Definition of Nonvolatile Markers for Flavor of Tomato (*Lycopersicon esculentum* Mill.) as Tools in Selection and Breeding

Peter Bucheli,^{*,†} Elisabeth Voirol,[‡] Rosa de la Torre,[§] Joaquin López,[§] André Rytz,[‡] Steven D. Tanksley,^{||} and Vincent Pétiard[†]

Nestlé Research Center, 101 Avenue Gustave Eiffel, F-37390 Notre Dame d'Oé, France; Nestlé Research Center, P.O. Box 44, CH-1000 Lausanne 26, Switzerland; Nestlé R&D Center Badajoz, E-06080 Badajoz, Spain; and Department of Plant Breeding, Cornell University, Ithaca, New York 14853

A methodology for flavor and composition assessment of processed tomato juice samples was developed using a wide range of commercial processing tomato varieties (*Lycopersicon esculentum*) grown in Spain and the United States. Fruitiness intensity was found by a trained panel to best describe overall tomato flavor. For two consecutive years, fruitiness intensity was significantly dependent on growing location and variety, and it was consistently linked to increased levels of glucose and reducing sugars and decreased glutamic acid content. Using the same procedure on a population of 176 breeding lines derived from the wild species of *Lycopersicon pimpinellifolium*, it was shown that tomato fruitiness intensity was significantly correlated to reducing sugars/glutamic acid ratio and glucose and glutamic acid contents. The definition of markers for tomato flavor of processed juice can provide the tomato breeder and processor with reliable analytical tools that can be applied in a straightforward way for the identification of raw materials that can be processed into juice with predictably high or low fruitiness.

Keywords: Tomato; L. pimpinellifolium; flavor; sugars; organic acids; glutamic acid

INTRODUCTION

Sugars and organic acids are major components of tomato fruit and account for \sim 60% of dry matter (Davies and Hobson, 1981). They not only contribute to soluble solids (°Brix), one of the key parameters for the tomato processor but are also essential factors in overall flavor intensity (Stevens et al., 1977; Jones and Scott, 1983). Both the absolute concentrations of sugars and organic acids and the balanced ratio between them are important factors in consumer acceptance. Processed tomato juice samples with a soluble solids/total acidity ratio of less than 10:1 or greater than 18:1 were found to be unacceptable for flavor (Gould, 1978). Increasing total sugar and organic acid levels of fresh tomato improved flavor acceptability (Malundo et al., 1995; Petersen et al., 1998), and a balanced sugar/organic acid ratio was preferred by a panel examining the flavor characteristics of cherry tomato (Hobson and Bedford, 1989). A study in The Netherlands on a wide range of fresh market tomatoes judged cherry tomatoes as having the best flavor (Jansen, 1994).

Flavor is also a function of aroma components. However, little is known about the \sim 400 volatile compounds identified in tomato (Pétro-Turza, 1986). The effect of genetic variation and growing conditions of tomato on aroma compounds is not understood. Reasons for this lack of information are the complexity of volatile analysis (Buttery et al., 1987), the difficulty in developing a consistent methodology for sensory evaluation, and the challenge to link these analytical tools to welldefined raw materials (Brauss et al., 1998). This is well illustrated by the work of Baldwin et al. (1991), who found significant differences in nine volatile components among six fresh market tomato varieties but could not assess whether these differences were effectively linked to perceived flavor.

Poor flavor in tomato fruit is a serious consumer concern (Hobson, 1988). Tomato flavor has declined as variety selection and tomato production has emphasized yield, fruit size, firmness, lack of defects, disease resistance, and processing performance (°Brix, consistency) and not the sensory aspects of fruit quality (Stevens and Rick, 1986). In addition, sensory parameters that could assist the breeders in an efficient selection for flavor have not been characterized. The definition and use of markers that correlate to tomato flavor could improve this situation and provide the breeder and processor with analytical tools for flavor enhancement of processed tomato raw materials (Bucheli et al., 1998).

This study describes a methodology to assess rheological, compositional, and sensory parameters of a large number of tomato raw materials. Juice samples were produced according to a standardized microwave heat treatment and sterilized for convenience and storability of the product. The samples were scored for fruitiness intensity, the attribute that characterized best overall flavor in sensory profiling by a trained panel. Using this approach, we demonstrate that it is possible to specify biochemical markers for tomato flavor and to use them to identify varieties and breeding materials that can be used to make juice with enhanced flavor.

MATERIALS AND METHODS

Commercial Tomato Materials. In 1991, 24 commercial processing tomato varieties were grown in Spain (Nestlé R&D

[†] Nestlé Research Center, Tours.

[‡] Nestlé Research Center, Lausanne.

[§] Nestlé R&D Center, Badajoz.

[&]quot; Cornell University.

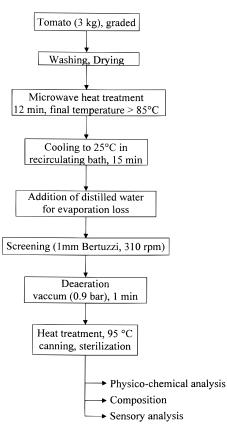


Figure 1. Description of tomato juice sample preparation.

Center Badajoz) and in the United States (Nestlé Food Co., Woodland, CA). Ten of the varieties were grown at both locations. In 1992, 18 varieties and types were used, and the genetic variability was considerably widened by including two cherry-type varieties and tomatillo (*Physalis philadelphica*). Nine of these varieties and tomatillo were grown at both locations. For both years, the trials consisted of processing varieties most commonly used by the industry in Spain and the United States.

Genetic Origin of *Lycopersicon pimpinellifolium* Breeding Materials. A single plant of the inbred wild species *L. pimpinellifolium* (originating from Peru) was hybridized to a single plant of the open pollinated processing inbred *Lycopersicon esculentum* variety M82 (Tanksley et al., 1996). Two F₁ hybrid plants were backcrossed to the related processing inbred E6203 to produce the BC₁ generation. A total of 27 BC₁ individuals selected for determinate growth, high fertility, and larger fruit size were backcrossed again to E6203, and ~10 BC₂ seeds from each of the selected BC₁ plants were used to produce a BC₂ population of 263 plants. Each BC₂ plant was crossed to the tester inbred S365 to produce BC₂F₁ hybrids that were grown in 1993 at the Nestlé R&D Center Badajoz (Spain).

Tomato Juice Sample Preparation. Only ripe tomatoes, from plants carrying 80–90% of mature fruits, were processed according to an adapted version of the method described by Wolcott and Merson (1990). The procedure recently presented by De la Torre et al. (1998) can be described as follows (Figure 1). Graded tomatoes (~3 kg) were washed and dried, cut in halves (only half kept), and then heat treated in a microwave oven (Moulinex 1100 W, 27 L; model 34 MGS 850/P00) for 12 min with a final temperature of >85 °C. Water loss due to evaporation was compensated with distilled water after rapid cooling to 25 °C. Tomatoes were then macerated, pulped (Bertuzzi, 1 mm screen, 310 rpm), and deaerated under vacuum (0.9 bar) for 1 min. The finished juice was sterilized in 400 g cans for 40 min at 95 °C and used as such for physical, chemical, and sensory analyses.

Sensory Analysis. The assessment of tomato juice was carried out at the Nestlé Research Center in Lausanne, Switzerland. A trained panel of 13-17 tasters used the following two sensory tests: (1) profiling on a 9-point scale on aroma (intensity, freshness, and fruity) and flavor attributes (sweetness, acid, bitter, salty, fruity taste, ripe, tomato concentrate, astringent, pungent, and off-flavor) that had been generated by the panelists in preliminary tests; (2) scoring tomato fruitiness intensity on a nonstructured 20-point scale (1 = not fruity, 20 = very fruity). The juices were presented to the tasters at random in small bowls under red light in series of 10-14 samples in two separate services within a session. The reference sample (variety N1401, a typical U.S. sample) was presented at each session. Three sessions were organized within a week for completion of the whole series of varieties. Each juice sample was tasted by the panel in triplicate for the 1991 and 1992 series and once for the 1993 samples (i.e., the 176 breeding lines from a L. pimpinellifolium derived population).

Physicochemical and Biochemical Analysis. Total soluble solids (°Brix) were estimated by measuring the refractive index with a Bellingham and Stanley RFM-81 refractometer. Bostwick flow was determined in a consistometer of 25 cm length. Enzymatic test kits from Boehringer Mannheim, Germany, were used according to the instructions of the manufacturer for the determination of glucose (GOD-Perid method, no. 124010), malic acid (no. 139068), citric acid (no. 139076), and glutamic acid (no. 139092). Reducing sugars were determined with the *p*-hydroxybenzoic acid hydrazide (PAH-BAH) assay (Lever, 1972). Concentrations of sugars and organic acids are expressed as percent (grams/100 g of juice).

Data Analysis. Sample means are compared using different analysis of variance models [Neter et al., 1990 (pp 818–860); Hintze, 1996]. When samples were compared on sensory data, a three-factor ANOVA was applied (product, taster, repetition), followed by a multiple-comparison test using Fisher's least significant difference (LSD). When variety and location effects on sensory and biochemical data were determined, a two-factor ANOVA was applied. Linear relationships between sample characterizations (sensory and biochemical) are summarized by simple correlation or correlation matrixes. Descriptive, more than predictive, models relating biochemical markers to sensory descriptors are built using an all-possible-regressions selection procedure. Mallows' Cp criterion was used to determine the best model (Neter et al., 1990; Hintze, 1996).

RESULTS AND DISCUSSION

Sensory Profiling of Commercial Varieties. The organoleptic tools for tomato flavor evaluation were developed on 16 commercial varieties grown in the United States and Spain in 1991. For most of the attributes, tasters scored the samples in a very consistent way over the three repetitions. The varieties grown in Spain tended to be sweeter, riper, and fruitier than the ones grown in the United States, which were more acid and pungent, except for Blazer, Pik Red, and the two repetitions of reference N1401, which were not very distinct. There were positive correlations between fruitiness and the attributes fresh (r = 0.74), sweet (r = 0.83), and ripe (r = 0.70) and negative correlations between fruitiness and the attributes bitter (r = -0.75), astringent (r = -0.52), and off-flavor (r = -0.84) (Table 1).

Definition of Fruitiness as a Key Attribute for Tomato Flavor. Because the attribute fruity was correlated with the majority of the key attributes of tomato flavor (Table 1), a scoring of the intensity of the fruity note was performed on the same samples that were used for sensory profiling and compared by correlation to the sensory profiling attributes fruity aroma (r = 0.61), sweet (r = 0.78), fruity taste (r = 0.80), and ripe (r = 0.84). As a result of these high correlation coefficients, the scoring of fruitiness intensity was defined as a tool for tomato flavor evaluation of pro-

 Table 1. Correlations^a between Sensory Attributes of 16 Commercial Processing Tomato Varieties Grown in 1991 in

 Spain and the United States

	aroma intensity	fresh	sweet	acid	bitter	salty	fruitiness	ripe	tomato concn	astringent	pungent	off-flavor
aroma intensity	_											
fresh		_										
sweet			_									
acid			-0.63	_								
bitter		-0.57	-0.74	-0.55	-							
salty	0.59		-0.71	0.84	0.67	_						
fruitiness		0.74	0.83		-0.75		_					
ripe		0.52	0.70				0.70	_				
tomato concn	0.56				0.48			0.51	-			
astringent	0.52		-0.72	0.81	0.84	0.81	-0.52		0.53	-		
pungent	0.52			0.91	0.49	0.69				0.80	_	
off-flavor		-0.85	-0.61		0.84		-0.84	-0.49		0.55		-

^{*a*} Only significant correlations at p < 0.05 are shown.

 Table 2. Fruitiness Intensity and Sugar and Organic Acid Data of Tomato Juices Produced from Commercial Tomato

 Varieties Grown in 1991 in Spain and the United States

variety	location	fruitiness intensity	multiple comparisons ^a	glucose, %g/g	reducing sugars, %g/g	citric acid, %g/g	malic acid, %g/g	glutamic acid, %g/g
Orion	Spain	12.43	k	1.51	3.26	0.19	0.03	0.21
Jet Star	U.S.	11.89	jk	1.41	3.34	0.23	0.06	0.29
813	Spain	11.57	ijk	1.78	3.32	0.23	0.03	0.20
reference 1	U.S.	11.41	ijk	1.42	3.03	0.31	0.05	0.25
7090	U.S.	11.18	hijk	1.33	3.07	0.23	0.03	0.25
Gemini	Spain	11.00	ghijk	1.58	2.77	0.18	0.02	0.20
Mesa	Spain	10.98	ghijk	1.40	3.05	0.23	0.03	0.15
5715	Ú.S.	10.64	fghijk	1.29	2.65	0.25	0.03	0.25
CSP-9	Spain	10.63	fghijk	1.30	2.77	0.21	0.02	0.21
reference 2	Ú.S.	10.61	fghijk	1.42	3.03	0.31	0.05	0.25
Sausalito	Spain	10.11	efghij	1.29	2.67	0.16	0.02	0.22
Vega	Spain	9.78	defghij	1.61	3.18	0.17	0.02	0.20
Blazer	Spain	9.71	defghij	1.33	2.71	0.31	0.03	0.20
La Rossa	Ú.S.	9.69	defghij	1.42	3.04	0.27	0.04	0.25
Pik Red	Spain	9.64	defghij	1.24	2.48	0.27	0.03	0.17
Jet Star	Spain	9.62	defghi	1.59	3.33	0.24	0.08	0.18
reference 3	Ú.S.	9.61	defghi	1.42	3.03	0.31	0.05	0.25
785	U.S.	9.46	defghi	1.18	2.70	0.25	0.07	0.24
Tierra	Spain	9.39	defghi	1.35	2.64	0.20	0.03	0.19
1643	Ú.S.	9.36	defghi	1.17	2.71	0.20	0.03	0.22
Blazer	U.S.	9.13	cdefgh	1.21	2.68	0.28	0.03	0.24
3075	Spain	9.11	cdefgh	1.33	2.87	0.20	0.03	0.20
N 1200	Ú.S.	9.06	bcdefgh	1.33	2.99	0.27	0.02	0.31
Viva	U.S.	9.06	bcdefgh	1.07	2.43	0.26	0.05	0.27
Viva	Spain	8.84	abcdefg	1.29	2.52	0.29	0.04	0.18
Tierra	Ú.S.	8.72	abcdef	1.03	2.15	0.20	0.06	0.22
CSP-7	Spain	8.51	abcdef	1.17	2.54	0.19	0.02	0.22
Vega	Ú.S.	8.37	abcdef	0.90	1.95	0.20	0.04	0.23
Pik Red	U.S.	8.26	abcde	1.15	2.47	0.24	0.06	0.20
Mesa	U.S.	8.22	abcde	0.84	2.03	0.24	0.04	0.20
3075	U.S.	8.21	abcde	0.93	2.03	0.24	0.04	0.19
P 512	U.S.	8.03	abcde	0.93	2.18	0.20	0.04	0.17
CSP-8	Spain	7.57	abcd	1.06	2.10	0.19	0.03	0.21
H 100	Ú.S.	6.94	abc	0.84	1.59	0.14	0.06	0.17
Gemini	U.S.	6.84	ab	1.01	2.30	0.17	0.04	0.20
Orion	U.S.	6.67	а	0.79	1.81	0.19	0.06	0.26

^{*a*} For fruitiness intensity, the same letter indicates that they are not significantly different (p < 0.05) according to Fisher's least significant difference (LSD = 2.26).

cessed juice and tested for three consecutive years on a wide range of commercial processing varieties and new breeding materials.

Effect of Variety and Location on Tomato Fruitiness Intensity. In 1991, a total of 36 tomato samples were evaluated for fruitiness intensity according to the described methodology. The mean fruitiness scores ranged from 6.7 to 12.4, with an overall mean of 9.4 (Table 2). According to Fisher's LSD, the tomato juices were significantly discriminated whenever their fruitiness intensity mean scores differed by at least 2.3. The observed range being 2.5 times greater than the LSD indicated the presence of a wide range of perceived fruitiness intensity among commercial processing varieties. For the 10 varieties grown in the United States and Spain, fruitiness intensity was variety (p < 0.01) and location dependent (p < 0.0001), with juices from Spain being higher in fruitiness (mean = 10.4 versus 8.3 for U.S. samples).

In 1992, the genetic variability of the materials assessed was widened by including nonprocessing tomato materials (cherry and tomatillo). A total of 28 tomato samples were scored by the same panel for fruitiness intensity, again in a very consistent way over the three repetitions. The mean fruitiness scores ranged from 4.0 to 12.2 (Table 3). According to Fisher's LSD,

Table 3. Fruitiness Intensity and Sugar and Organic Acid Data of Tomato Juices Produced from Commercial Tomato
Varieties, Cherry Tomato, and Tomatillo Grown in 1992 in Spain and the United States

variety	location	fruitiness intensity	multiple comparisons ^a	glucose, %g/g	reducing sugars, %g/g	citric acid, %g/g	malic acid, %g/g	glutamic acid, %g/g
CSP-13	Spain	12.24	k	1.50	3.30	0.31	0.09	0.14
CSP-11	Spain	11.00	ik	1.91	4.19	0.28	0.05	0.16
CSP-12	Spain	10.48	jk ij	1.63	3.70	0.42	0.04	0.08
Blazer	Spain	10.10	hij	1.63	3.46	0.40	0.03	0.10
Cherry K	Spain	10.08	hij	2.82	6.13	0.42	0.03	0.23
Orion	Spain	9.59	hij	1.27	2.80	0.30	0.03	0.13
Nema 1401	Spain	9.58	hij	1.43	3.15	0.30	0.03	0.13
Cherry	Spain	9.51	hij	2.17	4.90	0.43	0.05	0.21
CSP-14	Spain	9.24	ghi	1.49	3.33	0.34	0.03	0.12
CSP-15	Spain	9.21	ghi	1.14	2.69	0.33	0.03	0.12
Diablo	Spain	8.73	fgh	1.15	2.53	0.31	0.03	0.14
Exp854	Spain	8.54	fgh	1.14	2.58	0.24	0.03	0.10
CSP-13	Ú.S.	7.55	efg	1.29	2.99	0.31	0.07	0.23
CSP-10	U.S.	7.26	ef	1.51	3.54	0.66	0.03	0.28
CSP-11	U.S.	7.07	ef	0.88	2.35	0.38	0.07	0.25
CSP-8	Spain	6.47	de	1.12	2.65	0.28	0.03	0.11
CSP-12	U.S.	6.29	de	1.13	2.88	0.44	0.05	0.19
Blazer	U.S.	6.23	de	1.04	2.47	0.29	0.03	0.20
CSP-14	U.S.	6.10	cde	0.88	2.33	0.32	0.06	0.23
CSP-15	U.S.	5.86	bcde	0.87	2.35	0.33	0.05	0.19
SSD1-E41	U.S.	5.84	bcde	0.69	1.77	0.27	0.02	0.18
Rosso	U.S.	5.63	abcde	0.91	2.31	0.27	0.02	0.18
Orion	U.S.	5.57	abcde	0.85	2.15	0.25	0.03	0.16
Nema 1401	U.S.	5.26	abcd	0.87	2.06	0.20	0.03	0.19
Shako	U.S.	5.23	abcd	0.81	2.02	0.19	0.04	0.15
CSP-8	U.S.	4.44	abc	0.70	1.57	0.25	0.04	0.19
tomatillo	Spain	4.18	ab	1.11	2.97	0.95	0.06	0.26
tomatillo	U.S.	3.97	а	0.86	2.68	0.98	0.06	0.40

^{*a*} For fruitiness intensity, the same letter indicates that they are not significantly different (p < 0.05) according to Fisher's least significant difference (LSD = 1.73).

juices were significantly discriminated whenever the fruitiness intensity mean scores differed by at least 1.7. The observed range being 5 times greater than the LSD, this confirmed the presence of a wide range of perceived fruitiness intensity among commercial processing tomato varieties. For the 10 varieties produced in the United States and Spain, fruitiness intensity was variety (p < 0.0001) and location dependent (p < 0.0001) 0.0001), with juices from Spain [mean = 9.2, standard deviation (SD) = 1.9 being fruitier than those from the United States (mean = 5.9, SD = 1.0). There was no variety/location interaction (p = 0.58), indicating that the rankings of fruitiness intensity of the different varieties of tomato grown in Spain and the United States were similar. As an example, varieties CSP-13, CSP-11, and CSP-12 were the fruitiest tomato samples in both locations. Fruitiness intensity for the nine varieties and tomatillo grown in Spain was highly correlated (r = 0.93) with that of those grown in the United States.

Effect of Variety and Location on Sugars and Organic Acids. For the 10 varieties produced in the United States and Spain in 1991, glucose, reducing sugars, and malic and glutamic acid were not significantly variety dependent but were location dependent (Table 2). Spanish samples had more glucose (1.42 versus 1.03%) and more reducing sugars (2.88 versus 2.32%) but less malic (0.035 versus 0.048%) and glutamic acid (0.19 versus 0.23%) than U.S. samples. Citric acid was not location dependent but appeared to be highly variety dependent (ranging from 0.18% for Gemini to 0.30% for Blazer).

For the 10 varieties produced in the United States and Spain in 1992, glucose, reducing sugars, and malic and glutamic acid concentrations were also location dependent (Table 3). Samples from Spain had more glucose (1.46 versus 0.95%) and more reducing sugars (3.25 versus 2.35%) but less malic (0.040 versus 0.048%) and glutamic acid (0.12 versus 0.20%) than U.S. samples. Malic acid was variety dependent (ranging from 0.030% for Blazer, Orion, and Nema 1401 to 0.080% for CSP-13). Citric acid was only variety dependent [ranging from 0.25% (Nema 1401) to 0.43% (CSP-12), with an extreme of 0.97% for tomatillo].

Biochemical Markers for Tomato Fruitiness Intensity. The aim of this work was to define markers for flavor that could be used as simple and reliable tools to identify plant material for selection and breeding of better flavored tomato raw materials. The relationship between fruitiness intensity and the tomato constituents of glucose, reducing sugars, and citric, malic, and glutamic acid was examined (Table 4). For the 1991 trials, the best relationship was found between fruitiness intensity and the amount of reducing sugars (r =0.83) and glucose (r = 0.80). These correlations were location independent. Although a combination of reducing sugars and malic and glutamic acid contents did not better explain tomato fruitiness ($R^2 = 0.70$) than reducing sugars alone ($R^2 = 0.69$), malic acid appeared to discriminate tomatoes between the two growing locations. Citric acid content, which was variety dependent, did not explain very well perceived fruitiness (R^2 = 0.09).

In 1992, there was also a significant correlation between perceived fruitiness intensity and glucose (r =0.73) and reducing sugars (r = 0.65) (Table 4). Without the data for cherry and tomatillo, which had high concentrations of sugars and citric acid (Table 3), respectively, correlation with glucose (r = 0.86) and reducing sugars (r = 0.81) was better. Interestingly, the correlation between fruitiness and the ratio of reducing sugars/glutamic acid was high (r = 0.79), even with

Table 4. R^2 for Different Models of Prediction of TomatoFruitiness with Sugars, Organic Acids, and GlutamicAcid of Tomatoes Grown in 1991 and 1992 in Spain andthe United States

variable	1991	1992
glucose	0.64	0.53
reducing sugars	0.69	0.42
citric acid	0.09	0.06
malic acid	0.05	0.00
glutamic acid	0.03	0.28
reducing sugars/glutamic acid ratio	0.31	0.63
glucose + reducing sugars glucose + glutamic acid	0.70 0.66	0.69 0.74
reducing sugars + malic acid + glutamic acid	0.70	0.79
glucose + reducing sugars + glutamic acid	0.70	0.75
glucose + reducing sugars + citric acid + malic acid + glutamic acid +	0.71	0.84

reducing sugars/glutamic ratio

cherry and tomatillo, favoring the idea that tomato juice flavor is affected by the sugar-to-acid ratio (Gould, 1978). In fact, higher sugar and lower organic acid content in juices from varieties produced in Spain (and hence increased sugar-to-acid ratios) were associated with increased fruitiness intensity. The possibility that a combination of parameters could explain fruitiness intensity was examined, using multiple regression analysis (Table 4). The best model ($R^2 = 0.79$) combined reducing sugars and glutamic and malic acid, despite the facts that glutamic acid was negatively linked to fruitiness (r = -0.53) and that malic acid alone was not linked at all to fruitiness ($R^2 = 0.00$). To the authors' knowledge, this is the first report that glutamic acid can affect tomato flavor.

Biochemical Analysis and Fruitiness Evaluation of *L. pimpinellifolium* Breeding Lines. The juice samples of the 176 L. pimpinellifolium breeding lines produced in 1993 in Spain were scored only once by the panel (Figure 2), which had proven its consistency in 1991 and 1992. The juices were subjected to sugar and organic acid analysis (data not shown). The sensory screening showed a wide range of perceived fruitiness intensity (scores from 6.1 to 14.3, mean = 9.6). According to Fisher's LSD, tomato juices were significantly discriminated when their fruitiness intensity mean scores differed by at least 3.5. The juice composition of the breeding lines varied much more than for the commercial varieties tested in 1991 and 1992: glucose (0.60-2.88%; mean = 1.33%), reducing sugars (1.39-5.12%; mean = 2.73%), citric acid (0.14-0.47%; mean = 0.26%), malic acid (0.011-0.046%; mean = 0.028%), and glutamic acid (0.07-0.35%; mean = 0.16%). °Brix (4.18-5.98; mean = 5.16) and pH after heat processing (4.31-4.66; mean = 4.51) were also quite variable.

A simple correlation analysis over all data (for 176 samples, |r| > 0.14 is significant at a 5% significance level) revealed that the reducing sugars/glutamic acid ratio (r = 0.43), reducing sugars (r = 0.30), glucose (r = 0.26), and glutamic acid (r = -0.34) were the best individual markers for fruitiness intensity. Average fruitiness intensity was 10.5 for juices having a reducing sugar content >3.5%, confirming earlier findings with commercial varieties tested in 1991 and 1992 that samples with high glucose and reducing sugars and low glutamic acid contents were perceived as fruitier. By using multiple regression analysis, fruitiness intensity

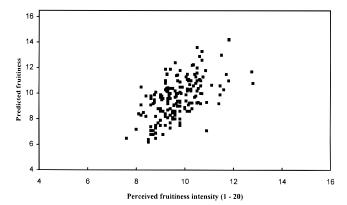


Figure 2. Comparison of perceived fruitiness intensity of a population of 176 breeding lines from *L. pimpinellifolium* with the calculated fruitiness intensity equation of fruitiness = $-19.38 - 35.59 \times \text{[malic acid]} + 0.084 \times \text{[reducing sugars/glutamic acid ratio]} + 5.53 \times \text{[pH after heat processing]} + 0.685 \times \text{[°Brix]}$ with $R^2 = 0.31$.

was better explained with a model including the parameters of (1) malic acid, (2) reducing sugars/glutamic acid ratio, (3) juice pH after heat processing, and (4) °Brix. These four parameters accounted for $R^2 = 0.31$, instead of $R^2 = 0.19$ for the reducing sugars/glutamic acid ratio alone, and a maximal $R^2 = 0.45$ for a total of 30 agronomic, technological, and compositional variables tested. The following equation described fruitiness intensity well with few variables: fruitiness = $-19.38 - 35.59 \times$ [malic acid] + $0.084 \times$ [reducing sugars/glutamic acid ratio] + $5.53 \times$ [pH after heat processing] + $0.685 \times$ [°Brix]. The plot of actual versus predicted fruitiness intensity is shown in Figure 2.

Summary. The scope of this study was the identification of reliable biochemical markers for the flavor of processing tomato. Variety dependence of the flavor of fresh market tomato is well documented (Stevens et al., 1979; Hobson and Bedford, 1989; Jansen, 1994), and it is clear that vine-ripened tomatoes have better flavor (Bisogni et al., 1976; Stevens et al., 1977). Regarding heat-inactivated processing tomato, to the authors' knowledge there are no published data available on the effect of genetic differences and location on flavor perception. Whether the fruitiness intensity differences found for varieties grown in Spain and the United States were due to agricultural practice, soil, or climatic conditions remains uncertain. However, they were clearly linked to compositional differences. Over a three year period, glucose, reducing sugars, reducing sugars/ glutamic acid ratio, and glutamic acid were consistently found to be the best individual markers for fruitiness intensity. The negative correlation of glutamic acid to tomato fruitiness was unexpected, in contrast to the well-known flavor enhancing ability of glutamic acid [reviewed by Raiten et al. (1995)]. The fact that glutamic acid is most effective as a flavor potentiator in the pH range 5.5–8.0 and not in the acid range of tomato juice (pH 4.0-4.6) may explain this observation. Sugars, organic acids, and glutamic acid being major constituents of tomato, and possibly directly interacting with flavor, their use as markers should permit considering flavor as well as compositional quality in current tomato breeding and thus contribute to the overall quality improvement of tomato.

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LITERATURE CITED

- Baldwin, E. A.; Nisperos-Carriedo, M. O.; Baker, R.; Scott, J.
 W. Quantitative analysis of flavor parameters in six Florida tomato cultivars (*Lycopersicon esculentum* Mill). *J. Agric. Food Chem.* **1991**, *39*, 1135–1140.
- Bisogni, C. A.; Armbuster, G.; Brecht, P. E. Quality comparisons of room ripened and field ripened tomato fruits. *J. Food Sci.* **1976**, *41*, 333–338.
- Brauss, M. S.; Linforth, R. S. T.; Taylor, A. J. Effect of variety, time of eating, and fruit-to-fruit variation on volatile release during eating of tomato fruits (*Lycopersicon esculentum*). *J. Agric. Food Chem.* **1998**, *46*, 2287–2292.
- Bucheli, P.; López, J.; Voirol, E.; Pétiard, V.; Tanksley, S. D. Definition of biochemical and molecular markers (Quality Trait Loci) for tomato flavour as tools in breeding. Abstract at the 3rd worldwide congress on the processing tomato, Pamplona, Spain, May 25–29, 1998. Acta Hortic. 1998, in press.
- Buttery, R. G.; Teranishi, R.; Ling, L. C. Fresh tomato aroma volatiles: a quantitative study. J. Agric. Food Chem. 1987, 35, 540–544.
- Davies, J. N.; Hobson, G. E. The constituents of tomato fruit the influence of environment, nutrition, and genotype. *CRC Crit. Rev. Food Sci. Nutr.* **1981**, Nov, 205–280.
- De la Torre, R.; Mangut, V.; Rodriquez, E.; González, C.; Latorre, A. Rapid method for stabilization of tomato variety trial juice samples for analysis. Abstract at the 3rd worldwide congress on the processing tomato, Pamplona, Spain, May 25–29, 1998.
- Gould, W. A. Quality evaluation of processed tomato juice. J. Agric. Food Chem. **1978**, 26, 1006–1011.
- Hintze, J. NCSS 6.0 User's Manual; Number Cruncher Statistical Systems: Kaysville, UT, 1996.
- Hobson, G. How the tomato lost its taste. *New Sci.* **1988**, Sept 19, 46–50.
- Hobson, G.; Bedford, L. The composition of cherry tomatoes and its relation to consumer acceptability. *J. Hortic. Sci.* **1989**, *64*, 321–329.
- Jansen, J. Geschmack von Tomaten; Gemüse: München, 1994; Vol. 30, pp 253–255.

- Jones, R. A.; Scott S. J. Improvement of tomato flavor by genetically increasing sugar and acid content. *Euphytica* **1983**, *32*, 845–855.
- Lever, M. A new reaction for colorimetric determination of carbohydrates. *Anal. Biochem.* **1972**, *47*, 273–279.
- Malundo, T. M. M.; Shewfelt, R. L.; Scott, J. W. Flavor quality of fresh tomato (*Lycopersicon esculentum* Mill.) as affected by sugar and acid levels. *Postharvest Biol. Technol.* **1995**, *6*, 103–110.
- Neter, J.; Wasserman, W.; Kutner, M. H. Applied Linear Statistical Models, 3rd ed.; Irwin, R. D., Ed.; Donnelley: 1990; pp 443–452, 818–860.
- Petersen, K. K.; Willumsen, J.; Kaack, K. Composition and taste of tomatoes as affected by increased salinity and different salinity sources. J. Hortic. Sci. Biotechnol. 1998, 73, 205–215.
- Pétro-Turza, M. Flavor of tomato and tomato products. *Food Rev. Int.* **1986**, *2*, 309–351.
- Raiten, D. J.; Talbot, J. M.; Fisher, K. D. In Analysis of Adverse Reactions to Monosodium Glutamate (MSG); American Institute of Nutrition: Bethesda, MD, 1995.
- Stevens, M. A.; Rick, C. M. Genetics and Breeding. In *The Tomato Crop. A Scientific Basis for Improvement*; Atherton, J. G., Rudich, J., Eds.; Chapman and Hall: London, 1986.
- Stevens, M. A.; Kader, A. A.; Albright-Holton M.; Algazi, M. Genotypic variation for flavor and composition in fresh market tomatoes. J. Am. Soc. Hortic. Sci. 1977, 102, 680– 689.
- Stevens, M. A.; Kader, A. A.; Albright-Holton M. Potential for increasing tomato flavor via increased sugar and acid content. J. Am. Soc. Hortic. Sci. 1979, 104, 40–42.
- Tanksley, S. D.; Grandillo, S.; Fulton, T. M.; Zamir, D.; Eshed, Y.; Pétiard, V.; López, J.; Beck-Bunn, T. Advanced backcross QTL analysis in a cross between an Elite processing line of tomato and its wild relative *L. pimpinellifolium. Theor. Appl. Genet.* **1996**, *92*, 213–224.
- Wolcott, T.; Merson, R. L. 1990 Processing Tomato Variety Trials; Department of Food Science and Technology, University of California: Davis, CA, 1990.

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