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Fine mapping of a quantitative trait locus (QTL) from *Lycopersicon hirsutum* chromosome 1 affecting fruit characteristics and agronomic traits: breaking linkage among QTLs affecting different traits and dissection of heterosis for yield

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Abstract The near-isogenic Line TA523, containing a 40-cM introgression at the bottom of chromosome 1 from *Lycopersicon hirsutum* acc. LA1777, affects several agronomically important traits. A set of recombinant lines (subNILs) derived from the original NIL TA523 were developed in order to fine-map, by substitution mapping, the genetic factors included within the original introgression. In the current experiment, TA523 showed redder, rounded, less pigmented shoulder, lower-weighted fruits and higher brix, whereas higher yield and brix*yield was observed only in the hybrid TA253×TA209 suggesting heterosis for these traits. By substitution mapping we mapped independent genetic loci affecting brix, yield and fruit shape, whereas fruit weight, shoulder pigmentation and external color mapped to a position coincident with the brix locus. Analysis of the subNILs revealed that the gene action of most of the QTLs was additive or nearly additive. The exception was for the yield QTL which was dominant (d/a=0.7), eliminating the possibility that yield increase is due to true overdominance at a single gene locus. However, no negative yield effects were detected in other regions of the introgressed segment, as would be predicted by a dominance complementation model. Therefore, epistatic interactions among genetic factors along the introgressed segment are suggested as the cause of yield heterosis. Results from this study, combined with previous experiments involving different tomato wild species, demonstrate that the base of chromosome 1 of tomato contains multiple QTLs affecting various agronomic and fruit traits and that these effects can not be attributed to the pleiotropic effects of a single locus.

Key words Heterosis · Fine mapping · Genetic interactions · Yield · Brix

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Introduction

The use of wild germplasm to introgress novel genetic variability into elite cultivars is one of the more important challenges for breeders. Wild germplasm has been demonstrated as useful to improve disease resistance (Yu et al. 1995; Mestre et al. 1997; Sandbrink et al. 1995; Stevens et al. 1995), salt tolerance (Monforte et al. 1996) and agronomic traits (Eshed and Zamir 1995; Grandillo and Tanksley 1996; Bernacchi et al. 1998a). Unfortunately, the introduction of exotic genes into an elite genome can lead to a decrease of desirable agronomic characteristics due to the linkage drag of genes with undesirable effects from the wild species. By using molecular markers and suitable statistical methods (Tanksley 1993) the chromosomal location of quantitative trait locus (QTL) involved in the character under study can be determinated and selected, allowing the breeder to discard other regions with undesirable effects after relatively few generations of marker-assisted selection (Young and Tanksley 1989). Furthermore, near-isogenic lines (NIL) with contain only one introgression from the wild species can be also constructed (Eshed and Zamir 1995).

NILs have been developed from diverse interspecific crosses in tomato (Eshed and Zamir 1995; Tanksley et al. 1996; Bernacchi et al. 1998b). Despite the fact that these NILs usually contain only a small percentage of the donor genome (less than 5%), they are often modified for several traits, including some undesirable effects. It is necessary to reduce the extent of the wild introgression in order to assess whether the undesirable effects are caused by the linkage drag of other genes or by pleiotropic effects of a single locus. Breaking the undesirable linkage effects can render introgressions more useful in breeding programs. This also has the added benefit of preventing the risk of losing desirable genes by recombination after the generations needed to incorporate them into the elite germplasm using marker-assisted selection.

Bernacchi et al. (1998b) developed a set of NILs from a cross between *Lycopersicon esculentum* cv E6203 and

Lycopersicon hirsutum acc. LA1777. One of them, TA523, contains the distal 40-cM introgression of chromosome 1, a region previously shown to affect several traits including brix, yield brix*yield and fruit color (Bernacchi et al. 1998a, b). The main objectives of the current study were to determine the number of loc which might be responsible for the phenotypic effects modified in TA523 and to distinguish between pleiotropy and linkage in the genetic control underlying the traits affected by this region by analyzing a new set of recombinant subNILs.

Materials and methods

Development of new recombinant lines (subNILs)

Two hundred and sixteen F_2 seeds were generated from the cross between TA523 NIL (see Fig. 1) and the processing inbred E6203 (TA209) (Bernacchi et al. 1998b). DNA from 216 F_2 plants was extracted, digested with *Eco*RI, electrophoresed in agarose gels, immobilized on Hybond N^+ nylon membranes (Amersham) and hybridized with TG161 and CT190 markers, which flanked the TA523 introgression (Bernacchi et al. 1998b). Three more markers (CT163, TG617 and TG267) internal to the introgressed segment were also scored in this population to verify the putative recombination break points. A molecular linkage-marker map of the base of chromosome 1 was generated from the TA523 F_2 population using the software package MAPMAKER 3.0 (Lander et al. 1987). The Kosambi mapping function (Kosambi 1944) was used to convert recombination frequencies of centriMorgans (cM). Eleven independent recombinat plants were selected and allowed to self. F_3 plants were scored with appropriate markers to fix the recombinant chromosomes. Additional markers were scored to determine the exact point of recombination within each one of the fixed 11 subNILs (Fig. 2).

Field evaluation

Five plants of each homozygous subNIL, ten of each hybrid with TA209, ten of both homozygous and hybrid original NIL TA523, and 50 control TA209 were transplanted to the field, allowing 1 m^2 for each plant, on June 1st 1998 in Ithaca, N.Y., in a complete randomized design. All the plants were harvested on October 5th. Due to field heterogeneity some plants did not grow properly and were excluded from the experiment. Ten traits were measured for each single plant: total yield, average fruit weight, solid-soluble concentration, brix*yield, internal and external color, shoulder pigmentation, epidermal reticulation and fruit shape. Total yield (TY) was evaluated as the weight in kilograms of all the tomatoes (green and mature) produced at the harvest time. Ten mature fruit of each plant were selected to evaluate the fruit traits. Average fruit weight (FW) was recorded in grams, soluble solids was measured as brix from juice for at least five mature fruits using a refractometer. Brix*yield (BY) was obtained as the product of TY and brix. Other fruit characteristics were scored visually using a scale from 1 to 5; internal and external fruit color (ICOL and ECOL): 1=pale–5=dark-red, shoulder pigmentation (SHO) as the extension of yellow pigmentation around the stem scar, 1=no pigmentation 5=extensive pigmentation; epidermal reticulation (ER): 1=smooth and bright 5=cracked; fruit shape (FS) 1=round 5=elongated. Shape was also measured as the ratio longitude/diameter (L/D) only in the homozygous subNILs.

Data analysis

The NIL TA523, its hybrid with TA209, and both homozygous and hybrid subNILs were compared with the TA209 isogenic control for each trait using Dunnett's contrast (Dunnett 1955) at $P \leq 0.05$ with the JMP V.3.1 software package for Macintosh (SAS Institute 1994). Genotypes with less than four replications were excluded from the analysis.

The subNILs also differed in marker-genotype composition, so QTL analysis could also be made by marker-genotype mean comparisons. This gives more replications for each chromosomal region and hence better estimates of the genetic effects. Marker-means comparisons by single-point analysis were performed using the software package QGENE (Nelson 1997). Due to linkage among markers, only seven different marker comparisons were done. The Bonferroni correction that gives an overall $\alpha=0.05$ has a threshold for each comparison $P \leq 0.05/7=0.007$ or $-\log(P) \geq 2.14$. Both strategies were combined to fine-map and estimate the effects of a QTL within the TA523 subNIL: (1) when a subNIL showed significant effects compared with the TA209 control according to Dunnett's contrast then a QTL was considered to be within the genomic region covered such subNIL. If several subNILs showed the effect, the QTL was located within the chromosomal region shared by the subNILs. (2) If the Dunnett contrast did not resolve the location of the QTL, it was estimated accordingly by a marker-mean comparison. (3) When a QTL was located within a specific chromosomal region, the additive effects (a), measured as a percentage of deviation from the TA209 control, and mode of gene action (d/a) of the QTLs were estimated according with marker means as a=100*[(HH–EE)/2EE], d/a={EH–[(HH+EE)/2]}/[(HH–EE)/2], where HH is the phenotypic mean of individuals homozygous for *L. hirsutum* alleles for the marker locus, EH the phenotypic mean of individuals heterozygous for the some locus, and EE is the TA209 mean.

Results

Original NIL performance

Table 1 shows the means, standard deviation and significance of the contrast among the original NIL (TA523), the recurrent inbred parent TA209 and the hybrid TA523×TA209. TA523 showed redder, rounded, less shoulder-pigmented, lower-weighted fruit and higher brix, whereas yield was not significantly different from the control. No significant effects were observed for internal fruit color, epidermal reticulation and stem scar (data not shown). Remarkably, the hybrid NIL showed a higher yield and brix*yield than either TA209 or the ho-

Table 1 Mean and standard error of quantitative traits for the control (TA209), the TA523 NIL and the F_1 significantly different from control at * $P<0.05$, ** $P<0.01$

Genotype	ECOL	SHO	Shape	L/D	FW	Brix	TY	B*Y
TA209	2.52±0.06	3.34±0.06	3.16±0.04	1.16±0.05	96.77±2.14	4.98±0.10	4.85±0.35	26.20±1.54
TA523×TA209	2.83±0.08*	2.55±0.23**	2.89±0.11**	1.06±0.10*	93.89±6.13	5.16±0.20	7.41±0.78**	39.23±4.25**
TA523	3.13±0.16**	2.77±0.18	2.83±0.08**	1.09±0.09*	82.04±4.18*	5.76±0.26**	4.65±0.91	28.04±4.78

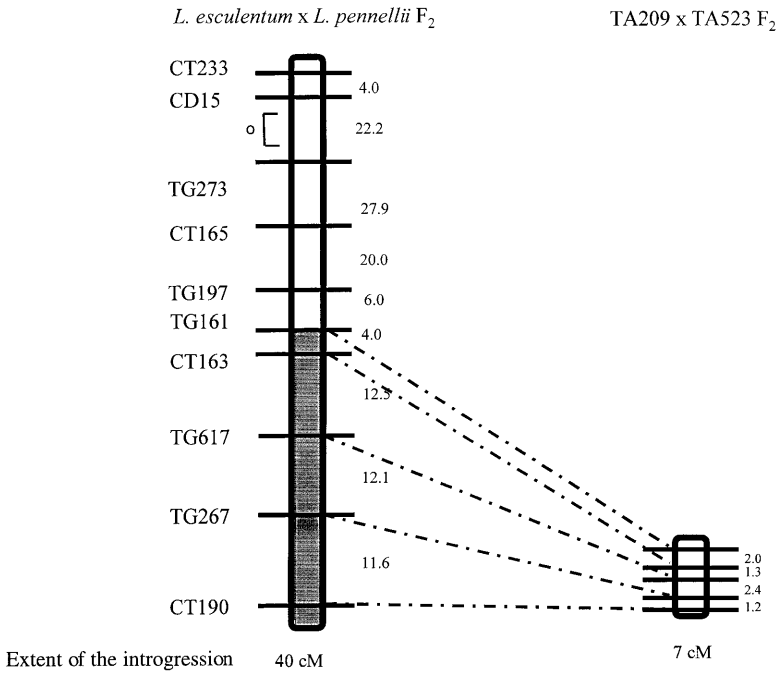


Fig. 1 Linkage-map comparisons for chromosome 1 developed from *L. esculentum*×*L. pennellii* F_2 (Tankley et al. 1992) and TA523×TA209 F_2. The *shadowed region* in the *L. esculentum*×*L. pennellii* map represents the extent of the introgression in the TA523 NIL. The position of the centromere is also indicated

mozygous TA523 NIL, suggesting overdominant gene action at the locus/loci underlying those traits. According to these results, the TA523 introgression region is expected to contain QTLs affecting external fruit color, fruit shape, shoulder pigmentation, fruit weight, brix, yield and brix*yield.

SubNIL developmen and recombination suppression in the introgressed segment

The TA523 NIL contains the distal 40 cM of chromosome 1 from *L. hirsutum*, corresponding to approximately 3% of the tomato genome (Fig. 1). In order to assess which effects shown by TA523 are controlled by linked genetic loci versus pleiotropy at one single locus, and to study the heterotic yield effects seen in the hybrid NIL, a set of recombinant subNILs were constructed from a F_2 population derived from a cross between TA523 and TA209 (NIL F_2). Based on this F_2, it was possible to construct a linkage map for the bottom of chromosome 1. Comparison of this linkage map with the published tomato linkage map from a cross between *L. esculentum* and *L. pennellii* (Tanksley et al. 1992) showed that there was an 80% recombination shrinkage in the NIL F_2 population (Fig. 1). Bernacchi and Tanksley (1997), however, did not observe recombination suppression within this region of the chromosome 1 in the original backcross population from which this NIL was developed.

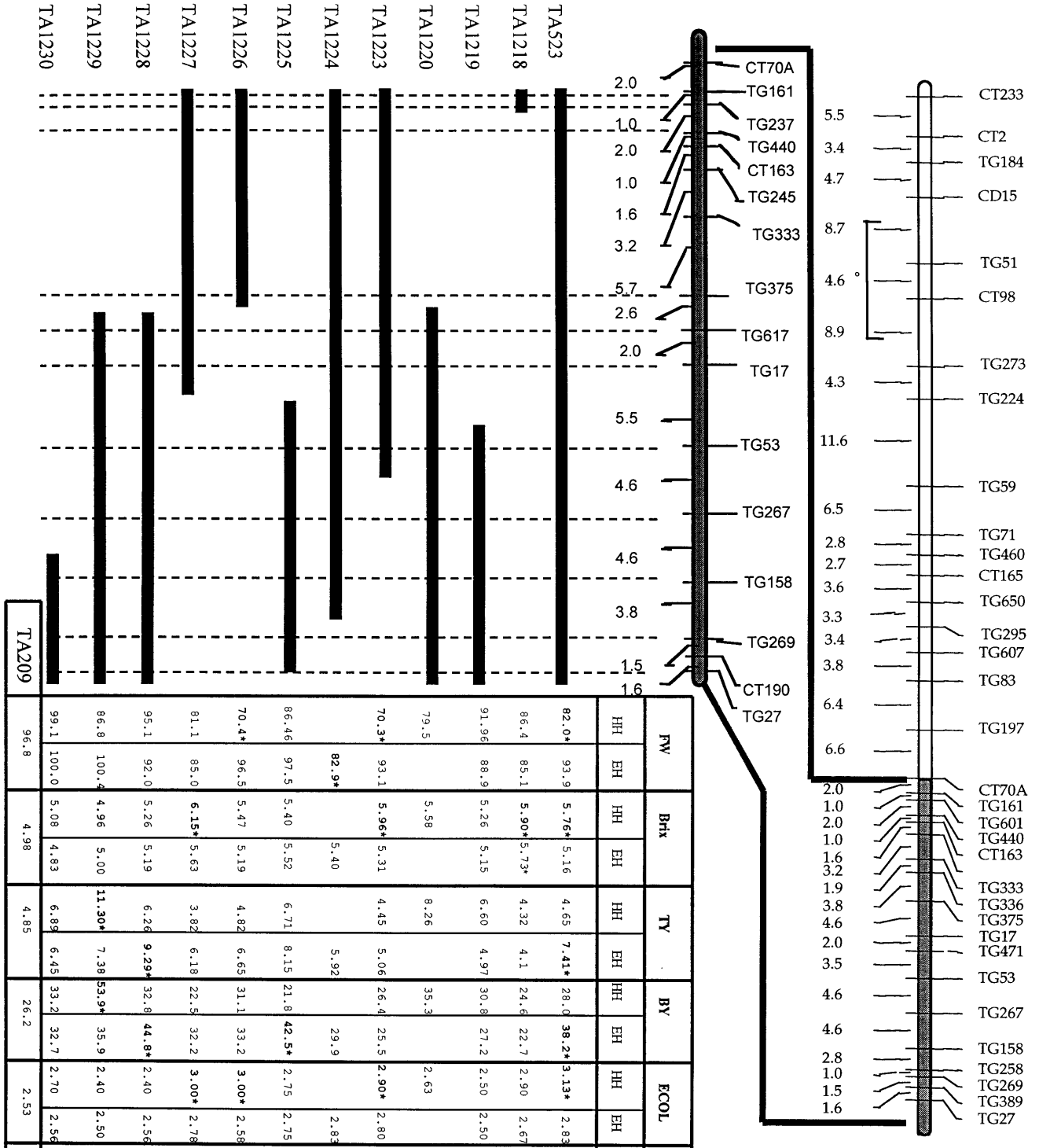
Recombination shrinkage in advanced generations of interspecific tomato crosses has been extensively reported (Rick 1969; Paterson et al. 1990; Ganal and Tanksley 1991; Alpert and Tanksley 1996; Grandillo et al. 1996); but it is not normally observed in ealy populations such as F_2 or BC1 (Rick 1969; Tanksley et al. 1992; de Vicente and Tanksley 1993; Bernacchi and Tanksley 1997). This phenomenon seems to be a consequence of the introgression of small regions of exotic DNA into the cultivated background, regardless of the wild species used. Sequence divergence between the wild and cultivated species has been proposed as the reason for this recombination shrinkage (Paterson et al. 1990). Selective crossing-over within a chromosome for more similar regions (non-introgressed) versus less similar (introgressed) would lead to recombination suppression in the introgressed region. This would not be the case in F_2 or backcross populations, since the entire length of the chromosome is heterozygous in the F_1 generation.

Fine mapping of QTLs within the TA523 introgression

Figure 2 depicts the subNILs and their means for the agronomic and fruit traits. Different subNILs were significantly different from the control (TA209) depending on the trait: subNILs sharing introgressions from TG161 to TG617 (TA1218, TA1223, TA1224, TA1226, TA1227) increased brix and external color, and decreased fruit weight and shoulder pigmentation. Only subNILs with introgressions from TG27 (distal marker on the chromosome), such as TA1225, TA1228 and TA1229, increased TY or BY in either the homozygous or heterozygous stage. Three subNILs (TA1223, TA1224 and TA1227) had significantly more-rounded fruit.

In order to increase the statistical power and obtain more precise estimates of the additive effects and the gene action of the QTLs, comparisons based on individual marker genotypes were also performed (Fig. 3).

Markers within the TG161–TG375 interval showed strong effects on brix. TA1218, and its corresponding



	FW		Brix		TY		BY		ECOL		SHO		Shape		L/D
TA209	HH	EH	HH	EH	HH	EH	HH	EH	HH	EH	HH	EH	HH	EH	HH
96.8	99.1	100.0	5.08	4.83	6.89	6.45	33.2	32.7	2.70	2.56	3.40	3.00	3.40	3.10	1.17
4.98	86.8	100.4	**4.96**	5.00	**11.30***	7.38	**53.9***	35.9	2.40	2.50	3.40	3.18	3.40	3.33	1.17
4.85	95.1	92.0	5.26	5.19	6.26	**9.29***	32.8	**44.8***	2.40	2.56	3.10	3.18	3.40	2.88	1.18
26.2	81.1	85.0	**6.15***	5.63	3.82	6.18	22.5	32.2	**3.00***	2.78	2.75	3.00	**2.70***	2.89	**1.05***
2.53	**70.4***	96.5	5.47	5.19	4.82	6.65	31.1	33.2	**3.00***	2.58	3.00	3.10	3.17	3.21	1.14
3.34	86.46	97.5	5.40	5.52	6.71	8.15	21.8	**42.5***	2.75	2.75		2.80	3.25	3.06	
3.16		**82.9***		5.40		5.92		29.9		2.83		2.60		**2.67***	
1.15	**70.3***	93.1	**5.96***	5.31	4.45	5.06	26.4	25.5	**2.90***	2.80	**2.30***	2.50	**2.50***	2.90	**1.06***
	79.5		5.58		8.26		35.3		2.63		2.75		3.00		1.12
	91.96	88.9	5.26	5.15	6.60	4.97	30.8	27.2	2.50	2.50	3.30	2.75	3.50	3.40	1.19
	86.4	85.1	**5.90***	5.73*	4.32	4.1	24.6	22.7	2.90	2.67	**2.30***	3.10	3.10	3.33	1.14
	82.0*	93.9	**5.76***	5.16	4.65	**7.41***	28.0	**38.2***	**3.13***	2.83	**2.77***	**2.55***	**2.83***	**2.89***	**1.09***

Fig. 2 Delineation of the introgressed segments and trait means among homozygous (*HH*) and heterozygous (*EH*) subNIL families. Means of the original TA523 and TA209 control lines are also shown. Statistically significant differences between subNILs and the control (Dunnett's method, $P<0.05$) are marked as *

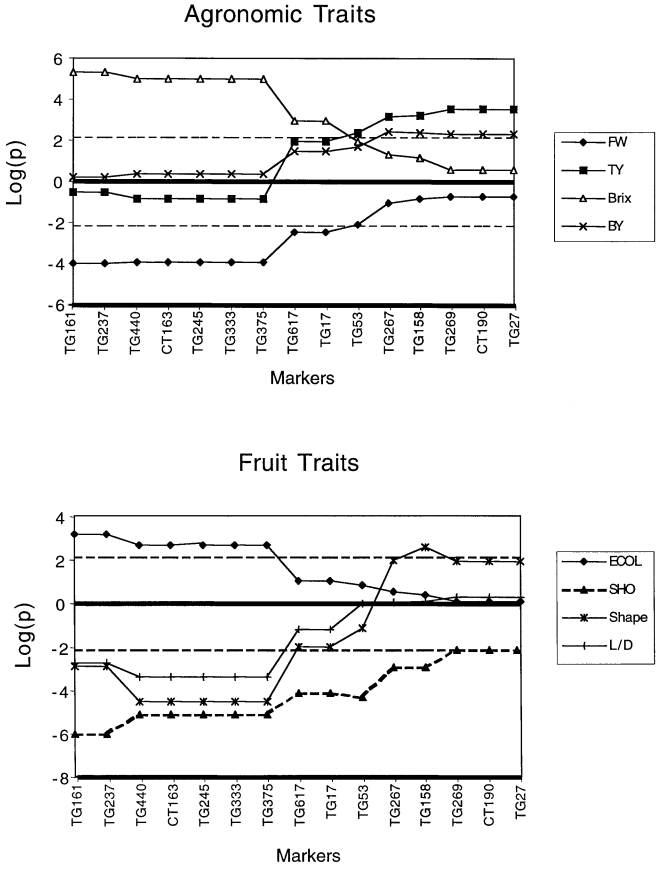


Fig. 3 Significance [*Log(p)*] of the marker-means comparisons within the subNIL population. Positive values indicate that *L. hirsutum* alleles increased the trait mean, whereas negative values mean that *L. hirsutum* alleles decreased the trait mean

hybrid, gave rise to the subNILs with a shorter introgression that increased brix, indicating that the brix locus is between the markers CT70A and TG440 (less than a 4-cM interval) and its mode of gene action is strictly additive (d/a=0, Fig. 4).

The marker-means comparison for FW were largely coincident with brix suggesting that the most likely position of the FW QTL is between TG161 and TG440 (Fig. 3), but we can reject the possibility that there is a FW QTL between the TG375 and TG617 markers. As for the brix QTL, the mode of gene action for FW was additive (d/a=0.2, Fig. 4).

A marker-means comparison showed that the most likely location of the TY QTL is at the end of the chromosome between TA158 and TG27 (about7 cM). The gene action of the TY QTL is dominant (d/a=0.7, Fig. 4). Overdominant gene action would have been expected for this QTL based on the results shown in Table 1 assuming that only one genetic locus influenced TY within the original introgression. Differences in BY among the control and the subNILs seem to be due to the effect of the TY QTL at the base of the chromosome.

Only TA1223, TA1227 and the hybrid TA1224× TA209 displayed clearly rounded fruits, evaluated either as a visual score or as the ratio L/D (only homozygous subNILs). TA1220, TA1226, TA1228 and TA1229, which had the recombination event between TG375 and TG617, produced blocky fruits like the TA209 control. The FS QTL is located between the markers TG375 and TG617 and its mode of gene action is additive (d/a=0.2, Fig. 4).

With regard to external fruit traits, subNILs with introgression extending from TG161 to TG375 had more red color and less shoulder pigmentation. A SHO QTL maps clearly within the TA1218 introgression but we can not rule out than an ECOL QTL is also within TA1218 or is in the TA523 introgression. No significant effects were found for ICOL, or ER (data not shown).

The locations, additive effects and mode of gene action of the QTLs included in the TA523 introgression are summarized in Fig. 4. The mode of gene action of the majority of the detected QTLs is additive, or close to additive, except for the TY QTL which showed dominant gene action. QTLs detected in the subNIL population are in accord with those predicted by the performance of the original TA523 NIL.

Discussion

Number of loci controlling QTL effects. Pleiotropy versus linkage

Paterson et al. (1990) proposed substitution mapping as a method for fine-mapping QTLs. The application of this method to characterize QTLs in diverse species (Eshed and Zamir 1995; Grandillo et al. 1996; Tuinstra et al. 1998; Lyman and Mackey 1998) has demonstrated that substitution mapping is a powerful tool to distinguish between pleiotropy versus linkage as well as pseudo-overdominance versus true-overdominance, eliminating the linkage drag and setting the basis for the map-based cloning of QTLs (Tanksley et al. 1995; Alpert and Tanksley 1996).

The TA523 introgression region has been reported to improve brix and BY and reduce FW (Bernacchi et al. 1998a, b). In the previous experiments only backcross populations or heterozygous NILs were evaluated, whereas in the present study we have analyzed both homozygous and heterozygous introgressions, thus allowing an estimate of the mode of gene action. Furthermore, a FS QTL not reported in the previous experiments was detected.

By substitution-mapping it was possible to distinguish between linkage and pleiotropy for several loci involved in the quantitative traits affected by the 40-cM introgression from *L. hirsutum*. Loci affecting brix and TY map to opposite extremes of the introgressed segment; thus these traits are clearly controlled by different genes. Brix and yield are usually negatively correlated, hampering the breeding of high-brix cultivars without a reduction in yield. As a result, breeders use the combination brix*yield as an indicator of the agronomic value of a to-

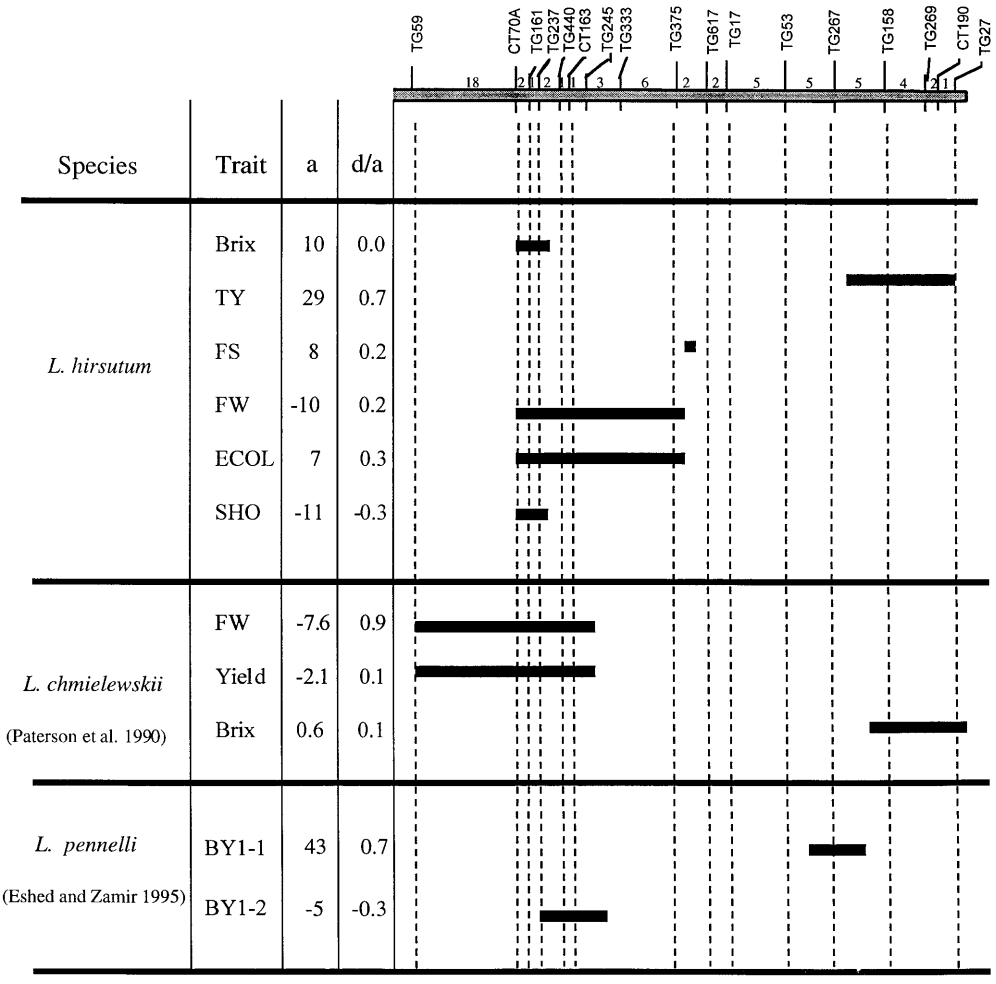


Fig. 4 Position, additive effect (*a*) expressed as % difference with the control and gene action (*d/a*) of the QTLs within the TA523 introgression. The position and genetic effects of the QTLs from *L. chmielewskii* and *L. pennellii* were calculated according to Paterson et al. (1990) and Eshed and Zamir (1995) respectively. The additive effects for *L. pennellii* QTLs are also expressed as a % difference with the control. The additive effects for *L. chmielewskii* QTLs are expressed as g for FW, Kg for Yield and Brix

mato variety. It is not clear whether this correlation is due to linkage or pleiotropy. In the current experiment, yield and brix were positively correlated in the TA523 NIL, due to separate loci located within the introgressed region. Substitution mapping revealed that the brix locus is located between CT70A and TG440, and interval less than 4-cM long, which, on average, should correspond to approximately 2.8 Mb and could be the target for map-based cloning.

The FS QTL is located in the center of the introgressed segment and is distinct from both the TY and brix loci. Effects on fruit shape have been observed previously in this region in advanced-backcross populations derived from crosses with *L. pimpinellifolium* (Tanksley et al. 1996), *L. peruvianum* (Fulton et al. 1997) and *L. chmielewskii* (Monforte et al., in preparation). The effect of this QTL is not as strong as the major fruit-shape QTLs *fs2.1* and *fs8.1* (Grandillo et al. 1996; Tankley et al. 1996). Interestingly, the direction of the allelic effect of the FS QTL in this region of chromosome 1 was different among species: *L. peruvianum* and *L. hirsutum* alleles induce rounded fruits, whereas *L. pimpinellifolium* and *L. chmielewskii* alleles produce elongated fruits. It would be interesting to test whether these effects are caused by the same genetic loci. If that was the case, the study of the alleles at this QTL would be an attractive system in which to analyze the physiological basis of tomato fruit-shape development.

The locus (loci) for ECOL, SHO and FW mapped to overlapping positions (between CT70A to TG617, Fig. 4) with brix, hence it was not possible determine if they are actually under different genetic control. External color and shoulder pigmentation are all part of fruit epidermal features, so it is reasonable that these traits could have the same genetic control. Similarly, brix and FW are usually negatively correlated, (i.e., larger fruits have higher water content within their cells, diminishing the soluble solid concentration); it is also possible that these two traits are under the pleiotropic effect of a single gene.

Paterson et al. (1990) using an accession of *L. chmielewskii*, and Eshed and Zamir (1995) using *L. pennellii*, also fine-mapped QTLs at the bottom of chromosome 1 by substitution mapping. According to Paterson et al. (1990) the brix QTL mapped to the end of the chromosome (between TG158 and TG27), unlike the findings of the current study. They also found that the FW and yield QTLs map between TG245 and TG295, whereas in the

current study only FW mapped to this region. As in the current study, Eshed and Zamir (1995) found an increase in BY associated with the end of the chromosome and, additionally, a decrease associated with the middle of the chromosome (Fig. 4). In that experiment it was not reported whether or not the increase of BY was due to an increase of brix or yield, or both. In the current report, the increase of BY associated with the base of the chromosome could be attributed solely to an increase in TY.

Even though it is difficult to compare the three experiments because they were performed in different years and locations, using different wild species and tomato cultivars as a genetic background, the locations of a yield or BY QTL at the end of the chromosome and a reduction of FW in the more interior regions are consistent in at least two experiments. Considering the results from the three experiments, we can conclude that the base of chromosome 1 contains multiple genetic loci involved in fruit and agronomic traits with different allelic effects.

Gene action and heterosis

The genetic basis of heterosis has been a subject of debate through this century (Schull 1908; East 1936; Sprague 1953; Crow 1993). The different hypothesis proposed to explain these phenomena are: overdominance at a single locus (Schull 1908; East 1936), pseudo-overdominance or dominance complementation, multiplicate gene action (Schenell and Cockerham 1992) and multilocus epistatic interactions (Allard 1996). In the current study, the heterosis found in the hybrid TA523×TA209 could not be attributed to a single overdominant locus. Homozygous subNILs carrying short introgressions at the bottom of the chromosome showed a high yield, and the TY QTL mode of gene action estimated in the subNILs population was partially dominant (d/a=0.7). These results agree with those reported by Eshed and Zamir (1995), except that they suggest that a BY QTL with a negative effect was located at the opposite end of the introgression, implying that the heterosis would be caused by the complemenentation of two loci in the heterozygous plants. We did not detect any negative effect on yield in that region. TA523 introgression showed a high yield only when heterozygous, but the gene action of the TY QTL had a strong additive component (a=+29%). Thus, two-loci dominance complementation can not explain the heterosis for TY observed in the hybrid TA523×TA209. This heterosis may be to be due to complex interactions among the TY locus and other genetic factors involved in the introgression, the effect of the TY locus being repressed when these factors are homozygous with repression being released in heterozygous plants.

Several models have been proposed to discriminate between over-dominance and dominance (see David 1997 and Deng 1998 for a detailed discussion). The best-documented example of heterosis attributable to over-dominance at a specific protein-encoding gene locus is the association of sickle-cell hemoglobin with malarial resistance. Very few other examples have been reported of a truly overdominant locus (Hall and Wills 1987; Hollick and Chandler 1998). On the contrary, when over-dominant effects at QTL loci have been studied more deeply, the single-locus overdominance hypothesis has been rejected. For example, Stuber et al. (1992) reported overdominant gene action in yield QTLs on chromosome 5 of maize, but re-analysis of the same data using a North Carolina Design III by Cockerham and Zeng (1996), and the use of NILs to fine map that QTL (Graham et al. 1998), demonstrated that the overdominant action reported in the previous experiment resulted from the expression of two dominant genes in repulsion-phase linkage. This agrees with Xiao et al. (1995) who suggested that dominance may be the genetic basis of heterosis in rice. Furthermore Yu et al. (1997) found that epistasis played a major role as the genetic basis of heterosis in a rice cross. From these studies, it may be concluded that true over-dominance is not the major cause of heterosis, especially for complex traits.

Hypotheses involving two or more loci may better explain the heterosis phenomenon, and especially in the case of models taking into account genetic interactions among the loci involved in the heterosis. In spite of the lack of success in the detection of epistatic interactions between QTLs in the early QTL mapping studies (Tanksley 1993), recent reports support the importance of digenic and higher-order interactions in the hereditance of quantitative traits, as well as the basis of heterosis (Allard 1996; Li et al. 1997; Monforte et al. 1997, 1999; Yu et al. 1997). Our data agrees with the hypothesis that heterosis is caused by interactions among different genetic loci, which may be closely linked, as we suggest in the current report.

Implications for breeding

The inbred line TA1218 carries a minimal introgression from *L. hirsutum* that increases brix when both heterozygous and homozygous and does not show any significant undesirable side effect. The small length of the introgression and the suppression of the recombination observed in the original NIL makes TA1218 a good choice to introduce a QTL for brix from *L. hirsutum* into the elite tomato germplasm with a minimal risk of introducing deleterious effects or loosing the gene(s) by recombination. In addition, several subNILs (e.g., TA1229) increase TY and could be used to improve the yield.

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