as in chr.2 (*umc1003* in bin2.05 placed in between markers in bin 2.06), chr.3 (*umc2261* in bin 3.04 placed in between markers in bin 3.05) and chr.10 (*umc1336* in bin 10.03 placed in between markers in bin 10.04).

QTL analysis

Popping expansion volume

Four QTLs were detected on chromosomes 1, 3, 8 and 10 (Table 2). One QTL on chr.1 in the interval between SSR markers, *umc1703* and *umc1661*, accounted for 30% of the phenotypic variance for PEV. The four QTLs detected for PEV contributed for 62% of $\sigma 2p$. The QTL on chromosome 10 (increasing PEV by 11%) was contributed by the flint corn parent, apart from which all the QTLs detected for increasing PEV were contributed by the popcorn parent. Two QTLs detected on chr.8 and chr.10 were showing over dominance, one on chr.1 showed dominance and the QTL on chr.3 showed partial dominance.

Flake volume (FV)

We identified four QTLs for the volume of 100 popped kernels (FV) on chromosomes 1, 5, 9 and 10, which accounted for 44% of the $\sigma^2 p$ with individual effects ranging from 4 to 18%. The QTL on chr.10 increasing the FV (at the same bin location as that of PEV) was contributed by the flint corn parent. Two QTLs detected on chr.1 and chr.10 showed over dominance, and the QTLs detected on chromosomes 5 and 9 showed dominance and partial dominance, respectively.

Percent unpopped kernels

For UPK, five QTLs were detected on chr.1, 3, 4, 5 and 9. All the QTLs for decreasing UPK were contributed by the popcorn parent except for one QTL on chr.4 in the marker interval phi096 and umc1299, which was contributed by the flint corn parent. Out of the five QTLs detected, three were over dominant, one on chr.4 was partially dominant and the one QTL on chr.9 was showing dominant gene action.

Table 1 Trait means, standard errors and heritability values for 194 F_3 families from the cross A-1-6 (popcorn) × V273, parents and F_1s based on combined data across two environments

Traits	Mean ± SE				Heritability (broad sense)
	F_3 families	A-1-6 (popcorn)	V273	F_1	
Percent unpopped kernels (UPK)	23.9 ± 0.56	0.5 ± 0.5	64.8 ± 7.8	24.5 ± 2.7	0.68 (0.60–0.74)
Flake volume (FV)	150.6 ± 2.2	310 ± 31.2	95.8 ± 16.6	157.5 ± 16.7	0.54 (0.42–0.63)
Popping expansion volume (PEV)	5.76 ± 0.10	18.4 ± 2.1	2.5 ± 0.4	5.67 ± 0.49	0.72 (0.65–0.78)

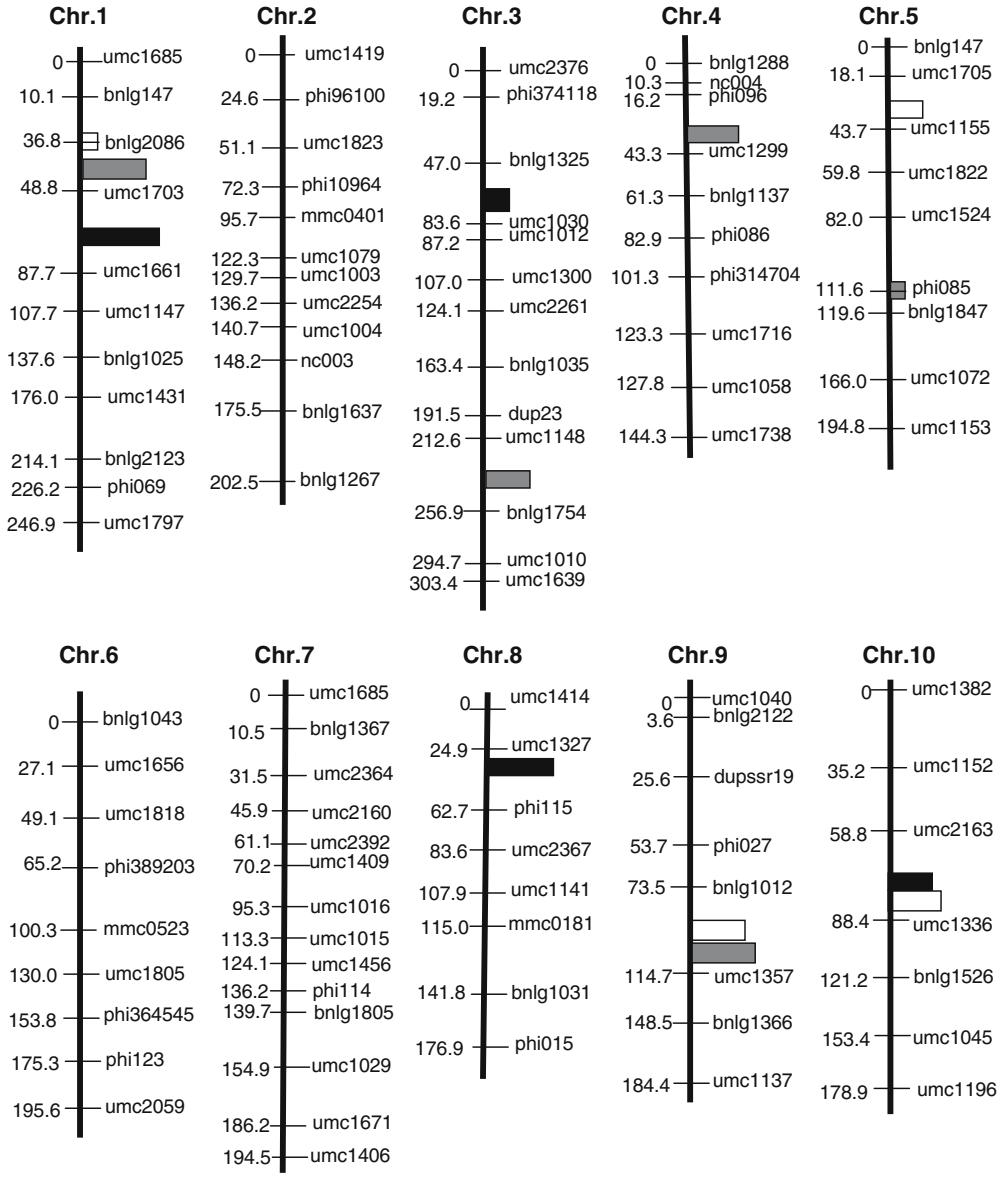

Fig. 1 Molecular linkage map of 101 SSR loci in an F_3 mapping population of A-1-6 × V273 and the location of QTLs for PEV, FV and UPK. *Numbers to the left* of the chromosomes indicate the cumulative distance in cM. Names of loci are indicated towards the *right* of the chromosomes. *Open bars* towards the right of the chromosomes depict QTLs for FV, *gray-colored bars* depict QTLs for UPK and *solid bars* depict QTLs for PEV. The *lengths of the bars* are indicative of the phenotypic variance explained by the QTLs

Discussion

Heritability, variance and correlation estimates for popping expansion volume and component traits

The distribution of means of the F_3 families for all the three traits PEV, FV and UPK reflected the quantitative genetic basis of these traits. None of the traits exhibited significant transgressive segregation suggesting that the popcorn parent (A-1-6) contained most of the favorable alleles with major effects. Even though some loci from the normal flint corn parent (V273) contributed popping expansion alleles, the population size of 194 F_3 families was too small to expect all the favorable alleles to be present in any single family. Heritability estimates for PEV (0.72) based on combined environment analysis was in close agreement with the reported heritability of PEV of 0.73 (Lu et al. 2003).

Significant positive correlations between PEV and FV and negative correlation between PEV and UPK coupled with detection of few identical QTLs for these traits in the present study substantiate that FV and UPK are important component traits of quantitative PEV. The weak correlation observed between the two component traits along with the discovery of different additional

Table 2 QTLs for PEV, FV and UPK based on combined data across environments for 194 F_3 families from the cross of A-1-6 × V273

Chrom/bin[a]	Position[b]	Interval	Max LOD[c]	R^{2d}	Gene effect[e]		Gene action[f]
					Additive	Dominant	
Popping expansion volume (PEV)							
1.05	67.0	*umc1703–umc1661*	4.4	30	1.8	−1.6	D
3.04	82.0	*bnlg1325–umc1030*	3.2	8	0.7	−0.2	PD
8.01	37.5	*umc1327–phi115*	2.6	28	1.7	−2.8	OD
10.04	70.8	*umc2163–umc1336*	3.6	11	−0.8	−1.8	OD
				62			
Flake volume (FV)							
1.05	36.8	*bnlg2086*	2.6	4	8.5	6.2	OD
5.03	24.1	*umc1705–umc1155*	2.8	10	22.6	−22.1	D
9.04	87.5	*bnlg1012–umc1357*	3.0	18	18.2	−11.6	PD
10.03	82.8	*umc1336–bnlg1526*	4.0	17	−19	−24.6	OD
				44			
Percent unpopped kernels (UPK)							
1.05	40.8	*bnlg2086–umc1703*	5.0	23	−6.9	−9.4	OD
3.07	215.3	*umc1148–bnlg1754*	3.2	10	−4.1	7.0	OD
4.06	32.0	*phi096–umc1299*	3.4	14	6.2	−2.8	PD
5.06	111.6	*phi085*	2.7	3	−1.7	−3.2	OD
9.04	94.5	*bnlg1012–umc1357*	2.6	20	−6.2	−7.0	D
				57			

[a]Bin locations of the nearest SSR marker
[b]Position of QTL peaks as indicated by the cumulative distance from the end of the short arm
[c]Likelihood ratio/4.6052. Critical thresholds established by permutation to give a genomewise error rate of $\alpha = 0.05$
[d]Percentage of phenotypic variance explained by the genotype class at LOD peak
[e]Additive and dominance gene effects at QTL peak; additive effects indicate the effects of A-1-6 alleles; dominance effects were doubled because $F_{2:3}$ lines were used rather than F_2 individuals
[f]Gene action was estimated by $(d)/(a)$: A (additive gene action) 0–0.20, PD (partial dominance) 0.21–0.80, D (dominance) 0.81–1.20 and OD (over dominance) > 1.20

QTLs for both the traits suggest scope for simultaneous improvement of these component traits in popcorn × flint corn crosses. Similar correlations among PEV, flake size and percent unpopped kernels have been reported by Dofing et al. (1990) in their study on genotype × popping method interaction for expansion volume in popcorn.

QTLs for popping expansion volume and its component traits

Our analysis revealed four QTLs each for PEV and FV and five for UPK. To determine if QTLs detected in this study for different traits may be equivalent, we used the bin concept. The bins in maize are generally 15–20 cM in length, essentially the same length as the confidence intervals for most QTLs (Jampatong et al. 2002). We considered QTLs for separate traits to be potential matches, if they occurred in the same or adjacent bins.

Popping expansion volume was found to be controlled by four genomic regions at bin locations 1.05, 3.04, 8.01 and 10.04 contributing 62% of the σ^2p for the trait. This result agrees with the earlier postulation that 3–4 genes may be involved in inheritance of PEV in popcorn (Lu et al. 2003). Of these QTLs, the one on 1.05 was contributing 30% of the σ^2p individually for the trait. This region was found to be significant for the two component traits, viz., FV and UPK as well. Hence what we identify as QTL for PEV may be due to the combined effects of increased flake volume and decreased number of unpopped kernels contributed by the popcorn parent. Lu et al. 2003 also identified a QTL for PEV explaining 16% of σ^2p at chr.1S in their experiment using a backcross breeding population of dent corn × popcorn in US environments. In another preliminary study, Lu et al. 2003 reported a QTL near the centromere of chr.1 for PEV using a different popcorn × dent corn cross, which was speculated to be the same as the one detected in the earlier study. Thus, this region in chr.1S appears to be an extremely significant region in the overall expression of PEV along with its component traits.

Both pericarp thickness and endosperm starch type (hardness) have been attributed to popping ability in popcorn (Hoseney et al. 1983; da Silva et al. 1993; Matz 1984). Dofing et al. (1990) suggested that popping phenomenon is a function of the physical structure of the entire kernel and the microscopic structure of the endosperm. The explosion part of the phenomenon is largely governed by the pericarp thickness and moisture in the kernel, while production of large flakes is associated with the ratio of hard (translucent) to soft (opaque) endosperm. These two aspects seemingly correspond to the two component traits considered in the present study, viz., UPK (with pericarp thickness) and FV (with ratio of hard to soft endosperm). Some earlier studies for mapping starch concentration have mapped significant QTLs at the same region (chr.1S) affecting this trait (Goldman et al. 1995; Berke and Rocheford 1995). In an attempt to map weevil resistance in maize, Bergvinson

(2000) identified QTLs for kernel hardness again on chr.1S. The results obtained in the present study along with earlier reports of QTLs for PEV and hard starch type strongly suggest that this part of the genome on chr.1S could be one of the most important regions responsible for popping traits in popcorn.

The QTL detected at bin location 10.04 contributing 11% of σ^2p for PEV seems to be the same QTL detected at the same location for FV. In both these cases, the QTL increasing the respective traits was contributed by flint corn parent. Though the recovery of popping expansion in popcorn × dent corn and popcorn × flint corn crosses had been found to be almost the same (Robbins and Ashman 1984), in our opinion faster and higher recovery of popping expansion should be possible in popcorn × flint corn crosses as there exists potential opportunity to introgress popping expansion alleles from the flint corn parent as well.

Out of the four QTLs detected for PEV, two were not detected for any of the contributing traits. Similarly some QTLs, like the ones on 5.03 and 9.04 for FV and 3.07, 4.06, 5.06 and 9.04 for UPK, were not observed for PEV. These findings along with weak correlation estimates between component traits justify our effort to break the quantitative PEV into component traits, viz., FV and UPK, which in turn should facilitate simultaneous selection for the component traits along with PEV. In the earlier QTL mapping study for popping ability in popcorn × dent corn cross by Lu et al. (2003), four QTLs were identified on chr.1S, 3S, 5S and 5L contributing 16, 16, 13 and 15% of σ^2p for the trait, respectively, and the QTL on 5L was considered the most significant as it was also detected in other independent mapping populations. We identified few QTLs either for PEV or for the component traits in these regions as well. Though chr.5L was not showing significant effect in our study on PEV as such, its effect was mostly detectable on FV and UPK. In a segregating population, a QTL can be identified only if both parental lines contributed different alleles at the QTL. It could be possible that the putative QTL on 5L could have been monomorphic in our parental lines, which may explain the lack of congruency between our study and that of Lu et al. (2003). Also, our analysis revealed QTLs in overlapping or mostly adjoining regions in the same chromosomes affecting the PEV and component traits. Most of the QTLs detected were clustered around specific regions in chromosomes 1, 3, 5, 8 and 10 than in a random manner.

Type of gene action for popping expansion and prospects for marker-assisted breeding

For PEV, two QTLs showed over dominant gene action, one QTL each showed partially dominant and dominant gene action. Similarly for the component traits, viz., FV and UPK, majority of the QTLs showed over dominant gene action. However, the earlier conventional genetic studies (Dofing et al. 1991) have reported significant additive gene action in popcorn × dent corn crosses for the PEV. Though additive gene action for PEV was more generally reported, there were notable exceptions such as inbred lines, Sg67, SA133 and J-2, which gave excellent popping expansions in specific combinations and mediocre results in others (Brunson 1955). Similar studies involving popcorn × flint corn crosses are scarcely reported. One possible reason for differences with respect to the type of gene action for popping expansion from conventional genetic studies could be due to the fact that estimate of the sum of dominance effects contributing to high PEV obtained from diallel analyses may not be very accurate because dominance for high and low popping volumes may cancel each other (Schon et al. 1993).

The relative efficiency (RE) of marker-based selection in comparison to phenotypic selection depends on the proportion of the additive variance explained by the markers and the heritability and can be estimated as $RE = [Q^2/h^2]^{1/2}$ (Lande and Thompson 1990). Estimates of relative efficiency for PEV, FV and UPK were 1.10, 1.22 and 1.11, respectively, in the present study. This indicates that marker-based selection, though marginal, could be relatively more efficient than conventional selection for all these traits. However, identification of potential candidate QTLs for MAS is crucial, which depends upon the consistency of QTLs across environments, the proportion of phenotypic variance they explain and the distance between the marker and QTL. Based on the results obtained in our study, the QTLs in chromosome bins 1.05 8.01 and 10.04 are good candidates for PEV. Similarly, QTLs in chromosome bins 10.03 and 9.04 for FV and 3.07, 1.05 and 9.04 for UPK seem to be potential candidates for MAS.

The most frequently adopted breeding approach to improve popcorns involves hybridization with field corns. Most field corns in temperate regions are soft dents, while many are hard flints in the tropics. Selection for popping expansion has been found to be effective in backcrosses to the popcorns (Johnson and Eldredge 1953) and equally effective among F_3 progenies from dent as well as flint crosses (Robbins and Ashman 1984). However, hybridization of popcorns with temperate dent varieties has often led not only to increased kernel weights and grain yields but also to greatly reduced popping expansions (Larish and Brewbaker 1999). Considering the practical difficulty that conditioning of ears for popping expansion tests and, subsequently, phenotypic selection for PEV are laborious and time consuming, MAS provides an attractive proposition and could effectively complement the recovery of PEV in recurrent backcrossing programs aimed at the improvement of popcorn germplasm. The discovery of different QTLs with significant genetic effects for PEV, FV and UPK in the present study provides ample scope for an effective pyramiding approach, in which candidate QTLs could be simultaneously selected using PCR-based

cost-effective marker systems in recurrent backcross generations and phenotypic selection for PEV could be deferred to later advanced generations. Besides, QTL detection results in our study demonstrate that field flint corn could also contribute favorable allele for increasing PEV (QTL in chromosome bin location 10.04 for PEV), which could potentially help in recovering high PEVs in backcross programs.

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