

Recovery and isomerization of carotenoids from tomato processing by-products

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Abstract

Industrial processing of tomato leads to by-products that represent a major disposal problem for the industry, intended mainly for animal feed or fertilizer. Carotenoids constitute an important component of tomato processing by-products, well credited with important health-promoting functions. This study examined the effect of several organic solvents (hexane, ethanol, acetone, ethyl acetate, ethyl lactate and their mixtures) and extraction temperature on the recovery and isomerization of carotenoids from tomato processing by-products. Another area under investigation was the stability of carotenoids, especially lycopene, as affected by drying and storage conditions of tomato by-products. HPLC-DAD analysis was applied for the efficient separation and analysis of the carotenoids and their *cis*-isomers. The identified carotenoids in all solvent extracts from dry tomato waste followed the order: lycopene > *b*-carotene > lutein, in their predominant *trans*-configuration, whereas 5-*cis* lycopene and 15-*cis-b*-carotene were the most abundant in all solvent extracts. The increase of extraction temperature increased the respective *all-trans* lycopene concentration in all solvent extracts, whereas a steadily high percentage of *cis*-isomer ($\approx 30.4\%$) was observed upon increasing the temperature from 25 °C to 70 °C in ethyl lactate extracts. Conversely, the increase of extraction temperature induced a reduction in total *b*-carotene and total lutein concentration in all solvents, but did not cause any further isomerization. The drying method affected significantly lycopene isomerization with a considerable increase of *cis*-isomers and a simultaneous reduction of *all-trans* configurations. A small percentage of isomerization was observed (2.4 %) during storage at -20 °C for 60 days.

Keywords: carotenoids, lycopene, tomato by-products, isomerization, organic solvent

Introduction

Industrial tomato processing generates large amount of low-value by-products, namely tomato seeds and peels, which represent 10–40% of total processed tomatoes [1, 2] and are primarily used as livestock feed or disposed of. The quantity of by-products generated during tomato processing, combined with the beneficial characteristics of the components of these by-products, justifies the great interest of researchers and manufacturers in extracting carotenoids, and mostly lycopene, from tomato wastes.

Organic solvent extraction is a well established method in the food industry. In a previous work [3] the efficiency of several organic solvents and solvent mixtures to extract carotenoids from dry tomato waste was examined as well as the effect of temperature, time and number of extraction steps. Three extraction steps of 30 min each were sufficient for all solvents, whilst the increase of temperature (from 25 to 70 °C) generally resulted in an increase of carotenoid yield.

Carotenoids predominantly occur in their *all-trans* configurations; however, processing may lead to the formation of *cis*-isomers, which are susceptible to oxidation and may present lower bioactivity than *all-trans* isomers. Bioactivity potency is dependent on the extent of biodegradation due to isomerization and oxidation [4]. The accumulation of lycopene *cis*-isomers was observed in biological fluids and tissues and raised the question as to their bioavailability [5, 6]. *Cis*-lycopene isomers are regarded as being more bioavailable because they are more soluble and better absorbed from the intestinal lumen than is the *all-trans* isomer [7], although the isomerization may also take place *in vivo*. The gastrointestinal lumen [8, 9], liver [10], and enterocytes [11] were identified as potential sites of lycopene isomerization *in vivo*. It is generally accepted that processing, especially heat

treatment, of food enhances lycopene bioavailability; however, the rate of absorption of the different lycopene *cis*-isomers still remains to be determined.

The objective of this study was to examine the extraction efficiency and degree of isomerization of carotenoids as affected by organic solvent and temperature of extraction. Also, to explore the stability of carotenoids, and especially lycopene, when tomato processing by-products are subjected to various drying and storage conditions before extraction for carotenoid recovery.

Materials and Methods

Materials

Tomato processing by-products, composed of skin and seeds, were kindly donated by NOMIKOS tomato-processing factory (Aliartos, Boeotia, Greece). The solvents used for sample preparation and extraction were of analytical grade and were obtained from Merck (Darmstadt, Germany). All solvents used for HPLC analysis (acetonitrile, 1-butanol and methylene chloride) were of HPLC grade and were obtained from Merck (Darmstadt, Germany). *All-trans* carotenoid standards (lycopene, β -carotene, lutein) were purchased from Sigma Chemical Co. (Sigma-Aldrich Company, St. Louis, MO).

Instrumentation

The HPLC (Hewlett Packard Series 1100, Waldbronn, Germany) system was composed of, a HP 1100 Quaternary Pump, an Agilent 1100 Series Micro Vacuum Degasser, a Rheodyne model 7010 Sample Injector, and a HP 1100 Series Diode Array Detector (DAD). It was equipped with a YMC (Tokyo, Japan) C₃₀ column (250x4.6 mm I.D., 5 μ m particle). The analysis of the chromatographic data was

carried out on a ChemStation for LC 3D software (Agilent Technologies 1999–2000, Waldbronn, Germany). A mobile phase of acetonitrile (A), 1-butanol (B), and methylene chloride (C) with the following gradient elution was used: 69.3% A, 29.7% B and 1.0% C, initially; increased to 67.2% A, 28.8% B and 4% C, in the first 10 min; 61.6% A, 26.4% B and 12% C, after 20 min; 49% A, 21% B and 30% C, after 40 min; and returned to 69.3% A, 29.7% B and 1% C, after 50 min. The flow rate was maintained at 2 mL/min and the column temperature at 25 °C.

Drying and storage pre-treatments

Tomato processing by-products were divided in 5 parts; the first part was stored for 60 days at -20 °C, the second part was air dried in open space at 25-30 °C, the third part was dried in an air circulation oven (Heraeus, Function Line UT20) at 70°C, the fourth part was dried in a vacuum oven (Büchivacutherm VT6025, Heraeus Instruments, Hanau, Germany) at 70 °C, and the last part was freeze-dried at -55 °C (Freeze-Dryer Christ Alpha, 1-4 LD Plus). The moisture content of samples was determined according to the AOAC method [12]. Dried samples were homogenized in a domestic blender and ground in a laboratory mill (Type ZM1, Retsch GmbH, Haan, Germany) at 0.5 mm particle size. The dry ground material was kept in glass jars wrapped with aluminium foil at -20 °C until analyzed.

Extraction and analysis of carotenoids

Carotenoids were extracted according to the method described by Strati and Oreopoulou [3], summarized as follows: Homogenized tomato by-products (10.0 g) were stirred with 100 mL of solvent in an extraction vessel, equipped with a vertical water cooler and placed in a water bath. The extraction time was kept constant at 30

min, while temperature varied from 25 to 70 °C. Limiting factor for the choice of the upper temperature value was the boiling point of solvent used. The mixture was vacuumfiltered; the solid residue was collected and re-extracted two more times, with fresh extraction solvent under the same conditions. The extracts were combined and centrifuged at 3000 rpm (HERMLE centrifuge Z380, Gosheim, Germany), for 10 min, to separate the supernatant. Then, the supernatant was evaporated to dryness in a rotary vacuum evaporator (Rotavapor RE 111, Switzerland), dissolved in 1 mL methylene chloride and transferred to a vial. The new solution was filtered through a 0.45 µm membrane filter and 20 µL were injected for HPLC analysis. All the procedure was performed under dimmed light.

The identification and quantification of *trans* and *cis* isomers of carotenoids was performed based on the method proposed by Strati et al. [13], by comparing retention times and absorption spectra with reference standards and absorption spectra characteristics and on the basis of calibration curves of all-*trans* lutein (447 nm), all-*trans*- β -carotene (455 nm) and all-*trans*-lycopene (476 nm), with a minimum of five concentration levels. *Cis* isomers of carotenoids were quantified using the standard curves of the corresponding all-*trans* carotenoids, because of the similarity in extinction coefficient [14]. Duplicate analyses were performed and the mean value was determined. The carotenoids of all samples were quantified on a dry weight basis of tomato by-product.

Results and Discussion

The effect of several organic solvents and solvent mixtures on the recovery and isomerization of carotenoids from air-dried tomato by-product is presented in Table 1. Three major carotenoids were identified in all extracts, in the following order:

lycopene > *b*-carotene > lutein, with the exception of ethanol and acetone extracts, where the total lutein (*all-trans* + *cis* isomers) yield was higher than the respective total *b*-carotene yield (Fig 1)

More specifically, the highest (166.4 µg/g dw) and the lowest (3.8µg/g dw) total lycopene yield were observed in ethyl lactate and ethanol extracts, respectively. In all other solvent extracts, total lycopene ranged from 19.1 to 30.2 µg/g dw, with hexane-ethyl acetate mixture presenting the best results..

Similarly, the highest (26.4 µg/g dw) and the lowest (0.6µg/g dw) total *b*-carotene yield were observed in ethyl lactate and ethanol extracts, respectively. In all other solvent extracts, total *b*-carotene ranged from 3.6 to 7.1µg/g dw, with best results obtained by mixtures of hexane with either ethanol or acetone.

Lutein was not detected in hexane extracts, and presented very low yield in hexane mixtures. Ethyl lactate showed the best results (10.8 µg/g dw), followed by acetone and ethyl acetate.

According to these results, ethyl lactate was the most efficient solvent for the recovery of carotenoids, and especially lycopene and *b*-carotene from tomato processing by-products. High yields from dry tomato powder were also reported by Ishida and Chapman [15]. As regards the rest examined systems, mixtures of hexane with medium or higher polarity solvents presented the best results for the recovery of lycopene and *b*-carotene, in agreement with other reported results [16, 17]. Acetone and ethyl acetate were efficient for the recovery of all three carotenoids, as these solvents penetrate the plant tissue and dissolve easily most carotenoids.

The *all-trans* configuration of lycopene, *b*-carotene, and lutein predominated in all solvent extracts (Table 1). The isomeric configurations of lycopene, 13-*cis*, 9-*cis* and 5-*cis*, were identified and quantified in hexane, ethyl acetate, acetone, hexane-

ethanol and hexane-acetone extracts. Moreover, 15-*cis* lycopene was identified only in hexane-ethyl acetate and ethyl lactate extracts. From all the identified lycopene isomers, 5-*cis*lycopene was the most abundant.

15-*cis* and 9-*cisb*-carotene isomers were detected in all extracts, except that of ethanol. Additionally, the concentration of 15-*cisb*-carotene was found to be almost three fold higher than the respective of 9-*cisb*-carotene in all extracts. Finally, 9-*cis* and 13-*cis* lutein isomers were identified in similar proportions in ethanol, ethyl acetate, acetone, hexane-ethyl acetate and ethyl lactate extracts, while no *cis* isomers were detected in the rest solvents (Table 1).

Table 2 presents the effect of extraction temperature on the yield of lycopene recovered from air dried tomato processing by-products. It is observed that the concentration of total (*all-trans* + *cis*) and *all-trans* lycopene generally increased with the increase of extraction temperature.

A substantial increase of the proportion of *cis*- to total (*all-trans* + *cis*) lycopene was noticed with the increase of extraction temperature in ethanol and acetone. In most of the other solvents, a noticeable increase was observed between 25 °C and 50 °C, while further increase in temperature did not provoke further isomerization. An almost constant and low (approximately 5 %) proportion of *cis*- to total lycopene was observed in hexane-ethyl acetate extract upon increase of extraction temperature from 25 °C to 70 °C. A similar trend was noticed in ethyl lactate extract, but with high *cis*-isomers proportion (≈30 %).

The above results are generally in agreement with the findings of other researchers. Lee and Chen [18] studied the stability of lycopene by heating standard lycopene (dissolved in HPLC grade hexane) at 50 °C, 100 °C, and 150 °C. They observed that at 50 °C, isomerization was the main reaction during the first hours of

heating, after which the degradation reaction dominated. Additionally, Shi et al. [19] showed that at moderate temperatures (60 °C), the *cis*-isomers were formed with a tendency to accumulate, while heating at 80 °C caused a slight decrease in total lycopene extraction. Nevertheless, the isomerization depends on the organic solvents and on the extraction conditions, reaching a high value of *cis*- to total lycopene ratio (47.0 %) with ethanol: hexane (4:3, v/v) as extraction solvent mixture, at 75 °C [20]. Calvo et al. [21] found that by using ethanol at high extraction temperatures, the principal mechanism was oxidative degradation, whereas by using ethyl acetate as extraction solvent, the main mechanism was isomerization. Both mechanisms are affected by the type of solvent and the extraction temperature.

As regards the rest identified carotenoids, a decrease of the concentration of total *b*-carotene and total lutein in all solvents was noticed, as the extraction temperature increased (Table 3). Accordingly, the concentrations of *all-trans* β -carotene and of *all-trans* lutein decreased with the increase of extraction temperature from 25 °C to 70 °C, in all solvent extracts. The *cis*- to total *b*-carotene and lutein ratios ranged from 23.9-39.1 % and from 36.9-48.7 %, respectively, in all extracts at 25 °C; however, the increase of temperature did not cause any further isomerization. It can be concluded that *b*-carotene and lutein isomers which were formed during the extraction process, were unstable and readily degraded. It should be mentioned that in ethyl lactate extracts, a considerable *cis*- to total *b*-carotene ratio (52.6 %) was noticed at 70 °C (Table 3).

The next field under investigation was the influence of drying and storage conditions of fresh tomato processing by-products on the recovery and stability of carotenoids, especially lycopene. The yield and distribution of lycopene isomers in fresh, compared to the different dehydrated tomato processing by-products is shown

in Table 4. The highest lycopene losses (26.3 % and 22.8 %) were observed after oven drying and air drying, respectively, and are possibly related to elevated temperature and to the presence of oxygen. The lowest lycopene loss (10.0 %) was noticed after freeze-drying, whereas vacuum oven drying caused a moderate lycopene loss (16.4 %), compared to fresh tomato processing by-products.

Cis-isomers were detected in trace amounts (0.3 %) in the fresh tomato processing by-products. A significant increase of *cis*-isomers with a simultaneous decrease of *all-trans* configurations was observed in the dehydrated samples through different dehydration methods, implying that the drying method affected significantly lycopene isomerization.

More specifically, the lowest amount of *cis*-isomers (4.5 %) was noticed in freeze-dried tomato by-products, whereas the greatest amount (15.6 %) was found in samples after oven drying (70 °C). The *cis*-isomers determined after dehydration with the other methods (air-drying and vacuum oven drying) were 5.6 % and 8.6 %, respectively.

Lycopene degradation and isomerization which occurs during dehydration is a complex procedure depending on many factors, such as oxygen exposure, moisture, temperature and/or the presence of prooxidant and antioxidant compounds. Our experimental results are close to those of other researchers [4], who compared lycopene degradation in tomatoes after vacuum-drying at 55 °C for 4-8 h, air drying at 95 °C for 6-10 h and osmotic vacuum drying. They found that with the above mentioned dehydration methods, total lycopene content remained essentially constant, but the distribution of *trans*- and *cis*-isomers changed; a significant increase of *cis*-isomers and, at the same time, a decrease of *all-trans* isomers was observed in dehydrated tomato samples through the dehydration methods used (osmotic vacuum

drying, vacuum-drying and air drying). Another study [22] examined the loss of lycopene after air drying at 80 °C and 110 °C and they concluded that drying at 80 °C did not cause any considerable loss of lycopene, whereas the maximum loss of lycopene (12 %) occurred at 110 °C. According to Anguelova and Warthesen [23] (2000), in fresh tomatoes *cis*-isomers of lycopene were detected in trace amounts, which were subsequently increased after spray drying. Conclusively, the drying process of tomatoes at moderate temperatures did not change significantly total lycopene content; on the other hand, a conversion of *all-trans* to *cis*-isomers of lycopene was observed in the dehydrated tomato products [24].

Finally, the storage of fresh tomato processing by-products at -20 °C for 60 days caused the degradation of lycopene by 13.5 % compared with total lycopene of fresh tomato by-products, while low amount of *cis*-isomers (2.4 %) was recorded. In a kinetic study of degradation of lycopene in tomato paste under different processing and storage conditions [25], it was reported that lycopene loss was observed even when stored in frozen state under vacuum and dark (16.2 % after 60 days), suggesting the possibility of an autocatalytic reaction.

Conclusions

Industrial tomato processing by-products can be valorized through the application of many extraction methods for recovering carotenoids; food technology is challenged to maximize bioavailability, while at the same time minimizing conversion of *all-trans* carotenoids into their *cis*-isomers.

From the results of this study, it can be concluded that the isomerization of carotenoids was affected by the extraction solvent. Ethyl lactate affected significantly the isomerization of lycopene (30.4 %), *b*-carotene (33.0 %) and lutein (44.4 %).

The use of non polar or medium-polarity solvents for the extraction of carotenoids resulted in limited isomerization, even at relatively high extraction temperatures. Especially, in hexane-ethyl acetate extract, an almost steady percentage ($\approx 5\%$) of *cis*-isomers of lycopene was observed, upon increase of the extraction temperature from 25 °C to 70 °C. Similarly, a steady but higher percentage ($\approx 30.4\%$) of *cis*-isomers of lycopene was observed in ethyl lactate extract. Therefore, ethyl lactate gave the highest carotenoid yield among the other solvents used; however, the degree of isomerization was greater.

Moreover, another interesting conclusion was that the carotenoids recovered from fresh tomato processing by-products are maintained relatively stable after industrial processing treatments. The drying method affected significantly lycopene isomerization, with the following order: freeze-drying (4.5 % *cis*-isomers) < air drying (5.6 % *cis*-isomers) < vacuum drying (8.6 % *cis*-isomers) < oven drying (15.6 % *cis*-isomers). There was a significant loss of carotenoids after the different drying methods used and after storage of tomato processing by-products at -20 °C for 60 days.

Conclusively, improved carotenoid yields could be achieved with direct extraction of fresh tomato processing by-products, at the tomato processing plant. If this is not feasible, drying should be performed at low temperatures and, preferably, in the absence of air, to prevent the loss of carotenoids.

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Table 1 Concentration of carotenoids and their isomers (µg/g dw) as affected by organic solvent extraction of air-dried tomato by-products

| SOLVENTS | LYCOPENE (µg/g dw) | | | β-CAROTENE (µg/g dw) | | | LUTEIN(µg/g dw) | | |
|-----------------------------------|---------------------------------|-------------------------------|-------|---------------------------------|-------------------------------|-------|---------------------------------|-------------------------------|-------|
| | <i>Cis-isomers</i> ^a | <i>All-trans</i> ^a | Total | <i>Cis-isomers</i> ^a | <i>All-trans</i> ^a | Total | <i>Cis-isomers</i> ^a | <i>All-trans</i> ^a | Total |
| Ethanol | nd | 3.8±0.0 | 3.8 | nd | 0.6±0.0 | 0.6 | 0.5±0.1 | 0.7±0.0 | 1.2 |
| Hexane | 1.0±0.1 | 19.9±0.2 | 20.9 | 0.9±0.1 | 2.7±0.0 | 3.6 | nd | nd | nd |
| Ethyl acetate | 1.1±0.2 | 21.7±0.2 | 22.8 | 1.1±0.3 | 3.2±0.5 | 4.3 | 1.6±0.2 | 2.1±0.0 | 3.7 |
| Acetone | 1.2±0.1 | 22.6±0.3 | 23.8 | 1.4±0.3 | 2.9±0.1 | 4.3 | 2.3±0.3 | 2.8±0.0 | 5.1 |
| Hexane-ethanol (50:50, v/v) | 1.0±0.1 | 18.1±0.3 | 19.1 | 1.8±0.2 | 5.3±0.3 | 7.1 | nd | 0.8±0.1 | 0.8 |
| Hexane-acetone (50:50, v/v) | 1.1±0.2 | 20.6±0.2 | 21.7 | 1.6±0.3 | 5.1±0.2 | 6.7 | nd | 0.9±0.1 | 0.9 |
| Hexane-ethyl acetate (50:50, v/v) | 1.7±0.2 | 28.5±0.3 | 30.2 | 1.1±0.2 | 3.2±0.2 | 4.3 | 0.7±0.0 | 0.9±0.1 | 1.6 |
| Ethyl lactate | 50.6±0.8 | 115.8±1.9 | 166.4 | 8.7±0.5 | 17.7±0.8 | 26.4 | 4.8±0.2 | 6.0±0.8 | 10.8 |

^aValues are means ± SD (n=2)

nd: not detectable

Extraction conditions: T=25 °C, solvent: dry tomato by-products=10:1 (v/w), particle size=0.5 mm, 3 extraction steps of 30 min each

Table 2 The effect of extraction temperature on the isomerization of lycopene recovered from air dried tomato processing by-products

| Solvents | Temperature (°C) | Lycopene concentration (µg/g dw) | | | |
|--------------------------------------|------------------|----------------------------------|---------------------------------|--|-----------------------|
| | | <i>all-trans</i> ^a | <i>Cis-isomers</i> ^a | Total (<i>all-trans</i> + <i>cis</i>) | <i>Cis</i> /Total (%) |
| Ethanol | 25 | 3.8±0.0 | nd | 3.8 | 0 |
| | 50 | 8.4±0.1 | 0.3±0.1 | 8.7 | 3.4 |
| | 70 | 9.3±0.2 | 2.3±0.1 | 11.6 | 19.8 |
| Hexane | 25 | 19.9±0.2 | 1.0±0.3 | 20.9 | 4.8 |
| | 50 | 20.7±0.2 | 1.3±0.1 | 22.0 | 5.9 |
| | 60 | 26.9±0.4 | 2.0±0.1 | 28.9 | 6.9 |
| Ethyl acetate | 25 | 21.7±0.2 | 1.1±0.2 | 22.8 | 4.8 |
| | 50 | 28.0±0.3 | 2.1±0.2 | 30.1 | 7.0 |
| | 70 | 31.4±0.3 | 2.4±0.3 | 33.8 | 7.1 |
| Acetone | 25 | 22.6±0.3 | 1.2±0.1 | 23.8 | 5.0 |
| | 50 | 23.4±0.4 | 4.9±0.3 | 28.3 | 17.3 |
| Hexane-ethanol (50:50, v/v) | 25 | 18.1±0.3 | 1.0±0.1 | 19.1 | 5.2 |
| | 50 | 20.1±0.2 | 1.7±0.3 | 21.8 | 7.8 |
| | 70 | 23.9±0.2 | 1.8±0.2 | 25.7 | 7.0 |
| Hexane-acetone (50:50, v/v) | 25 | 20.6±0.2 | 1.1±0.3 | 21.7 | 5.1 |
| | 50 | 21.6±0.5 | 2.0±0.4 | 23.6 | 8.5 |
| | 70 | 22.3±0.4 | 1.9±0.2 | 24.2 | 7.9 |
| Hexane-ethyl acetate (50:50, v/v) | 25 | 28.5±0.3 | 1.7±0.2 | 30.2 | 5.6 |
| | 50 | 32.4±0.2 | 1.7±0.3 | 34.1 | 5.0 |
| | 70 | 37.3±0.3 | 1.6±0.3 | 38.9 | 4.1 |
| Ethyl lactate | 25 | 115.8±1.9 | 50.6±0.8 | 166.4 | 30.4 |
| | 50 | 121.2±1.5 | 47.1±0.8 | 168.3 | 28.0 |
| | 70 | 134.4±1.8 | 57.6±0.6 | 192.0 | 30.0 |

^aValues are means ± SD (n=2)

Extraction conditions: solvent: dry tomato by-products=10:1 (v/w), particle size=0.5 mm, 3 extraction steps of 30 min each

Table 3 The effect of extraction temperature on the isomerization of β -carotene and lutein recovered from air dried tomato processing by-products

| SOLVENTS | Temperature(°C) | β -carotene concentration (µg/g) | | | | Lutein concentration (µg/g) | | | |
|-----------------------------|-----------------|--|---------------------------------|---|-----------------------|-------------------------------|---------------------------------|---|-----------------------|
| | | <i>all-trans</i> ^a | <i>cis-isomers</i> ^a | Total (<i>all-trans</i> + <i>cis</i>) | <i>Cis</i> /Total (%) | <i>all-trans</i> ^a | <i>cis-isomers</i> ^a | Total (<i>all-trans</i> + <i>cis</i>) | <i>Cis</i> /Total (%) |
| Ethanol | 25 | 0.6±0.0 | 0.3±0.0 | 0.9 | 33.3 | 0.7±0.0 | 0.5±0.1 | 1.2 | 41.7 |
| | 50 | nd | nd | nd | - | nd | nd | nd | - |
| | 70 | nd | nd | nd | - | nd | nd | nd | - |
| Hexane | 25 | 2.7±0.0 | 0.9±0.1 | 3.6 | 25.0 | nd | nd | nd | - |
| | 50 | 2.1±0.0 | nd | 2.1 | 0 | nd | nd | nd | - |
| | 60 | 1.6±0.1 | nd | 1.6 | 0 | nd | nd | nd | - |
| Ethyl acetate | 25 | 3.2±0.5 | 1.1±0.2 | 4.3 | 25.6 | 2.1±0.0 | 1.6±0.1 | 3.7 | 43.2 |
| | 50 | 2.9±0.1 | nd | 2.9 | 0 | 1.9±0.1 | nd | 1.9 | 0 |
| | 70 | nd | nd | nd | - | nd | nd | nd | - |
| Acetone | 25 | 2.9±0.1 | 1.4±0.2 | 4.3 | 32.5 | 2.8±0.0 | 2.3±0.2 | 5.1 | 45.1 |
| | 50 | nd | nd | nd | - | 2.5±0.1 | nd | 2.5 | 0 |
| Hexane-ethanol (50:50, v/v) | 25 | 5.3±0.3 | 1.8±0.2 | 7.1 | 25.3 | 0.8±0.1 | nd | 0.8 | 0 |
| | 50 | 3.3±0.2 | nd | 3.3 | 0 | nd | nd | nd | - |
| | 70 | 1.1±0.2 | nd | 1.1 | 0 | nd | nd | nd | - |
| Hexane-acetone (50:50, v/v) | 25 | 5.1±0.2 | 1.6±0.1 | 6.7 | 23.9 | 0.9±0.1 | nd | 0.9 | 0 |
| | 50 | 4.6±0.2 | nd | 4.6 | 0 | nd | nd | nd | - |
| | 70 | 2.5±0.1 | nd | 2.5 | 0 | nd | nd | nd | - |
| Hexane-ethyl acetate | 25 | 3.2±0.2 | 1.1±0.2 | 4.3 | 25.6 | 0.9±0.1 | 0.7±0.1 | 1.6 | 43.7 |

| | | | | | | | | | |
|---------------|----|----------|---------|------|------|---------|---------|------|------|
| (50:50, v/v) | | | | | | | | | |
| | 50 | 1.8±0.1 | nd | 1.8 | 0 | nd | nd | nd | - |
| | 70 | nd | nd | nd | - | nd | nd | nd | - |
| Ethyl lactate | 25 | 17.7±0.8 | 8.7±0.3 | 26.4 | 33.0 | 6.0±0.8 | 4.8±0.1 | 10.8 | 44.4 |
| | 50 | 12.6±0.5 | 8.1±0.3 | 20.7 | 39.1 | 5.8±0.7 | 3.4±0.1 | 9.2 | 36.9 |
| | 70 | 7.4±0.2 | 8.2±0.2 | 15.6 | 52.6 | 3.9±0.5 | 3.7±0.2 | 7.6 | 48.7 |

^aValues are means ± SD (n=2); Extraction conditions: solvent: dry tomato by-products=10:1 (v/w), particle size=0.5 mm, 3 extraction steps of 30 min each

Table 4 The effect of drying and storage conditions on lycopene isomerization

| Drying/Storage conditions | | Moisture content (g /100 g ww) ^a | Total lycopene (µg/g dw) ^b | Lycopene loss (%) | <i>All-trans</i> isomers (%) | <i>Cis</i> -isomers (%) |
|---------------------------|--------------------|---|---------------------------------------|-------------------|------------------------------|-------------------------|
| Fresh processing | tomato by-products | 80.48±0.35 | 39.1±0.25 | 0 | 99.7 % | 0.3 % |
| Air drying | | 5.65±0.21 | 30.2±0.61 | 22.8 | 94.4 % | 5.6 % |
| Oven drying | | 4.86±0.19 | 28.8±0.52 | 26.3 | 84.4 % | 15.6 % |
| Vacuum drying | oven | 4.41±0.11 | 32.7±0.34 | 16.4 | 91.4 % | 8.6 % |
| Freeze drying | | 3.87±0.32 | 35.2±0.20 | 10.0 | 95.5 % | 4.5 % |
| Storage days | -20°C/60 | 81.21±0.17 | 33.8±0.17 | 13.5 | 97.6 % | 2.4 % |

^a: Values are means ± standard deviation (n=3)

^b: Values are means ± standard deviation (n=2)

Extraction conditions: hexane-ethyl acetate mixture (50:50, v/v), T=25 °C, solvent: tomato by-products =10:1 (v/w), particle size=0.5 mm, 3 extraction steps 30 min each

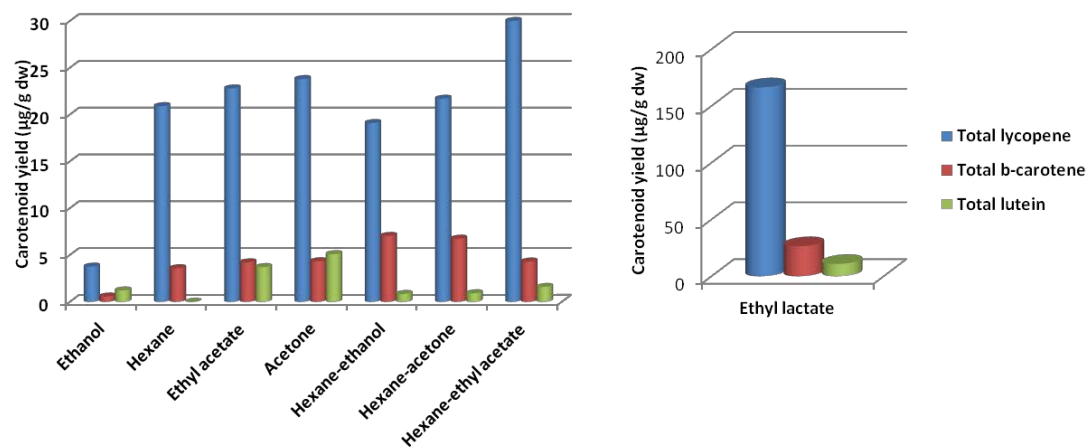


Figure captions

Fig. 1 The effect of organic solvents on the recovery of total carotenoids from tomato processing by-products