Theor Appl Genet (1996) 92:213–224

S. D. Tanksley · S. Grandillo · T. M. Fulton
D. Zamir · Y. Eshed · V. Petiard · J. Lopez
T. Beck-Bunn

Advanced backcross QTL analysis in a cross between an elite processing line of tomato and its wild relative *L. pimpinellifolium*

Received: 5 July 1995 / Accepted: 4 August 1995

Abstract Approximately 170 BC2 plants from a cross between an elite processing inbred (recurrent parent) and the wild species *Lycopersicon pimpinellifolium* LA1589 (donor parent) were analyzed with segregating molecular markers covering the entire tomato genome. Marker data were used to identify QTLs controlling a battery of horticultural traits measured on BC2F1 and BC3 families derived from the BC2 individuals. Despite its overall inferior appearance, *L. pimpinellifolium* was shown to possess QTL alleles capable of enhancing most traits important in processing tomato production. QTL-NIL lines, containing specific QTLs modifying fruit size and shape, were subsequently constructed and shown to display the transgressive phenotypes predicted from the original BC2 QTL analysis. The potential of exploiting unadapted and wild germplasm via advanced backcross QTL analysis for the enhancement of elite crop varieties is discussed.

Key words Molecular markers · Introgression · Plant breeding · Quantitative trait loci

Communicated by G. Wenzel

S. D. Tanksley (✉) · S. Grandillo · T. M. Fulton
Department of Plant Breeding and Biometry, 252 Emerson Hall, Cornell University, Ithaca, NY 14853, USA

D. Zamir · T. Eshed
Hebrew University; The Faculty of Agriculture; Department of Field Crops; Kennedy Lee Building; Rm 222; Box 12; Rehovot 76-100, Israel

V. Petiard
Francereco; 101 Avenue Gustave Eiffel; 37390 Notre Dame D'OE; Tours, France

J. Lopez
Hispareco S.A.; Centro de Investigacion y Desarrollo en Alimentacion; Apartado 435; E-06080; Badajoz; Spain

T. Beck-Bunn
Peto Seeds; 37437 State Highway 16; Woodland, CA 95695, USA

Introduction

In a companion paper we have proposed a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines (Tanksley and Nelson 1995). The method, termed advanced backcross QTL analysis (AB-QTL analysis), utilizes a BC2 or BC3 population from a cross between an elite line and an unadapted accession for the discovery and mapping of valuable donor QTL alleles. Simulations indicate that advanced backcross QTL analysis will be most efficient for: (1) uncovering non-epistatic QTLs from the donor line having a dominant, partially dominant or overdominant gene action, and (2) allowing selection of elite QTL-NILs for variety prouction. This method is also projected to help reduce linkage drag around targeted QTLs (Tanksley and Nelson 1995).

The present paper describes the results of applying AB-QTL analysis to a BC2 population derived from a cross between an elite processing tomato inbred and the wild species *Lycopersicon pimpinellifolium* LA1589. Approximately 170 BC2 individuals were characterized with molecular markers covering the entire tomato genome. The corresponding BC2F1/BC3 families were evaluated in the field at four locations worldwide and the data used to uncover QTLs controlling a variety of important horticultural traits including fruit size, fruit shape, soluble solids, fruit color, fruit firmness and yield. Results from QTL mapping in the advanced backcross population are compared with: (1) theoretical expectations for AB-QTL analysis based on computer simulations (Tanksley and Nelson 1995), and (2) results obtained from a QTL study of the same interspecific cross conducted in the BC1 generation (Grandillo and Tanksley 1995a).

Materials and methods

Population development

A single plant of the inbred wild species *L. pimpinellifolium* (LA1589)

(originating from Peru) (hereafter referred to as *PM*) was hybridized to a single plant of the open-pollinated processing inbred *L. esculentum* cv M82. Two F1 hybrid plants were backcrossed to the related processing inbred E6203 to produce the BC1 generation. Sixty-three BC1 individuals were grown in the greenhouse at Cornell University in February 1992. The RFLP marker TG279 was probed onto this population to select for individuals homozygous for *esculentum* alleles at the *sp* locus on chromosome 6. The *sp/sp* genotype confers a determinate growth habit, a requirement for the field evaluation of processing tomato varieties. TG279 is approximately 3 cM from the *sp* locus (Grandillo and Tanksley 1995b). A total of 32 plants were determined to be homozygous for TG279. Phenotypic selection for high fertility and larger fruit size was used to narrow the selection further to the best 27 individuals. Each selected BC1 individual was backcrossed again to E6203 and approximately ten BC2 seed from each of the selected BC1 plants were sown in the greenhouse in September 1992 to produce a BC2 population of 263 plants. The BC2 were backcrossed again to E6203 to produce the BC3 generation. In addition each BC2 plant was also crossed to the tester inbred S365 to produce BCF1 hybrid seed lots. Previous experience had shown that E6203 and S365 have good specific combining ability and produce a commercially acceptable hybrid processing tomato variety (D. Zamir, unpublished data). DNA was extracted from each BC2 plant for RFLP analysis. Phenotypic selection was exercised again on the BC2 plants for fertility and large fruit and BC3F1 and BC3 seed from the best 171 individuals was selected for field testing.

Field trials

BC2F1 hybrid plots

The 171 BC2F1 families, derived from the test crosses with S365, were evaluated in California, Spain and Israel in the summer of 1993. A subset of 142 BC3 families, derived from crosses of the same BC2 plants to E6203, were evaluated in Israel. Planting/harvesting dates and cultural conditions for each location are described in Table 1.

Trait evaluations

A total of 21 traits were evaluated for each BC2F1 and BC3 plot in one or more locations in the 1993 field trials. Details for each trait are given below.

Soluble-solids content

The soluble solids in tomato fruit are comprised mainly of sugars and, to a lesser degree, organic acids. The higher the soluble solids the greater the amount of product (paste, catsup, etc) that can be extracted from a fixed quantity of freshly harvested fruit. In these experiments, the soluble-solids content was measured in all locations with a refractometer (°Brix). In California and Israel, measurements were made on serum from a raw, de-aerated cold-break puree derived from a random sample of more than 40 randomly harvested fruit per plot. In Spain, measurements were made on serum from a pulped canned sample derived from approximately 5 kg of fruit per plot which had been heated in a microwave for 15 min, cooled to 30 °C and heated to 80 °C prior to canning.

Fruit weight

Average weight (grams) per fruit was determined from a random sample of at least 40 ripe fruit per plot in all locations.

Fruit color

Intense red pigmentation of fruit is a desirable trait in processing tomatoes. The degree of red fruit color was measured differently in each location. In California the level of redness was quantified with an Agtron (LA/B) on raw de-aerated puree. A lower value indicates more intense red color. In Spain color was evaluated on the same canned sample used for solids measurements using a Gardner Colorgard 2000/05 optical sensor (A/B) calibrated with Black Tile and Red Tile B.C.R. 801. For this measurement, a higher value indicates more intense color. In Israel the internal color of tomatoes was evaluated subjectively (scale of 1–5, 5 = more intense color) by viewing at least 40 fresh ripe fruit per plot which had been cut transversely.

Fruit pH

In all locations, fruit pH was measured on the same samples used for solids measurements (see above).

Yield

In California, yield potential was estimated visually (scale 1–5, 5 = greatest yield potential) on July 16. In Spain and Israel, fruit from the entire plots were harvested and weighed. Total yield in all cases refers to all fruit (red and green) whereas red yield refers to red fruit only. Brix*red yield is the product of the brix reading multiplied by the red yield and is an estimate of the amount of processed product (e.g., paste, catsup) that can be expected from a plot.

Fertility

Fertility of the plots was evaluated visually in California (July 16) and Spain (July 8) in 1993. A high fertility rating indicates that the majority of the plants in the plot had a heavy fruit set. A low fertility rating indicates that many of the plants had reduced fruit set, indicating subnormal fertility.

Puffiness

Puffiness is a measure of the amount of free air space observed in the locules of transversely cut fresh fruit. This trait was evaluated visually (scale of 1–5, 1 = very puffy, 5 = non-puffy) in Israel on at least 40 fresh fruit. A value of 1 indicated much puffiness (much free air space, undesirable) and a value of 5, little or no free air space (desirable).

Table 1 Field-trial information for BC2F1 plots (with S365 as tester) and BC3 plots (E6203 = recurrent parent). F = furrow irrigation, S = sprinkle irrigation, D = drip irrigation

Trial	Tester	Location	Soil type	Irrigation	Plants/ plot	Bed spacing, cm,	Plant spacing, cm	Transplant data	Harvest data
California	S365	Woodland, Calif.	Sandy loam	F/S	40	150	30	Apr 7	Jul 20
Spain	S365	Badajoz, Spain	Sandy	F	40	150	25	Apr 15	Aug 2–11
Israel.1	S365	Akko, Israel	Clay loam	D	40	196[a]	25	Mar 20	Jul 17
Israel.2	E6203	Akko, Israel	Clay loam	D	40	196[a]	25	Mar 20	Jul 17

[a] Double-rowed beds (35 cm between rows within a bed)

Shoulders

The degree of uneven color in ripe fruit cut transversely near the stem end was evaluated in Israel on at least 40 ripe fruit. A value of 1 indicated much mottled coloring (undesirable) and a value of 5 indicated little or no mottling (desirable).

Veins

The amount of white vascular veins visible in cut ripe fruit cut transversely was evaluated in Israel on at least 40 ripe fruit. A value of 1 indicated much venation visible (undersirable) and a value of 5 indicated little or no venation visible (desirable).

Viscosity

Viscosity was measured only in Spain on pulped canned sample derived from approximately 5 kg of fruit per plot which had been heated in a mcirowave for 15 min, cooled to 30 °C and heated to 80 °C prior to canning. A modified Bostwick was utilized with the viscosity measurement being expressed in cm of migration over a 30-s period at 25 °C. A low value indicated high viscosity (desirable).

Fruit shape

The shape of ripe fruit was evaluated visually on at least 40 fruit per plot in Israel. A rating of 1 indicated round fruit whereas a rating of 3 indicated more elongated, blocky shaped fruit (typical for processing tomatoes).

Percent stem release

In processing tomatoes it is desirable that the stems detach from the fruit at harvest to avoid puncture damage during shipping. The proportion of harvested ripe fruit releasing the stem was calculated from a sample of 100 ripe fruit per plot only in Israel.

Percent non-rotted fruit

The percentage of healthy ripe fruit (no rot) was calculated from a sample of 100 ripe fruit per plot in Israel.

Fruit firmness

Firm fruit are desirable to ensure maintenance of fruit quality during harvest and shipping. In Israel fruit firmness was evaluated subjectively by hand squeezing of at least ten fully ripe fruit per plot and given a numerical rating of 1 (soft) to 5 (very firm).

Plant growth

The amount of vegetative growth of each plot was evaluated visually in California (scale of 1–5, 1 = minimal growth, 5 = excessive growth) on June 11, 1993. Similar measurements were made in Israel at harvest.

Sun scald

Sun scald describes the phenomenon of fruit damage due to excessive exposure to the sun which causes bleaching of the fruit. Plants with poor vegetative cover tend to have higher levels of sun scald. In Spain sun scald was measured as the proportion of red fruit with sun scald damage at the time of harvest.

Cover

The degree to which fruit are protected from the sun by leaves is referred to as cover. Cover was evaluated visually (scale of 1–5, 1 = poor cover, 5 = good cover) in California on July 16, 1993 and in Spain (same scale as California) on July 8, 1993. Cover was evaluated on a scale of 1–3 in Israel at time of harvest.

Fruit ripening

The number of days from transplanting to first ripe fruit was recorded in California in 1993. This parameter was estimated in Spain by rating the level of fruit ripening (scale of 1–4, 1 = no ripe fruit, 4 = many ripe fruit) on July 8, 1993.

Marker analysis

BC2 plants were evaluated for 121 segregating molecular markers at approximately 11 cM intervals thorughout the genome as described by Grandillo and Tanksley (1995).

Data analysis

Trait correlations

Pearson correlation coefficients were calculated for each trait/location combination based on BC2F1/BC3 field data using the JMP V3.0 software package for Macintosh (SAS Institute 1994).

QTL analysis

One-way analysis of variance was used to determine the effects of each marker on each trait measured on the BC2F1 and BC3 plots using StatView 4.02 software (Abacus). Separate probabilities were calculated for each trait/location combination. The presence of a QTL near a marker locus was judged to be likely if one or more of the following criteria were met: (1) a significant effect was observed for a single marker/trait combination at a single location with $P < 0.001$; (2) significant effects were observed, in the same direction (i.e., the *PM* allele for the QTL in question was associated with either all positive or all negative effects in all locations), for a marker/trait combination at two or more locations with $P < 0.01$; (3) significant effects were observed, in the same direction, for a marker/trait combination at three or more locations with $P < 0.10$.

The additivity percentage (A%) of each significant QTL is herein as the additivity (AB − AA) divided by the midpoint [(AB + AA)/2] multiplied by 100 = 200(AB − AA)/(AB + AA), where AA = phenotypic mean for individuals homozygous for *esculentum* alleles at specified markers, AB = phenotypic mean for heterozygotes (*esculentum/pimpinellifolium*). However, since only one-half of the individuals in any given BC2F1 or BC3 testcross plot derived from a heterozygous BC2 individual would be heterozygous for a given QTL locus, the resulting value was multiplied by two. Hence, for these experiments, A% = 400(AB − AA)/(AB + AA).

Selection of QTL-NILs

BC3 plots were grown in Israel in 1993. BC3S1 seeds were bulk harvested from each BC3 plot and saved for the selection of QTL-NILs. Using BC2 marker information, BC3S1 progeny were identified which were most likely to yield homozygous QTL-NILs for selected regions of the genome using HyperGene™ software (Young and Tanksley 1989). Up to 100 plants were assayed for the presence of the target donor segment using RFLP markers encompassing that region. Plants determined to be homozygous for *PM* alleles in the target segment were then assayed with markers corresponding to unlinked regions of the genome known to be segregating for non-target *PM* alleles based on marker data for all lcoi which were

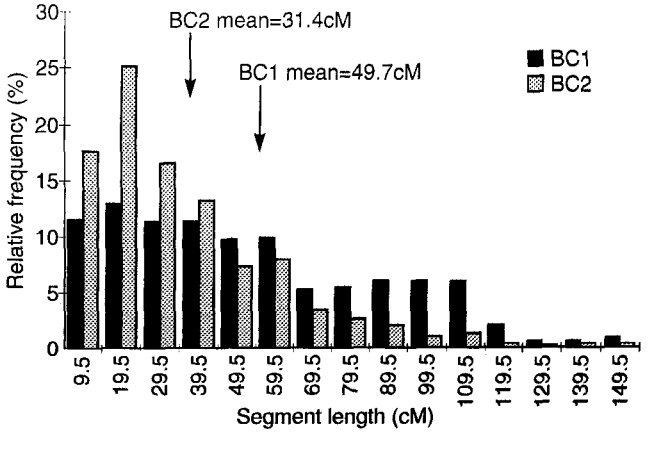
available for each corresponding BC2 plant. Plants containing the *PM* target region, but otherwise possessing *L. esculentum* alleles throughout their genome (based on marker data), were considered to be be nearly-isogenic for the targeted QTL. Each QTL-NIL was hybridized to the recurrent parent (E6203) and hybrid progeny were evaluated for the phenotype expected to be modified by the *PM* QTL allele.

Results and discussion

Average size of donor chromosomal segment in BC2: comparison with BC1

Shorter average donor segments should result in reduced linkage drag and potentially in greater resolution in the mapping of QTLs. Simulations have indicated that donor chromosome segments should average approximately 40 cM in the BC1 versus 27 in the BC1 versus 27 in the BC2 – a 34% reduction in size (Tanksley and Nelson 1995). To test the prediction of the simulations, the distribution of donor segment lengths was calculated for both the BC1 (Grandillo and Tanksley 1995b) and the BC2 generations derived from the *L. esculentum* × *L. pimpinellifolium* cross (Fig. 1). Chromosome 6 was not included in this calculation since selection against the Sp^+ allele in BC1 eliminated a large portion of this chromosome (Grandillo and Tanksley 1995). The average donor segment length observed was 31 cM in the BC2 versus 50 cM in the BC1. Both of these values are greater than predicted from the simulations; however, the percentage decrease from BC1 to BC2 was close to what was predicted – 37% versus 34%. The predicted average segment lengths were less than those observed, perhaps because the simulations were based on a set of theoretical chromosome 100 cM in length (Tanksley and Nelson 1995), whereas in the *L. esculentum* × *L. pimpinellifolium* cross the chromosomes range in length from 85 cM to 150 cM (Grandillo and Tanksley 1995b).

Fig. 1 Distribution of donor segment lengths as calculated for both the BC1 (Grandillo and Tanksley 1995) and the BC2 generations derived from the *L. esculentum* × *L. pimpinellifolium* cross



Correlations among traits

Figure 2 displays the correlation matrix among all traits measured on the BC2F1/BC3 families in all locations. It is impractical to discuss all the implications of these correlations; however, we highlight some of the significant points.

A negative correlation was found between soluble solids and fruit weight in all locations ($r = -0.18$ to $r = -0.36$), a result in agreement with many previous findings (Goldenberg and van der Pahleen 1966; Ibarbia and Lambeth 1971; Paterson et al. 1988, 1991). Soluble solids and yield were negatively correlated. This is also consistent with previous finding (Stevens 1976; Stevens and Rudich 1978). Soluble solids and growth were positively correlated in California and Israel ($r = 0.24$; $r = 0.40$ and $r = 0.39$). This positive relationship may be explained by the increased leaf area per fruit ratio which is consistent with earlier findings (Davis et al. 1958; Emery and Munger 1970; Fisher 1975; Hewitt and Stevens 1981). For processing tomato this may be an undesirable character because large leaf area is often associated with scattered fruit set (Hewitt and Stevens 1981). Negative correlations were found between fruit weight and fruit color in all locations ($r = 0.17$, $r = -0.19$, $r = -0.37$ and $r = -0.35$). Larger fruits resulted in correlated higher yield in all locations but in Spain ($r = 0.34$, $r = 0.26$ and $r = 0.19$). In Israel firmness was positively correlated with fruit shape (e.g., blocky shape associated with firmer fruit) in both trials ($r = 0.37$ and $r = 0.52$)

Detection of QTLs based on BC2F1/BC3 data

A total of 21 horticultural traits were evaluated on BC2F1/BC3 hybrid plots in California, Spain and Israel. Putative QTLs for each trait are listed in Table 2 and briefly described below.

Soluble-solids content

Twelve significant genomic regions were detected for soluble-solids content. The *L. pimpinellifolium* allele increased soluble solids in 11 of these cases. For the QTL on chromosome 4 (*ssc4.1*) the *PM* allele decreased soluble solids. None of the detected QTLs were significant in all locations. Six were significant in three locations, four in two locations and two in only one location.

ssc7.1 on chromosome 7 had the greatest overall effect, the *PM* allele significantly increasing the soluble-solids content in three of the four locations. The maximum phenotypic effect exerted by *ssc7.1* was in Israel (with S365 as the tester) where it accounted for 18% of the phenotypic variation. The region of chromosome 7 containing *ssc7.1* also showed a significant negative effect on yield, fertility, maturity and fruit ripening (see following sections). It seems likely that the soluble-solids

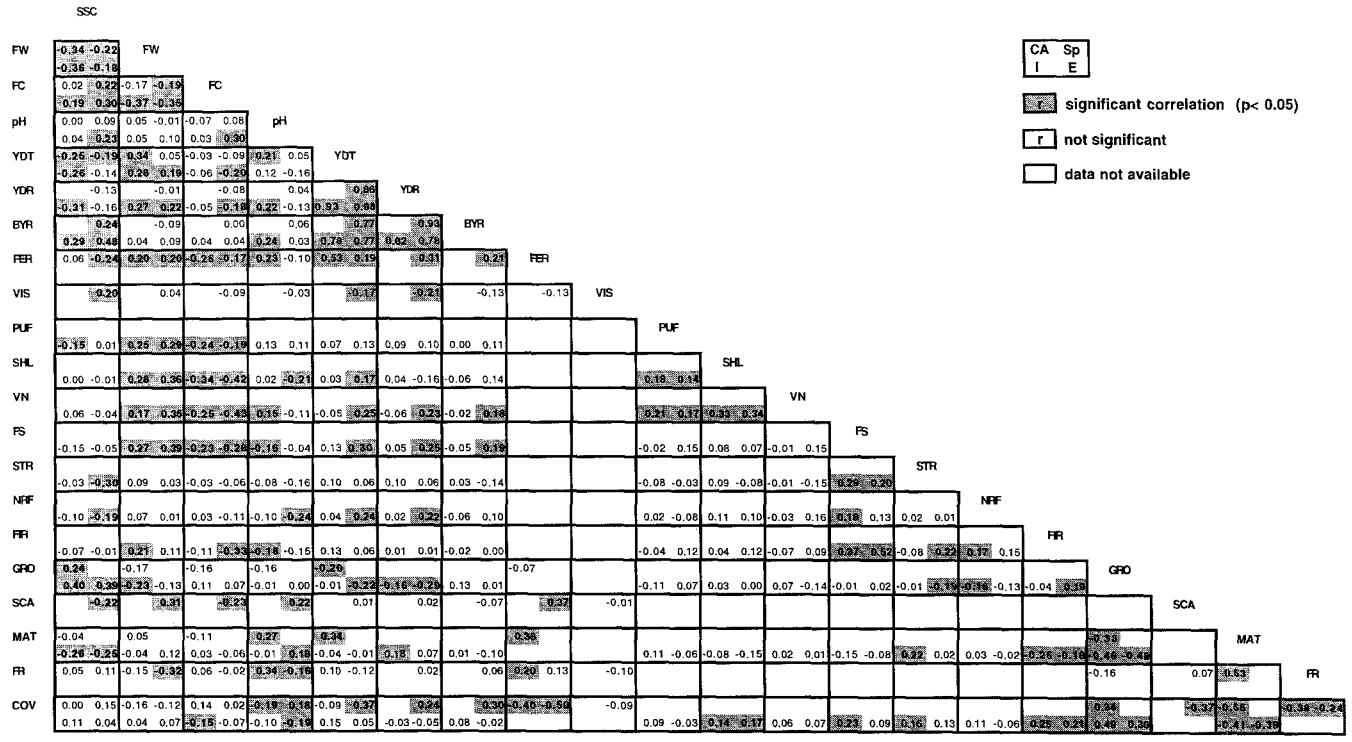


Fig. 2 Correlations between traits in the BC2F1 and BC3 populations. The sign of the correlation coefficients reflects the real relationship between the traits analyzed regardless of the scale used to measure the characters. *Gray cells* indicate correlations significant at $P < 0.05$. *CA* = $BC2F_1$ tested in Woodland, California; *Sp* = BC2F1 tested in Badajoz, Spain; *I* = $BC2F_1$ tested in Akko, Israel; *E* = BC3 tested in Akko, Israel. *SSC* = soluble-solids content, *FW* = fruit weight, *FC* = fruit color, *pH* = fruit pH, *YDT* = total yield,. *YDR* = red yield, *BYR* = Brix*red yield, *FER* = fertility, *VIS* = viscosity, *PUF* = puffiness, *SHL* = fruit shoulders, *VN* = fruit venation, *FS* = fruit shape, *STR* = % stem release, *NRF* = percent non-rotted fruit, *FIR* = fruit firmness, *GRO* = plant growth, *SCA* = sun scald, *MAT* = maturity, *FR* = fruit ripening, *COV* = cover

Table 2 List of putative QTLs detected from data collected from BC2F1 and BC3 field plots. C = California, S = Spain, I = Israel (plots with S365 as tester), E = Israel (BC3 plots). $*P < 0.1$, $**P < 0.01$, $***P < 0.001$, $****P < 0.0001$. Boxed area = location for which % var and A (%) were calculated. A (%) = 400(AB − AA)/(AB + AA) where AA = phenotypic mean for individuals homozygous for *esculentum* alleles at specified marker, AB = phenotypic mean for heterozygotes, % PV = percent phenotypic variance estimated from regression of marker against phenotypic. na = not available

Trait	QTL	Chrm.	marker	BC2F1			BC3	A(%)	%PV
				C	S	I	E		
Soluble solids	*ssc2.1*	2	TG337	**+	**+	***+	ns	8	6
	ssc3.1	3	TG388	***+	ns	ns	*+	7	9
	ssc3.2	3	TG129	*+	***+	ns	*+	7	7
	ssc4.1	4	CT157	ns	***−	ns	ns	−10	12
	ssc5.1	5	TG441	***+	ns	ns	ns	7	7
	ssc5.2	5	CT93	*+	*+	**+	ns	8	4
	ssc5.3	5	TG185	ns	*+	****+	ns	12	10
	ssc6.1	6	TG590	*+	*+	*+	ns	7	3
	ssc7.1	7	TG342	ns	**+	****+	*+	16	18
	ssc8.1	8	TG176	ns	****+	ns	*+	9	12
	ssc11.1	11	I29	ns	ns	**+	*+	8	6
	ssc12.1	12	CT211A	**+	ns	*+	*+	7	10
Fruit weight	*fw2.2*	2	TG492	****−	****−	****−	****−	−38	20
	fw3.1	3	TG388	**−	*−	*−	**−	−18	9
	fw3.2	3	CT85	***−	***−	*−	ns	−21	10
	fw4.1	4	TG574	*−	***−	ns	*−	−21	8
	fw5.1	5	CT167	*−	*−	*−	*−	−16	5
	fw7.1	7	TG20A	*−	****−	*−	****−	−22	9
	fw7.2	7	TG156	ns	*−	**−	***−	−29	10
	fw9.1	9	TG291	ns	*+	**+	*+	14	3
Fruit color	*fc2.1*	2	TG492	ns	ns	****+	**+	36	7
	fc3.1	3	TG525	***+	ns	*+	*−	4	10
	fc4.1	4	CT157	**−	***−	ns	ns	−2	7
	fc7.1	7	TG342	ns	****+	*+	ns	4	6
	fc8.1	8	CD40	***−	*−	*+	ns	−4	10

Table 2 (Continued)

Trait	QTL	Chrm.	marker	BC2F1			BC3	A(%)	%PV
				C	S	I	E		
Fruit pH	*ph1.1*	1	TG460	ns	**–	**–	ns	–2	4
	ph3.1	3	TG214	ns	ns	ns	****+	4	12
	ph5.1	5	CT172	****–	ns	ns	****–	–5	16
	ph7.1	7	CT52	***–	ns	ns	*–	–3	8
	ph12.1	12	CT211	ns	***+	ns	**+	3	9
Total yield	*ydt2.1*	2	TG151	ns	ns	***–	*–	–15	10
	ydt3.1	3	CT82	ns	***+	ns	ns	17	8
	ydt3.2	3	TG214	ns	***–	*–	ns	–15	7
	ydt7.1	7	TG20A	*–	ns	*–	*–	–6	3
	ydt7.2	7	TG342	****–	*–	****–	****–	–25	12
	ydt9.1	9	CT143	ns	**–	*–	ns	–12	4
Red yield	*ydr2.1*	2	TG151	na	ns	***–	*–	–14	6
	ydr7.1	7	TG342	na	**–	****–	****–	–34	18
Brix*red yield	*byr3.1*	3	CT82	na	***+	ns	ns	17	6
	byr3.2	3	TG214	na	**–	**–	ns	–16	5
	byr7.1	7	TG342	na	*–	**–	***–	–25	8
	byr9.1	9	CT143	na	*–	**–	ns	–11	4
Fertility	*fer5.1*	5	TG185	****–	**–	na	na	–41	14
	fer7.1	7	TG342	****–	****–	na	na	–67	30
Puffiness	*puf2.1*	2	CT176	na	na	***+	ns	32	6
	puf2.2	2	TG14	na	na	*+	**+	28	5
	puf8.1	8	TG349	na	na	**+	*+	20	3
	*puf*9.1	9	TG291	na	na	****–	*–	–32	7
	puf11.1	11	TG393	na	na	*+	**+	22	5
Viscosity	*vis9.1*	9	TG421	na	***+	na	na	13	8
Fruit shape	*fs1.1*	1	CT149	na	na	***+	ns	38	6
	fs1.2	1	TG245	na	na	***+	**+	34	7
	fs2.1	2	TG492	na	na	****–	****–	–53	17
	fs8.1	8	CD40	na	na	****–	****–	–80	37
% Stem release	*str1.1*	1	TG301	na	na	ns	***–	–31	9
	str2.1	2	TG337	na	na	***–	ns	–26	7
	str3.1	3	CD51	na	na	****–	****–	–32	13
	str10.1	10	OPV-20	na	na	***+	ns	35	7
Firmness	*fir2.1*	2	TG154	na	na	****–	****–	–45	11
	fir3.1	3	TG525	na	na	***+	*+	36	7
	fir4.1	4	CT157	na	na	**–	*–	–31	4
	fir8.1	8	CD40	na	na	***–	*–	–47	11
Growth	*gro1.1*	1	TG70	ns	na	**–	**–	–35	7
	gro5.1	5	TG351	*+	na	***+	**+	29	6
	gro7.1	7	TG342	*+	na	****+	****+	52	20
	gro8.1	8	TG302	ns	na	**–	*–	–24	4
	gro9.1	9	CT143	***–	na	***–	ns	–31	7
	gro11.1	11	12–9	*+	na	**+	*+	18	4
Sunscald	*sca7.2*	7	TG342	na	****+	na	na	158	9
	sca8.1	8	CD40	na	****–	na	na	–78	7
Maturity	*mat2.1*	2	TG492	**+	na	**+	ns	22	7
	mat3.1	3	TG388	ns	na	ns	****–	–41	12
	mat5.1	5	TG185	*–	na	****–	****–	–42	10
	mat7.1	7	TG342	****–	na	****–	****–	–70	15
	mat8.1	8	TG349	**+	na	*+	**+	22	7
	mat9.1	9	CT143	ns	na	****+	ns	34	15
	mat9.2	9	TG551	*+	na	***+	ns	37	8
Cover	*cov5.1*	5	CT172	na	**+	***+	ns	32	7
	cov6.1	6	CT216	na	**+	*+	ns	28	4
	cov7.1	7	TG342	na	****+	*+	*+	58	16
	cov8.1	8	TG176	na	ns	**–	*–	–24	4
	cov8.2	8	TG302	na	ns	****–	*–	–30	8
	cov9.1	9	CT143	na	***–	*–	ns	–33	6
Fruit ripening	*fr2.1*	2	TG492	****+	****+	na	na	54	20
	fr4.1	4	TG574	*+	****+	na	na	39	10
	fr7.1	7	TG342	*–	***–	na	na	–48	8
	fr8.1	8	CD40	*+	****+	na	na	36	10

increase observed for *ssc7.1* is a pleiotropic effect of semi-sterility which would decrease fruit set making a greater proportion of the sugars available to fewer fruit, hence an increase soluble-solids content. *ssc7.1* is therefore unlikely to be of economic benefit since, even though if increases soluble solids significantly, it greatly decreases yield and brix*yield. *ssc5.3* on chromosome 5 was also associated with lower overall fertility and later maturity.

Three soluble solids QTLs (*ssc2.1*, *ssc3.1*, *ssc5.1*) were also associated with lower overall fruit weight. *ssc3.2*, *ssc4.1*, *ssc5.2*, *ssc6.1*, *ssc8.1*, *ssc11.1* and *ssc12.1* appear to be independent of reduced fertility or fruit size.

Fruit weight

Fruit weight was affected by eight significant QTLs. For seven of these (88%) the *PM* allele reduced fruit size. This result is consistent with the phenotype of *L. pimpinellifolium* which has an average fruit size of approximately 2 g compared with 60–80 g for the *L. esculentum* parent. An effect opposite of that expected was observed for *fw9.1* on chromosome 9. At this locus the *PM* allele increased fruit size by approximately 14%.

The strongest effect was observed for *fw2.2* which accounted for 20% of the phenotypic variance and was associated with as much as a 38% reduction in fruit weight (Table 2). Other fruit-weight QTLs had significantly lesser effects, accounting for 3–10% of the phenotypic variance each. The *PM* allele for *fw2.2* was also associated with increased fruit color, higher soluble solids, early maturity and fruit ripening, decreased yield, rounder fruit shape and softer fruit in one or more locations. Some of these associations may be due to pleiotropic effects.

Fruit color

Five QTLs were detected for fruit-color intensity. *fc2.1* had a very strong effect on this trait with the *PM* allele increasing color in both Israel trials, but had no detectable effect in California or Spain. Alternatively, *fc4.1* had a significant effect in California and Spain, with the *PM* allele decreasing color, but was not significant in Israel. The difference in these results probably reflects the manner in which fruit color was evaluated in the three locations. In California and Spain, color was determined spectrophotometrically on post-processing puree. In Israel, however, fruit color was based on a visual rating of fresh fruit slices. It seems likely that these different methods may be measuring different parameters and hence different QTLs were detected. The *PM* allele for *fc2.1* significantly increased visual fruit color in Israel. The same region of chromosome 2 was also associated with a major decrease in fruit size (*fw2.2*). The increased color attributed to *fc2.1* may be due to a pleiotropic effect of decreasing fruit size, hence concentrating pigmentation.

Fruit pH

All of the five QTLs detected for pH were effective in only one or two locations. The effects were mixed and for some QTLs the *PM* allele increased pH (e.g., *ph3.1*) whereas in others it decreased pH (e.g., *ph5.1*). The strongest effect was observed for *ph5.1* which greatly decreased pH in California and in the Israel BC3 plots but had a non-significant effect in Spain and in Israel S365 plots.

Total yield

Six QTLs were detected for total yield. For five (83%) the *PM* allele caused a decrease in yield. However, for *ydt3.1* on chromosome 3, a significant increase in yield was observed in Spain. *ydt7.2* had the overall greatest and most-consistent efffect (across locations) on yield. In this case the *PM* allele caused a substantial decrease in yield. As mentioned earlier, this region of chromosome 7 was also associated with sterility which probably accounts for the yield reduction. For *ydt2.1* the *PM* allele also decreased yield and was associated with small fruit (see previous section).

Red yield

Two QTLs (*ydr2.1* and *ydr7.1*) were detected for red yield and correspond to the two total yield QTLs described earlier (*ydt2.1*, *ydt7.2*). In both instances the *PM* allele decreased red yield.

*Brix*red yield*

This parameter measures the potential amount of processed tomato paste, catsup, etc., that a line will produce. The QTLs detected for brix*red yield map near QTLs detected for soluble solids (e.g., *ssc3.1*) or QTLs for yield (e.g., *ydt3.1*, *ydt3.2*, *ydt7.2*, *ydt9.1*). Three of these QTLs (*byr3.2*, *byr7.1*, *byr9.1*) decreased brix* yield. *byr3.1* near the middle of chromosome 3 increased brix*yield in Spain by 17% ($P = 0.001$) but did not show a significant effect in either trial in Israel. Brix*red yield was not evaluated in California.

Fertility

Two QTLs were detected which significantly affected fertility–*fer5.1*, chromosome 5, and *fer7.1*, chromosome 7. In both instances the *PM* allele reduced fertility. As discussed earlier, the same regions of these two chromosomes were also found to be associated with QTLs for increased soluble solids, later maturity, and in the case of *fer7.1*, reduced yield. All are likely to be pleiotropic effects of partial sterility imparted by the *PM* allele.

Puffiness

The *PM* allele decreased fruit puffiness for all but one of the five QTLs detected. *puf 9.1*, which increased puffiness, maps to the same region of chromosome 9 as *fw9.1* which is the only QTL affecting fruit weight for which the *PM* allele increases fruit size. One possible explanation is that the increase in fruit size causes an associated increase in locular air, hence greater fruit puffiness.

Shoulders and veins

Evaluations of fruit shoulders and veins were made only in Israel. No significant QTLs were detected.

Viscosity

Viscosity was measured only in Spain and a single QTL on chromosome 9 (*vis9.1*) was detected. The *PM* allele for *vis9.1* increased the viscosity of the processed tomato puree by approximately 13% (Table 2).

Fruit shape

Fruit shape was evaluated only in Israel where four QTLs were identified. Three of the QTLs (*fs1.2*, *fs2.1*, *fs8.1*) were associated with significant effects in both Israeli trials. *fs1.1* was significant only in the Israel trial with S365 as the tester. For two of the QTLs (*fs2.1*, *fs8.1*) the *PM* allele caused the fruit to be more rounded (less elongated). This result is consistent with the phenotype of *L. pimpinellifolium* fruit which are rounder than the *L. esculentum* lines which have blocky, elongated fruit typical of processing tomatoes (Fig. 4). *fs1.1* and *fs1.2* had allelic effects opposite of that predicted by the *L. pimpinellifolium* phenotype. In both instances, the *PM* allele caused the fruit to be less round and more elongated (Table 2, and see Fig. 4).

Percent stem release

Stem release was also measured only in the Israeli trials. Four QTLs were detected and only one of these (*str3.1*) was significant in both Israeli trials. For three of the QTLs, the *PM* allele reduced the percentage of stem release. The QTL on chromosome 2 maps to the same position as *fw2.2* and a variety of QTLs affecting other traits, all of which may be associated with pleiotropic effects of a change in fruit size and shape (Table 2, Fig. 3).

Percent non-rotted fruit

The percentage of healthy ripe fruit with no rot symptoms was evaluated only in the Israel trials and no significant QTLs were detected.

Fig. 3 Linkage map of markers used for BC2F1/BC3 QTL analysis. Map based on the cross *L. esculentum* cv M82 × *L. pimpinellifolium* (LA1589) backcrossed to *L. esculentum* cv E6203 (Grandillo and Tanksley 1995). Positions of significant QTLs indicated on the left. *Underlined* QTLs may be in common with those discovered in the BC1 generation (Grandillo and Tanksley 1995)

Fruit firmness

Fruit firmness was measured only in Israel and four QTLs were detected. For three of these the *PM* allele caused the fruit to be softer. The *PM* allele for *fir3.1* on chromosome 3, however, significantly increased fruit firmness in both Israel trials. None of the fruit-firmness QTLs detected in these experiments mapped near major genes known to affect fruit firmness, such as *rin*, *nor* or *Pgal* (gene encoding polygalacturonidase) (Tanksley et al. 1992).

Plant growth

Six significant QTLs were detected which affected plant growth. The *PM* allele increased overall growth in three instances. Two of these QTLs, *gro5.1* and *gro7.1*, were also associated with reduced fruit set (e.g., lower fertility, see previous section) which may account for the increase in vegetative growth.

Sun scald

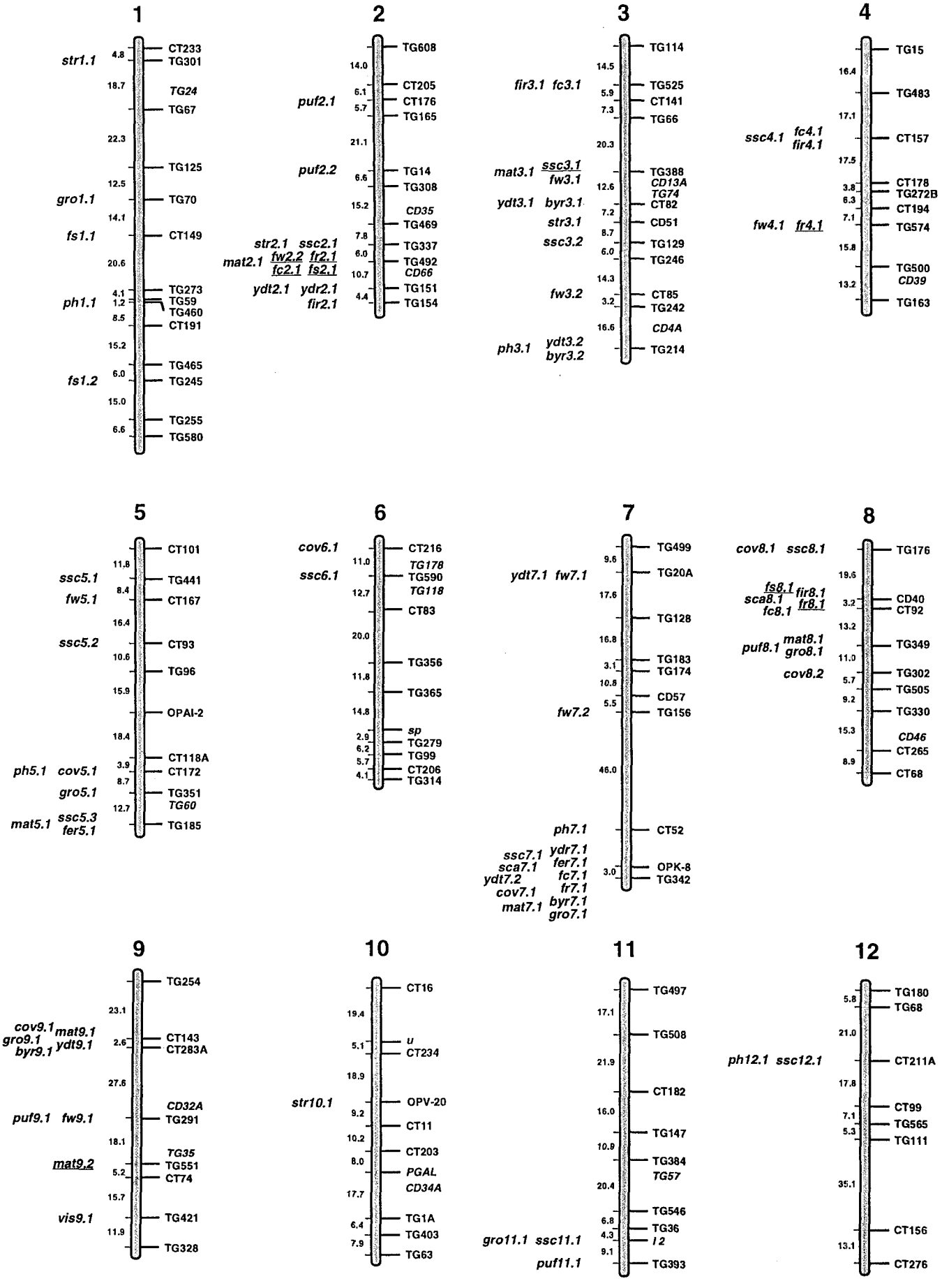
Sun scald was measured only in Spain and two significant QTLs were observed. For *sca7.1*, the presence of the *PM* allele decreased sun-scald damage. *sca7.1* maps to the same chromosomal position as *gro7.1*, *fer7.1* and *cov7.1* which decrease fruit set, increase vegetative growth and cover – all of which might lead to a reduction in sun-scald damage. For *sca8.1*, the *PM* allele was associated with increased sun-scald damage.

Maturity

Seven QTLs were found for maturity. In approximately half of these cases the *PM* allele resulted in earlier maturity. For *mat8.1* on chromosome 8 the *PM* allele advanced maturity in all locations where it was measured. For *mat5.1* and *mat7.1* the *PM* allele delayed maturity in all locations. These latter two QTLs were also associated with reduced fertility and increased vegetative growth (see previous sections).

Cover

Six QTLs were detected for cover. As might be expected, many of these were coincident with QTLs for growth.


1
str1.1
CT233
TG301
TG24
TG67
TG125
gro1.1
TG70
fs1.1
CT149
TG273
ph1.1
TG59
TG460
CT191
TG465
fs1.2
TG245
TG255
TG580
2
puf2.1
TG608
CT205
CT176
TG165
puf2.2
TG14
TG308
CD35
TG469
str2.1 ssc2.1
mat2.1 fw2.2 fr2.1
fc2.1 fs2.1
TG337
TG492
CD66
ydt2.1 ydr2.1
fir2.1
TG151
TG154
3
fir3.1 fc3.1
TG114
TG525
CT141
TG66
mat3.1 ssc3.1
fw3.1
TG388
CD13A
TG74
ydt3.1 byr3.1
CT82
str3.1
CD51
ssc3.2
TG129
TG246
fw3.2
CT85
TG242
CD4A
ph3.1 ydt3.2
byr3.2
TG214
4
TG15
TG483
ssc4.1 fc4.1
fir4.1
CT157
CT178
TG272B
CT194
fw4.1 fr4.1
TG574
TG500
CD39
TG163
5
CT101
ssc5.1
TG441
fw5.1
CT167
ssc5.2
CT93
TG96
OPAI-2
CT118A
ph5.1 cov5.1
CT172
gro5.1
TG351
TG60
mat5.1 ssc5.3
fer5.1
TG185
6
cov6.1
CT216
TG178
ssc6.1
TG590
TG118
CT83
TG356
TG365
sp
TG279
TG99
CT206
TG314
7
ydt7.1 fw7.1
TG499
TG20A
TG128
TG183
TG174
CD57
fw7.2
TG156
ph7.1
CT52
ssc7.1 ydr7.1
sca7.1 fer7.1
ydt7.2 fc7.1
fr7.1
cov7.1 byr7.1
mat7.1 gro7.1
OPK-8
TG342
8
cov8.1 ssc8.1
TG176
fs8.1 fir8.1
sca8.1 fr8.1
fc8.1
CD40
CT92
puf8.1 mat8.1
gro8.1
TG349
cov8.2
TG302
TG505
TG330
CD46
CT265
CT68
9
TG254
cov9.1 mat9.1
gro9.1 ydt9.1
byr9.1
CT143
CT283A
CD32A
puf9.1 fw9.1
TG291
TG35
mat9.2
TG551
CT74
vis9.1
TG421
TG328
10
CT16
u
CT234
str10.1
OPV-20
CT11
CT203
PGAL
CD34A
TG1A
TG403
TG63
11
TG497
TG508
CT182
TG147
TG384
TG57
TG546
TG36
gro11.1 ssc11.1
I2
puf11.1
TG393
12
TG180
TG68
ph12.1 ssc12.1
CT211A
CT99
TG565
TG111
CT156
CT276

For example, *cov5.1* and *cov7.1* mapped to the same chromosomal locations as highly significant growth QTLs. *cov5.1* and *cov7.1* were also associated with QTLs affecting fertility. QTLs for cover frequently, but not always, mapped near QTLs affecting soluble solids. For half of the QTLs detected, the *PM* allele increased cover.

Fruit ripening

The time to first ripe fruit was estimated only in California and Spain. Four QTLs were detected and in three of these instances the *PM* allele reduced the number of days required for the first fruit to ripen. The most significant effect was observed for *fr2.1* on chromosome 2. *fr2.1* maps coincidental with *fw2.2*, the most significant QTL detected for fruit weight. It seems likely that the *PM* allele decreases fruit size and hence reduces the number of days required for fruit maturity and ripening.

Comparison with QTLs detected in the BC1 population

A BC1 population, derived from the same cross *L. esculentum* × *L. pimpinellifolium* LA1589, has been studied independently to detect QTLs for several of the same traits described above including fruit weight, fruit color, soluble solids, fruit shape and days to first ripe fruit (Grandillo and Tanksley 1995a). A number of common QTLs were detected for these characters in the BC1 and BC2F1/BC3 generations.

fw2.2 on chromosome 2 was the most significant QTL affecting fruit weight in both studies. In both instances, the *PM* allele decreased fruit weight by approximately 30–40%. *fw2.2* has also been shown to be a major QTL controlling fruit weight in other *Lycopersicon* species (Alpert et al. 1995; Eshed and Zamir 1995). The same region of chromosome 2 also had a significant effect (most likely due to pleiotropy) on the days to first ripe fruit, fruit color and fruit shape in both studies.

A QTL affecting soluble solids near TG66/TG388 on chromosome 3 (*ssc3.1*) was also detected in both studies. The *PM* allele increases soluble solids in both instances, but in the BC2F1/BC3 population, this same region of chromosome 3 was found to be associated with a QTL reducing fruit weight (*fw3.1*). No fruit weight QTL was detected in the BC1 generation.

A QTL for which the *PM* allele reduced the time to first ripe fruit was also detected in both studies near the middle of chromosome 4 (*fr4.1*). The region of chromosome 4 was also found to be associated with small fruit in the BC2F1 population, but not in the BC1 population.

Chromosome 8 has two likely common QTLs between the BC2F1/BC3 and BC1 populations. The *PM* allele for *fs8.1* caused fruit to be rounder (versus blocky and elongated) in both studies. *fr8.1* (a QTL for which the *PM* allele decreases the time to first ripe fruit) was detected in the BC2 study. The same region of this chromosome was found to be associated with the length of time required for fruit to ripen. This region of chromosome 8 was associated with smaller fruit (*PM* allele) in the BC1 study, but not in the BC2F1/BC3 study.

mat9.2 is a putative QTL detected on chromosome 9 in the BC2F1 populations which decreases the number of days to maturity (*PM* allele). A QTL affecting the length of time required for fruit to ripen was observed in the BC1 population, mapping near the same point in chromosome 9 (near TG551). This same region of chromosome 9 was also associated with increased soluble solids (*PM* allele) in the BC1 but not in the BC2F1/BC3.

In general, more putative QTLs (for the same traits) were observed in the BC2F1/BC3 populations than in the BC1 population. For example, 12 putative QTLs affecting soluble-solids content were observed in the BC2F1/BC3 whereas only three QTLs were observed for the same trait in the BC1. Seven putative QTLs affecting maturity date were observed in the BC2F1/BC3 and only three in the BC1. Eight fruit-weight QTLs were observed in the BC2F1/BC3 and seven in the BC1. Also, for all of the fruit-weight QTLs observed in the BC1, the *PM* allele decreased fruit weight (Grandillo and Tanksley 1995). In the BC2F1/BC3, one putative fruit-weight QTL was observed for which the *PM* allele increased fruit weight (*fw9.1*). This QTL was not detected in the BC1 population. A nearly-isogenic line subsequently constructed for *fw9.1* supports the validity of this QTL (see next section).

Several factors may have contributed to the overall observation of a larger number of QTLs in the BC2F1/BC3 than in the BC1. First, the phenotypes of the BC2F1/BC3 were recorded over plots of at least 30 plants each in several different locations, whereas the phenotypes of the BC1 were measured on single plants in a single location. Plot measurements are likely to give a better overall assessment of the phenotypic effect of any given genotype and to increase the statistical ability to detect a QTL. Tending to counter this effect, however, is the fact that the BC2F1/BC3 plots were segregating (in an approximate 1:1 ratio) for individuals homozygous versus heterozygous for donor alleles – a situation which would tend to cause an underestimate of the phenotypic effects of heterozygous individuals.

A second difference in these studies concerns the statistical thresholds for declaring the effect of a QTL to be significant. Since the BC1 was evaluated in only a single location, probability thresholds were set at relatively stringent levels ($P < 0.001$, LOD > 2.4). For the BC2F1/BC3 the effects of the same QTL in different locations were taken into consideration before a QTL was declared to be significant (see Materials and methods). This method of evaluation is likely to be biased toward the detection of QTLs with minimal QTL*environment effects.

Construction of NILs for selected QTLs affecting fruit characteristics

Whether an apparent QTL is genuine can be assessed only through subsequent experimentation. One strategy is to construct nearly isogenic lines (NILs) for specific regions of the genome declared contain a QTL. Such lines would contain a segment of the *PM* genome thought to contain a QTL, but would otherwise have *L. esculentum* alleles throughout the rest of their genome. We will refer to such lines as QTL-NILs. QTL-NILs were selected from BC3S1 seed for three QTLs affecting the fruit characteristics described below.

fs8.1 (chromosome 8), which affects fruit shape, was detected in both the BC1 and BC2F1/BC3 generations. The QTL-NIL for this region of chromosome 8 clearly displayed the expected change in fruit shape in all of the locations where it was evaluated (Table 3, Fig. 4). *fs1.2* (chromosome 1) is another QTL affecting fruit shape, but in this instance, the *PM* allele was projected to cause the fruit to become more elognated than the *L. esculentum* recurrent parent. This is a phenotypic change opposite to that expected from the *L. pimpinellifolium* parent which has much rounder fruit than the *L. esculentum* parent (Fig. 4). The QTL-NIL for this region of chromosome 1 displayed the phenotype predicted from the BC2F2/BC3 data, producing fruit which were more elongated than the *L. esculentum* isogenic parent (Table 3, Fig. 4). The *PM* allele for *fw9.1* on chromosome 9 (detected only in the BC2F1/BC3 population) was predicted to cause an increase in fruit weight. Again, this change in phenotype is opposite to that expected from *L. pimpinellifolium* fruit which are approximately 2% the weight of the *L. esculentum* parent (Fig. 4). QTL-NILs for this region of chromosome 9 did possess fruit of greater weight (approximately 8% greater than the recurrent parent) confirming the prediction of the BC2F1/BC3 analysis (Table 3, Fig. 4).

The QTL-NILs described above confirm the validity of the corresponding QTLs deduced from BC2F1/BC3 data. Evaluation of additional QTL-NIL lines should reveal what proportion of the putative QTLs detected in the BC2F1/BC3 plots are genuine. QTL-NILs (and corresponding hybrids) for yield, soluble solids and other production chracteristics are currently being

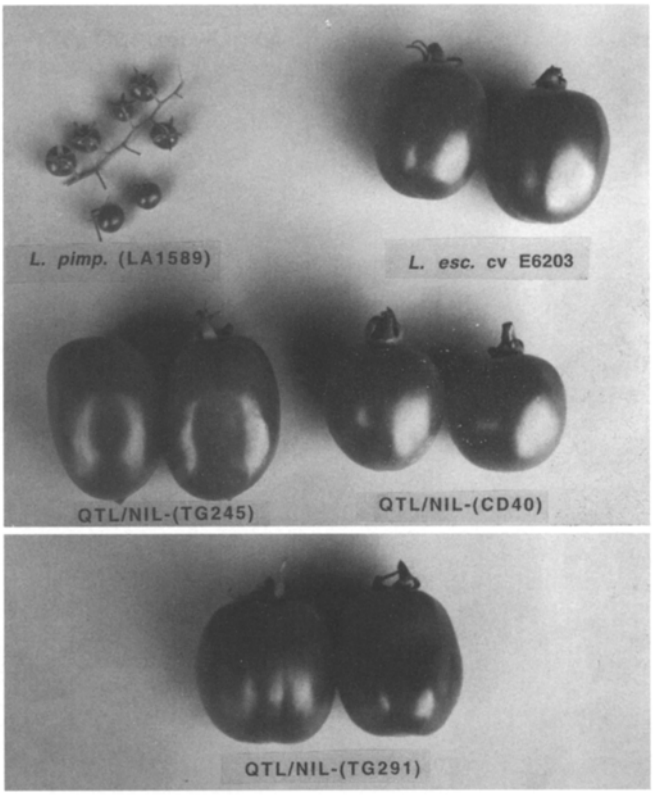


Fig. 4 Comparison of fruit characteristics for the donor (*L. pimpinellifolium* LA1589), recurrent parent (*L. esculentum* cv E6203), and derived QTL-NILs generated transferring single chromosomal regions from *L. pimpinellifolium* into an otherwise E6203 genetic background. Each QTL-NIL contains a single heterozygous chromosomal segment from *L. pimpinellifolium* in an otherwise E6203 background. QTL-NILs were generated via backcrossing and marker-aided selection (see text for details). QTL-NIL-(CD40) contains a small portion of chromosome 8 deduced to contain *fs8.1*, a QTL for fruit-shape modification. Note that QTL-NIL-(CD40) possesses round fruit like *L. pimpinellifolium*. QTL-NIL-(TG245) contains a small portion of the bottom chromosome 1 deduced to contain *fs1.2*, a QTL for fruit-shape modification. However, in contrast to QTL-NIL-(CD40), QTL-NIL-(TG245) possesses more elongate fruit – an effect predicted from the BC2F1/BC3 QTL analysis but opposite that which would have been expected from the *L. pimpinellifolium* phenotype. QTL-NIL-(TG291) contains a small portion of the chromosome 9 deduced to contain *fw9.1*, a QTL affecting fruit weight. Note that QTL-NIL-(TG291) has fruit larger than either the donor parent or recurrent parent–an effect predicted from the BC2F1/BC3 QTL analysis but opposite to that which would have been expected from the *L. pimpinellifolium* phenotype

Table 3 Performance of heterozygous QTL/NILs containing specific segments of the *L. pimpinellifollium* genome in an otherwise isogenic (*L. esculentum*) background. Statistics refer to fruit characteristic listed under "expected phenotype" and were derived from data collected from at least 30 pairs of NIL plants. e/e = homozygous for *esculentum* alleles at target QTL, e/p = heterozygous for *pimpinellifolium* allele at target QTL. Values for fruit shape are ratio of fruit length to fruit width. % control = percentage increase/decrease in target phenotype of QTL/NIL with respect to *L. esculentum* isoline control, *P* = probability derived from t-test comparing QTL/NILs and *L. esculentum* control

NIL(s)	Segment	Chrom.	Target QTL	Expected phenotype	NIL (e/p)	Control (e/e)	e/p – e/e	% Control	*P*
93T579.13,39	TG176-CD40	8	*fs8.1*	Round fruit	1.64	2.67	−1.03	−38%	<0.00001
93T577.29	TG245-TG580	1	*fs1.2*	Elongate fruit	2.85	2.54	0.31	12%	<0.005
93T582.4, 93T579.11,30	TG291	9	*fw9.1*	> Fruit mass	65.5 g	60.4 g	5.1 g	8%	<0.00001

evaluated in larger trials in several locations. The results of these trials will be the subject of a future report. However, that the QTL-NIL lines for fruit size and fruit shape, described above, performed as predicted suggests that a significant portion of the QTLs identified in the BC2F1/BC3 generations are geniune.

Conclusions

Advanced backcross QTL analysis has been proposed as a method for combining marker-based QTL detection with elite variety improvement (Tanksley and Nelson 1995). The method is specifically designed to: (1) selectively introduce valuable QTLs into elite varieties from unadapted germplasm and (2) minimize the time from QTL discovery to variety release. Simulations have been used to evaluate statistical properties of AB QTL analysis (Tanksley and Nelson 1995). The current study was executed to determine empirically whether QTLs could actually be detected using AB analysis and to compare the results with a more conventional BC1 QTL study utilizing the same breeding material (Grandillo and Tanksley 1995a). The results show that a number of the QTLs discovered in the BC1 population were also discovered in the advanced backcross populations (BC2F1/BC3). However, a number of additional QTLs were observed in the advanced backcross populations. Using marker information, it was possible to select QTL-NIL lines (from BC3S1 seeds) in a single generation (6 months) after discovery of the putative QTLs in the BC2F1/BC3 populations. Three NILs containing putative QTLs for fruit characteristics (shape and size) have been evaluated and performed as predicted. Two of these QTL-NILs exhibited phenotypes (fruit weight and fruit elongation) transgressing the recurrent parent lines, even though the phenotype of the donor line (*L. pimpinellifolium*) was deficient in these characters. Overall, these results indicate that advanced backcross QTL analysis can be used for quick discovery and transfer of QTLs from unadapted germplasm, leading to transgressive improvement of elite breeding lines. In this regard, the method may help open the door to more efficient utilization of unadapted germplasm and thereby to the expansion of the genetic base of crop plants.

Acknowledgements This work was supported in part by grants from the National Research Initiative Cooperative Grants Program, Plant Genome Program USDA (No. 58-1908-5-001) and by the Binational Agricultural Research and Development Fund (No. US-2427-94). Silvana Grandillo was supported in part by the N.Y. State's Centre for Advanced Technology (CAT) in Biotechnology (Biotechnology Program) and in part by the Italian CNR (National Research Council). We acknowledge the assistance of G. Gera from the Akko Experiment Station in conducting the field trials and H. van Oss and T. Bloch for laboratory analyses. Thanks to Dr. Clare Nelson for helpful comments and discussions

References

Alpert K, Grandillo S, Tanksley SD (1995) *fw2.2*: a major QTL controlling fruit weight is common to both red and green-fruited tomato species. Theor Appl Genet (in press)

Davies JN, Massey DN, Winsor GW (1958) The effect of defoliating tomato plants on fruit composition. Rpt Glasshouse Crops Res Inst 1957:53–66

Emery GC, Munger HM (1970) Effects of inherited differences in growth habit on fruit size and soluble solids in tomato. J Am Soc Hort Sci 95:410–412

Eshed Y, Zamir D (1995) An introgression line population of *Lycopersicon pennellii* in the cultivated tomato enables the identification and fine mapping of yield-associated QTLs. Genetics 141:1147–1162

Fisher KJ (1975) Effects of the amount and position of leaf tissue on the yield of single-truss tomatoes. Scien Hort 3:303–308

Goldenberg JB, von der Pahleen A (1966) Genetic and phenotypic correlation between wieght and dry matter content of tomato fruits and their heritabilities. Bol Genetico 2:1–15

Grandillo S, Tanksley SD (1995a) QTL analysis of horticultural traits differentiating the cultivated tomato from the closely related species *Lycopersicon pimpinellifolium*. Theor Appl Genet (in press)

Grandillo S, Tanksley SD (1995b) Genetic analysis of RFLPs, GATA microsatellites and RAPDs in a cross between *L. esculentum* and *L. pimpinellifolium*. Theor Appl Genet (in press)

Hewitt JD, Stevens MA (1981) Growth analysis of two tomato genotypes differing in total fruit solids content. J Am Soc Hort Sci 106:723–727

Ibarbia EA, Lambeth VN (1971) Tomato fruit size and quality interrelationships. J Am Soc Hort Sci 96:199–210

Paterson AH, Lander ES, Hewitt JD, Peterson S, Lincoln SE, Tanksley SD (1988) Resolution of quantitative traits into Mendelian factors by using a complete linkage map of restriction fragment length polymorphisms. Nature 335:721–726

Paterson AH, Damon S, Hewitt JD, Zamir D, Rabinovitch HD, Lincoln SE, Lander ES, Tanksley SD (1991) Mendelian factors underlying quantitative traits in tomato: comparison across species, generations, and environments. Genetics 127:181–197

SAS Institute, Inc (1994) JMP users guide: version 3.0 of JMP. SAS Institute, Carry, N.C.

Stevens MA (1976) Inheritance of viscosity potential in tomato. J Am Soc Hort Sci 101:152–155

Stevens MA, Rudich J (1978) Genetic potential for overcoming physiological limitations on adaptability, yield, and quality in the tomato. Hort Science 13:673–678

Tanksley SD, Nelson JC (1995) Advanced backcross QTL analysis: a method for the simultaneous discovery and transfer of valuable QTLs from unadapted germplasm into elite breeding lines. Theor Appl Genet (in press)

Tanksley SD, Ganal MW, Prince JP, de Vicente MC, Bonierbale MW, Broun P, Fulton TM, Giovannoni JJ, Grandillo S, Martin GB, Messeguer R, Miller JC, Miller L, Paterson AH, Pineda O, Röder MS, Wing RA, Wu W, Young ND (1992) High-density molecular linkage maps of the tomato and potato genomes. Genetics 132:1141–1160

Young ND, Tanksley SD (1989) Restriction fragment length polymorphism maps and the concept of graphical genotypes. Theor Appl Genet 77:95–101