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Full Length Research Paper

Use of factorial experimental design for analyzing the effect of storage conditions on color quality of sun-dried tomatoes

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Color of sun-dried tomato is an important quality criteria and color stability of sun dried tomatoes is affected by storage conditions such as temperature, time and packaging type. The objective of this study was to investigate the effects storage time, storage temperature and packaging type on color stability of sun-dried tomatoes by using factorial experimental design. In this study, the 2^3 factorial design was used to represent mathematical relationship between the storage conditions and color values (L and a/b) and used to verify whether or not factor variables of storage conditions are effective on color. Results in experimental model showed that all factors had negative effects on the color values which indicated the reducing stability of color. The main effect was found to be the time having the highest coefficient (-3.3 for L and -0.12 for a/b) in design models and storage temperature was found to be the least effective factor. Design model and response surface plots revealed that storage temperature could be kept around 20°C for 9 months and storing under vacuum packaging should be selected to reduce color losses for the safe storage conditions.

Key words: Sun-dried tomatoes, color, storage, factorial experimental design, quality.

INTRODUCTION

Tomato is an important commercial vegetable with the highest production and consumption volumes in the world (Cernişev, 2010; Geboloğlu et al., 2011). Drying is an important process for food preservation (Esehaghbeygi and Basiry, 2011) where water is removed from the foods. In drying process, fresh tomato can be cut in different shapes such as halves, thin slices and quarters and dried tomato products have different applications such as they can be used in pizza and in different vegetable and spicy dishes (Zanoni et al., 1999). Many undesirable quality changes could occur in the dried products during drying and storage (Maskan, 2000, 2001). The color of tomato is an important quality index concerning consumer acceptance (Dermesonlouoglou et al., 2007; De Sousa et al., 2008). A common problem in dried samples is browning and brown color products are not attractive and are not desired by the consumers (Demirbüker et al., 2004; Cemeroğlu and Acar, 1986). To reduce undesirable quality changes during drying and storage, NaCl and sulphite were usually used as microbial and chemical preservatives and/or as color stabilizers in sun-dried tomatoes (Babalik, 1996; Yurdagel et al., 1996; Baloch et al., 1997; Shi and Le Maguer, 2000; Demirbüker, 2001; Latapi and Barrett, 2006; Lavelli and Torresani, 2011). Lycopene is the main coloring agent for the red color of the tomatoes (D'Sousa et al., 2008; Lavelli and Scarafoni, 2011). Lycopene oxidation is reported to be the most important reason for the color loss during storage of tomatoes (Demirbüker, 2001). Nguyen and Schwartz (1999) indicated that lycopene is susceptible to chemical changes when exposed to light and heat because of the presence of a long chain of conjugated carbon–carbon double bonds.

Oxygen plays an important role on non-enzymatic or

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enzymatic browning of the products and if oxygen in the package of the product increase color changes accelerates during storage period. Additionally, temperature and light have increasing effect on browning of carotenoids (Cemeroğlu and Acar, 1986). Expression of color values by Hunter or CIElab color space with L, a and b coordinates is common quality indicator of tomato and tomato based products. L (lightness variable) is also measured to indicate relative darkness (0) or lightness (100), a and b are chromaticity coordinates which represents color quality. Positive a value corresponding to the degree of redness while a negative value corresponds to the degree of greenness whereas positive b value represents to the degree of yellowness and the negative value represents the blueness. Additionally, a/b ratio value is commonly used as a redness index to report the color quality (brightness of red color) of tomato and tomato products (Babalik, 1996; Yurdagel et al., 1996; Shi et al., 1999; Demirbüker, 2001; López and Gómez, 2004; Batu, 2004; Ali et al., 2004; Sacilik et al., 2006; Liu et al., 2009). During drying, the moisture content is reduced and deterioration of the dried products is prevented within a period of time regarded as the safe storage period. Therefore, considering the safe storage conditions for the reduction of quality losses, storage conditions such as temperature, time and packaging are important factors for the color stability. Zanoni et al. (1999) reported that lycopene appeared to be stable during drying; however it was not stable during the storage of the dried product. Since the quality losses in the dried products may have adverse economic effects (Sacilik et al., 2006; Lahsasni et al., 2004), there is a need for safe storage condition settings that helps retain the color characteristics of tomato. Thus, proper storage conditions for dried tomato could be designed to reduce quality losses. However, reports about setting storage conditions for sun-dried tomatoes are limited.

Considering the aforementioned features, the objective of this study was to investigate the effects storage time, storage temperature and packaging type on color stability of sun-dried tomatoes by using factorial experimental design. Factorial experimental design was used to give mathematical relationship between the factors and the response. It has been frequently used in different food research experiments (Gardeur et al., 2007; Al-Asheh et al., 2003; Wormbs et al., 2004; Januszkiewicz et al., 2008). This design provides the best information for the effects of independent variables and their interactions on an experimental model (Dehghan et al., 2010; Forte et al., 2003); thus factors and their interactions can be estimated with minimal number of runs which is also important from the economical point of view (Cavaliere et al., 2010; Dos Santos et al., 2005; Montgomery, 2001; Forte et al., 2003). Models helped to analyze the safe storage conditions and were used to verify whether or not variables of storage conditions are effective on color response where both L and a/b values were analyzed separately.

MATERIALS AND METHODS

Raw materials and pre-drying treatments

Rio Grande variety harvested at red stage of maturity was used as raw material. Tomatoes were washed, cut into halves perpendicularly with stainless steel knife. Tomato halves were dipped into 6% NaCl, and 15% $Na_2S_2O_5$ solution stirred for 3.5 min until they contained 500 to 1000 ppm SO_2 and 10 to 13% NaCl. The tomato/dipping solution ratio was 1/3. Processed tomato halves left to dry under direct sun light for 6 days at 22.4 to 42.6°C and 32.0 to 39.4% relative humidity (RH). The drying was carried out to final moisture content of 10.5 to 11.0% from initial 4.5 to 5.2° Bx dry matter. Atmospheric conditions were monitored by ON SET HOBO H8 RH/Temp, 2 channels Data Logger during sun drying process. Total soluble solids were measured by a hand-held refractometer (Atago Co, Japan). Moisture content was determined according to the AOAC method (1995) at 70°C under 700 mmHg vacuum with vacuum oven (NUVE EV018). Total sulphur dioxide content was measured by Reith Williams Method modified by Ural et al. (1990) (Babalik, 1996). NaCl content was measured with Mohr method (Yurdagel et al., 1996).

Packaging and storage

Dried samples of approximately 100 g from each combination of the pre-drying treatments were used for packaging. In order to minimize degradative effects of light, light-proof aluminum laminated bags were selected for packaging which were kindly provided by Bakambalaj (Izmir, Turkey). Three layers of the bag were consists of polyester/Al/PE lamination. Thickness of the layer were polyester (12 µm), Al (9 µm) and PE (70 µm). Gas permeability of the bag was (O_2: 3.6 ml/m^2 day.atm at 23°C, 0% RH and vapour: 0.01 ml/m^2 day.atm at 38°C, 90% RH. Sun dried tomatoes were packaged under two atmospheric conditions as normal (thermal sealing in normal atmospheric conditions) and vacuum (thermal sealing under 40 mb vacuum) packaging. A combined system of Multivac Sepp Haggenmüller KG D-8941 Wolfertschwenden gas mixer (Multivac GMBH, Wolfertschwerden, Germany) and a Kramer Grebe vacuum apparatus (Kramer Grebe GMBH, Biedenkopf-Wallau, Germany) were used for packaging. Packaged samples were stored at two conditions as +4°C (at 37±3 RH) and +20°C (52±3 RH) for 9 months as well.

Experimental design: Factors, levels and response

To design proper storage conditions for sun-dried tomato to reduce color losses, factorial design was used. To set a mathematical model between response and factors, two responses were under investigation for color prediction; L and a/b value. To investigate the color changes, experimental design was carried out choosing three factors (process variables) effective on responses, namely: storage temperature, storage time and packaging type. These three factors were coded as A, B and C, respectively. For each of the factor, two different levels were set. They correspond to low and high levels of treatment conditions and coded as (-1) and (+1), respectively. Factors and levels were given in Table 1.

Experimental design methodology

A 2^3 full factorial design was carried out to set the mathematical

Table 1. Experimental factors and levels.

Factors	Parameter values	
	Low level (-1)	High level (+1)
A: Storage temperature (°C)	+4	+20
B: Storage time (month)	0	9
C: Packaging type	Vacuum package (under 40 mBar vacuum)	Normal package (atmospheric;1023 mBar)

Table 2. Design matrix for the experiments.

Std	run	Factor 1 A: storage temperature	Factor 2 B: storage time	Factor 3 C: packaging type	Response 1: L value	Response 2: a/b value
14	1	-1	1	1	24.97	1.94
16	2	1	1	1	24.63	1.92
6	3	-1	1	-1	27.58	2.4
5	4	-1	1	-1	27.64	2.42
11	5	1	-1	1	32.78	2.38
13	6	-1	1	1	24.98	1.94
4	7	1	-1	-1	32.84	2.4
9	8	-1	-1	1	32.78	2.38
1	9	-1	-1	-1	32.78	2.38
3	10	1	-1	-1	32.78	2.38
10	11	-1	-1	1	32.84	2.4
8	12	1	1	-1	27.62	2.29
2	13	-1	-1	-1	32.84	2.4
7	14	1	1	-1	27.65	2.31
15	15	1	1	1	24.63	1.93
12	16	1	-1	1	32.84	2.4

relationships and to represent how color of sun-dried tomatoes depends on storage time, temperature and packaging type. An experimental matrix was shown in Table 2. Running order for each run was randomized in order to minimize possible systematic errors. Coded levels were given as (-1) and (+1) on columns 3 to 5 and responses (L and a/b) of each run with two replicates were given in columns 6 and 7. To get information about the effects of storage conditions, a mathematical model was established which gave the relation between the response (Y) and the factors A, B and C. Thus, 2^3 factorial design model was set for each responses; L and a/b separately. All factors and their interaction terms were taken into account. Model can be expressed as follows:

$$Y = \beta o + \beta 1A + \beta 2B + \beta 3C + \beta 12AB + \beta 13AC + \beta 23BC + \beta 123ABC \quad (1)$$

Where βo was the constant; β1, β2 and β3 were coefficients for the coded variables A, B and C, respectively; β12, β13, β23 and β123 were the interaction effects between variables. In Figures 1 and 2, cube plots were presented to aid data interpretation which showed the combination of there factors with highest and lowest value of each effect at each corner. Response surface was graphed and used to graphically represent the values of factors, thus, it will be helpful to plan new experiments setting proper storage conditions even more. All design and analyzes of experiments were performed using design expert (6.0.6 trial). The main effects and interactions of factors on color were determined. As well as standard error of estimate, sum of squares of the errors, F statistics, p value, contour and surface plots were analyzed.

Color measurements

Color was measured by Minolta CR 310 chromameter (model CR 300 series; Minolta Co. Ltd., Japan) with xenon arc lamp for D65 light source and with ø50 mm measuring area for wide area illumination/0 viewing angle to obtain CIELab values L (lightness variable), a and b (chromaticity coordinates) values which represents color quality. Each reading was the average of five flashes and readings were calibrated with CR-A43 calibration plate. Expression of lightness were recorded in L and redness values were recorded in a/b. To avoid high standard deviations during CIELab measurements, tomato halves were measured from skin side which was the opposite side of interior (eliminating full of seeds and to have non uniform color structure). There were 20 halves of (approximately 100 g) dried tomato per package. 10 halves of tomatoes in each package have been brought together, skin side of the halves faced upwards and flattened by hand pressing with a petri dish. Then, measurement was taken from the skin side of each 10 halves of flattened dried tomato for every experimental set.

Two packages of each storage condition were measured as duplicate and one measurement for each halve was the average of

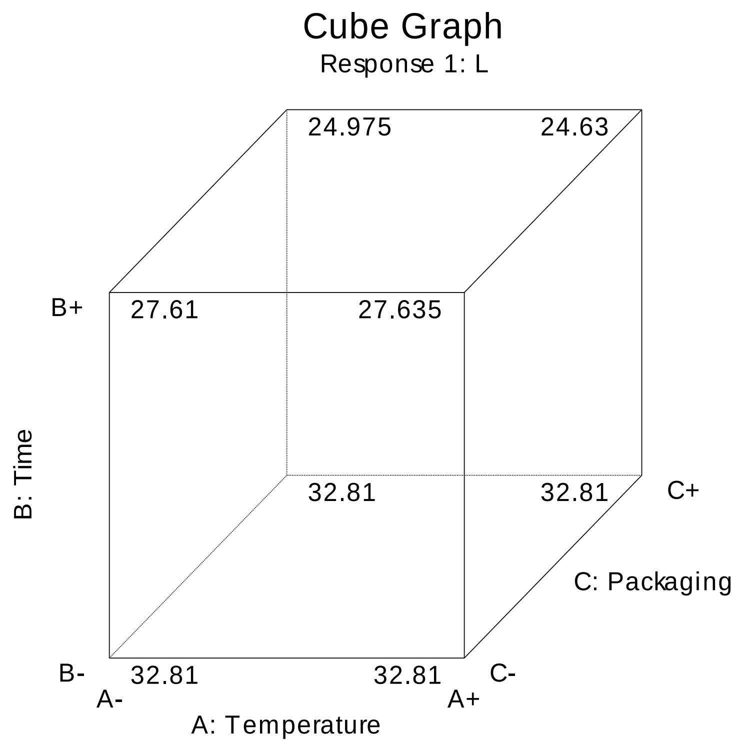

Figure 1. Cube plot showing the predicted values from the coded model for the combinations of the –1 and +1 levels of any three factors for response 1: L.

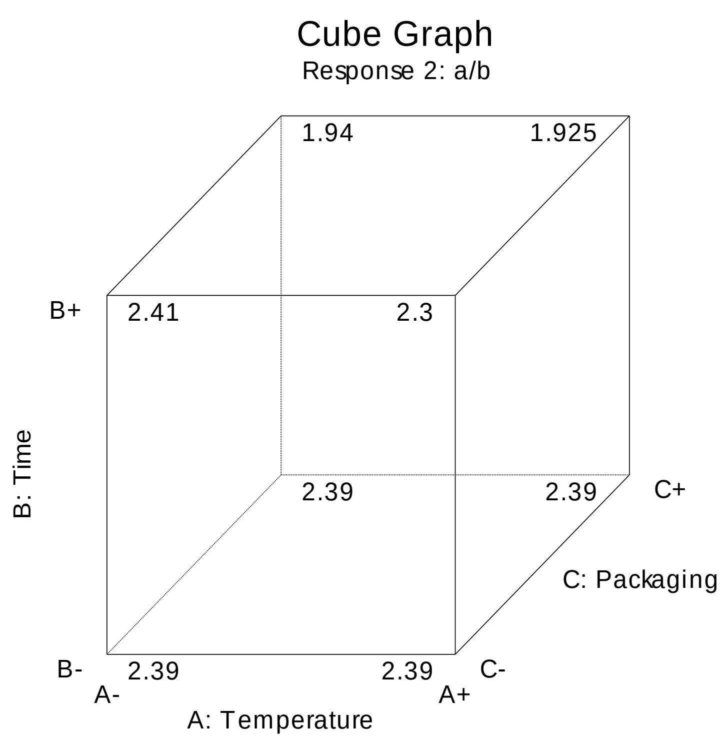

Figure 2. Cube plot showing the predicted values from the coded model for the combinations of the –1 and +1 levels of any three factors for response 2:a/b.

Table 3. Analyses of variance for the factorial model (Response 1: L value).

Source	Sum of squares	DF	Mean square	F value	Prob > F
Model	190.1325	7	27.16178	22873.08	< 0.0001
A	0.0256	1	0.0256	21.55789	0.0017
B	174.108	1	174.108	146617.3	< 0.0001
C	7.9524	1	7.9524	6696.758	< 0.0001
AB	0.0256	1	0.0256	21.55789	0.0017
AC	0.034225	1	0.034225	28.82105	0.0007
BC	7.9524	1	7.9524	6696.758	< 0.0001
ABC	0.034225	1	0.034225	28.82105	0.0007
Pure error	0.0095	8	0.001188		
Cor total	190.142	15			

Table 4. Analyses of variance for the factorial model (response 2: a/b value).

Source	Sum of squares	DF	Mean square	F value	Prob > F
Model	0.611894	7	0.087413	559.4457	< 0.0001
A	0.003906	1	0.003906	25	0.0011
B	0.242556	1	0.242556	1552.36	< 0.0001
C	0.178506	1	0.178506	1142.44	< 0.0001
AB	0.003906	1	0.003906	25	0.0011
AC	0.002256	1	0.002256	14.44	0.0052
BC	0.178506	1	0.178506	1142.44	< 0.0001
ABC	0.002256	1	0.002256	14.44	0.0052
Pure Error	0.00125	8	0.000156		
Cor Total	0.613144	15			

5 flashes of chromameter.

RESULTS AND DISCUSSION

Experimental factorial design for color changes

Experimental factorial design is a cost-effective method with a minimum number of trials (Yann et al., 2005). Using experimental factorial design, mathematical model was set to analyze the effects of storage conditions to retain color quality of sun-dried tomatoes. Factors for the storage conditions were represented as A, B and C in the model and they were storage temperature, storage time and packaging type, respectively. The results of ANOVA for two color measurements were given in Tables 3 and 4. ANOVA results indicated that two model were both significant with the F values of 22873.08 and 559.45. P value is the lowest level of significance and it is important for the rejection of the null hypothesis (Abdel-Ghani et al., 2009; Montgomery, 2001). Results showed that all of the three factors in both models were found to be statistically significant at $P<0.05$ in 95% confidence interval. The values of "Prob > F" for each factor were found less than 0.05. This result indicated that model terms A, B, C, AB, AC, BC and ABC in both models were all statistically significant.

Fitted regression models for the responses L and a/b was expressed in the following equations:

$$\text{Response 1 (L)} = 29.51 - 0.004\,A - 3.3\,B - 0.71\,C - 0.04\,AB - 0.46\,AC - 0.7\,BC - 0.046\,ABC \qquad (2)$$

$$\text{Response 2 (a/b)} = 2.27 - 0.016\,A - 0.12\,B - 0.11\,C - 0.016\,AB + 0.12\,AC - 0.11\,BC + 0.012\,ABC \qquad (3)$$

These functions described the effect of experimental variables and their interactions on the response (color) (Bingol et al., 2010; Lundstedt et al., 1998; Katsoni et al., 2008; Yann et al., 2005). Results showed that all three factors in both models had negative effect on color values. This result revealed that increase in storage time, increase of the amount of air (oxygen) in packaging material and increase in the storage temperature of sun-dried tomatoes reduced the L and a/b values. Lycopene is the main coloring agent for the red color of the tomatoes (D'Sousa et al., 2008; Lavelli and Scarafoni, 2011). It was reported that Hunter L*, a* and b* values of

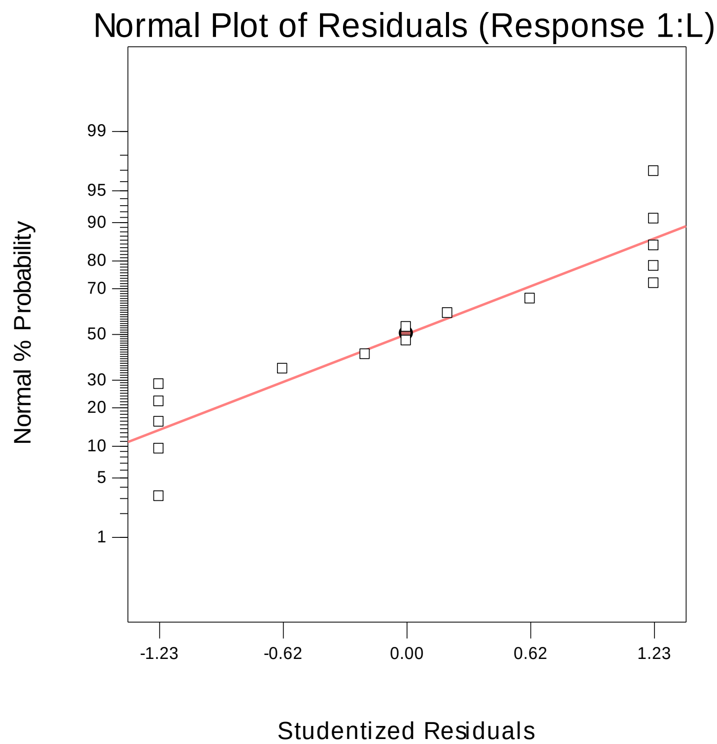

Figure 3. Normality plot of residuals for response 1:L.

the tomato surface were related with tomato lycopene concentration where a*/b* ratios were used to predict the lycopene content (D'Souza et al., 1992; Arias et al., 2000). Color reductions in sun-dried tomatoes might be due to the isomerization and autoxidation of lycopene (Anguelova and Warthesen, 2000). Additionally, that color changes might be attributed to non-enzymatic browning or Maillard reactions (Cernîşev, 2010).

In both models, storage time was found to be the most effective factor on color of sun-dried tomatoes. Its effect was easily observed in the model with its highest coefficient and especially, it was more effective on L value (with coefficient of -3.3). Since it has negative correlation coefficient, increase in storage time reduced both L and a/b values. Similar results were found by different researches. For example, Shi et al. (2008) found that the value of a*/b* decreased significantly in tomato puree during 12 day and Sharma and Le Maguer (1996) showed that lycopene loss was about 76% in dried tomato samples after 4 months of storage at room temperature. Baloch et al. (1997) found that carotenoid loss was above 50% after 20 days storage in a dark place at 40°C. Highest effect of storage time on color was followed by packaging type and storage temperature in both models. It was important to note that effect of temperature was found to be very low compared to other factors (-0.004 and -0.016 for the response L and a/b, respectively). Results showed that color change was almost preserved at the range of +4 and 20°C where similar results were reported by Anguelova and Warthesen (2000). They found that stability of lycopene in tomato powders was not significantly affected by the increase in storage temperature from 6°C to room temperature. Thus, design model pointed out that storage temperature might be kept around 20°C instead of lower temperatures to protect color quality which was also important in economical point of view.

In both models, interactive effects of storage time-packaging type (BC) was the most effective factor on both L and a/b values as they have higher interaction coefficients. Other interactive factors of AB and ABC on a/b values have positive coefficients. However, their numeric values were substantially low. It was important to remind that in ANOVA, all model parameters were found as significant and they were included in the model. Although they could be considered numerically as non-significant for the model, no reduced model was designed by ignoring those terms; they kept in the model to investigate the effect of all terms (factors) on response. A normal probability plot of the residuals were shown in Figures 3 to 4 and they were used to determine either the data was distributed normally or not (Safa and Bhatti, 2011). Plots indicated that residuals reasonably aligned suggesting a normal distribution. R^2 values using sum of squares (SS_{model}/SS_{total}) were calculated and they were found as 0.99 for both responses. Thus, results showed

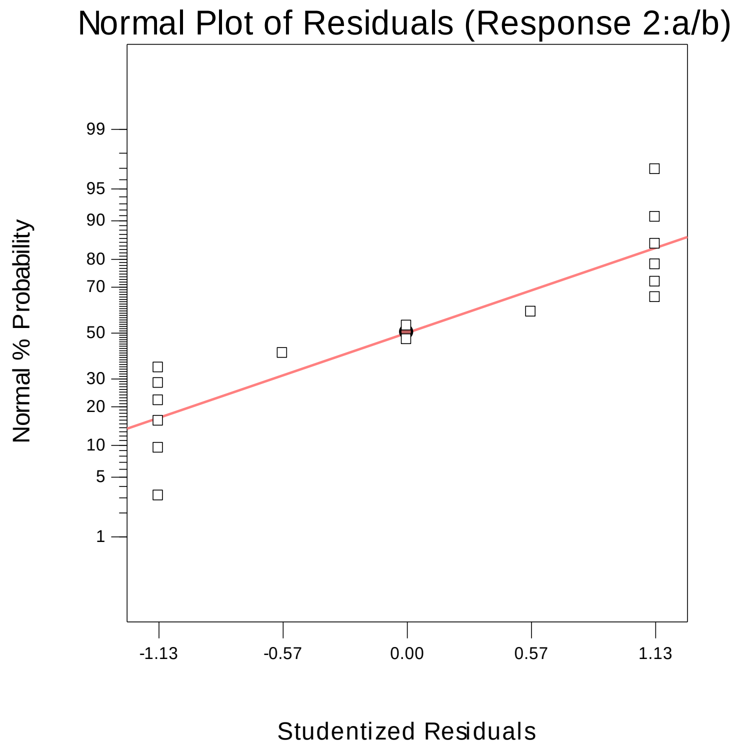

Figure 4. Normality plot of residuals for response 2: a/b.

that selected model was considered to be adequate to describe the observed data (Montgomery, 2001; Abdel-Ghani et al., 2009).

Factor effects

Perturbation plots were given to compare the effect of all three factors at mid point (zero) as reference point (Figures 5 and 6). Thus, responses L and a/b were given by changing one factor while the other two were kept constant similar to single factor effect. Those plots showed that storage time (B) was the main effective factor on color L and a/b values. Very high change with factor (B) was observed especially in response 1 (L) with a steep slope (Figure 5) which indicated that L (brightness) of the samples was very highly sensitive to the storage time. After factor B, second effective factor on color values was the type of packaging. Oxygen plays an important role on non-enzymatic or enzymatic browning of fruit and vegetable products (Cemeroğlu and Acar, 1986). Therefore, air in the package might have accelerated the color changes in dried tomatoes due to the presence of higher oxygen in the packaging material. Similarly, Wong and Bohart (1957) showed that air-packed samples had the lowest lycopene levels and lycopene was significantly loss throughout the storage period all air-packed samples. With careful selection of storage conditions in experimental design such as storing under vacuum, it was possible to retain initial color levels.

Color sensitivity was found to be the lowest with the factor temperature where more flat lines with factor A was obtained compared to the other two factors. Results were also in good agreement with the experimental model.

3-D response plots

Response surface plots were widely used to graphically represent the various combinations of factors and main effects of the factors (Cojocaru et al., 2007). Response graphs (Figures 7 to 12) for color values were plotted and variations of color values for storage temperature, time and packaging type were presented. In Figures 7 and 9, response surface plots were nearly plane and contours had nearly parallel straight lines due to the mathematically low interaction coefficients. On contrary, twists in all response surface graphs were easily observed especially with interaction terms having higher coefficients in mathematical model. Referring to the 3D graph of combination effect, results showed that time played most significant effect towards color rather than temperature and packaging. Surface results indicated that higher the storage time was, the lower L and a/b values. Similarly, higher the air (oxygen) in packaging was, the lower L and a/b values were obtained. These

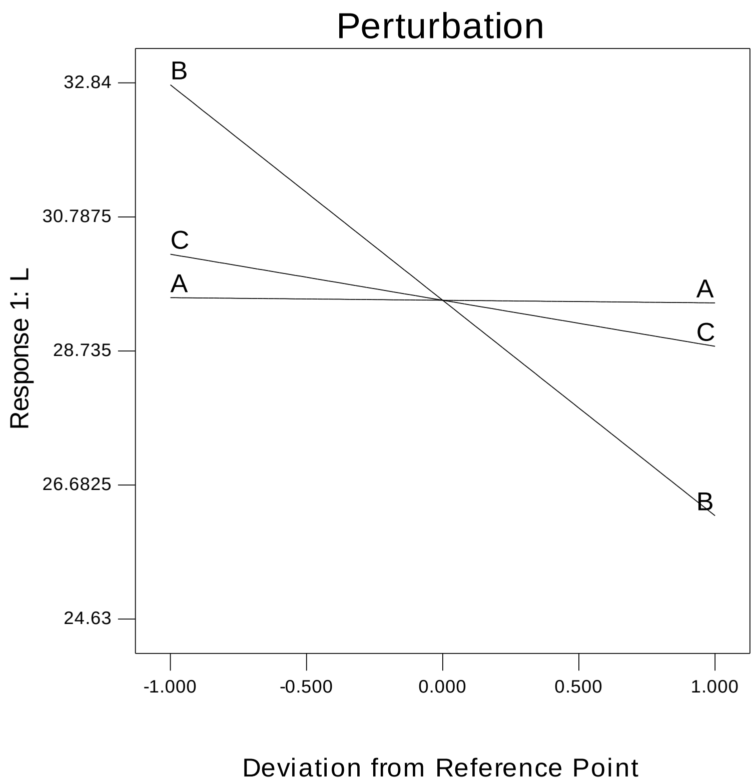

Figure 5. Perturbation plots for the factor effects (A: temperature, B: time, C: packaging type on response 1: L).

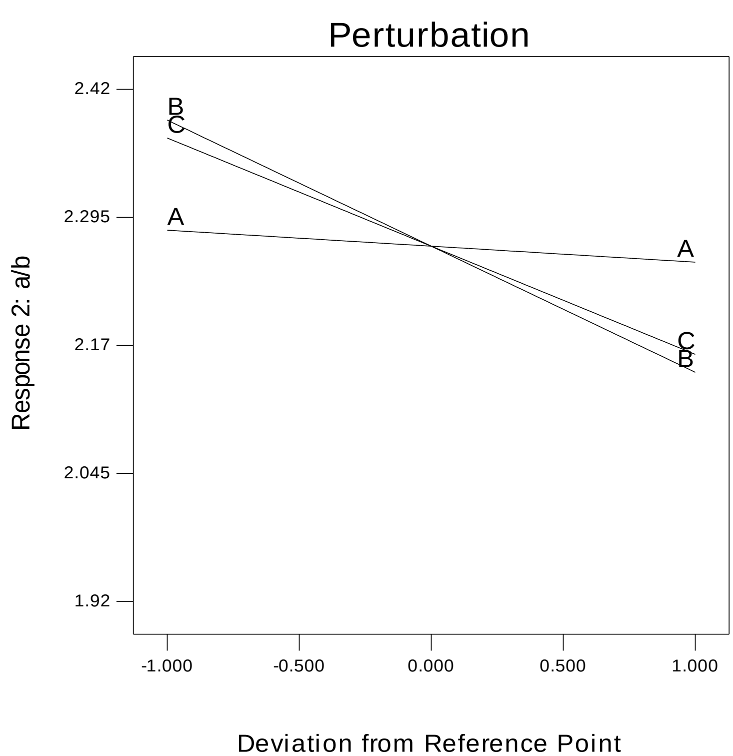

Figure 6. Perturbation plots for the factor effects (A: temperature, B: time, C: packaging type on response 2: a/b).

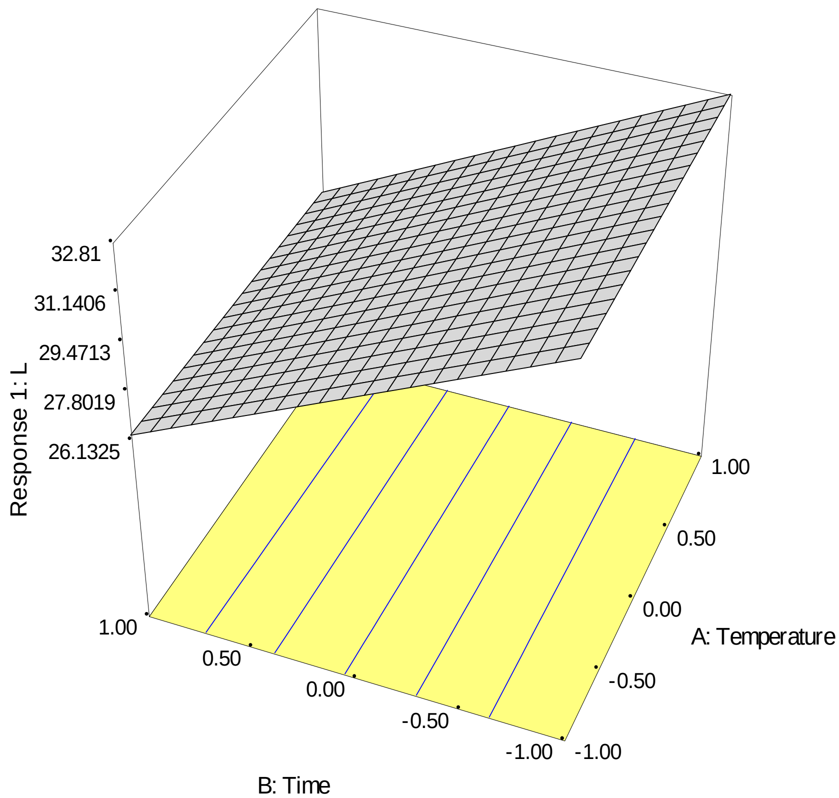

Figure 7. Surface plot for AB for response 1: L.

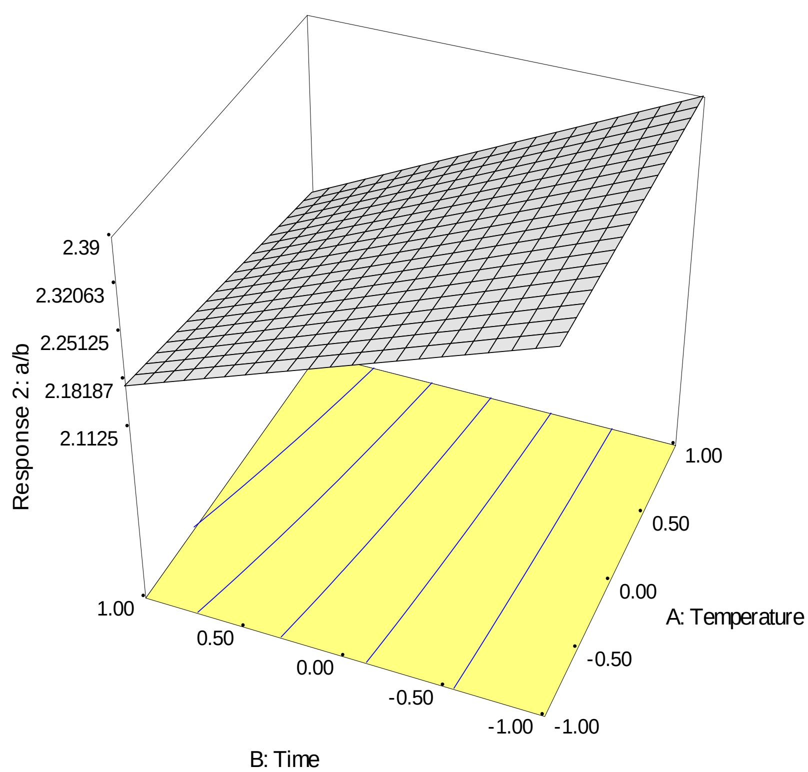

Figure 8. Surface plot for AB for response 2: a/b.

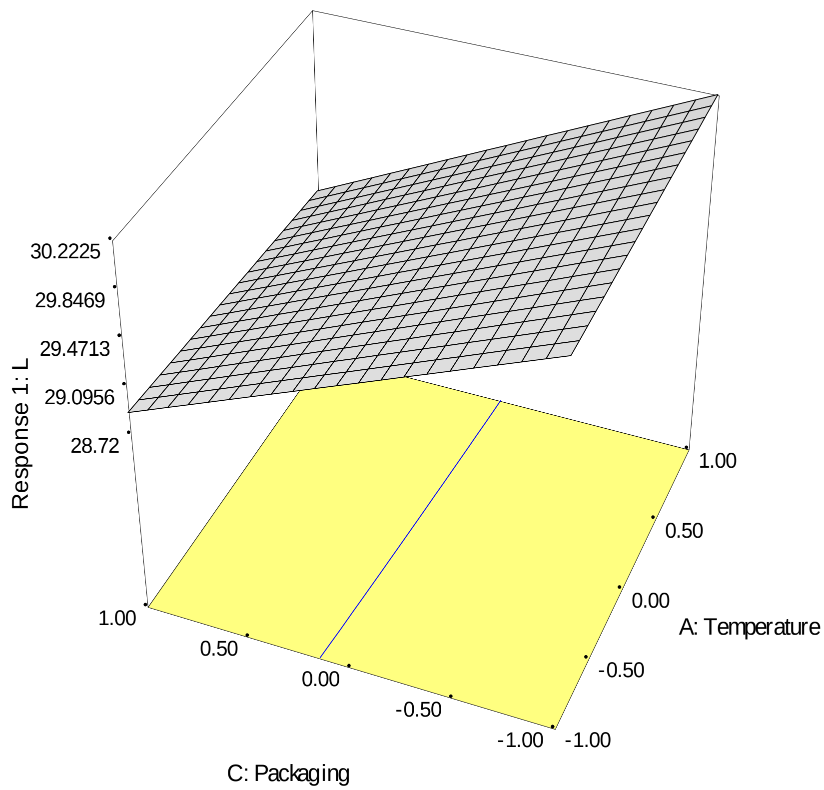

Figure 9. Surface plot for AC for response 1: L.

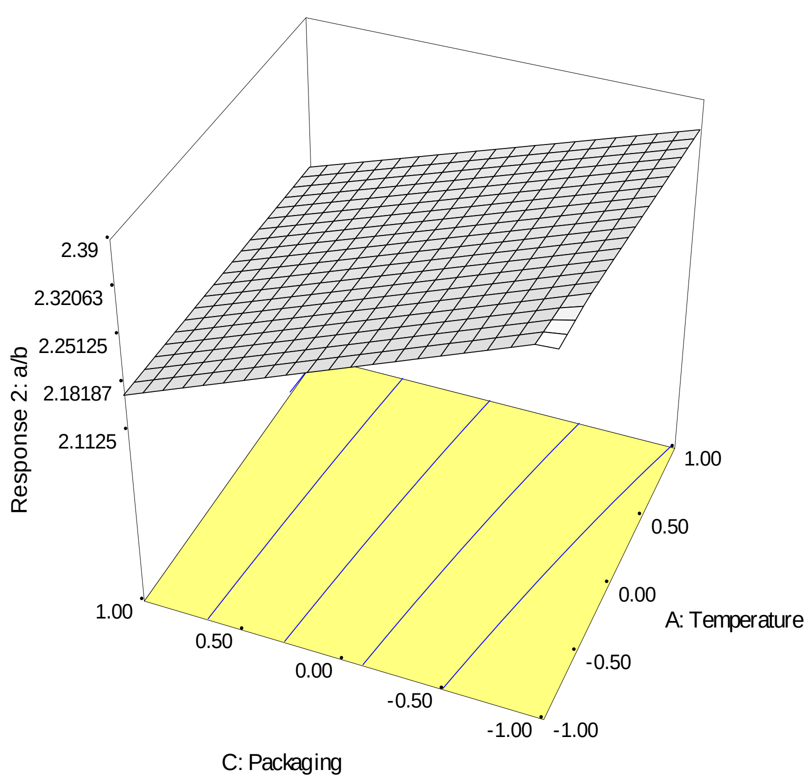

Figure 10. Surface plot for AC for response 2: a/b.

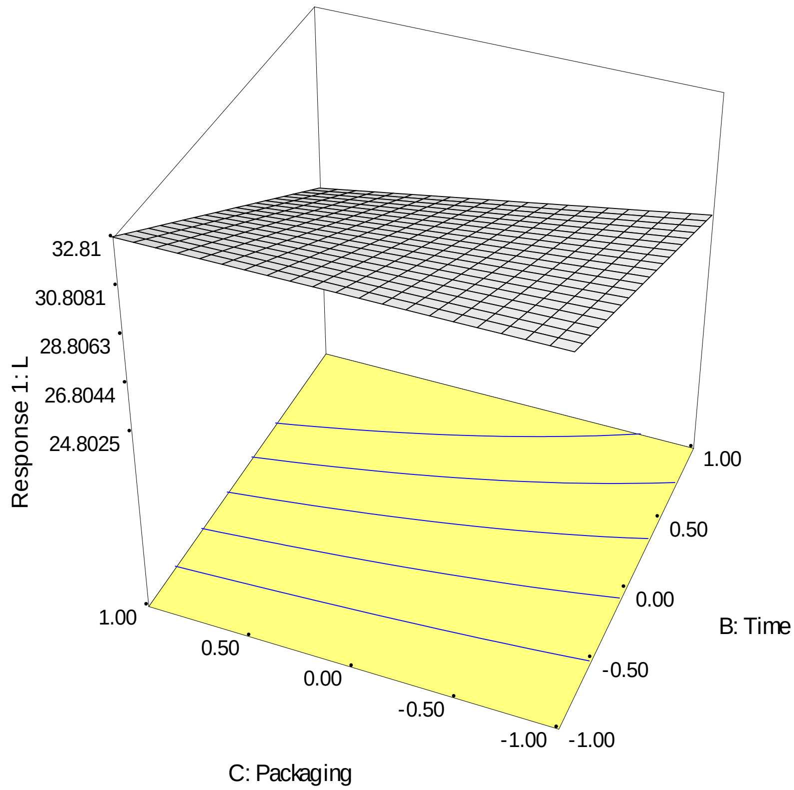

Figure 11. Surface plot for BC for response 1: L.

results were consistent with the coefficients in the statistical model. Results of 3D modeling and their contours easily represented the direction of the sensitivity of color changes. Results showed that higher storage temperatures (in the experimental working range of 4 to 20°C) would be effective to retain colors of sun-dried tomatoes up to 9 months of storage where it was used combined with vacuum-packaging. However, sharp twists were observed when (air) oxygen in packaging material was increased (Figure 12) and curved contour lines were observed due the high interaction effects (Bingol et al., 2010; Montgomery, 2001).

Results indicated that color quality was significantly and negatively affected during storage where contours of the response plots showed the direction of sensitivity of color changes.

Conclusion

The 2^3 factorial design was used to investigate the effect storage time, storage temperature and packaging type on color stability of sun-dried tomatoes. Experimental model indicated the fact that sun-dried tomato products are very sensitive to color fading during storage which was mostly due to lycopene isomerization, oxidation and non-enzymatic browning. Results showed that all three factors negatively affected the color values of both L and a/b. The main effect was found to be the storage time (B) with the highest coefficient of -3.3 and -0.12 in the mathematical models for L and a/b, respectively. Second effective factor was the packaging type where results revealed that air in the package accelerated the color changes in dried tomatoes due to the presence of higher oxygen in the packaging material. However, effect of temperature was found to be the least effective factor and color change was almost retained when temperature was increased from 4 to 20°C. Results from this study indicated that with careful selection of the storage conditions, color quality could be retained.

Design model and 3D visualization pointed out that storage temperature could be kept around 20°C during 9 months and storing under vacuum packaging should be selected to reduce color losses.

ACKNOWLEDGEMENTS

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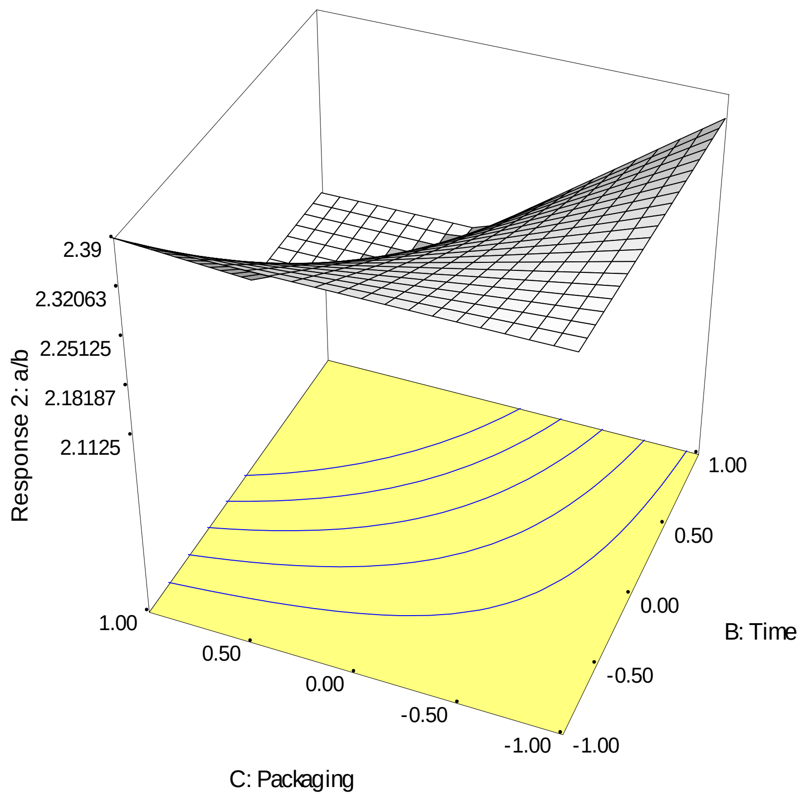

Figure 12. Surface plot for BC for response 2: a/b.

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