Phytochemical and antioxidant composition in *Lycopersicum esculentum*

J. Sravanthi, S. Gangadhar Rao

Abstract

The antioxidant activities were investigated in tomato, (DPPH, ABTS, FRAP), and the phytochemicals such as ascorbic acid, β carotene, total carotenoids were also determined the results showed that highest antioxidant activity was because of phenols and FRAP (46 ± 0.5 mg/g, 20 ± 0.02 mg/g), There was a significant correlation between phenol and FRAP scavenging, it is considered as good sources of antioxidants as shown by the values obtained for antioxidant activity, phenolic, contents.

Keywords: *Lycopersicum esculentum*, antioxidant, phytochemicals Phenols, FRAP, DPPH, ABTS activity.

1. Introduction

*Lycopersicum esculentum* commonly known as in (English – tomato, Hindi – tamatar, Telugu – tomato), for the present study Arka Vikas variety has been used for the analysis of phytochemical compounds. Tomato plants grows upto the height of 1–3 meters (3–10 ft) in height and have a weak stem that often sprawls over the ground and vines over other plants, it is a perennial plant and it’s a native habitat an average common tomato weighs 102–105 grams it contains antioxidant phytochemicals such as the carotene and lycopene. Our body is constantly exposed to a variety of oxidizing agents and the body is equally inbuilt with antioxidants to cater for the free radicals generated from the oxidants thus maintaining a balance between the production of free radicals and neutralization by antioxidants. When there is imbalance between formation and neutralization of free radicals by antioxidants, it results to oxidative stress \[1-3\]. Oxidative stress has been implicated in the etiology of diseases such as cardiovascular diseases and lung cancer \[4-7\]. They are good sources of natural antioxidants which include carotenoids, vitamins, phenolic compounds, flavonoids, dietary glutathione, and endogenous metabolites and have been shown to scavenge singlet and triplet oxygen, free radicals, enzyme inhibitors and decompose peroxides \[9-10\]. Phenolic compounds are secondary metabolites in fruits and vegetables. Carotenes have been proved to possess antioxidant activity due to their ability to quench singlet oxygen and inhibit lipid peroxidation \[12\]. Thus, this study was set to determine the antioxidant activity and phytochemical contents.

Chemicals and reagents required.

Ferric reducing antioxidant property (FRAP), 2, 2 – Diphenyl - 2-picryl hydrazyl (DPPH), Folin ciocalteau, TPTZ(2,4,6-Tris(2-pyridyl)-s-triazine), ABTS(2,2’-azinobis-3-ethylbenzthiazoline-sulphonic acid), aluminium chloride, was obtained from Sigma – Aldrich Co., St. Louis, USA. 2, 6 – dichlorophenolindophenol (DCIP), methanol Folin ciocalteau, phenol, sodium carbonate. Ascorbic acid, metaphosphoric acid (MPA), acetic acid, potassium persulphate, aceton, sodium hydroxide, All chemicals used were of GR grade only.

Material and Methods.

Determination of ascorbic acid

Ten millilitres of the sample (fruit) was titrated against standard 2, 6-dichlorophenolindophenol dye \[11\] which was already standardized against standard ascorbic acid. Results were expressed in percentage.

Determination Of β – carotene

10g of the sample was lyophilized to remove the moisture content. Resulting dried samples were powdered using blender. These ground samples were extracted twice with a total volume...
of 100 ml of 70% aqueous methanol. The mixture was shaken on an orbital shaker for 75 min at 2500rpm and then filtered through Whatman No 1 filter paper [12] the combined methanolic extract was then evaporated at 55 °C using water bath and dried to powder in a lyophilizer. β- Carotene was determined according to the method [14] the dried methanolic extract (100mg) was vigorously shaken with 10ml of acetone – hexane mixture (4:6) for 1 min. The absorbance of the filtrate was measured at λ = 453, 505, 645 and 663 nm by Shimadzu 116 A UV-VIS Spectrophotometer. Contents of β- carotene were calculated according to the following equations:

\[ \beta-\text{Carotene} = 0.216 \times A_{663} - 0.304x A_{395} + 0.452 \times A_{453}. \]

The values are expressed as mg/g of extract.

Where,

\[ A = \text{absorbance recorded at specific wavelengths} \]

### Determination of phenols

The 10gms of the sample (pulp) were sliced, frozen into liquid nitrogen and stored at -80 °C until the analyses were carried out. Frozen pulverized samples were weighed and mixed with 2.5 ml of the extract solution (3% MPA and 8% acetic acid for MPA-acetic acid extraction and 0.1% oxalic acid for oxalic acid extraction). The mixture was homogenized in a high-speed blender at 18000 g (in ice and darkness) for 1 min and then centrifuged at 9000 rpm (refrigerated at 4 °C) for 20 min. This procedure was repeated twice and the two resulting supernatants were mixed together. All extractions were carried out in triplicate. Several precautions were taken in order to perform all the operations under reduced light and at 4 °C temperature [13].

### Ferric reducing antioxidant power (FRAP) assay

The FRAP Reagent was prepared from sodium acetate buffer (300 mM, and pH 3.6), 10 mM TPTZ Solution (40 mM HCl as a solvent) and 20 mM iron (Fe³⁺) chloride solution in a volume ratio of 10:1:1, respectively. The FRAP Reagent was prepared freshly and warmed to 37°C in a water bath before use, One hundred µL of the diluted sample was added to 3 ml of the FRAP reagent, The absorbance of the reaction mixture was then determined at 593nm using Shimadzu 160 A UV-VIS double beam Spectrophotometer, after 4 min in room temperature. The standard curve was constructed using FeSO₄ solution and the results were expressed in µg/g dry wt of plant material.

### Determination of 2, 2′-azinobis 3-ethylbenzthiazoline-sulphonic acid (ABTS) radical scavenging activity

The ABTS⁺ cation radical scavenging activity of the extracts was determined according to the modified method of [15] a stock solution of ABTS was produced by mixing 7 mM aqeous solution of ABTS with potassium per sulfate (2.45 mM) in the dark at ambient temperature for 12–16 h before use. The radical cation solution was further diluted until the initial absorbance value of 0.7± 0.005 at 734 nm was reached. For assaying test samples, 0.98 mL of ABTS solution was mixed with 0.02 ml of the plant extracts. The decrease in absorbance was recorded at 0 min and after 6 min. Scavenging ability relative to the reaction control (without plant extract as 100%) was calculated by using the formula: ABTS⁺ radical scavenging activity (%) = [(Initial reading-final reading)/Initial reading] x 100, where initial reading is absorbance at 0 min and final reading is absorbance at 6 min.

### Determination of 2, 2′-Diphenyl -2-pieryl hydrazyl (DPPH) radical scavenging activity

The DPPH• radical scavenging activity was estimated by measuring the decrease in the absorbance of methanolic solution of DPPH• [16] in brief, to 5 mL DPPH• solution (3.3 mg of DPPH in 100 mL methanol), 1mL of each plant extracts were added, incubated for 30 min in the dark and the absorbance (A1) was read at 517 nm. The absorbance (A0) of a reaction control (methanol instead of plant extract) was also recorded at the same wavelength. Ascorbic acid (5-50 µg/ml) was used as a standard. Scavenging ability (%) was calculated by using the formula: DPPH• radical scavenging activity (%) = [A0− A1)/A0] × 100, where A0 was the absorbance of reaction controls and A1 was the absorbance of extracts or standards.

### Statistical analysis

All results are expressed as mean± Standard deviation. All results are means of three replicates. The data were correlated using Pearson correlation coefficient at P<0.05 Correlations among data obtained were calculated using Pearson’s correlation coefficient (r) and P<0.05 was considered significantly different; SPSS 15 Version was used for the statistical analysis.

### Results and Discussion

**Table 1: Phytochemical content in Lycopersicum esculentum L.**

<table>
<thead>
<tr>
<th>S.NO</th>
<th>Plant Name</th>
<th>Parts Used</th>
<th>Phytochemical compounds</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Tomato</td>
<td>pulp</td>
<td>Ascorbic acid</td>
<td>0.21±0.002</td>
</tr>
<tr>
<td>2</td>
<td>Tomato</td>
<td>pulp</td>
<td>Beta carotene</td>
<td>1.23±0.001</td>
</tr>
<tr>
<td>3</td>
<td>Tomato</td>
<td>pulp</td>
<td>Total carotenoids</td>
<td>2.9±0.007</td>
</tr>
</tbody>
</table>

Each value in the table is represented as mean ±SE (n=3) of triplicates, statistically significant (p<0.05) and are expressed in mg/g dry.

**Table 2: Phytochemical content in Lycopersicum esculentum L.**

<table>
<thead>
<tr>
<th>S.NO</th>
<th>Plant Name</th>
<th>Parts Used</th>
<th>Phytochemical compounds</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Tomato</td>
<td>pulp</td>
<td>Phenols</td>
<td>46±0.5</td>
</tr>
<tr>
<td>2</td>
<td>Tomato</td>
<td>pulp</td>
<td>FRAP</td>
<td>20±0.02</td>
</tr>
<tr>
<td>3</td>
<td>Tomato</td>
<td>pulp</td>
<td>ABTS</td>
<td>4.5±0.01</td>
</tr>
<tr>
<td>4</td>
<td>Tomato</td>
<td>pulp</td>
<td>DPPH</td>
<td>13.2±0.05</td>
</tr>
</tbody>
</table>

Each value in the table is represented as mean ±SE (n=3) of triplicates, statistically significant (p<0.05) and are expressed in mg/g dry.

![Graph showing Phytochemical content in Lycopersicum esculentum](image-url)
Discussion

The ascorbic acid content was observed to be 0.21 ± 0.002 mg/g (table 1, figure 1). Vitamin C scavenges the harmful free radicals produced in the body and also enhances the antioxidant defense mechanism in body. Recent studies have shown that intake of sufficient amount of vitamin C is highly protective to prevent stroke and heart attack consuming diet rich in vitamin C from fruits and vegetables provides protection against cancer. There is no correlation between total ascorbic acid and total antioxidant activities and phenolic. According to [18] it is normal when total ascorbic acid do not correlate with the total antioxidant activities since total ascorbic acid made little or no contribution to the total antioxidant activities of vegetables. Ascorbic acid (vitamin C) is a familiar molecule because of its dietary significance, it is not only an important antioxidant, it also appears to link flowering time, developmental senescence, programmed cell death and responses to pathogens through a complex signal transduction network [19].

The total carotenoid content was observed to be 2.9 ± 0.007 mg/g the values are significant (p<0.05). (Table 1, figure 1) Carotenoids exhibit a central role against cancers, cardiovascular diseases and HIV infection and other age-related disorders [20-21]. Carotenoids exhibit a central Role against cancers, cardiovascular diseases and HIV infection and other age-related disorders.

β-Carotene possesses the ability to scavenge singlet oxygen. Consumption lycopene and β-Carotene have been reported to be inverse with incidence of cancer 1.23 ± 0.001 mg/g (table 1, figure 1) Similar work has been done but the results do not coincide [22]. This could be attributed to the close association of β-carotene to chlorophyll. In a similar investigation, reported higher concentration of β-carotene in spinach and lettuce, which are green leafy vegetables, as against carrot. This is an indication that vegetables further suggests the direct association between β carotene and vitamin A. β-Carotenoid studies.

The phenol content in tomato fruit was observed to be 46 ± 0.5 mg/ g, (table 2, figure 2) High phenolic contents obtained in this study showed that they could serve as nutritional sources for anticancer, antiviral and anti-inflammatory the values are significant (p<0.05) many plant extracts have been reported to have multiple biological effects, including antioxidant properties due to their phytoconstituents including phenolics the antioxidant activity of phenols is mainly due to their redox properties, which can play an important role in adsorbing and neutralizing free radical quenching singlet and triplet oxygen or decomposing peroxides [21] showed the total phenolic content of different vegetable varieties an increase in phenol contents in tomatoes high correlations have often been observed between phenol and antioxidant capacity and our results indicate that phenol has highest correlation coefficient with high antioxidant capacity.

The antioxidant activity was observed to be 20 ± 0.02% (table 2, figure 2) in tomato fruit reductants in the sample reduce a ferric tripyridyltriamine complex, present in the stoichiometric excess, to the blue ferrous form the change of absorbance at 593 nm over 4min is proportional to the combined FRAP value. FRAP Ferric reducing antioxidant power [24] The scavenging activity was observed to be 4.5 ± 0.01 %, (Table 2, figure 2) The values are significant (p<0.05). In this assay, ABTS is converted to its radical cation by addition of potassium per sulfate. This radical cation is blue in color and absorbs light at 734 nm. The ABTS radical cation is reactive towards most antioxidants including phenolics, thiols and ascorbic acid. During this reaction, the blue ABTS radical cation is converted back to its colorless neutral form. Shows ABTS radical scavenging activity of standard ascorbic acid [25] ABTS 2, 2’-azinobis3-ethylbenzthiazoline-sulphonic acid and DPPH2, 2’-Diphenyl -2-pieryl hydrazyl radical scavenging activity by [26-27].

The activity was 13.2 ± 0.05% (table 2, figure 2) the values are significant (p<0.05). The stable DPPH radical model is a widely used, relatively quick and precise method for the evaluation of free radical scavenging activity. DPPH is a stable free radical and accepts an electron or hydrogen radical to become a stable diamagnetic molecule. Antioxidant on interaction with DPPH both transfer electron or hydrogen atom to DPPH and thus neutralizing its free radical character and convert it to 1-1, diphenyl-2-pieryl hydrazine and the degree of discoloration indicates the scavenging activity of the drug. The reduction capacity of DPPH radical is determined by the decrease in its absorbance at 517 nm induced by antioxidants the decrease in absorbance of DPPH radical caused by antioxidants because of the reaction between antioxidant molecules and radical progress which results in the scavenging of the radical by hydrogen donation. It is visually noticeable as a change in color from purple to yellow, hence DPPH is usually used as a substance to evaluate the antioxidant activity shows DPPH radical scavenging activity. This assay is a sensitive way to survey the antioxidant activity (Singh et al. 2012) which is dependent on solvent type, pH and temperature of the system [28-29].

Conclusions

The antioxidant activities, total phenol, β-Carotene, and ascorbic acid content in tomato which is commonly consumed and the chemical composition were assessed and considered as good sources of natural antioxidants since their extract were found to possess high antioxidant activity.

Acknowledgements

I sincerely thank the Department of Botany, University College of Science, Osmania University, Telangana, Hyderabad, India, for sponsoring the RFSMS (UGC- United Grants Commission, New Delhi) fellowship.

Reference

2. Aruoma OI. Methodological considerations for