Determination of the Influence of Variety and Level of Maturity on the Content and Development of Carotenoids in Tomatoes

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Abstract: The influence of variety and the level of maturity on the content and composition of carotenoids in tomatoes were monitored in a field experiment in the year 2007. A total of 11 varieties were submitted to this experiment – Albertovské žluté, Bejbino F1, Cristina F1, Bonset F1, Dominato F1, Monika F1, Orkado F1, Start S F1, Stupické polní rané, Tornádo F1 and Rougella F1. A significant difference in the ability of tomatoes to synthesise carotenoids in relation to variety was found. At the mature phase of fruits an average content of carotenoids was determined as 665 μg/g dry mass. The varieties with high content of carotenoids were Bonset F1, Start S F1 and Cristina F1. On the contrary, the varieties with low ability in carotenoids production are Albertovské žluté, Bejbino F1 and Tornádo F1. The main carotenoids determined at the red-fruit tomatoes were lycopene, β-carotene and lutein. In the yellow-fruited variety – Albertovské žluté, only β-carotene and lutein was found.

Keywords: carotenoids; tomatoes; varieties; ripening; HPLC

INTRODUCTION

Tomatoes contain a number of compounds, which affect positively the human organism. Besides the high content of vitamins and minerals, they are an important source of antioxidants. Important group of antioxidants in tomatoes are carotenoids, which participate on the cell protection against the free radicals. Carotenoids are naturally occurring, nonwater-soluble pigments that generally have 40 carbon atoms. They possess very special and remarkable properties, and are effective on all kinds of living organisms. To the most important function belong their antioxidant and provitamin activity. Carotenoids have essential role in plant organisms as well, where they participate on the process of photosynthesis (Gross 1991). Recent evidence has shown carotenoids can be effective antioxidants for inhibiting the development of heart disease (Kritchevsky 1999) and for reducing risk of some types of cancer (Franceschi *et al*. 1994). The development of red pigmentation is one of the most recognisable features of ripening in most tomato fruits. The major carotenoids that accumulate in ripe red tomato fruits are lycopene (~ 90%), β-carotene (5–10%), and lutein (1–5%) with trace amounts (< 1%) of other carotenoids (Ronen *et al*. 1999). Lycopene and β-carotene are the main pigments responsible for the characteristic colour of ripe fruits. These carotenoids largely influence the quality perception of fresh tomatoes because consumers prefer tomatoes with intense red colour. According to Hart and Scott (1995) and Sahlin *et al*. (2004) the carotenoids and the antioxidant content of tomato mostly depends on cultivars, stage of maturity, environmental factors and growing conditions. The aim of this study is to evaluate the effect of variety and stage of maturity on the content and composition of carotenoids in tomatoes.

MATERIALS AND METHODS

Plant material. Tomato plants were cultivated in an experimental field of the Czech University of

Life Sciences in Prague. A total of 11 varieties were submitted to this experiment – Cristina F1, Bonset F1, Dominato F1, Monika F1, Orkado F1, Start S F1, Stupické polní rané, Tornádo F1 and Rougella F1. Simultaneously one yellow-fruit tomato – Albertovské žluté, and one variety of cherry tomato – Bejbino F1 were cultivated. During the ripening, four sample takings were carried out at different levels of maturity. The fruits were taken at these development levels: Breaker, Pink, Light Red and Red (Californian Tomato Commission 2008). The tomato samples were placed in air-tight plastic bags and frozen immediately. The samples were then freeze-dried, homogenised and placed in oxygen barrier bags until analyses of monitored parameters.

Determination of total carotenoids. About 0.25 g of homogenised tomato samples were weighted and placed in 50 ml beakers and extracted for 2 days in dark with 30 ml 100% acetone. After the extraction the samples were for 15 minutes ultrasonicated and then filtered through the Buchner funnel under vacuum with filter paper. The filtrate was quantitatively transfered to 50 ml flask and completed to 50 ml volume with acetone. Analyses of samples were caried out using UV-VIS spectrophotometer Spectronic Hελios γ (Spectronic Unicam, UK). The absorbance of acetone extracts was measured at 662 nm, 645 nm and 470 nm. The total content of carotenoids was calculated from the equations (Lichtenthaler & Wellburn 1983) for 100% acetone (in μg/ml of plant extract)

$$C_a = 11.75\ A_{662} - 2.35\ A_{645}$$

$$C_b = 18.61\ A_{645} - 3.96\ A_{662}$$

$$C_{x+c} = (1000\ A_{470} - 2.27\ C_a - 81.4\ C_b)/227$$

where:

C_a – content of chlorophyll *a*

C_b – content of chlorophyll *b*

C_{x+c} – content of carotenoids

Separation of individual carotenoids. For determination of individual carotenoids about 0.3 g of powdered freeze-dried tomato samples were extracted twice with 10 ml ethylacetate, followed by ultrasonication and centrifugation at 3000 rcf for 10 minutes. Supernatants were combined and the solvent was evaporated to dryness in a rotatory evaporator at 40°C. The organic residue was than redissolved in 5 ml mixture of ethanol:acetone (6:4) with addition 0.2% BHT. Samples were filtered through a 0.45 μm membrane filter and duplicates of 20 μl for each extract were analysed by HPLC. For separation of carotenoids, C18 OmniSpher column (250 × 4.6 mm, S-5 μm, VARIAN) was used with C18 guard column (10 × 3 mm). The isocratic elution consisted of 56% methanol, 40% acetonitrile and 4% tetrahydrofuran for 30 min at a flow rate of 2 ml/min. The runs were monitored with UV-Visible photodiode array detector at 444 nm, 450 nm and 470 nm. Lutein, lycopene and β-carotene were used as standards, eluting at 2.2 min, 14.0 min and 23.4 min, respectively.

RESULTS AND DISCUSSION

The carotenoids of 11 tomato varieties were investigated. The average values of total carotenoids in assessed varieties according to individual

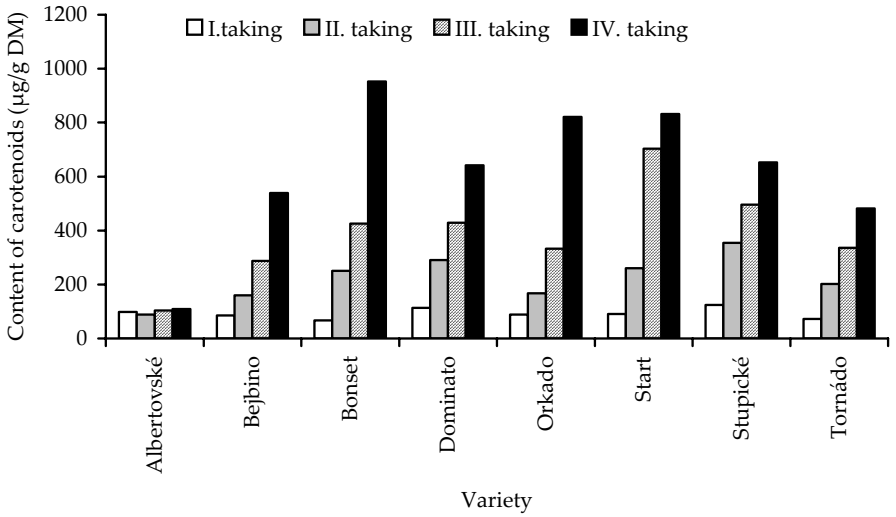

Figure 1. The content of total carotenoids in monitored varieties according to individual stages of maturity (values are calculated from triplicate determinations)

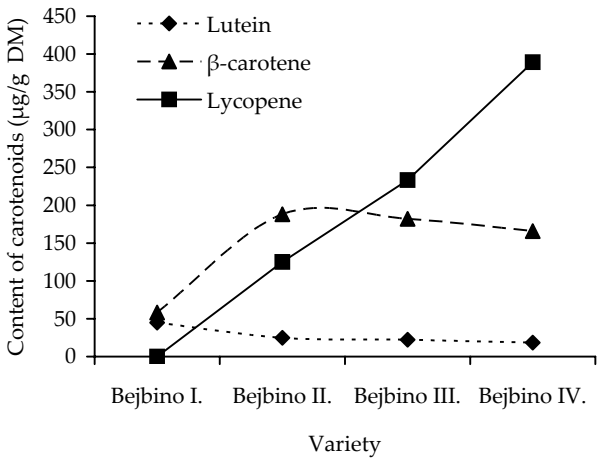

Figure 2. The changes in content of individual carotenoids during ripening (red-fruit variety Bejbino F1)

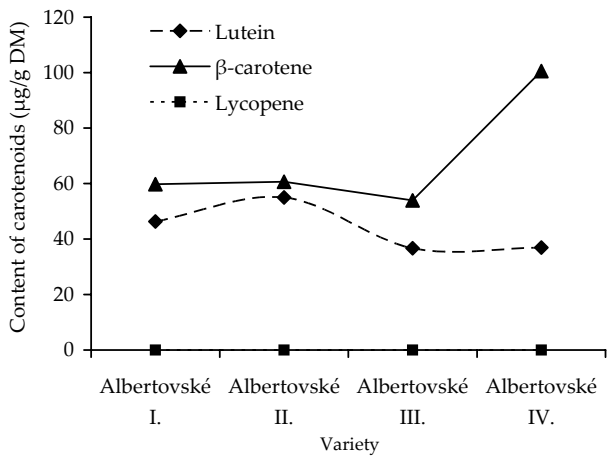

Figure 3. The changes in content of individual carotenoids during ripening (yellow-fruit variety Albertovské žluté)

stages of maturity are recorded in Figure 1. The measured values were processed by the analysis of variance method (ANOVA) using Tukey's test for more detailed evaluation. From the results of statistical analysis emerged, that there is a significant difference in the ability of tomatoes to synthesise carotenoids in relation to variety. Significant differences ($P < 0.05$) were found both amongst varieties and different levels of fruit maturity. At the mature phase of fruits an average content of carotenoids was determined as 665 µg/g dry mass in 2007. The varieties with high content of carotenoids were Bonset F1, Start S F1 and Cristina F1. On the contrary, the varieties Albertovské žluté, Bejbino F1 and Tornádo F1 had low ability in carotenoids production during the maturing process. Content and the composition of individual carotenoids were analysed by high performance liquid chromatography (HPLC-DAD). The main carotenoids determined at the red-fruit tomatoes were lycopene, β-carotene and lutein. In total carotenoid content lycopene abundance was on average 70%, β-carotene 26%, and lutein 4%. Burns *et al*. (2003) recorded that content of total carotenoids in tomatoes averaged 908 µg/g dry mass, where lycopene represented 522.5 µg/g, β-carotene 56.1 µg/g and lutein 94.9 µg/g. The content of lycopene has been increasing evenly during the all four stages of fruit maturity. On the contrary, β-carotene and lutein were intensively synthesised between the first and the second stage of maturity, and their content has not increased in further stages. This is in accordance with those reported by Biacs *et al*. (1987) but not with other authors, who observed a constant increase in β-carotene as ripening progressed (Abushita *et al*. 1997). The changes in carotenoid content during ripening process are showed in Figures 2 and 3. In the yellow-fruit variety – Albertovské žluté, only β-carotene (72%) and lutein (28%) was found. The synthesis of lutein was similar as in red-fruit tomatoes, the most intensive between the first and the second stage of fruit maturity. On the contrary, the synthesis of β-carotene was the most intensive at the final stage of fruit maturity.

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