

Mixture Approach for Optimizing Lycopene Extraction from Tomato and Tomato Products

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A simple mixture process design based on the comparison of both quadratic and special cubic models and involving three mixture components (hexane/acetone/ethanol) as a solution for extracting lycopene from raw tomato, tomato sauce, and tomato paste was used to confirm the hypothesis that lycopene extraction rates are a function of the solvent used during the extraction process. Conventional criteria ($p \leq 0.15$) were used to identify influencing effects in each model. Although the major component used in lycopene extraction was hexane, there was a positive secondary synergistic interaction of hexane with ethanol (all sample types) and with acetone (tomato paste samples); this suggests that a mixture including all three components is essential for optimizing the extraction process. The partial special cubic model yielded three stationary points, indicating the concentrations of hexane, acetone, and ethanol required to optimize lycopene extraction in raw tomato, tomato sauce, and paste.

KEYWORDS: Raw tomato; lycopene extraction; tomato sauce and paste; statistical design of experiments

INTRODUCTION

Lycopene is the major carotenoid in tomatoes and tomato products, which are considered an important source of this compound in the human diet (1, 2). The tomato lycopene content varies considerably, reflecting the influence of variety (generally genetic factors), maturity, and both agronomic and environmental conditions during growing (2–8). Generally speaking, tomato products tend to be concentrated, thus also concentrating the lycopene content (9–11). Processed tomatoes additionally appear to increase the lycopene absorption by body tissues, due to enhanced bioavailability attributed to geometric isomer variation during processing (*cis*-isomers are more available) and to changes in the composition and structure of the food, which may increase the release of lycopene from the tomato tissue matrix (2).

Several epidemiological studies report that lycopene-rich diets have beneficial effects on human health (12, 13). A possible role has been suggested for tomatoes and tomato products in preventing cardiovascular disease and protecting against some types of cancer (based on lycopene content) (7, 14) as well as against ultraviolet light-induced erythema (15). The quantification of lycopene content is thus of considerable nutritional interest and is essential for determining the potential health benefits of tomatoes and tomato products.

Recent studies have described a lycopene extraction process based on supercritical CO₂, which enables the extraction of over 60% of lycopene from tomato waste (16, 17). However, because lycopene is fat soluble, it is more commonly extracted with organic solvents such as ethanol, acetone, petroleum ether, hexane, benzene, chloroform, etc. prior to chemical analysis for quantitative determination (9, 11, 18–23). A mixture of hexane with acetone and ethanol or methanol is often used (2, 23, 24) because (i) other components such as diethyl ether and tetrahydrofuran may contain peroxides that react with carotenoids (24), (ii) recovery rates with mixtures including ethyl acetate are very low (23), and (iii) the stability of lycopene extracts obtained with hexane/acetone or hexane/ethanol is higher than that of extracts obtained with other organic solvents such as chloroform, methanol, or dichloromethane (22). Lycopene can subsequently be quantified spectrophotometrically or by high-performance liquid chromatography, but in both cases, a good rate of lycopene extraction from samples is essential for accurate results. To reduce the analysis time, other methods have also been developed, based on certain color parameters (CIE Lab system) of tomato and tomato products (25, 26). Although these methods are rapid, inexpensive, and require no hazardous chemicals and could thus be used for lycopene screening by growers and industrial processors, lycopene is quantified using a number of different equations, which could give rise to errors when used in scientific studies.

A statistical design of experiments (DOE) is a well-established concept for the planning and execution of informative experiments. One major application of DOE is in the preparation and

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Table 1. Moisture, Soluble Solid, pH, and Color Parameters in Raw Tomatoes, Tomato Sauce, and Tomato Paste^a

sample	moisture (%)	SS ^b (%)	pH	color parameters						
				L*	a*	b*	a*/b*	(a*/b*) ²	H ^c	C ^d
raw tomato	94.5	4	4.24	39.12	4.92	6.20	0.79	0.62	0.31	64.70
tomato sauce	82.97	13	4.25	37.39	12.95	10.97	1.18	1.39	0.88	289.61
tomato paste	76.21	29	4.14	34.71	14.17	6.29	2.25	5.06	2.10	242.06

^a Mean values of three measurements. ^b Soluble solids. ^c Hue angle values. ^d Saturation or metric chroma values.

modification of mixtures; this involves the use of mixture designs (27).

The aim of this study was to confirm the hypothesis that lycopene extraction rates are a function of the solvent ingredients included in the extraction mixture; if this hypothesis were true, i.e., if the null hypothesis (H_0), which states that the response (extracted lycopene) does not depend on the mixture components, were rejected, the study would then seek to develop the best solvent mixture including hexane/acetone/ethanol to extract lycopene from samples studied. To achieve this, the problem was approached as a general mixture problem because the measured response was assumed to depend only on the proportions of the ingredients present in the mixture and not on the amount of the mixture.

MATERIAL AND METHODS

Samples. The samples used in the present study were raw tomatoes (commercial variety: Canario, size 57/67, first class), tomato sauce, and tomato paste, all purchased in a local supermarket. On the day of purchase, the raw tomatoes were cleaned, homogenized, and stored at -80°C in plastic bottles until analysis. Common brands of tomato sauce and tomato paste were sampled directly from the containers. Before the lycopene content was determined, soluble solids, moisture, pH, and color parameters were analyzed in all samples (Table 1).

Experimental Design. A simplex centroid mixture design {3,2} was used, increased by six central points for three-component mixtures, according to Cornell (28), as shown in Figure 1. Each of the 13 extractant mixtures was prepared for immediate use. The proportion of each reagent (mixture component) varied between 0 (reagent not present) and 1 (extractant mixture comprising a single component). Thus, a mixture system consisting of q components (X_1, X_2, \dots, X_q) satisfied the following constraints (28):

$$0 \leq X_i \leq 1 \quad (1)$$

$$\sum_{i=1}^{q-1} x_i + x_2 + \dots + x_q \quad (2)$$

The selection of reagents to be included in the extraction mixture (factors) was based on data in the scientific literature (2, 22–24), and one response (extracted lycopene) was measured in raw tomato, tomato sauce, and tomato paste.

In the same way that in factorial experimental design, it is considered advisable to transform natural variables into coded variables, usually defined as dimensionless with mean zero and the same spread or standard deviation (29), in mixture experimental design, the pseudo-components are defined as combinations of the original components; the reason for introducing pseudo-components is that usually both the construction of designs and the fitting of models are much easier when done in the pseudo-component system than when done in the original component system (28). Thus, for each reagent included in the extraction mixture (hexane, acetone, and ethanol), one pseudo-component was considered, i.e., X_1, X_2 , and X_3 , respectively.

The basic analysis for a response surface in a simplex (the experimental region of a problem mixture with p components) consists

Experimental design

Point	Coordinate
1	(1, 0, 0)
2	(0, 1, 0)
3	(0, 0, 1)
4	(0, 1/2, 1/2)
5	(1/2, 0, 1/2)
6	(1/2, 1/2, 0)
7	(1/3, 1/3, 1/3)
8	(1/6, 5/12, 5/12)
9	(5/12, 1/6, 5/12)
10	(5/12, 5/12, 1/6)
11	(2/3, 1/6, 1/6)
12	(1/6, 2/3, 1/6)
13	(1/6, 1/6, 2/3)

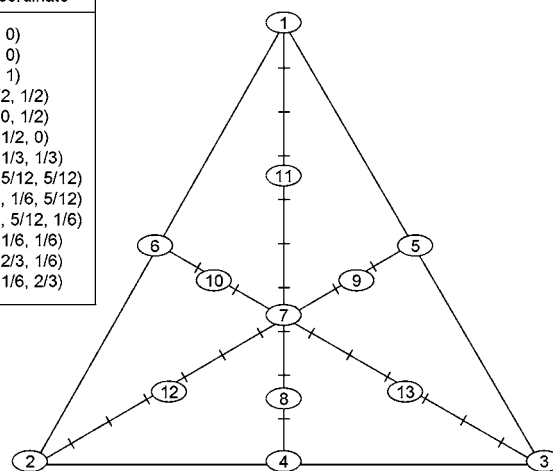


Figure 1. Thirteen-point augmented simplex-centroid points to support quadratic or special cubic models for three-component mixtures.

of fitting a quadratic model of the type

$$Y = \sum_{i=1}^p b_i X_i + \sum_{i=1}^{p-1} \sum_{j=i+1}^p b_{ij} X_i X_j + \epsilon \quad (3)$$

where Y is each response, the first summation is the linear blending portion, the second represents the excess response from the quadratic model over the linear model (30), and ϵ represents the error of model. This model was fitted using the PLS (partial least squares projections to latent structures) regression technique. PLS has been extensively described in the literature (28).

Although the second-degree model provided information on each of the components individually (main effects) as well as on pairs of components (secondary effects), to locate stationary points, data were fitted by PLS regression to a special form of polynomial equation developed by Scheffe and generally known as the special cubic model (31), which includes a third-order term ($X_1 \times X_2 \times X_3$) to reveal the three-component interaction, if any, according to the following model.

$$Y = \sum_{i=1}^3 b_i X_i + \sum_{i=1}^2 \sum_{j=i+1}^3 b_{ij} X_i X_j + b_{123} X_1 X_2 X_3 + \epsilon \quad (4)$$

To compare the quadratic and special cubic models, it was necessary from the start to obtain both theoretical models.

The Design-Expert (Stat-Ease, Inc., Minneapolis) and Statistical (StatSoft, Inc., Tulsa) software packages were used to generate designs, fit the response surface model to the experimental data, and draw response surface figures.

Determination of Lycopene. For lycopene extraction, 1 g of raw tomatoes and of tomato sauce and paste was weighed into a 125 mL flask wrapped with aluminum foil to exclude light. Fifty milliliters of a mixture of hexane/acetone/ethanol, in different proportions according to the experimental design described below and shown in Table 2, was added to the flask to solubilize carotenoids. The samples were shaken for 30 min, and then, 10 mL of distilled water was added. The solution was left to separate into a distinct polar layer and a nonpolar

Table 2. Results Obtained for Each of 13 Extraction Mixtures Studied in Raw Tomato (R), Tomato Sauce (S), and Tomato Paste (P) as a Function of Hexane (X_1), Acetone (X_2), and Ethanol (X_3) Mixtures

trial	run	pseudo-components			lycopene (mg/100 g)		
		X_1	X_2	X_3	R	S	P
11	1	1	0	0	1.24	7.61	31.1
1	2	0	1	0	0.00	0.00	0.00
13	3	0	0	1	0.00	0.00	0.00
5	4	1/2	1/2	0	0.97	4.57	47.46
2	5	1/2	0	1/2	3.35	11.36	41.74
7	6	0	1/2	1/2	0.00	0.00	0.00
10	7	2/3	1/6	1/6	3.78	10.46	53.90
12	8	1/6	2/3	1/6	0.67	2.85	13.58
3	9	1/6	1/6	2/3	2.92	3.11	35.52
8	10	1/3	1/3	1/3	4.72	14.71	53.78
6	11	5/12	1/6	5/12	2.82	11.53	46.76
9	12	5/12	5/12	1/6	3.11	7.31	40.81
4	13	1/6	5/12	5/12	0.65	1.06	11.47

layer containing lycopene. The total lycopene content was obtained by measuring the absorbance of the lycopene hexane solution at 472 nm (20, 21). Pure lycopene (Sigma, St. Louis, MO) was used for the preparation of calibration curves.

Chemical Parameters of Samples. The moisture content was analyzed in all samples by oven drying at 105 °C to a constant weight. The pH was determined using a Crisson 2000 pHmeter (Barcelona, Spain), and the soluble solids were quantified in homogenized samples using a Leica Abbe Mark II Refractometer (Buffalo, NY) following Board (32).

Color Parameters. Three color readings were taken for each sample after homogenization of whole fruit, using a Minolta Chromameter Reflectance II CR-2000 (Minolta Limited, Milton Keynes, United Kingdom). The values a^* (red-green) and b^* (yellow-blue) were used to calculate the hue angle [$H = \tan^{-1}(a^*/b^*)$] and metric chroma [$C = (a^2 + b^2)^{1/2}$], which provided information about the color index of the samples (33).

RESULTS AND DISCUSSION

Table 2 shows the full experimental design and the amounts of lycopene extracted (expressed as mg/100 g) from raw tomatoes, tomato sauce, and tomato paste. In **Table 2**, the "trial" column shows the order in which the experiments were carried out (a randomized order) while the "run" column shows the formal or systematic order developed to obtain the experimental design. Randomization by these means is essential to ensure that the average influence of noise factors, such as environmental factors, is lessened (34).

The lycopene content ranged from 0.65 to 4.72 mg/100 g in raw tomato, 1.06 to 14.71 mg/100 g in tomato sauce, and 11.47 to 53.90 mg/100 g in tomato paste. In trial without hexane, the lycopene content was not quantified because the different layers could not be separated. The type of solvent used for extraction yielded different amounts of lycopene from the various samples, as reported by other authors (22, 23); however, comparison of samples showed that the lycopene content increased significantly as a function of heat processing. Khachick et al. (9) observed that the total carotenoid content after stewing the raw tomatoes remained unchanged, whereas heat processing applied during the manufacture of tomato paste led to an increase in total carotenoids due to concentration; however, the qualitative distribution remained identical for raw and stewed tomatoes.

The effect of each mixture ingredient (X_1 , hexane; X_2 , acetone; and X_3 , ethanol) on lycopene extraction from raw tomato, tomato sauce, and tomato paste, using both the quadratic and the special cubic models, is shown in **Table 3**. An effect was considered strong, and thus termed an influencing effect

Table 3. Effects in Terms of Pseudo-Components on Response (Extracted Lycopene) Fitted to Both Quadratic and Special Cubic Models and Statistical Significance Obtained for Each^a

terms of model	raw		sauce		paste	
	effect	p	effect	p	effect	p
quadratic model						
X_1	1.15	0.28	2.42	0.04 ^b	3.45	0.01 ^b
X_2	-0.36	0.73	-0.16	0.88	-0.44	0.67
X_3	0.00	0.99	-0.33	0.74	0.26	0.79
$X_1 \times X_2$	1.24	0.25	0.85	0.42	3.41	0.01 ^b
$X_1 \times X_3$	3.01	0.01 ^b	2.96	0.02 ^b	3.34	0.01 ^b
$X_2 \times X_3$	0.39	0.71	1.36	0.89	-0.00	0.99
special cubic model						
X_1	1.40	0.21	2.57	0.04 ^b	3.36	0.01 ^b
X_2	-0.24	0.82	-0.04	0.96	-0.35	0.74
X_3	0.15	0.89	-0.22	0.83	0.32	0.76
$X_1 \times X_2$	0.44	0.67	0.19	0.85	2.54	0.04 ^b
$X_1 \times X_3$	2.13	0.07 ^b	2.09	0.08 ^b	2.48	0.04 ^b
$X_2 \times X_3$	-0.36	0.73	-0.45	0.67	-0.33	0.75
$X_1 \times X_2 \times X_3$	1.51	0.18	1.18	0.28	0.66	0.53

^a A strong effect was considered when $p \leq 0.15$. ^b Considered as IE.

(IE), at $p \leq 0.15$. In general, different IEs were observed for lycopene extraction in raw tomato and tomato sauce and paste, although similar IEs were identified in both quadratic and special cubic models (**Table 3**), despite certain differences. The synergic hexane \times ethanol interaction was of less importance in the special cubic model, whereas in the quadratic model it was considered an IE for all three samples.

With regard to individual reagent effects on lycopene extraction, the effect of hexane was found to depend on the degree of sample processing (1.15, 2.42, and 3.45 for raw tomato, tomato sauce, and tomato paste, respectively, using the quadratic model, and 1.40, 2.57, and 3.36, respectively, using the special cubic model); hexane was an IE for tomato sauce and tomato paste (**Table 3**). The strong effect of hexane in processed samples increased with the intensity of heat processing (stronger in tomato paste than in tomato sauce). This suggests that the quantitative importance of hexane in lycopene extraction is related to tomato product processing.

Although acetone was not an IE in any substrate, its inclusion in the extractant mixture had a negative effect over the experimental range studied (**Table 3**); the addition of this reagent could therefore lead to a reduction in lycopene extraction (**Figure 2**). Hakala and Heinonen (35) found that the extraction of lycopene from tomato purée did not require water soluble penetration solvents, such as acetone, because when carotenoids are extracted with relatively unpolar solvents the amounts of polar xanthophylls decrease and the proportion of lycopene relative to total carotenoids increases from 76 to 87%, improving lycopene extraction. The negative effect of acetone on lycopene extraction would account for the results obtained by Lin and Chen (23), who extracted 27% more lycopene in tomato juice in the absence of acetone. It would therefore appear that acetone should be excluded from the solvent mixture. However, this is not necessarily the case; the secondary effects obtained with both models revealed a positive synergistic effect of hexane and acetone, with higher values than those obtained for the negative effect of acetone (**Table 3**). In fact, for tomato paste, the hexane/acetone interaction was considered an IE, with values of 3.41 and 2.54 in the quadratic and special cubic model, respectively. The exclusion of acetone in the extractant mixture might eliminate this positive effect on the lycopene extraction efficiency; the inclusion of a small fraction of acetone might reduce the negative primary effect and achieve the positive

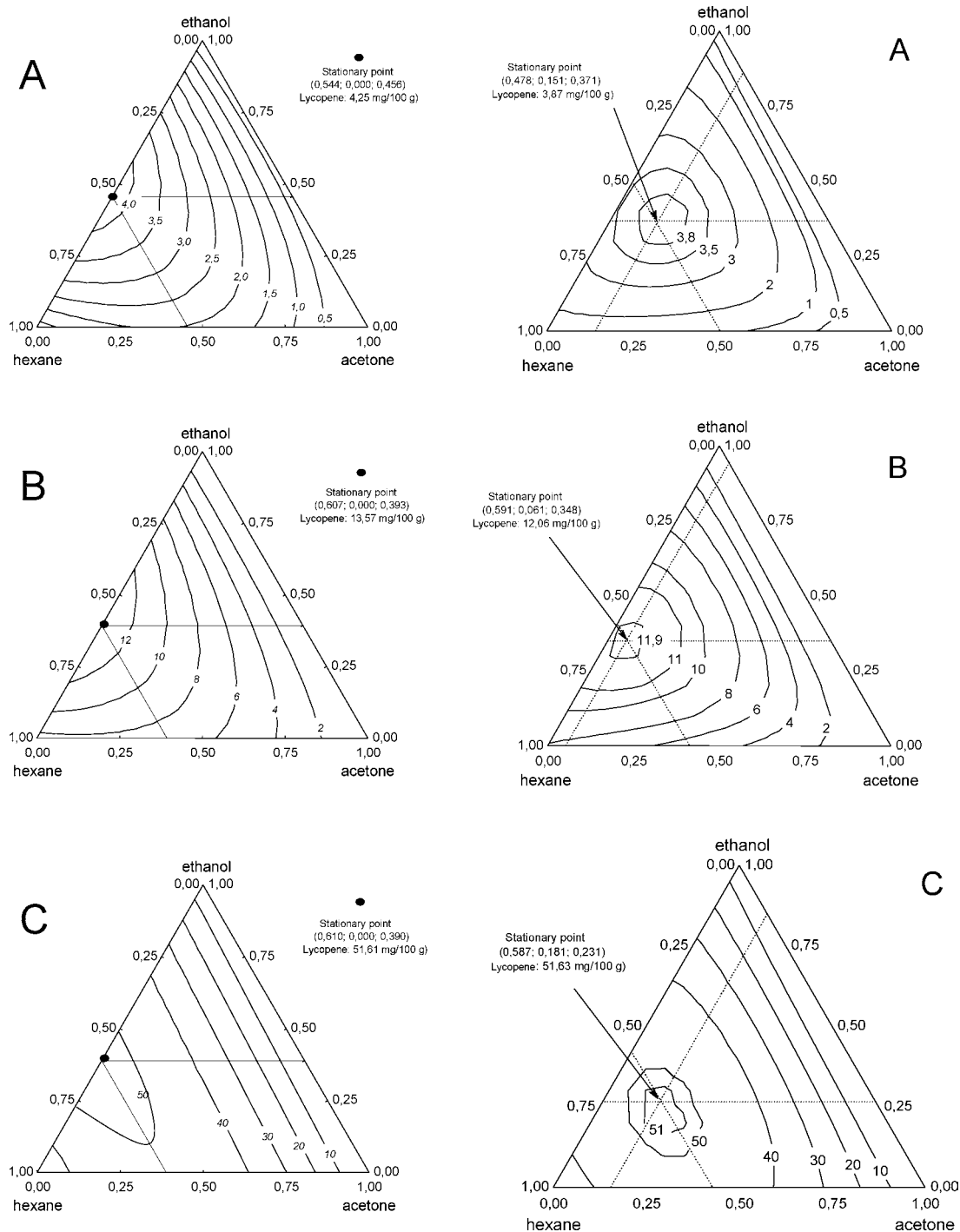


Figure 2. (a) Surface contours of estimated lycopene extraction from raw tomato (A), tomato sauce (B), and tomato paste (C) as a function of the extractant mixture used according to the quadratic model. (b) Surface contours of estimated lycopene extraction from raw tomato (A), tomato sauce (B), and tomato paste (C) as a function of the extraction mixture used according to the special cubic model.

secondary effect. Other authors report conflicting effects of acetone on lycopene extraction: Lin and Chen (23) found that the acetone/hexane mixture (3:5, v/v) led to lower lycopene extraction rates in tomato juice, whereas Tanugbodhitam et al. (22) reported that the same mixture (4:6, v/v) was as efficient as the ethanol/hexane mixture (4:3, v/v).

Ethanol was not an IE for lycopene extraction from any of the tomato samples (Table 3). The effect of ethanol on lycopene extraction from raw tomato was negligible (0.0, $p < 0.99$, Table 3), which might suggest that it could be excluded from the extractant mixture. However, a positive synergistic secondary effect was found for hexane/ethanol in all three samples, suggesting that the inclusion of ethanol would improve extrac-

tion rates. Similar findings are reported by Tanugbodhitam et al. (22) and Lin and Chen (23), who observed that the ethanol/hexane mixture (4:3, v/v) increased lycopene extraction in tomatoes.

The observation of different IEs for lycopene extraction from the different types of sample might be ascribed to differences in compounds extracted, since—like other carotenoids—lycopene occurs in various geometrical isomers (36) and displays varying degrees of oxygenation (37). In fact, differences may be due to the effects of heat processing on changes in chemical isomers and on the release of carotenoids from the matrix. The kinetics of tomato lycopene degradation by heating includes the oxidation and isomerization of all *trans*- to *cis*-isomers. Heat treatment

Table 4. Lycopene Content (mg Lycopene/100 g) in the Confirmatory Experiments, Using the Optimum Extraction Mixtures Obtained via Both Quadratic and Special Models, in Raw Tomato (R), Tomato Sauce (S), and Tomato Paste (P)

sample	mixture	quadratic			special cubic			CV
		estimated	found	CV	estimated	found	CV	
				full model				
R	54.4/0.0/45.6	4.25	3.14	21.24	47.8/15.1/37.1	3.87	3.28	11.67
S	60.7/0.0/39.3	13.57	10.78	16.20	59.1/6.1/34.8	12.06	12.43	2.14
P	61.0/0.0/39.0	51.61	43.03	12.58	58.7/18.1/23.1	51.63	46.52	7.36
				partial model ^a				
R	54.4/0.0/45.6	4.70	3.14	28.14	47.8/15.1/37.1	3.87	3.28	6.58
S	60.7/0.0/39.3	14.42	10.78	20.43	59.1/6.1/34.8	12.06	12.43	3.55
P	61.0/0.0/39.0	51.60	43.03	12.81	58.7/18.1/23.1	51.63	46.52	6.02

^a For R and S samples excluding both X_1X_2 and X_2X_3 terms and for C samples excluding the X_2X_3 term and for special cubic model in all cases excluding the $X_1X_2X_3$ term.

induces *cis/trans* isomerization (e.g., formation of 5-*cis*-lycopene), and a high proportion of *cis*-isomers in tomato-based products results in a less intense red color (38). Wiese and Dalmasso (39) report an increase in the hue angle of tomato juice after processing and storage, indicating a loss of red color. A hue angle of 0° indicates red color; rising values indicate decreasing color purity. The ratios a^*/b^* and $(a^*/b^*)^2$ have indeed been used to determine the red color in tomatoes and to estimate the lycopene content (25). In the present study, samples displayed an increase in hue angle, a^*/b^* , and $(a^*/b^*)^2$ values as a function of heat processing (Table 1). The lycopene content and the ratio of *trans*- to *cis*-isomers may have caused the a^*/b^* values, since a less pure red color in tomato and tomato products indicates a greater content of lycopene *cis*-isomers in the sample. However, trends in lycopene degradation and color parameters have not been widely studied or correlated.

Regardless of changes in lycopene isomers, heat processing, e.g., by cooking, and mechanical texture disruption lead to a breakdown of tomato cell wall structures, disrupting chromoplast membranes and reducing cellular integrity, thus rendering several phytochemicals, including lycopene, more accessible to extraction (2, 24). It has been reported that food processing, including cooking and grinding, might improve lycopene bioavailability for humans by breaking down cell walls and increasing the ratio of *cis/trans* isomers (2, 24). Although differences in the IEs observed here for different tomato samples may well suggest that the compounds extracted in each sample were not identical, a number of authors (2, 9, 10, 24) report that tomato sauce and tomato paste contain about 90% *trans*-isomers, so that the proportion of *cis*-isomer lycopenes could be considered low after processing.

Clearly, given their positive synergistic interaction with hexane, both ethanol (in all samples) and acetone (in tomato paste) are essential elements of the extraction mixture, and neither should be excluded. For that reason, most lycopene extraction methods use two or three of the reagents selected here. To establish the most efficient reagent combination to optimize lycopene extraction, we must turn to the theoretical model, which describes the extraction process as a function of pseudo-components, and locate on the graph the stationary point X_0 at which the derivative of a function $f(X)$ vanishes, i.e., $f'(X_0) = 0$.

Table 4 shows the estimated lycopene concentrations using both quadratic and special cubic models, considering in each case the full and the partial model. For raw tomato, the quadratic model ($F = 3.45$, $p < 0.07$, $R^2 = 0.71$) yields a stationary point at which lycopene extraction is optimized using a mixture of

Table 5. Proposed Extractant Mixture Composition for Raw Tomato (R), Tomato Sauce (S), and Tomato Paste (P) as Compared to Reported Mixtures of Hexane/Acetone/Ethanol^a

reference	extractant mixture component		
	hexane	acetone	ethanol
18		100	
19			100
8, 20, 25, 26, 40	50	25	25
35		100	
41	33.33	33.33	33.33
22	43		57
42	50	25	25
43; n = method	40	60	
43; n = method	60		80
44; n = method	60	40	
23	26.57		35.42
23	38.75		23.25
23	31	10.33	20.66
proposed by sample type			
raw tomato	47.80	15.1	37.1
tomato sauce	59.10	6.1	34.8
tomato paste	58.70	18.11	23.10

^a All components are expressed as percentages (v/v).

54.4% hexane and 45.6% ethanol (excluding acetone); this gives an estimated concentration of 4.25 mg/100 g (Figure 2). However, because of the synergistic effect between hexane and acetone (Table 3, 1.24, $p < 0.25$), the special cubic model ($F = 3.79$, $p < 0.06$, $R^2 = 0.79$) yielded a stationary point at which lycopene extraction is optimized using 47.8% hexane, 15.1% acetone, and 37.1% ethanol, giving an estimated lycopene concentration of 3.87 mg/100 g (Figure 2).

The same procedure was carried out for tomato sauce and tomato paste. In tomato sauce, the stationary point for the quadratic model ($F = 4.56$, $p < 0.03$, $R^2 = 0.76$) gave 60.7% hexane and 39.3% ethanol, while that of the special cubic model ($F = 4.24$, $p < 0.05$, $R^2 = 0.81$) gave 59.1% hexane, 6.1% acetone, and 34.8% ethanol. In tomato paste, the stationary point for the quadratic model ($F = 10.27$, $p < 0.00$, $R^2 = 0.88$) gave 61.0% hexane and 39.0% ethanol, while that of the special cubic model ($F = 7.94$, $p < 0.01$, $R^2 = 0.88$) gave 58.7% hexane, 18.1% acetone, and 23.1% ethanol.

To determine which model best fit the data obtained from laboratory extraction, a series of complementary experiments were carried out. The lycopene contents found in the confirmatory experiments were 3.14, 10.78, and 43.03 mg/100 g and 3.28, 12.43, and 46.52 mg/100 g for raw, tomato sauce, and tomato paste, extracted with the stationary point of quadratic and special cubic models, respectively (Table 4). The best

results were obtained using the special cubic model on a partial form, displaying the lowest coefficient of variation between estimated and observed values. The stationary points obtained for each sample type with the special cubic model should therefore be used to optimize lycopene extraction (**Figure 2b**). In addition, a positive correlation was found between lycopene content and color parameters $[(a^*/b^*)^2]$ and hue angle values] (**Table 1**), with a correlation coefficients of $r = 0.99$ and $r = 0.97$ ($p < 0.001$), as have been described previously by other authors (5, 25). The proposed extraction mixtures are shown in **Table 5**, in comparison to a number of hexane/acetone/ethanol mixtures reported in the literature.

In summary, the results obtained here bear out the hypothesis of van den Berg et al. (24), who postulated that the choice of solvent depends on the nature of the matrix. In the present study, which used samples subjected to different forms of industrial processing, the use of DOE methodology with mixtures enabled identification of the best solvent mixture composition for each type of tomato sample.

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Received for review April 22, 2004. Revised manuscript received June 28, 2004. Accepted June 29, 2004. We are grateful to the European Union (FEDER) and to the Murcia Regional Government Department of Agriculture, Water and the Environment for providing financial support for this research, through Project AGR/21/FS/02.

JF049345H