# Comparison of lycopene and β-carotene content in tomatoes determined with chemical and non-destructive methods

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Abstract. Tomatoes are one of the most popular vegetables due of their wide use as food. Tomatoes are not only tasty fruit, but one of its benefits - high carotenoids content is well-known. Non-destructive analyses methods are used more and more in different industries. It is cheaper, faster and environmentally friendly way of analyse than traditional chemical methods. But these methods need references to the traditional ones. The aim of this study was to find the correlation between lycopene and  $\beta$ -carotene content in tomatoes determined with reflectance spectrometer and extraction of pigments. Content of two carotenoids (lycopene and  $\beta$ -carotene) was determined in 27 varieties of tomatoes. Red, pink, orange, yellow and brown fruits were included in experiment. Reflectance spectrums of tomatoes fruits were obtained with remote sensing portable spectroradiometer RS-3500 (Ltd.Spectral Evolution). Tetrahydrofuran was used for extraction of pigments. Absorption spectra of extract were obtained by spectrophotometer UV-Vis -1800 (Ltd. Shimadzu). Linear regression analyses were performed to correlate spectral data with lycopene and  $\beta$ -carotene concentrations measured by pigment extraction. The best reflectance region for lycopene spectral detection was 570  $\pm$  5 nm, but for  $\beta$ -carotene 487  $\pm$  5 nm. Reflectance indexes for both pigments were worked out. High linear correlation ( $R^2 > 0.9$ ) between spectral parameters and lycopene concentration was detected. Correlation between results obtained with methods used for  $\beta$ -carotene determination was lower and depended of colour of tomatoes fruits.

Key words: Lycopersicon esculentum, reflectance spectrum, reflectance index.

# **INTRODUCTION**

Tomatoes (*Lycopersicon esculentum* Mill.) are second most consumed vegetable in the world (Clément et al., 2015). Tomatoes differ in size, shape, color, ripening rate, firmness and composition determinating quality and taste (Gastélum-Barrios et al., 2011). They are consumed fresh and in many processed forms Tomatoes and their products are the world richest source of lycopene (Dias, 2012). The average daily intake of lycopene in human diet is about 25 mg and 85% of that is obtained from tomatoes.

Tomatoes contain significant amounts of carotenes and they are the forth leading source of provitamine A in the American diet (Arab & Steck, 2000). Lot of researches demonstrate that lycopene and  $\beta$ -carotene can act as free- radical quencers, prevent aging, tissue damage, heart diseases and certain cancers (Pedro & Ferreira, 2005).

Standart biochemical methods to assay lycopene and  $\beta$ -carotene content are time consuming and use hazardous organic solvents (Davis et al, 2003, Pedro et al., 2005).

Non-destructive analyses are used more and more in different industries. Itis cheaper, faster and environmentally friendly way of analyse than traditional chemical methods. But these methods need references to the traditional ones. They allow to follow on the biochemical changes of the fruits during the ripening process and harvest plants at the optimum time. Non-destructive analyses are used to clarify tomatoes and their products quality as well. Pedro & Ferreira (2005) reported about satisfactory prediction abilities of total solids, lycopene and  $\beta$ -carotene in tomatoe products. Clement et al. (2008) reported about simultaneously measured quality parameters of tomato in a nondestructive manner using vis-NIR reflectance spectroscopy and chemometrics. Results showed well predicted tomatoe color index. Ciaccheri et al. (2018) clarified that lycopene prediction models were dependent on cultivar and season. Clement et al. (2015) informed of determination of lycopene content in vine or pink beaf-type tomatoes and obtained coefficient of determination was 0.65. Davis et al. (2003) reported about light absorbance measurements with scanning xenon flash colorimeter/spectrophotometer, correlation coefficients for lycopene in pureed fresh tomatoes was 0.97, for tomatoes products- 0.88.

The aim of this study was to clarity the correlation between lycopene and  $\beta$ -carotene content in fruits of tomatoes determined with reflectance spectrometer and extraction of pigments.

### **MATERIALS AND METHODS**

#### Samples

Measurements were done with 27 varieties of tomatoes. Tomatoes were grown in plastic film greenhouse without additional lighting. All fruits were harvest at fully ripening stage. In the experiment red fruit varieties 'Amaneta F1', 'Aurea F1', 'Bellastar F1', 'Berberana F1', 'Conchita F1', 'Elegance F1', 'Gardener's Delight F1', 'Gaurmandia F1', 'Lancelot F1', 'Nectar F1', 'Pozano F1' and Sunstream F1', pink ones 'Cipars F1', 'Dimerosa F1', 'DRK936 F1', 'Fuji Pink F1', 'Pink Wonder F1', 'Rhianna F1', and 'Rosastar F1', orange – 'Apressa F1', 'Beorange F1', 'Organza F1', and 'Oranjstar F1', yellow – 'Bolzano F1' and 'Gualdinjo F1' and brown – 'Black Cherry F1' and Chocomate F1' were included. Average tomatoe fruit weight varied from 8.6 g ('Bellastar F1') till 212.0 g ('Pink Wonder F1').

#### *Non-destructive determination of lycopene and* $\beta$ *-carotene*

Reflectance spectrums of tomatoes fruits were obtained with remote sensing portable spectroradiometer RS-3500 (Ltd.Spectral Evolution). 4 – tomatoes fruits depending on fruit size were used for analyses. Totally 16 reflectance spectrums for each variety were obtained. Spectrums were represented as averages for each variety.

## *Extraction and determination of lycopene and* $\beta$ *-carotene*

The same tomatoes fruits were used for biochemical analyses. All the chemicals used were with the analytical grade.

Fresh tomatoes were homogenised,  $1 \pm 0.005$  g of sample was placed in test tube, filled up to 10 mL with tetrahydrofurane (THF), mixed and shacked 30 min in the dark. Then the light absorption values (A) at 663, 645, 505 and 453 were noted by UV-VIS spectrophotometer (UV-1800 Shimadzu Corporation, Japan). Equations 1 and 2 were used to calculate lycopene and  $\beta$ -carotene content (Nagata & Yamashita, 1992). Lycopene and  $\beta$ -carotene content was expressed as mg 100 g<sup>-1</sup> of fresh matter (FM).

$$Lyc = 0.0458A_{663} + 0.204A_{645} + 0.372A_{505} - 0.0806A_{453}$$
(1)

$$\beta car = 0.216A_{663} - 1.22A_{645} - 0.304A_{505} + 0.452A_{453} \tag{2}$$

Biochemical analyses were performed in three replicates.

#### Data analyses

Obtained data were processed by Excel software (Anova and Correlation analyses), data were expressed as means of three replicates for biochemical analyses and 16 replicates for non-destructive analyses.

## **RESULTS AND DISCUSSION**

The reflectance spectrums of tomatoes are shown in Fig. 1. Tomatoes were grouped by their colour and the significant differences between differently coloured fruits are clearly seen in the visible light wavelength (400–700 nm). The major peak of reflectance for red coloured tomatoes fruits was at 652 nm, for pink ones at 643 nm, brown – 636 and for orange and yellow at 612 nm. Decrease of reflectance spectrums in the area between 670–680 nm (minimum at 678 nm for brown coloured tomatoes fruits) indicates chlorophylls presence in tomatoes fruits. Similar evidence of presence of chlorophylls was obtained by Ciaccheri et al. (2018). Rapid increase of reflectance was observed for yellow tomatoes at 457 nm, for orange at 490 nm, but for red, purple and brown ones at 583–587 nm wavelength zone.

Wavelengths area from 1,800–2,500 is less informative due to low reflectance signal (Fig. 1)

Reflectance spectrums at visible light area (400–700 nm) were used to check the correltion of lycopene and  $\beta$ -carotene content determined with biochemical analyses (Fig. 2). The highest coefficient of correlation for lycopene content was detected at the wavelength 570 nm |r| = 0.864, but for  $\beta$ -carotene at the wavelength 487 nm (|r| = 0.682). These wavelengths were used for development of normalized difference indexes for determination of both carotenoids. Normalized difference index is popular in remote sensing and based on ratio of difference and sum of selected wavelengths thus include the effect of average spectra height of sample (Choudhary et al., 2009). In our experiments as reference spectrum 630 nm (cross-point of lycopene and  $\beta$ -carotene correlation curves) was used and equations for index of lycopene (LYC) (Eq. 3) and  $\beta$ -carotene CAR (Eq. 4) was drown up, where RW reflectance at specified wavelength.

$$LYC = \frac{RW_{630} - RW_{570}}{RW_{630} + RW_{570}}$$
(3)



Figure 1. Reflectance spectrums of tomatoes (determined with RS3500); A – in the wavelength region 400–2.500 nm< B – in the wavelength region 400–800 nm.



Figure 2. Coefficients of correlation between reflectance at different wavelength and lycopene and  $\beta$ -carotene determined by biochemical methods.

Obtained results showed that calculated lycopene index from reflectance spectrums (LYC) highly correlated with results of biochemical analyses (coefficient of determination  $R^2 = 0.889$ ). There are no differences depending on tomatoes fruit colour. (Fig. 3).



Figure 3. Correlation of lycopene index (LYC) and biochemically tested lycopene content in tomatoes fruits (mg 100  $g^{-1}$  FM).

Correlation between calculated carotene index and biochemical analyses is lower. Coefficient of determination is  $R^2 = 0.553$ . Differences between tomatoes fruit colour are recognized. Pink tomatoes fruits have less value of indexes in comparison with red ones (Fig. 4).



**Figure 4.** Correlation of  $\beta$ -carotene index (CAR) and biochemically tested 1  $\beta$ -carotene content in tomatoes fruits (mg 100 g<sup>-1</sup> FM).

Several different non-destructive spectroscopic methods have been used to detect lycopene content in tomatoes. Our results corresponds to the same prediction level ( $R^2 = 0.889$ ). Pedro & Ferreira (2005) reported about  $R^2 = 0.999$  for lycopene in tomatoes products, Ciaccheri et al. (2018)  $0.73 < R^2 < 0.81$ , depending on sensor and tomatoes cultivar, Choudhary et al. (2009)  $R^2 = 0.88$  for tomatoes puree, Clement et al. (2008)  $0.92 < R^2 < 0.98$  for fresh tomatoes, Davis et al. (2003)  $R^2 = 0.97$  for fresh tomatoes and 0.88 for tomatoes products, Tilahun et al. (2018) - 0.85 for intact tomatoes fruits. Results depended on used sensors, obtained spectral data, tomatoes cultivars,

growing conditions, but generally the correlation level with biochemical analyses is rather high, that during plant cultivation, fruit storage and processing non- destructive methods are useful for lycopene quantification.

Less information and lower coefficients of determination was reported for  $\beta$ -carotene content. Pedro & Ferreira (2005) reported about R<sup>2</sup> = 0.996 for  $\beta$ -carotene in tomatoes products, Tilahun et al. (2018) – 0.77 for intact tomatoes fruits. Our results confirm that future investigations of non-destructive carotene determination is requred.

## CONCLUSIONS

Elaborated lycopene index (LYC) can be used with an adequate precision for nondestructive assessment of lycopene in tomatoes fruits.

For assessment of  $\beta$ -carotene content with non-destructive methods future investigations are needed.

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