EVALUATION OF BIOCHEMICAL POTENTIAL IN TOMATO (Solanum lycopersicum) GERMPLASMS

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A total of 185 diverse genotypes of tomato (*Solanum lycopersicum* L.), including 33 genotypes of *Lycopersicum* var. *Cersiforme* (cherry tomatoes), were grown under shade house at National Agriculture Research Council (NARC), Islamabad Pakistan. The genotypes were harvested at fully ripened stage to evaluate the minerals and antioxidant properties. Genotypes were evaluated for antioxidants (ascorbic acids, lycopene, β -carotene and total soluble solids), mineral contents (Na, K, Ca, Mg and P) and trace elements (Fe, Cu, Mn, and Zn). Significant variations were recorded for antioxidant compounds, minerals and trace elements. Lycopene contents ranged from 1.57 to 23.24 mg 100g⁻¹, ascorbic acid from 11.64 to 29.11 mg 100g⁻¹, total soluble solids from 3.33 to 6.46 mg 100g⁻¹ while beta-carotene ranged from 1.32 to 7.6 mg 100g⁻¹. The contents of potassium, magnesium, calcium, iron, and zinc were ranged from 780 to 3260 mg kg⁻¹, 97.37 to 315 mg kg⁻¹, 30.89 to 164.2 mg kg⁻¹, 4.15 to 15.67 mg kg⁻¹and 1.1 to 12.8 mg kg⁻¹ respectively. Cherry genotypes (W-C 1653,W-C 1654,W-C 1656, W-C 1655,W-C 1658, W-C 1657,W-C 1660,W-C 1659,W-C 1666, W-C 16661, W-C 16667,W-C 16668,W-C 1670,W-C 1671,W-C 1673) were found rich in antioxidants, minerals and trace elements. The results reported that quality and anti-oxidants of tomato fruit indicate high potential for genetic improvement of tomato through utilization of identified genotypes for important traits.

Keywords: Solanum lycopersicum, antioxidant, minerals, trace elements, rawalakot.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is a horticultural crop generally consumed either fresh or processed such as tomato ketchup, puree, soup and salsa (Helyes *et al.*, 2009; Ray *et al.*, 2011). Tomatoes and their products are popular nutritive food and are considered good source of minerals, vitamins and antioxidants (Afzal *et al.*, 2013). Macro and microelements along with vitamins A and C are present in tomato fruit that strongly support health of human hence its consumption is increasing globally (Nour *et al.*, 2013). The fresh tomato fruit (100g) is rich in vitamins i.e. A (28%), C (21%); mineral nutrients i.e. potassium (5%), sodium (1%), calcium (1%), magnesium (6.5%), zinc (1.5%), iron (4%) manganese (3.5%) and water 95% (Mark *et al.*, 2013; USDA, 2017).

Tomato is used in daily food in a variety of dishes due to its nutritive values and is also an excellent source of antioxidant compounds such as β -carotene, lycopene and polyphenols (Samad *et al.*, 2017). Lycopene is the most robust antioxidant while β -carotene compound is source of vitamin A in human diet (Luthria *et al.*, 2006; Borguini and Torres, 2009). Epidemiological studies revealed that lycopene alleviates

heart diseases, stomach and prostate cancer (Fernández-Ruiz et al., 2011). Additionally, tomato has been reported effective in the treatment of eye diseases, inflammation and osteoporosis (Singh et al., 2014). Lycopene compound is a member of carotenoids family and it is a lipophilic in nature that dissolves in organic solvent (Bungheze et al., 2011). Most prominent phytochemicals in tomato are carotenoids, of which lycopene is the most abundant in the ripened fruit, accounting for approximately 80-90% of the total pigments (Hernández-Suárez et al., 2007; Helyes et al., 2009). Tomato contains a multitude of vitamins and minerals that act to support human health. They are an excellent source of vitamin C, potassium, and trace elements, i.e. selenium, copper, manganese and zinc, which are cofactors of antioxidant enzymes (Borguini and Da Silva Torres, 2009; Luthria et al., 2006).

Nutritional quality of fruit is associated with various factors, i.e. environment (temperature, light, water, air composition and nutrient availability), genetics (genotype-environment interaction), agriculture practices (plant growth regulators, varieties, irrigation, training system and ripening stage) and storage conditions (Marsic *et al.*, 2011; Haider *et al.*, 2013,

2014). To meet the increasing demands of the customers, it is pre-requisite to collect, evaluate, conserve and exploit the crop germplasm of tomato. For industrial use, precise documentation of germplasm is inevitable keeping in view the status and quality of this crop. Due to import of tomato seed in bulk quantity, most of the germplasm remains unexploited and unidentified for morphological, biochemical, quality and molecular traits in Pakistan. In general, little efforts have been taken to evaluate the various vegetables including tomato crop in Pakistan.

Nutritional gaps between tomato cultivars and germplasm reveals that it is indispensable to design breeding program to improve the cultivars rich in antioxidant as well as high quality fruit traits and yield (Dar and Sharma, 2011). Recently new improved tomato varieties have been developed with respect to nutritional and health benefits keeping fruit yield in consideration (Pinela *et al.*, 2012). Consequently, it is important to estimate nutritional status in terms of mineral contents of tomato germplasm for exploitation in the breeding programs (Fernández-Ruiz *et al.*, 2011; Pinela *et al.*, 2012).

The present study was carried out to explore 185 genotypes of tomato collected from different countries of world grown under shade houses at NARC Islamabad, Pakistan to determine the available antioxidants, minerals and trace elements.

MATERIALS AND METHODS

Plant material: One hundred and eighty-five accessions of tomato were obtained from the national Gene Bank, Bioresources Conservation Institute (BCI), National Agricultural Research Centre (NARC), Islamabad, Pakistan. The nursery was raised in the green-house. The ranges of temperature were maintained between 70-80°F during day and 60-64°F at night while humidity levels were 80-90% during day and 65-75% for night time. The fifteen seeds were sown in plastic

pots of 10 cm length on first week of February during 2017. The equal proportion of farm yard manure, sand and soil were used as a substrate in pots and the pots were kept in greenhouse at 25°C temperature and irrigated with the help of sprinkler at alternate days. One-month old seedlings were transplanted with layout of Augmented Design in the shaded field house. Each genotype was planted in two rows keeping 75 cm row spacing and 50 cm plant spacing with ten plants per entry in the shade-house (130 feet length, 30 feet width and 9 feet height) at Crop Science Institute (CSI), NARC, Islamabad. All the recommended cultural practices were followed during the growing season of crop. Well rotten FYM @30,000kg per hectare and NPK @ 150:75:75kg per hectare was applied. Whole quantity of phosphorus and half of nitrogen and Potash was applied at the time of seed bed preparation and remaining half N and K was applied two weeks interval after transplanting of rice nursery in five split doses. Insecticides (Cypermethrin mixed with Chlorpyriphos or Permethrin) were applied whenever required to control the attack of chewing and sucking insects.

One eighty-five tomato genotypes including thirty cherry tomato genotypes were considered for estimation of minerals and antioxidant analysis (Table. 1). All genotypes of tomato were grown in NARC at Islamabad, Pakistan in shade houses. Transplanting date of seedling of all plants of total genotypes was March 10th. During the growing season all cultivation procedures (nutrition supply, irrigation and plant protection) were conducted according to technological expectations like transplantation was practiced in the evenings to avoid transplant shock. Pruning of lower leaves and weeding was done just after the establishment of transplants. Two weeks after transplantation, earthing up was done followed by supporting of plants with vertical strings after.

Sampling: Data was recorded on randomly based selection. Three fruits of tomato from each genotype were washed and seeds were removed by cutting fruit into halves for estimation

Table 1. Analysis of Variance, mean, range and standard deviations of minerals and antioxidants.

Characters	Mean <u>+</u> SE	Standard	Variance	Range	
		Deviation (SD)		Minimum	Maximum
Sodium (mg kg ⁻¹)	83.14±1.0	20.88	436.050	36.36	134.48
Potassium (mg kg ⁻¹)	2189.71±47.0	652.33	425531.640	780.00	3260.00
Calcium (mg kg ⁻¹)	87.00 <u>+</u> 2.0	29.89	893.310	30.89	164.20
Magnesium (mg kg ⁻¹)	182.92 <u>+</u> 3.0	54.21	2939.140	97.37	315.26
Phosphorus (mg kg ⁻¹)	231.21 <u>+</u> 3.0	51.70	2672.460	122.00	298.00
Iron (mg/kg)	9.40 <u>+</u> 0.2	3.01	9.050	4.15	15.67
Manganese (mg kg ⁻¹)	1.43 <u>+</u> 0.0	0.52	0.270	0.51	2.87
Copper (mg kg ⁻¹)	4.31 <u>±</u> 0.1	2.55	6.480	0.46	10.72
Zinc (mg kg ⁻¹)	8.00±0.2	2.86	8.200	1.10	12.80
Lycopene (mg 100g ¹)	6.17±0.3	5.15	26.526	1.52	23.24
Beta carotene (mg 100g ⁻¹)	3.30±0.1	1.49	2.243	1.32	7.61
Ascorbic acid (mg 100g ⁻¹)	20.77±0.3	4.20	17.668	11.64	29.11
Total soluble solids (mg 100g ⁻¹)	4.22±0.0	0.61	0.374	3.33	6.46

of mineral contents. Pericarp and mesocarp of fruits were first dried in oven at 70°C for 48 hours then sun dried, grounded for one minute in grinder and stored in plastic bottles. The standard protocol for estimation mineral contents were followed (Ryan *et al.*, 2001). For antioxidant analysis, randomly selected fully ripened fruits were washed with running and distilled water and then dried. The fruits were crushed and homogenized in a domestic blender at maximum speed. All the analysis for estimation of antioxidants was triplicated to minimize error.

Determination of minerals and trace elements (Wet *digestion*): For wet digestion of tomato sample, mixture of (2:1) nitric-perchloric acid (HNO₃:HClO₄) was used. One gram of grounded sample mixed in 10 ml of acid mixture, was added in (50 ml) digestion flask. The contents of sample and acids were mixed continuously by spinning of flask and left for overnight. The flask was placed in a digestion chamber on hot plate and temperature was increased gradually up to ~ 230 °C. The flask was heated up until the production of NO2 brown fumes finishes and condensed white fumes of HClO4 in the flask. Moreover, the contents were evaporated till the volume was reached to almost 3-5 ml and precaution was taken to avoid desiccation of volume. The digestion was completed when color of liquid became transparent and after cooling of flask 20 ml of double distilled water was added. Volume was made up with distilled water and the solution was transferred into 25 ml plastic vials and this solution was further used for the estimation of P, K, Fe, Zn, Mn and Cu (Ryan et al., 2001). Iron, zinc, manganese and copper were assessed in tomato fruit by atomic absorption spectroscopy (spectrophotometer UV-1800, Shimadzu, Tokyo, Japan) (Wright and Stuczynski, 1996).

Phosphorus in digest: Phosphorus contents were determined by spectrophotometer following the method of Ryan *et al.* (2001). After digestion of nitric and per chloric acid (HNO₃:HClO₄). Phosphorus was determined calorimetrically as molybdo vanadate phosphoric acid.

Calcium and magnesium in digest: Specific conditions were taken in the cases of calcium and magnesium. A total of 0.5% of lanthanum chloride was added in solution for calcium and magnesium determination in order to avoid interference (Foster and Sumar, 1997). For magnesium determination additional water dilutions were also made.

Potassium and sodium in digest: Potassium and sodium were examined by flame photometer (Jenway PFP7 Dunmow, UK) with the help of air-propane flame. For sodium and potassium determination, an appropriate water dilution was used. The wavelength of 769.9 nm was used for K analysis, 589.0 nm in Na, 422.7 nm in Ca, 324.8 nm in Cu, 286.2 nm in Mg, 248.3 nm in Fe, 213.9 nm in Zn and 279.5 nm in case of Mn and results were expressed in mg kg⁻¹(AOAC, 1990; Hernández *et al.*, 2005).

The total soluble solids (TSS) was estimated by placing one drop of extracted juice of tomato samples on digital refractometer in accordance with Association of Official Analytical Chemists (AOAC, 1994) methods at room temperature. The prism plate completely dried and cleaned before carried out each fruit sample and readings were noted in milligram 100 g^{-1} .

Ascorbic acid (AA) was estimated following the method of Klein and Perry (1982). One gram of sample homogenized with 10 ml of 1% oxalic acid for 45 minutes and filtered excellently. Three ml filtrate was assorted with 1 ml of 2, 6-dichloroindophenol and absorbance was recorded by UV-visible spectrophotometer (UVmini 1240, Schimadzu, China) at 521 nm. The ascorbic acid contents examined in mg 100 g⁻¹ of fresh weight.

For Beta carotene, one-gram sample was placed into a beaker and marinated with 10ml mixture of n-hexane and acetone (1:1) and filtered as well. A total of 10ml of 50% (NH4)₂SO₄ solution was also added in the mixture then shaked thoroughly and allowed to settle it in bottom. After that the upper layer was pipetted off carefully and the absorbance read in double beam Spectrophotometer (PyeUnicam) at 450nm while hexane was run as blank (Alexander and Griffiths, 1993).

The lycopene content was determined according to Noshad et al. (2016). Tomato juice was extracted from 5-10 g pulp with acetone. The acetone extracts were placed in a separate funnel then 20 ml of petroleum ether and 20 ml sodium sulphate solvent (5%) was also added in a same funnel and gradually mixed. Subsequently, two phases were formed, upper phase pipetted off and the lower aqueous phase was again reextracted with other petroleum ether till the colorless aqueous phase was formed. Petroleum ether extracts was transferred in a brown bottle having 10 g anhydrous sodium sulphate. After leaving this extract, for ten minutes, the petroleum ether extract was poured through a funnel in 100 ml flask. The final volume was made up and the absorbance was read at 503 nm in UV-visible double beam spectrophotometer (Shimadzu-UV-160) using petroleum ether as blank. The results of lycopene were expressed as milligram 100 g⁻¹ of sample.

Statistical analysis: Descriptive statistics (mean, standard error, standard deviation, range and frequency distribution) were computed for all the quantitative morphological and biochemical parameters to estimate the genetic diversity present in the germplasm of tomato. Data analyses were carried out using the R software. Means were compared with the help of least significant test (LSD).

RESULTS AND DISCUSSION

Tomatoes consumption mainly contributes to the intake of antioxidant compounds as well as fiber. It is also a good source of some minerals. Tomatoes are rich in vitamins and minerals including potassium, magnesium and phosphorus. There are about 36 calories in 200 g of tomato (Institute of Medicine, 1997; 2005).

Significant variations in minerals were recorded among all genotypes except manganese content (Table 1). Potassium

exhibited abundant concentration among macro elements such as phosphorus while magnesium, calcium, irons and sodium were also present in higher amount in studied genotypes. Potassium ranged from 780 to 3260 mg kg⁻¹ with means value of 2189 mg kg⁻¹. Maximum potassium contents (3260 mg kg⁻¹) were observed in Cherry genotype W-C 1657 followed by Florida (3240 mg kg⁻¹), Yellow Round Tomato (3226 mg kg⁻¹), Zhongza No. 4 (3220 mg kg⁻¹), W-C 1656 (3208 mg kg⁻¹) genotypes with significant variations to the rest of germplasm whereas, genotype TH-10-0007 had minimum concentration of potassium followed by Black from Tula (815 mg kg⁻¹), W-C 16667 (864 mg kg⁻¹), TH-15-095 (867 mg kg⁻¹), and Tomato Grande Vermeiho (887 mg kg⁻¹). These findings related to potassium content were in the best agreement with those described by (Nour et al., 2013; Fernández-Ruiz et al., 2011; Costa et al., 2011; Guil-Guerrero and Rebolloso-Fuentes, 2009).

Magnesium content varied from 97.37 to 315 mg kg⁻¹among studied germplasm of tomato with mean values of 182.92 mg kg⁻¹(Table 1). Higher concentration of magnesium was found in W-C 16668 genotypes tailed by WK 015 (313 mg kg⁻¹) and pair of genotype Tom 10, W-C 1666 (312 mg kg⁻¹), 17904, TH-15-109 (311 mg kg⁻¹), however lower concentration was found in Merbein monarch (100 mg kg⁻¹), Burnley Bounty (100 mg kg⁻¹) and Rhodade, TH-10-5-0030 (99 mg kg⁻¹)genotypes. These values are similar to the findings of Nour *et al.* (2013) but higher than previous data stated by some authors (Chavez-Servia *et al.*, 2018; Guil-Guerrero and Rebolloso-Fuentes, 2009; Costa *et al.*, 2011; Hernández-Suárez *et al.*, 2007).

Calcium content with mean values of 87 ranged between 30.89 to 164.2 mg kg⁻¹ (Table.1). Genotype Black Cherry showed higher concentration of calcium followed by 6233 (162 mg kg⁻¹), W-C 16667 (161 mg kg⁻¹), and W-C 1673 (155 mg kg⁻¹) genotypes. However, Jersey Devil produced low calcium concentration followed by TH-15-096 (34.56 mg kg⁻¹), TH-10-5-0011 (34.87 mg kg⁻¹), CN-87 (35.57 mg kg⁻¹), and Cherokee Purple (36.44 mg kg⁻¹). These results were in good support of the studies conducted by Guil-Guerrero and Rebolloso-Fuentes (2009), Nour *et al.* (2013) and Takač *et al.* (2016).

Sodium content ranged between 36.36 to 134.8 mg kg⁻¹ with 83.14 mg kg⁻¹ mean values. Maximum sodium contents were found in College Abundant genotype followed by the genotypes 1369 (128.38 mg kg⁻¹), Vendor (123.43 mg kg⁻¹) Beef tomato 2015 (123.00 mg kg⁻¹) and Oahu (122.65 mg kg⁻¹). The minimum sodium contents were detected in Scoresby Dwarf followed by Cherokee Purple (37.26 mg kg⁻¹), TH-10-0004 (45.23 mg kg⁻¹), and TH-10-0018 (48.44 mg kg⁻¹). These results are in good consistent with Nour *et al.* (2013).

Phosphorus varied significantly among total germplasm of tomato ranged from 122 to 298 mg kg⁻¹ with mean value of 231.21 mg kg⁻¹. Genotypes TH-15-095 and TH-15-103 showed a maximum value (298 mg kg⁻¹) of phosphorus

followed by TH-10-5-0021(mg kg⁻¹), TH-15-109 (mg kg⁻¹), TH-10-5-0015 (291 mg kg⁻¹) genotypes. Merbein Monarch presented lower phosphorus value followed by Takiis Gem (128 mg kg⁻¹), Beef Tomato 2015 (129 mg kg⁻¹), and Santa (130 mg kg⁻¹). Our values of phosphorus are similar with those reported by Hernández-Suárez *et al.* (2007). Microelements elements Fe (iron), Mn (manganese), Zn (zinc), and Cu (copper), were also estimated in our study (Table. 1).

Tomatoes contain higher iron concentrations than fish and chicken (Bhattarai *et al.*,2016). Iron was principal element among trace elements ranged from 4.15 to 15.67 mg kg⁻¹ with 9.40 mean values. Higher concentrations of iron (15.67 mg kg⁻¹) was found in W-C 1660 genotype followed by Roma (15.61 mg kg⁻¹), YRF1 (15.46 mg kg⁻¹), W-C 2341(15.26 mg kg⁻¹), and W-C 1659 (15.17 mg kg⁻¹). Minimum contents of iron were detected in Cra (4.15 mg kg⁻¹) followed by B147 (4.88 mg kg⁻¹), Hillbilly potato Leaf (5.22 mg kg⁻¹), Yubily (5.22 mg kg⁻¹), and TH-15-109 (5.39 mg kg⁻¹). Our results regarding iron coinciding with those values reported by Saha *et al.* (2010) (6.1 to 17.7 mg kg⁻¹) but slightly higher than the results of Nour *et al.* (2013) (5.5 to 9.7 mg kg⁻¹).

Copper varied from 0.46 to 10.72 mg kg⁻¹ with mean value of 4.31 mg kg⁻¹. Higher contents of copper were observed in genotype W-C 2365 (10.72 mg kg⁻¹) followed by W-C 1657 (10.66 mg kg⁻¹), W-C 1654 (10.34 mg kg⁻¹), W-C 16661 (9.62 mg kg⁻¹), and Black cherry (9.41 mg kg⁻¹) whereas, lower contents (0.46 mg kg⁻¹) observed in Burnley Bounty, Basketvee and Merbein Monarch genotypes followed by Yellow Round Tomato (0.53 mg kg⁻¹), VF-Roma (0.68 mg kg⁻¹), Fire steel (0.67 mg kg⁻¹), Santa (0.67 mg kg⁻¹), and Fukuju (large) (0.86 mg kg⁻¹). Our values are similar with the findings of Geboloğlu et al. (2011) but Hernández-Suárez et al. (2007) stated 1.8 to 3.0 mg kg⁻¹ whereas Fernández-Ruiz et al. (2011) reported 4.3 to 7.4 mg kg⁻¹in terms of copper. Zinc contents varied from 1.1 to 12.8 mg kg⁻¹ with 8.00 mean values. Genotypes W-C 16668 exhibited maximum values $(12.8 \text{ mg kg}^{-1})$ of zinc followed by W-C 1671 (12.6 mg kg $^{-1}$), W-C 1654 (12.5 mg kg⁻¹), W-C 1658, W-C 1660, W-C 1654 (12.4 mg kg⁻¹) and Black Cherry (12.3 mg kg⁻¹) while minimum values found in Taturaa dwarf globe and bowen M8 (1.1 mg kg⁻¹) followed by Striped Cavern (2.2 mg kg⁻¹), TH-15-104 (2.7 mg kg⁻¹), Bowen R3, TH-15-109, Sub-Arctic Cherry (3.1 mg kg⁻¹), Hatif de cologne (3.3 mg kg⁻¹), J Morgan EC6582, APO-12 (3.6 mg kg⁻¹), and Potentate (3.8 mg kg⁻¹). Present results of our work are slightly greater than those reported by Fernández-Ruiz et al., 2011; Geboloğlu et al. (2011) and Nour et al. (2013).

Manganese contents showed less variability among all tomato genotypes ranged from 0.51 to 2.87 mg kg⁻¹ with mean value of 1.43 mg kg⁻¹. Greater contents of manganese were observed in TH-15-115 (2.87 mg kg⁻¹) followed by Jersey Devil, TH-15-112 (2.86 mg kg⁻¹), TH-10-5-0030 (2.76 mg kg⁻¹), Hongza No.20A (2.54 mg kg⁻¹), and W-C 1658 (2.25 mg

kg⁻¹) whereas lower contents detected in LA-4285 genotype $(0.51 \text{ mg kg}^{-1})$ followed by Kazkemet 815 $(0.54 \text{ mg kg}^{-1})$, CN-87 $(0.66 \text{ mg kg}^{-1})$, TH-10- 0024 $(0.55 \text{ mg kg}^{-1})$, and TH-15-104 $(0.73 \text{ mg kg}^{-1})$ genotypes. Present values investigated for Mn are in agreement with the findings of Fernández-Ruiz *et al.* (2011) and Hernández-Suárez *et al.* (2007) but lower than Geboloğlu *et al.* (2011). It has been observed that in present study largest fruit genotypes contain less concentration of minerals. On the other hand, accessions of smallest fruit size like Cherry genotypes contain higher concentration of minerals. Our results are in consistence with Costa *et al.* (2011) who reported that cherry tomatoes exhibited the highest concentration of minerals among the studied germplasm of tomato.

The studied tomato germplasm depicted enough variation, Table 3 showed selected tomato genotypes for their better performance on the basis of minerals content (mg kg⁻¹). Out of one hundred and eighty-five genotypes, 64 genotypes were identified for potassium content (>2500mg kg⁻¹), 33 genotypes for calcium (>110 mg kg⁻¹), 35genotypes for magnesium (>230 mg kg⁻¹) and 69 genotypes for iron content (>10 mg kg⁻¹). We suggested that these genotypes can be used in future breeding either through direct exploitation or through hybridization program.

Cluster analysis: Germplasm of tomato for minerals contents was grouped into four clusters. Cluster I consisted of 22 genotypes, cluster II of 23, Cluster III of 52 and cluster IV of 82 genotypes (Table 2, Fig. 1). Mean values along with standard deviation for each cluster described that genotypes in cluster I were high in sodium (87.0±21 mg kg⁻¹), calcium (82.1±27 mg kg⁻¹), magnesium (196.2±64 mg kg⁻¹), phosphorus (243.3 \pm 50 mg kg⁻¹), iron (9.7 \pm 3.4 mg kg⁻¹) and zinc content (7.2±2.8 mg kg⁻¹), medium in potassium $(1510.0\pm122 \text{ mg kg}^{-1})$ and copper content $(3.3\pm2.5 \text{ mg kg}^{-1})$. Germplasm from Pakistan and cherry genotypes distributed in all clusters. Genotypes of cluster II had high mean calcium (103.1±36 mg kg⁻¹), magnesium) 199.5±60 mg kg⁻¹), phosphorus (232±44 mg kg⁻¹) and zinc content (9.4±2.6 mg kg⁻¹), whereas low means were observed for sodium (76.3 \pm 20 mg kg⁻¹), potassium (1034.9±138 mg kg⁻¹). The unique

Characters	Cluster I	Cluster II	Cluster III	Cluster IV
Total genotypes	22	23	52	82
Sodium	87.0±21	76.3±20	87.1±21	81.2±20
Potassium	1510.0±122	1034.9±138	2913.2±200	2184.3±22
Calcium	82.1±27	103.1±36	84.0±26	85.9±29
Magnesium	196.2±64	199.5±60	175.8±47	179.7±53
Phosphorus	243.3±50	232.0±44	223.6±53	233.2±52
Iron	9.7±3.4	8.9±3.0	9.1±2.9	9.7±3.0
Manganese	1.3±0.6	1.5 ± 0.5	1.5 ± 0.5	1.4 ± 0.5
Copper	3.3 ± 2.5	$4.4{\pm}2.4$	4.7 ± 2.8	4.3±2.4
Zinc	7.2 ± 2.8	9.4 ± 2.6	7.7 ± 2.7	8.0±3.0



Figure 1. Cluster diagram based on minerals contents (mg kg⁻¹) of tomato germplasm.

Table 3. Selected tomato genotypes for better performance on the basis of minerals content (mg kg⁻¹).

I raits	Kange	Genotypes Identified	Total
			Genotypes
Potassium	>2500	Hillbilly Potato Leaf, Cherokee Purple, TH-15-112, TH-15-114, TH-10-5-0011, TH-10-5-	64
$(mg kg^{-1})$		0012,TH-10-5-0026,TH-10-5-0030,TH-10-5-0044,Yellow Round Tomato, Beef Tomato 2015,	
		Zhongza No. 4, Hongza No.20A, W-C 1654, W-C 1656, W-C 1655, W-C 1657, W-C 1660, W-C	
		1673, Merit, Rhodade, Oahu, GeraldtonSmmoth Skin, Stakeless, Florida,W-C 2404,W-C	
		2405, Takiis Gem, Big Girl VF, Rey de Lops Tempranos, B 147, Harvestvee, Li Cun, Daydream,	
		Alton, Kootenai, La Rochape, Scores by dwarf, Latah, APO-12, Basketvee, Rodade, Money	
		Maker, Accession 1369, Fukuju (large), Sadong (19850), Castlemorll, Roma, Burneiy Gem,	
		Molokai, Red rock, Tomato 004, TH-10-0018, TH-10-0035, CN-87, TH-15-106, W-C 1646, W-	
		C2365,W-C 1665, Bulgaria, Preslav, ST 23S. Klave, Blagoevgrad, S. Milanovo, Shumen	
Calcium	>110	Black from Tula, Roma, TH-10-5-0044, G 28504, W-C 1656, W-C 1658, W-C 1657, W-C	33
(mg kg ⁻¹)		1660,W-C 1659,W-C 1666, W-C 16661,W-C 16667,W-C 1670,W-C 16669,W-C 2404,W-C	
(88)		2405, Burnley Bounty, Bowen R3, Kootenai, Master No.2, Yubily, Salad Special, VF-Roma, W-	
		C 1642, W-C 1643, W-C 1644, W-C 1645, W-C 1646, W-C 1647, W-C 1649, W-C 1651, W-	
		C2365,W-C 1665	
Magnesium	>230	17904,TH-15-096, TH-15-109,TH-15-117,TH-10-5-0015, TH-10-5-0043,Tom 10, Sub-Arctic	35
$(mg kg^{-1})$		Cherry, Hongza No.16, W-C 1653, W-C 1658, W-C 1657, W-C 1660, W-C 1666, W-C 16661, W-	
		C 16667,W-C 16668,W-C 1670,W-C 1671,W-C 1673,W-C 16669,W-C 2405, LA-4285, LA-	
		0314, LA-1969, WK 015, W-C 1643, W-C 1644, W-C 1645, W-C 1647, W-C 1649, W-C 1651, W-	
		C 2330,W-C2365,W-C 1665	
Iron	>10	Striped Cavern, Principe Borghese, Black Cherry, LA 4285A, TH-15-096, TH15-115, TH-15-	69
(mg kg ⁻¹)		116,TH-10-5-0011, Verigated Striped Tomato, Hongza No.16,Hongza No.20A, W-C 1653,W-C	
		1654,W-C 1656, W-C 1655,W-C 1658, W-C 1657,W-C 1660,W-C 1659,W-C 1666, W-C 16661,	
		W-C 16667,W-C 16668,W-C 1670,W-C 1671,W-C 1673, W-C 16669,Vendor, College	
		Abundant, Oahu, Stakeless, Florida, W-C 2405, Merbein monarch, Harvestvee, LiCun,	
		Alton, Master No. 2, Salad Special, Bowen M8, Bonnyvee, YRF1, Punhong (19842), Hongza	
		No.20, Roma, J Morgan EC6582, Burneiy gem, Ponderosa, Homestead (T3), TH-10-0013, TH-	
		10- 0024,TH-10-0038, TH-10-0009, TH-10-0037,TH-15-101,TH-15-107,W-C 1642,W-C	
		1643,W-C 1644,W-C 1645,W-C 1646,W-C 1647,W-C 1649, W-C 1651,W-C 2330,W-C	
		2341,W-C2365,W-C 1665, Preslav	

features of Cluster III had highest potassium content (2913.2±200 mg kg⁻¹), iron (9.1±2.9 mg kg⁻¹) content and medium mean values for sodium (87.1±21 mg kg⁻¹), calcium (84.0±26 mg kg⁻¹), phosphorus (223.6±53 mg kg⁻¹), zinc content (7.7±2.7 mg kg⁻¹).Cluster IV had medium mean values for all studied traits except high mean vales were observed in iron (9.7±3.0 mg kg⁻¹) and zinc content (8.0±3.0 mg kg⁻¹).

From the present results, we can conclude that the mineral contents are associated to the water present in fruit as well as translocation and absorption of macro and microelements in the plant (Chávez-Servia et al., 2018). It is necessary to highlight the evidences that the tomato fruit composition of mineral contents depend on a series of factors such as genotype, species, planting or harvesting season, organic or conventional crop system, fertilizer application, and field or greenhouse growing conditions (Chávez-Servia et al., 2018; Aghili et al., 2012). The maturation stage of plant, environmental conditions, and genetic variations were also the potential description for inconsistencies observed. Very limited work has been reported on mineral analysis of tomato fruit. Knowledge of chemical composition of tomato fruit is important for nutritive and dietary values, yield, quality and behavior of raw materials during processing, conservation as

well as storage influenced by chemical composition of fruit (Turhan *et al.*, 2009). It is observed in present investigation that genotypes not differ only with respect of morphological traits but also varied significantly in terms of mineral concentration of tomato fruit. It is important to describe that plants of genotype from which fruits were harvested for the analysis were handled properly in the field. It is indispensable to record that there are complications associated to comparability among different aspects of studies but investigation contributes together and provides valuable evidence to articulate strategies for exploitation and preservation of germplasm.

Tatal

Antioxidants contents: The significant variance was observed among genotypes for all traits with respect to antioxidant potential (Table 1). The level of lycopene content of tomato fruit depends upon the genetic potential of genotypes. The lycopene content ranged from 1.57 to 23.24 mg 100g⁻¹ with grand mean values of 6.17 mg 100g⁻¹ for whole population. The mean values of all antioxidants traits of tomato fruit displayed in Table 2. Maximum lycopene contents (23.24 mg 100g⁻¹) were observed in Black Cherry which was at par with Juane Flamme (22.32 mg 100g⁻¹), Black from Tula (20.56 mg 100g⁻¹), Brandy Wine (20.12 mg 100g⁻¹), and W-C 1660 (18.68 mg 100g⁻¹) genotypes.

However the minimum content of lycopene $(1.57 \text{ mg } 100\text{g}^{-1})$ was recorded in Colonial (1.52 mg 100g⁻¹) followed by College abundant (1.57 mg 100g⁻¹), Victoria dwarf, Sadong (19850) no.1 (1.60 mg 100g⁻¹), TH-15-106, Vendor (1.67 mg 100g⁻¹), and Bulgaria (1.70 mg 100g⁻¹). Our findings are similar with Hanson et al. (2004) who investigated lycopene contents in 53 genotypes of tomato for antioxidants. However, our findings are higher than the values of Kaur et al. (2017) who estimated variability in lycopene content of 55 elite lines of tomato and Pinela et al. (2012) for tomato genotypes grown in Italy (2.33-16.9 mg 100g⁻¹) and farmer varieties of Portuguese grown in home gardens (10.9-18.6 mg 100g⁻¹), respectively. A large variation in the lycopene contents of genotypes depend upon growing and environmental conditions mainly light and temperature. Similarly, different accessions of tomato possessed variation for lycopene content Abushita et al. (2000); Toor and Savage (2005) and Fanasca et al. (2006).

The content of ascorbic acid of analyzed tomato fruit ranged from 11.64 to 29.11 with mean values of 20.73 mg 100g⁻¹. The highest ascorbic acid content (29.11 mg 100g⁻¹) was noted in Black Cherry which are at par with Principe Borghese (28.51 mg 100g⁻¹), W-C 1642 (28.42 mg 100g⁻¹), Delicious (28.2 mg 100g⁻¹), and LA-4285A (27.91 mg 100g⁻¹)genotypes while lower level of ascorbic acid was registered in G 28504 genotypes (11.64 mg 100g⁻¹) trailed by TH-10-5-0044 (14.86 mg 100g⁻¹), Variegated Striped Tomato (15.17 mg 100g⁻¹), and 17904 (17.02 mg 100g⁻¹) genotypes of population. These results are in agreement with Nour *et al.* (2013) who described similar contents of ascorbic acid (9.9-34 mg 100g⁻¹) in tomato genotypes developed in southwestern Romania.

On the other hand, our results are slightly higher than Mostapha *et al.* (2014) who explored antioxidants of eight tomato genotypes grownup in Algeria. George *et al.* (2004) stated that ascorbic acid contents of 12 tomato cultivars varied from 8.4 to $32.4 \text{ mg } 100\text{g}^{-1}$. It is observed that main factor

Table 4. Clusters, means along standard deviations for antioxidant traits of tomato germplasm.

Traits	Number of genotypes	Cluster I	Cluster II	Cluster III	Cluster IV
Lycopene mg 100g ⁻¹	37	2.43±0.90	3.58±1.4	21.9±1.6	12.7±3.3
Beta carotene mg 100g ⁻¹	94	$4.04{\pm}1.45$	$2.52{\pm}1.1$	4.2±0.3	4.2 ± 1.4
Ascorbic acid mg 100g ⁻¹	3	22.72 ± 2.58	18.80 ± 11.60	27.9±1.1	21.7±3.0
Total soluble solids mg 100g ⁻¹	51	4.17±0.39	3.94±0.3	4.3±0.5	4.8 ± 0.8



Cluster Diagram Antioxidants

Figure 2. Cluster diagram based on antioxidants compounds of tomato germplasm.

which effects ascorbic acid content of genotypes is light intensity in studied population. This could be a valid reason for explaining the relative higher content of ascorbic acid of genotypes who receive much light with respect to other accessions in the field.

Total soluble solids ranged from 3.33 to 6.46 mg $100g^{-1}$ with mean value of 4.22 mg $100g^{-1}$. Genotype W-C 16668 exhibited higher content of total soluble solids (6.46 mg $100g^{-1}$) followed by W-C 16667 (6.39 mg $100g^{-1}$), W-C 1642 (5.97 mg $100g^{-1}$), and W-C 1643 (5.95 mg $100g^{-1}$) genotypes. Genotype TH-10-0004 had minimum soluble solids (3.33 mg $100g^{-1}$) concentrations followed by Cra, W-C 1665 (3.36 mg $100g^{-1}$), Taturaa dwarf globe (3.42 mg $100g^{-1}$), YRF1 (3.46 mg $100g^{-1}$) and Harvestvee (3.56 mg $100g^{-1}$). It is observed that smaller fruits of cherry genotypes contain higher concentrations of total soluble solids than large fruited genotypes. These findings are slightly lower than the values of Hanson *et al.* (2004), but slightly higher than Kaur *et al.* (2017).

Mamatha *et al.* (2017) reported 2 to 5 brix total soluble solids in tomato varieties of India. Our results regarding this trait are in consistence with Rai *et al.* (2016) who reported 3-6.42 mg $100g^{-1}$ total soluble solids in different tomato varieties of tomato.

Beta-carotene ranged from 1.32 to 7.6 mg 100g⁻¹ with mean value of 3.30 mg 100g⁻¹. TH-10-5-0021 genotype registered the higher value of beta carotene (7.6 mg 100g⁻¹ followed by Merbein Monarch (7.53 mg 100g⁻¹), TH-10-5-0026 (6.85 mg

100g⁻¹), Taturaa Dwarf Globe (6.74 mg 100g⁻¹), and TH-10-5-0030 (6.55 mg 100g⁻¹). Lower beta carotene was recorded in Merit and TH-15-109 genotypes (1.32 mg 100g⁻¹followed by Florida (1.34 mg 100g⁻¹), Bowen M8 (1.47 mg 100g⁻¹), GeraldtonSmmoth Skin (1.48 mg 100g⁻¹), W-C 2405 (1.56 mg 100g⁻¹). Similar findings were stated by Sonam and Hussain (2017) who reported that beta-carotene was ranged from1.6 to7.61 (mg 100g⁻¹) in studied tomato germplasm of Jammu Kashmir. A high concentration of lycopene and antioxidant activity enhances nutritional values of germplasm and improves our diet by higher antioxidants contents (George *et al.*, 2004).

From studied germplasm, some of tomato genotypes were selected for better performance on the basis of antioxidants content shown in Table 5. Lycopene content were found >8 mg $100g^{-1}$ in 50 genotypes while 52 genotypes exhibited >21 mg $100g^{-1}$ ascorbic acid and 47 genotypes were selected for >4mg $100g^{-1}$ beta-carotene. These selected genotypes can be used for further research in future.

Cluster Analysis: A tree diagram was constructed for antioxidant compounds of tomato based on Nei's coefficient matrix method (Table 4, Fig. 2). Dendogram was distributed into four main clusters at a linkage distance of 15. Means with standard deviation and variance for their particular clusters given in Table 4. Thirty-seven genotypes were grouped in cluster I. Genotypes of cluster I had maximum mean values for ascorbic acids ($22.72\pm2.58 \text{ mg } 100\text{g}^{-1}$) and medium mean beta carotene ($4.04\pm1.45 \text{ mg } 100\text{g}^{-1}$). Low means were

Table 5. Selected tomato genotypes for better performance on the basis of antioxidants content.

Traits	Range	Genotypes identified	Number of genotypes
Lycopene (mg 100g ⁻¹)	>8	Black Cherry, Brandy Wine, Black from Tula, Cherokee Purple, JuaneFlamme, Roma, LA- 1969A, LA-0314A, LA-4285A, TH-10-5-0011, TH-10-05-0009, TH-10-5-0015, TH-10-5-0012,	50
		TH-10-5-0021, TH-10-5-0026, TH-10-5-0030, Yellow Round Tomato, Beef Tomato 2015, W-C	
		1055, W-C 1054, W-C 1050, W-C 1055, W-C 1058, W-C 1057, W-C 1000, W-C 1059, W-C 16666 W-C 16667 W-C 16667 W-C 16668 W-C 1670 W-C 1671 W-C 1673 W-C 16669 W-C	
		1665. Geraldtonsmoth skin. Big Girl VF. Rougr de marnande. TH-15-099. W-C 1642. W-C	
		1643,W-C 1644, W-C 1645, W-C 1646, W-C 1647,W-C 1649,W- 1651, W-C 2330,W-C 2341,W-C2365	
Beta carotene >4 (mg 100g ⁻¹)	>4	Yellow Stuffer, Striped Cavern, Principe Borghese, Brandy Wine, Cherokee Purple, Juane	47
		Flamme, TH-10-05-0009, TH-10-5-0021, TH-10-5-0026, TH-10-5-0030, Tom 10, Sub-Arctic	
		Cherry, Zhongza No. 4, W-C1654, W-C 1656, W-C 1655, W-C 1658, W-C 1660, W-C 1659, W-C	
		16667, W-C 1670, W-C 16669, Vendor, Big Girl VF, Merbein Monarch, ZhongShu No. 6, B 147,	
		Early Chatham, Yellow Wattle, Victoria Dwarf no.1, Santa, Taturaa Dwarf Globe, Colonial,	
		Molokai, LA-1969, Homestead (T3), TH-10-0018, LA0314 ,W-C 1642,W-C 1643,W-C	
		1644,W-C 1645,W-C 1646, W-C 1647, W-C 1649, W-C 1651,W-C 2341	
Ascorbic acid	>21	Black Cherry, Principe Borghese, Brandy wine, Black from Tula, JuaneFlamme, Delicious,	52
(mg 100g ⁻¹)		Jersey Devil, Burgess Stuffing, Roma, LA-1969A, LA-0314A, LA-4285A, TH-15-109, TH-10-	
		5-0011,TH-10-5-0012,TH-10-5-0043,Yellow round tomato,Beef tomato 2015, Hongza No.16,	
		Zhongza No. 4, Hongza No. 20A, W-C 1656, W-C 1655, W-C 1658, Cherokee, College	
		Abundant, Big Girl VF, Merbein Monarch, ZhongShu No. 6, B 147, Rougr de Marnande,	
		Yellow Wattle, Bowen R3, Scoresby Dwarf, Bonnyvee, MoneyMaker, Colonial, Sadong	
		(19850), Molokai, TH-10-0018, LA-4285, TH-15-099, TH-15-106, WK 015, W-C 1642, W-C	
		1643,W-C 1644,W-C 1645,W-C 1646, W-C 1647,W-C 2341, A. KostaPerchev, Vidin	

observed for lycopene $(2.43\pm0.9 \text{ mg } 100\text{g}^{-1})$ content. Cluster II comprised94 genotypes and was largest cluster. It contained mixed population of different sources and difficult to differentiate cluster with respect to origin. The significant features of this cluster were lowest mean lycopene content $(3.58\pm1.4 \text{ mg } 100\text{g}^{-1})$, beta carotene $(2.52\pm1.1 \text{ mg } 100\text{g}^{-1})$, total soluble solids $(3.94\pm0.3 \text{ mg } 100\text{g}^{-1})$ and medium ascorbic acids $(18.8\pm11.6 \text{ mg } 100\text{g}^{-1})$ content. Genotypes Black Cherry, Brandy wine and Juane Flamme contributed in cluster III.

The unique characteristics of this well separated cluster were highest in mean lycopene ($21.9\pm1.6 \text{ mg } 100\text{g}^{-1}$), ascorbic acid content ($27.9\pm1.1 \text{ mg } 100\text{g}^{-1}$), medium mean beta carotene (4.2 ± 0.3) and total soluble solids ($4.3\pm0.5 \text{ mg } 100\text{g}^{-1}$). Cluster IV consisted of 51 genotypes. Twenty-nine cherry genotypes out of total 34 participated in this cluster. Genotype had high lycopene content ($12.7\pm3.3 \text{ mg } 100\text{g}^{-1}$), ascorbic acids ($21.7\pm3.0 \text{ mg } 100\text{g}^{-1}$), medium beta carotene ($4.2\pm1.4 \text{ mg } 100\text{g}^{-1}$) and medium to high total soluble solids ($4.8\pm0.8 \text{ mg } 100\text{g}^{-1}$).

The results reported in the manuscript for tomato fruit quality and anti-oxidant indicate high potential for genetic improvement of tomato through utilization of identified genotypes for important traits. Therefore, this study can be proven a bench-mark for breeders in targeting future tomato improvement for yield, quality, anti-oxidants and microminerals either through direct selection or through hybridization for developing pure lines or hybrids.

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