Lycopene degradation and isomerization in tomato dehydration

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Abstract

Lycopene is an important nutrient, since it appears to provide protection against a broad range of epithelial cancers. Tomatoes and tomato products are the major source of lycopene, and are considered to be an important source of carotenoids in the human diet. Biodegradation of lycopene not only affects the attractive color of the final products, but also their nutritive value. The main cause of lycopene degradation in tomato dehydration is isomerization and oxidation. The objectives of this study were to determine the retention of total lycopene and isomerization in different dehydration methods, and to optimize processing technology for the retention of lycopene biological potency in the tomato products. Experiments were carried out to compare the effect of osmotic treatment, vacuum-drying, air-drying and their combination on the retention of lycopene bioactivity. Firstly a skin treatment was applied to the tomatoes, following an osmotic treatment at 25°C in 65°Brix sucrose solution for 4 h, then vacuum-drying at 55°C for 4–8 h, or air-drying at 95°C for 6–10 h. In the fresh tomato samples, lycopene content is 75.5 μg/100 g on dry weight basis. Lycopene occurs in nature primarily in the more stable all-*trans* form. A significant increase in the *cis*-isomers with simultaneous decrease in the all-*trans* isomers can be observed in the dehydrated tomato samples in the different dehydration methods. The *cis*-isomers increased with temperature and processing time. In the osmotic treatment, the predominating mechanism is isomerization of lycopene. Since the total lycopene content remained essentially constant, but the distribution of *trans*- and *cis*-isomers changed. In the air-drying processing, isomerization and oxidation (autoxidation) as two strong factors affected simultaneously the decrease of total lycopene content, distribution of *trans*-and *cis*-isomers, and biological potency. A possible explanation of this result is that sugar enters the tomato matrix and strengthen the binding force on lycopene in the tomato matrix. Osmotic solution (sugar) remaining on the surface layer of the tomato prevents oxygen from penetrating and oxidizing lycopene. The osmotic treatment could reduce lycopene losses in comparison with other dehydration methods. © 1999 Canadian Institute of Food Science and Technology. Published by Elsevier Science Ltd. All rights reserved.

1. Introduction

Lycopene is the principal pigment found in tomatoes and is responsible for the characteristic deep-red color of ripe tomato fruits and tomato products. Lycopene is the most abundant carotenoid in tomatoes, and represent about 83% of the total pigments present (Gould, 1992). Tomatoes and tomato products are the major source of lycopene, and are considered to be an important contributor of carotenoids to the human diet. The wide-spread use of tomato paste as a colorant makes lycopene a commercially important natural pigment.

Recently there has been growing interest in the ability of lycopene to act as a cancer-preventative agent.

Although it has no provitamin A activity, lycopene is able to function as an antioxidant and to quench singlet oxygen in vitro. The quenching constant of lycopene was found to be more than double that of β-carotene and 10 times more than that of α-tocopherol, which makes its presence in the diet of considerable interest (Di Mascio, Murphy & Sies, 1991; Ribaya-Mercado, Garmyn, Gilchrest & Russel, 1995). Levy et al. (1995) studied the inhibitory effect of lycopene comparing it with that of α-and β-carotene on the growth of several human cancer cells and found lycopene to be the most potent inhibitor. Lycopene can be considered an important nutrient, since it appears to provide protection against lung cancer and a broad range of epithelial cancers at other sites such as the digestive tract, pancreas and bladder (Gerster, 1997; Levy et al.; Micozzi, Beecher, Taylor & Knachik, 1990). The number of servings of lycopene-rich food, such as tomatoes, tomato sauce,

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and pizza, significantly correlated with a low risk for prostate cancer (Giovannucci et al., 1995).

Deep-red fresh tomato fruits are considered to contain high concentrations of lycopene. Processing conditions such as high temperature, long processing time, light, and oxygen have been shown to have effects on lycopene degradation. Degradation of lycopene not only affects the attractive color of the final products, but also their nutritive value. Losses of lycopene during the dehydration of tomatoes are of commercial significance. Although processing of tomatoes by cooking, freezing, or canning does not usually cause significant changes in the total lycopene content, in the conventional processing of tomatoes, much of the bioactivity of lycopene can be lost due to the conversion of the all-*trans*-isomers to *cis*-isomers which have lower bioactivity and are susceptible to oxidation. These changes are mainly due to heat stress imposed by the relatively harsh thermal process required to achieve the shelf-life stability of processed tomato products. Coupled with exposure to oxygen and light, heat treatment which disintegrates tomato tissue can result in destruction of lycopene (Cole & Kapur, 1957). Heat, light, acids and other factors have been reported to cause isomerization of lycopene (Nguyen & Schwartz, 1998; Schierle et al., 1997). It is generally accepted that the all-*trans*-isomers have the highest bioactivity and the *cis*-isomers have the lowest bioactivity. Bioactivity potency is dependent on the extent of biodegradation due to isomerization and oxidation. Thus, the isomerization can lead to a reduction of lycopene bioactivity. It is generally assumed that lycopene in fresh tomatoes are in the all-*trans* form, and isomerized from all-*trans*- to *cis*-isomers of less bioactivity when processed tomato products are subjected to processing (Emenhiser, Sander & Schwartz, 1995; Khachik, Beecher, Goli, Luby & Smith, 1992; Sies, Stahl & Sundquist, 1992; Stahl & Sies, 1996; Wilberg & Rodriguez-Amaya, 1995). A true assessment of the nutritional quality of dehydrated tomatoes depends not only on the total lycopene content but on the distribution of lycopene isomerization. The characterization and quantification of isomers would be desirable to more accurately assess the bioactivity than just the total lycopene content with no knowledge of its isomeric composition. However, actual determination of the degree of isomerization resulting from processing tomatoes are very few. The color changes, usually used as a quality index, cannot be explained by a change in lycopene content nor by isomerization to *cis*-isomers. In the dehydration procedure, oxidation of lycopene is complex and depends on many factors such as moisture, temperature, presence of pro- and anti-oxidants, and lipids. This study was designed to define the changes of lycopene content and to determine the distribution of the all-*trans*- and *cis*-isomers in the tomato products in the different dehydration methods and steps for retention of lycopene bioactivity in tomato products.

2. Material and methods

2.1. Material

Mature and firm tomatoes, *Lycopersicum esculentum*, var. *Heinz* 9478, were obtained from the Greenhouse and Processing Crop Research Center, Agriculture and Agri-Food Canada, Harrow, Ontario, and stored at 5°C before use. Damaged and over-mature fruits were discarded.

2.2. Methods

2.2.1. Skin treatment

Tomatoes were perforated with a set of fine needles to create pin holes on the tomato surface. The pin hole density was 20 holes/cm^2 (Shi, Maguer, Wang & Liptay, 1998).

2.2.2. Dehydration treatments

Two kinds of samples, intermediate moisture (IM) and dried samples were prepared. After skin treatments, the dehydration treatments of the ripe tomatoes were designed as following in the three methods: (a) tomatoes by conventional air-drying at 95°C for 6–10 h; (b) tomatoes by vacuum-drying at 55°C for 4–8 h; (c) tomatoes were first dehydrated with an osmotic treatment at 25°C in 65°Brix sucrose solution for 4 h, followed by vacuum-drying at 55°C for 4–8 h. The final moisture content of IM and dried samples were 50–55% and 3–4%, respectively.

2.2.3. Sample preparation for HPLC analysis

Five tomatoes were selected and blended into puree in a Waring Blendor for 3 min. The puree was homogenized with Poytron (PT2000, Kinematica Ag, Littau, Switzerland). Ten grams of reconstituted tomato puree (8–9° Brix) were precisely weighted and transferred into a 125 ml flask. Flasks were wrapped with aluminum foil to exclude light. One hundred millilitre of hexane–acetone–ethanol solution (2:1:1 v/v/v) was added into the flask to solubilize lycopene, which was agitated for 10 min on a wrist action shaker until the lycopene is completely extracted. Further, 15 ml water was added followed by another 5 min on the shaker. The solution was separated into 65 ml polar and 55 ml non-polar layers, which was then followed by vacuum filtration through 0.22 μm filter paper. The upper hexane layer was collected for HPLC analysis. The entire procedure was performed in dim light. All extracts were stored in the freezer at −20°C before HPLC analysis.

2.2.4. Instrument and chromatography

The content of lycopene in fresh and processed tomato samples were determined by high performance liquid

chromatography (HPLC). HPLC was performed with a Waters 600E System Controller incorporating a Waters Associates Model 700 Satellite WISP, a Waters Model U6K injector, a Model 410 differential refractometer detector, a Waters Associates Model 486 Tunable Absorbency ultraviolet detector (Spectra-Physics) set at 254 nm, and a Fisher Revordall Series 5000 recorder (Millipore Co. Milford, ON, Canada).

Each sample was analyzed in triplicate. Analyses were performed under dim light to prevent sample degradation by photooxidation. HPLC analysis of lycopene was done with an analytical 3-µm polymeric C_{30} column (C_{30} isocratic separation 4.6 mm i.d.×250 mm, S-5) (YMC, Inc. Wilmington, NC, USA). A mobile phase of methanol methyl-butyl ether (MTBE)(62:38 v/v) was used at a flow of 1 ml/min. Prior to HPLC analysis, the mobile phase was degassed by a supersonic bath and filtered through a 0.22 µm filter paper. A sample volume of 30 µl was injected into the column. The detector had 460 nm absorption to record the maximum of lycopene in this mobile phase. The column temperature and mobile phase were maintained at 25°C.

2.3. Standard solution

All-*trans*-lycopene standard were purchased from Sigma Chemical Co. (St. Louis. MO). All reagents were of HPLC grade. Before chemical analyses, the standard solutions were filtered on Millex GV 0.22 µm (Millipore); the buffer solution was filtered on a Millipore membrane filter HA 0.45 µm. Double distilled deionized water was used to prepare solutions. Four concentrations of lycopene standard in the range 0.25–2 mg/ml were injected for quantification by peak area to determine lycopene content. Lycopene content in tomato samples were identified by comparing peak retention time. The retention time of lycopene averaged 35 min. Quantification was accomplished by determination of the areas under the chromatographic peak and calculation of the level of each component on the basis of standard curves generated with pure compounds (Emenhiser et al., 1995).

2.4. Color parameter measurement

The color parameter measurement of fresh and dehydrated tomatoes was determined by direct reading with a Minolta Chroma meter (CR200, Minolta, Japan). The tomato puree samples were carefully placed in a special black cup covered with optical glass for color measurement. The instrument had an area of view of 25.4 mm and was used with a D65 illuminant as a reference at an observation angle of 10°. The CIF lab color parameters were calculated from the reflectance data: luminance (L^*), red saturation index (a^*) and yellow saturation index (b^*).

3. Results and discussions

3.1. The effect of dehydration techniques on lycopene retention

Total lycopene content in the fresh and dehydrated tomatoes is shown in Table 1. The dehydration of tomato slices is typically carried out at high temperature over an extended period under vacuum. The general tendency of lycopene retention in samples slightly decreased during the dehydration processes. During osmotic dehydration, lycopene content remained essential constant. After osmotic-vacuum drying, total lycopene retention in tomatoes was greater than those using vacuum-drying. A probable explanation is that the sugar solution keeps oxygen from the tomatoes and reduces the oxidation of lycopene in the tomato tissue matrix at low operating temperature. Conventional air drying decreases lycopene retention greatly in tomato samples. This was attributed to the influence of heat and oxygen. Heat treatment disintegrated tomato tissue and increased exposure to oxygen and light, which resulted in the destruction of lycopene.

3.2. Influence of dehydration on lycopene isomerization

The distribution of isomers in the different dehydrated tomato samples are shown in Table 1. In the fresh tomato samples, lycopene content is 75.52 µg/g on a dry weight basis. *Cis*-isomers were not detected in the fresh tomato samples. Lycopene occurs in nature primarily in the more stable all-*trans* configuration (Zechmeister, 1962). Chandler and Schwartz (1978), Rodriguez-Amaya and Tavares (1992) also reported no *cis*-isomers in the fresh tomato samples after HPLC analysis. The *cis*-isomers appeared in processed tomato samples (Fig. 1). A significant increase in the *cis*-isomers with simultaneous decrease in the all-*trans* isomers was observed in the dehydrated tomato samples through different dehydration methods.

Degradation of lycopene not only affects the attractive color of final products, but also their nutritive value. The main cause of lycopene biodegradation in tomato dehydration is isomerization and oxidation. It is widely presumed that lycopene in general undergoes isomerization with thermal processing. This isomerization resulted in conversion of the all-*trans* isomers to the *cis*-isomers. It was observed that fewer *cis*-isomers were present in osmotically-dehydrated tomatoes as compared to those directly air-dried and vacuum-dried. Tomatoes after osmotic treatment showed very little isomerization. Other dehydration methods, especially in conventional air-drying, produced greater isomerization. The highest amount of *cis*-isomers were found in air-dried tomato samples. This supports the assumption of Miers, Wong, Harris and Dietrich (1958), that the

Table 1
Total lycopene and *cis*-isomer content in the dehydrated tomato samples[a]

Sample	Total lycopene (µg/g dry basis)[b]	Lycopene loss (%)	All-*trans*-isomers (%)	*Cis*-isomers (%)
Fresh tomato	75.5a	0	100	0
Osmotic treatment	75.5a	0	100	0
Osmo-vac dried	73.7b	2.4	93.5	6.5
Vac-dried	73.1c	3.2	89.9	10.1
Air-dried	72.6d	3.9	84.4	16.6

[a] Data presented as means of triplicate determinations.
[b] Means in a column not sharing a common letter (a–d) are significantly different ($p < 0.01$).
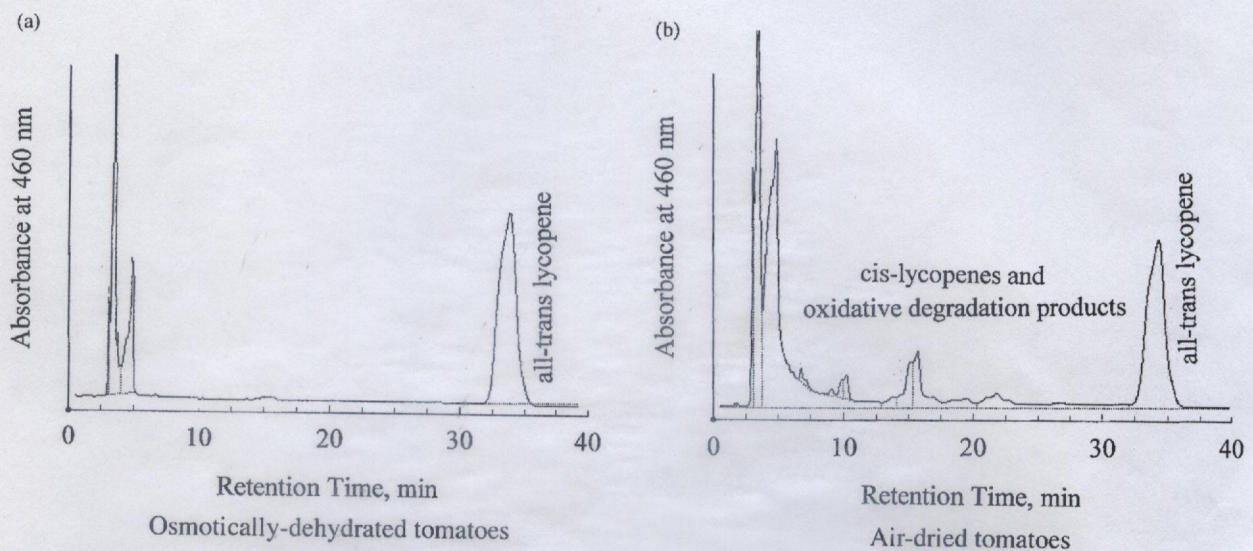

Fig. 1. HPLC chromatograms of *trans*- and *cis*-isomers of lycopene in osmotically-dehydrated tomatoes (a) and in air-dried tomatoes (b).

amount of *cis*-isomers exceeds those present in the initial tomato material and osmotically-treated tomato samples when dehydrated by conventional methods. The *cis*-isomers were formed in processed tomato samples and increased with temperature and time during dehydration. An increase in *cis*-isomers indicates a loss of biopotency of lycopene. Each sample dehydrated by different methods had a negative factor favoring isomerization and /or oxidation of the lycopene, e.g. oxygen permeability, light exposure and presence of some metals in the processing system. A large loss of lycopene during processing would indicate a longer and more drastic procedure, particularly in the thermal dehydration steps. Dehydration of tomatoes at mild temperature does not usually cause significant losses in total lycopene content (Nguyen & Schwartz, 1998), but the conversion of *trans*- to *cis*-isomers always occurred in the dehydrated products. In the osmotic treatment, the predominating mechanism may be isomerization of lycopene. Because the total lycopene content remained almost constant, only the distribution of all-*trans*- and *cis*-isomers was changed. In air-drying, isomerization and oxidation (autoxidation) were two strong factors

that simultaneously affected the total lycopene content, distribution of *trans*- and *cis*-isomers, and biological potency.

It should be pointed out that lycopene is a polyene component having 13 double bonds, of which 11 are conjugated double bonds and seven of which can isomerize from the *trans*-form to the *cis*-form or visa-versa under the influence of heat, light, mechanical action, and other factors. The changes in lycopene content and the distribution of *trans-cis* isomerization will result in a reduction in biological potency, when tomato-based products are subjected to processing (Emenhiser et al., 1995; Khachik et al., 1992; Stahl & Sies, 1996; Wilberg & Rodriguez-Amaya, 1995; Zechmeister, 1962).

Osmotic solution (sugar) remaining on the surface layer of tomato prevents oxygen from penetrating and oxidizing lycopene. A possible explanation of this result is that sugar enters the tomato matrix and strengthens the binding force on lycopene in the tomato matrix. Osmotic treatment could reduce lycopene losses in comparison with other dehydration methods. These results will be useful to develop new dehydration techniques and improve product quality.

3.3. Lycopene degradation and color changes of tomato products

A number of publications have reported the tendency of lycopene compounds to isomerize from one form to another with accompanying color changes (Miers et al., 1958; Wong & Bohart, 1957). Lycopene is located in chromoplasts dispersed throughout the tomato fruits. Lycopene appears as solid microcrystals so that the light reflected from them gives the tomato its typical bright red color. When lycopene is dissolved in lipids or other solvents, its color is yellow or dark orange, but not red. It seems possible that the naturally occurring all-*trans* lycopene isomerizes to the less red partly cis-isomers with a corresponding change in absorption spectra during processing of tomato products (Miers et al.). Color evaluation of whole fresh tomatoes have traditionally been presented as Hunter L*, a*, b* values. Results of color parameters L*, a*, and b* together with the ratio a*/b* and the overall color difference (E) of the dehydrated tomato products are presented in Table 2. Tomatoes with osmotic treatment had more red color than those treated by air-drying and vacuum-drying, which indicated there was more lycopene in the samples. For the overall color difference ΔE,

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

Wiese and Dalmasso (1994) reported an increase in the hue angle of tomato juice after processing and storage, indicating loss of red color. Color retention in tomato products is better at lower temperatures (Sherkat & Luh, 1976; Villari et al., 1994). The main cause of lycopene biodegradation in foods is oxidation. In processed tomato products, oxidation is a complex process and depends upon many factors, such as processing conditions, moisture, oxygen, temperature, light, and the presence of pro- or autoxidants and lipids. The large surface exposed to air and metal enhanced oxidation of pigments of tomato products (Miers et al., 1958). The amount of sugar, acids (pH), and amino acids, as well as time of processing also affected the color of processed tomato products by causing the formulation of brown pigments (Gould, 1992).

From Table 2, a slightly better color can be observed in the samples dehydrated at low temperatures. The color differences between the samples were not readily discernible by visual evaluation. There was no significant difference between Hunter color value a* of different dehydrated tomatoes. This was attributed to the lycopene crystals formation in the tomato tissue matrix after heating in the dehydration processes. Upon heating, the spectrum did not change significantly. But there was a significant difference ($p = 0.01$) in the ratio of all-*trans* to cis-isomers. The color measurement did not show the relative composition of all-*trans* and *cis*-isomers. An increase in *cis*-isomers would indicate a loss in lycopene bioactivity, but would not show up as a significant difference in color. Lycopene content and the ratio of *trans*- to *cis*-isomers may have caused the a*/b* value to stay at a higher level (Wong & Bohart, 1957). The color quality, a*/b* values, remained essentially unchanged during the osmotic treatment, but there were lower values of a*/b* in the conventional air-dried sample. Product color showed progressive deterioration of overall color quality (ΔE) in conventional air-drying.

Yeatman (1969) indicated that the value b*L*/a* provided a high linear correlation with visual color scores of processed tomato products. The overall mean b*L*/a* value for the osmotic treatment, osmo-vac-drying, were 16.42 and 18.34, respectively. The average color reflectance reading for osmotically dehydrated fruits had color characteristics close to those of the fresh material. The L* and a* values decreased in the other dehydration treatment. A comparison of lycopene biodegradation

Table 2
Color values of dehydrated tomato samples[a]

Samples and dehydration condition	Color parameters					
	L*	a*	b*	a*/b*	ΔE	L*b*/a*
Fresh material	38.4a	37.7a	16.1a	2.3a	56.2a	16.4a
IM Tomatos						
Osmotic dehydration at 25°C	38.4a	37.7a	16.1a	2.3a	56.2a	16.4a
Osmo-Vac drying at 55°C	36.7a	35.2a	16.9a	2.1a	53.6a	17.2a
Vacuum-drying at 55°C	34.2a	34.3a	17.4a	1.9a	15.5b	17.4b
Air-drying at 95°C	29.9[b]	33.2b	19.9b	1.7b	48.9c	18.1c
Dehydrated tomatos						
Osmo-Vac drying at 55°C	31.4c	36.4c	18.3b	1.7b	48.1b	18.3c
Vacuum drying at 55°C	28.3d	25.6d	18.2b	1.4b	42.3c	20.1d
Conventional air drying at 90°C	25.6c	23.2e	18.9b	1.2b	39.4d	20.9e

[a] Data presented as means of triplicate determinations.
[b] Means in a column not sharing a common letter (a–e) are significantly different ($p < 0.01$).
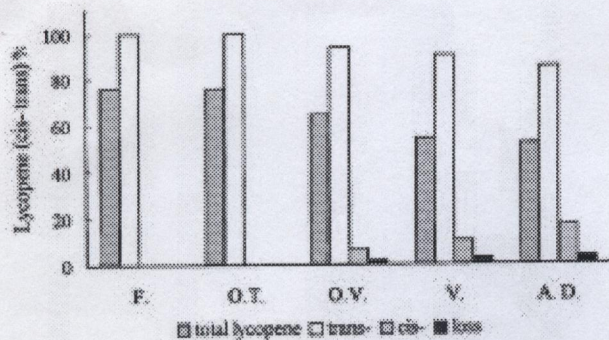
Fig. 2. Comparison of lycopene degradation in the different dehydration processes (F-fresh tomato, O.T.-osmotic treatment, O.V.-osmo-vacuum-drying, V-vacuum-drying, A.D.- air-drying).
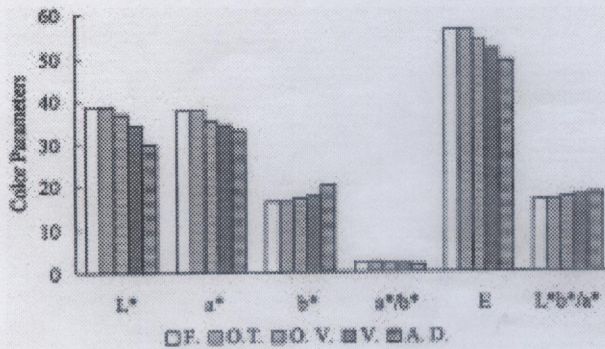

Fig. 3. Comparison of color changes in the different dehysdration processes (F-fresh tomato, O.T.-osmotic treatment, O.V.-osmo-vacuum-drying, V-vacuum-drying, A.D.- air-drying).

tendency and color parameters in the different dehydrated tomato products are shown in Figs. 2 and 3. The change tendencies are not parallel. Tomatoes with osmotic treatment had more red color than those treated by air-drying and vacuum-drying, which indicated there was more *trans*-isomer lycopene in the samples. Osmotically-dehydrated tomatoes appeared to be a promising, though new processing technique to keep the fresh natural reddish color.

4. Conclusions

Conservation of lycopene during tomato processing of tomato products is of commercial significance. Degradation of lycopene not only affects the attractive color of tomato products, but also their nutritive value and flavor. Four dehydration methods produced slight differences in the total lycopene content, but resulted in quite different distribution of the isomer composition. Osmotic treatments retained more total lycopene and induced only slight changes in the distribution of all-*trans*- and *cis*-isomers. The osmotic-vacuum treatment had less effect on lycopene loss and isomerization than vacuum-drying and conventional air-drying. Heat

treatment under atmospheric conditions in the dehydration processes accounts for the lycopene degradation through isomerization and oxidation.

In the osmotic treatment, the predominating mechanism of change may be isomerization of lycopene. Because the total lycopene content remained essentially constant, only the distribution of all-*trans*- and *cis*-isomers was changed. In the air-drying, isomerization and oxidation (autoxidation) were two factors that simultaneously affected the decreased of total lycopene content, distribution of *trans*-and *cis*-isomers, and biological potency. Osmotic treatment could reduce lycopene losses in comparison with other dehydration methods. These results will be useful to develop new dehydration technologies and improve product quality.

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