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Spectrophotometric Analysis of Different Genotypes of Tomato Fruit for Different Pigments

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ABSTRACT
Consumption of tomato products has been associated with decreased risk of many diseases and may exerts positive effects on human health due to the presence of varied antioxidants in it. Therefore, an experiment was carried out to evaluate the biochemical characteristics of tomato in different genotypes. A total of 22 genotypes were studied in this experiment using spectrophotometer at different wavelength i.e., 663 nm, 505 nm and 453 nm for simultaneous study of β-carotene, lycopene, chlorophyll a, chlorophyll b and total carotenoid at 480 nm. Significant differences were observed for these pigments among the various genotypes. The result revealed that Beta-carotene ranged between 0.032 mg/100ml to 0.27 mg/100ml, lycopene from 0.028 mg/100ml to 0.48 mg/100ml, chlorophyll a from 0.003 mg/100ml to 0.068 mg/100ml, chlorophyll b 0.004 to 0.25mg/100ml and total carotenoid between 0.101mg/100g to 0.531 mg/100g.
***Keywords**: β-carotene, lycopene, chlorophyll a, chlorophyll b, Total carotenoid*

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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, non-water soluble pigments that generally have 40 carbon atoms. They possess very special and remarkable properties, and are effective on all kinds of living organisms [8]. Tomato has high nutritional value due to presence of carotenoids, lycopene and β-carotene attributed to their antioxidant and provitamin activity. Lycopene is the most powerful antioxidant of carotenoid family known to prevent cancer, blindness and certain other illnesses. Red fruited varieties tend to possess more of this antioxidant. Together with carotenoids, lycopene helps to protect cells and other structures in the human body from harmful oxygen-free radicals. β-carotene is known for its provitamin A activity and lutein for reduced risk of lung cancer [18]. Deficiency of vitamin A can cause xerophthalmia, blindness and premature death [11].Vegetables are easily accessible to the poorer sections of the society. Tomato being acceptable to people as a food could be a good source of nutrients. The consumption of a diet rich in fresh fruits and vegetables has been associated with a number of health benefits including the prevention of chronic diseases [16, 7, 5]. Therefore, the present investigation undertaken to estimate the beta carotene content, lycopene, chlorophyll, and other nutritional quality attributes of different genotypes of tomato.

MATERIALS AND METHODS

Plant Material

The experiment was conducted in an open field. The investigations included 22 tomato genotypes from different sources. During its growing season all standard growing measures have been applied to researched tomato genotypes. For the purposes of this research, fruits were harvested at full maturity stage. After harvesting, the samples were analyzed for the content of β-carotene, lycopene, chlorophyll a, b and total carotenoid.

Determination of Pigments

β-carotene, lycopene, chlorophyll a and b were determined according to the method of Nagata and Yamshita [13]. It is a simple method for simultaneous determination of pigments in tomato. One gm sample of tomato was taken in a tube and 20 ml of acetone-hexane (4:6) solution was added and then vertexing and centrifugation was done for 10 and 15 minutes (3000 rpm) respectively. Supernatant was collected and filtered through Whatman No.4 filter paper. The absorbance of the filterate was measured at 663nm, 645nm, 505nm and 453nm by spectrophotometer at the same time. Contents of β-carotene, lycopene, chlorophyll a and b were calculated according to the following equations:

Chlorophyll a (mg/100ml) = $0.999\,A663 - 0.0989\,A645$

Chlorophyll b (mg/100ml) = $-\,0.328\,A663 + 1.77\,A645$

Lycopene (mg/100ml) = $-\,0.0458\,A663 + 0.204\,A645 + 0.372\,A505 - 0.0806\,A453$

β-carotene (mg/100ml) = $0.216\,A663 - 1.22\,A645 - 0.304\,A505 + 0.452\,A453$

Determination of Total Carotenoid

Total Carotenoids was determined according to the method of Harborne J.B. [6]. 100 mg of fresh crushed plant tissue was taken in a tube and 10 ml of 80% acetone was added and centrifuged at 3000 rpm for 10 minutes. Supernatant was taken in a separate tube and volume make up was done upto a known volume of 10 ml with 80% acetone. The optical density of supernatant was measured at 480 nm in UV spectrophotometer. Total Carotenoid was calculated according to the following equations:

Calculation –

$$\text{Amount of Carotenoids in 100 mg plant tissue} = \frac{4 \times \text{OD Value} \times \text{Total volume of sample (i .e. we have make the Supernatant volumes as 10 ml)}}{\text{Weight of fresh plant tissue (i.e, we have taken 100 mg plant tissue to grind)}}$$

RESULTS AND DISCUSSION

The mean performance of 22 genotypes regarding pigments are given in the table 1.1. lycopene is a pigment responsible for red colour of the mature tomato and its products [17]. In the present investigation, lycopene content ranges from 0.028 mg /100ml- 0.483 mg/100ml. The findings were in accordance with Burns *et al.*, [2], who reported lycopene content of 522.5µg/g. Mladenovic *et al.* [12] also reported similar results (0.031mg /100g-4.330mg/100g).

β-carotene ranges from 0.032mg/100ml – 0.268mg/100ml. Results were similar as reported by Hallmann *et al.*, [7] that tomato fruits contained 0.26mg/100g fw of β-carotene, while in 2009 it was 0.21mg/100g fw. Kotikova *et al.*, [8] also reported similar results. Abushita *et al.,* [1] found that the β-carotene content was between 2.9mg/kg- 6.2mg/kg. It is believed that the differences among the contents depend upon the growing methods and climate conditions [15], but on the traits of the researched tomato genotypes, too.

Chlorophyll a and Chlorophyll b ranges between 0.003mg/100ml- 0.068 and 0.004 mg/100ml-0.255mg/100ml, respectively. Watada *et al.*, [19], reported that the average chlorophyll content decreased from 13.45µg/g fw in immature green fruit to 0.3µg/g fw in partially ripe fruit. The range of total carotenoid was 0.101mg/g-0.531mg/g. Raffo *et al.*, [16] reported that carotenoids content of tomato were very low at the breaker stage (1.08mg/100g) which increased 10-fold during ripening and reached 12.705mg/100g at full ripening stage. The carotenoids content increased during storage in tomato because of advancement of ripening stage, chlorophyll degradation and increase in the carotenoids synthesis [14]. Carotenoid concentrations in fruits and vegetables were shown to vary with plant variety, degree of ripeness, time of harvest and growing and storage conditions [10].

Apart from that, environmental factors such as temperature, plant nutrition, and light can also considerably affect the biosynthesis of carotenoids. Optimum temperature range for lycopene synthesis is between 12 and 32°C. Temperature below 12° C strongly inhibit the biosynthesis and above 32° C obstruct the process altogether [3, 4]. The increase in lycopene content along with more intense nitrogen fertilization is justified by Lacatus *et al.,* [9] nitrogen is the main element that forms Acetyl-CoA enzyme which plays a central role in the synthesis of carotenoid pigments and converts beta-carotene into lycopene. Dadomo *et al,.* [3] found that with the increased dose of nitrogen the yield of red and uniformly stained fruits as well as the number of fruits per unit of cultivation area were higher.

Table 1.1 : Mean Performance of Beta carotene, Lycopene, Chlo.a, Chlo.b and Total carotenoid in Different Genotypes of Tomoto

GENOTYPES	Beta Carotene (mg/100ml)	Lycopene (mg/100ml)	Total Carotenoids (mg/g)	Chlo.a (mg/100ml)	Chlo.b (mg/100ml)
2013/TODVAR-1	0.056	0.055	0.131	0.010	0.007
2013/TODVAR-2	0.087	0.032	0.355	0.011	0.016
2013/TODVAR-3	0.086	0.267	0.255	0.007	0.021
2014/TODVAR-1	0.032	0.051	0.108	0.004	0.004
2014/TODVAR-2	0.072	0.159	0.122	0.007	0.007
2014/TODVAR-3	0.268	0.483	0.371	0.014	0.109
2014/TODVAR-4	0.082	0.132	0.251	0.003	0.005
2014/TODVAR-5	0.073	0.121	0.168	0.011	0.167
2014/TODVAR-6	0.112	0.118	0.101	0.048	0.080
2015/TOCVAR-1	0.078	0.033	0.162	0.007	0.164
2015/TOCVAR-2	0.103	0.094	0.238	0.007	0.009
2015/TOCVAR-3	0.083	0.120	0.190	0.068	0.255
2015/TOCVAR-5	0.104	0.052	0.223	0.022	0.032
2015/TOCVAR-6	0.151	0.071	0.207	0.035	0.025
2015/TOINDVAR-1	0.088	0.042	0.228	0.007	0.015
2015/TOINDVAR-2	0.049	0.028	0.168	0.031	0.105
2015/TOINDVAR-3	0.040	0.041	0.160	0.007	0.015
2015/TOINDVAR-4	0.064	0.089	0.159	0.012	0.008
2015/TOINDVAR-5	0.207	0.232	0.531	0.008	0.011
H-86	0.170	0.142	0.210	0.035	0.108
ARKA VIKAS	0.128	0.071	0.114	0.016	0.013
SWARNA RATAN	0.063	0.050	0.319	0.006	0.019
CD at 5%	**0.01**	**0.02**	**0.04**	**0.004**	**0.01**
CV (%)	**9.88**	**15.94**	**12.29**	**15.47**	**13.61**

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