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Biochemical and quality attributes of grafted tomato (*Solanum lycopesicum* L.)

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Abstract
Tomato is universally considered as "Protective food" due to the presence of health promoting compounds. Interference of biotic and abiotic stresses drastically reduces the yield and quality of tomato. Considering this bottlenecks, cleft grafting was attempted in tomato involving three wild *Solanum* species *viz.*, *Solanum torvum*, *Solanum sisymbrifolium* and *Solanum capsicoides* and two tomato hybrids *viz.*, TNAU tomato hybrid CO 3 and Shivam and evaluated under shade net condition for yield and quality. The results of the experiment revealed that TSS and titrable acidity was superior (79 °Brix and 0.79%) in TNAU tomato hybrid CO 3 grafted onto *Solanum sisymbrifolium* rootstock and ascorbic acid (37.92 mg/100g), total chlorophyll (2.81 mg/g), pH (5.63), lycopene (5.69 mg/100g) was higher in Shivam grafted on *Solanum sisymbrifolium.* Similarly, total phenol content was high in TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock (3.73 mg/g). Enzyme activity such as peroxidase was higher (1.34 g/min) in immature fruits of TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock and matured fruits of Shivam grafted on to *Solanum torvum* (0.91 g/min) rootstock. Increased catalase activity(1.92 µg of H_2O_2/g/min) was noticed in immature fruits of Shivam grafted on *Solanum torvum* rootstock and in matured fruits of TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* rootstock (1.32 µg of H_2O_2/g/min). Among the graft combinations solasodine content was observed more in Shivam grafted on to *Solanum torvum* rootstock (0.028%).The biochemical and quality parameters is not altered by grafting and from this study it could be interfered that the above traits are determined by the scion not by the rootstocks.

Keywords: Grafted tomato, cleft grafting, peroxidase, catalase, Solanum rootstocks

Introduction
Tomato is universally considered as "Protective food" due to its health promoting recognition among the people. Tomato fruits are loaded with vitamins, minerals and bioactive compounds, which includes carotenoids (lycopene and β-carotene), ascorbic acid, tocopherols and polyphenols. In India, tomato is cultivated in an area of 7.89 lakh ha with the production of 197.59 lakh tonnes (Horticulture Statistics at a Glance, 2018) [6]**.** In India, major tomato growing states are Andhra Pradesh with a production of 27.44 lakh tonnes with a share of 13.90% followed by Madhya Pradesh and Karnataka. In Tamil Nadu, total production of tomato is about 8.87 lakh tonnes with a share of 4.49% (Horticulture Statistics at a Glance, 2018) [6]. In 2020, India's export of tomato accounted for approximately 93.62 thousand metric tonnes of fresh and chilled tomatoes which has been notably decreased from previous years may be due to COVID 19 pandemic.

Consumption of tomato helps in preventing chronic diseases like cancer and cardio vascular problems has been underlined in many epidemiological studies (Klipstein Grobusch *et al.*, 2000 and Giovanelli *et al.*, 2002) [8, 4]. In addition to the above, Tomato is rich in lycopene, which acts as an antioxidant by quenching out the freely available toxic reactive oxygen species (ROS), thereby avoiding cell injury. Processed tomato products also play a vital role in export market as Americans alone consume 12 kg of processed tomato per year per person excluding ketchup and sauces (Thakur *et al.*, 2009) [23]. The cultivation of tomato encountered many field problems *viz.*, pest and diseases, which leads to yield losses. The mitigation of these biotic and abiotic factors requires repeated application of fungicides and pesticides which in turn leads to threats to ecological safety. However, breeding is considered as a viable option to develop varieties/hybrids with resistance to these stresses, but it is difficult to achieve targeted variety/hybrid of expected results. Hence, grafting may serve as an alternative approach to overcome these problems within short period of time.

Grafting is one of the successful proven technology which is being widely followed in other horticultural crops (Tamilselvi and Pugalendhi, 2017; Pugalendhi *et al.*, 2019) [21, 14]. Considering the importance of the crop, there is an ultimate need to enhance the productivity of tomato through grafting. The biochemical parameters are the key parameters to be assessed in grafted plants for biotic and abiotic stress tolerance. Similarly, the effect of rootstocks on the quality parameters of the fruits is essential for the consumer preference and nutritional security. Under these circumstances, two commercial tomato hybrid *viz.*, TNAU tomato hybrid CO 3 and Shivam were cleft grafted on to three wild *Solanum* species and were evaluated for biochemical and quality parameters under shade net condition.

Materials and Methods

Study area and grafting of tomato

The experiment was carried out during the year 2019-2020 at College orchard situated at 11˚N latitude, 77˚E longitude and an altitude of 426.6m above Mean Sea Level (MSL) in Horticultural college and Research Institute, Tamil Nadu Agricultural University, Coimbatore. The performance of three wild *Solanum* rootstocks *viz.*, *Solanum torvum*, *Solanum sisymbrifolium* and *Solanum capsicoides* grafted on to two commercial tomato hybrid *viz.*, TNAU tomato hybrid CO 3 and Shivam and were evaluated under shade net condition. Fruits of three rootstock species were collected from various places of Tamil Nadu and the seeds are extracted for raising rootstock. Shivam hybrid and TNAU tomato hybrid CO 3 are used as a scion for grafting. The successful tomato grafted plants was planted during Feb - July under insect proof net house for evaluation. The experiment was laid out through Randomized Block Design (RBD) with eight treatments and three replications.

Assessment of Biochemical and quality attributes

Fruits of the three wild rootstocks, tomato fruits from grafted and non-grafted tomato plants were collected at maturity stage and used for analyzing the presence of phytochemicals. Translocation of phytochemicals from the rootstock to scion was also assessed. Quality traits *viz.*, pH, total soluble solids (°Brix), titrable acidity (%), ascorbic acid (mg/100g), lycopene and β-carotene (mg lycopene in 100 g of sample), total phenol (µg/g of sample), total carotenoids, catalase (µg of H_2O_2 min^{-1} g^{-1}), peroxidase (OD at 420 nm min^{-1} g^{-1}), polyphenol oxidase (OD at 495 nm min^{-1} g^{-1}) and solasodine (percentage) was analyzed by following the standard procedures suggested by Ranganna (1986) and Sadasivam and Manickam (1992) [15, 17].

Statistical analysis

The statistical analysis was done by adopting the standard procedures of Panse and Sukhatme (2000) [13]. The critical difference was worked out at five per cent (0.05) probability. The mean analysis was carried out with AGRES software package and MS Excel spreadsheet.

Results and Discussion

Performance of tomato grafted plants on fruit quality parameters

The values of pH were significantly different among all the treatments. Comparing non-grafted plants, Shivam recorded higher level of pH (5.19) whereas TNAU tomato hybrid CO 3 showed pH value of 4.38. Higher TSS content was noticed in the fruits harvested from TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* (5.79˚Brix) rootstock followed by Shivam grafted on to *Solanum sisymbrifolium* rootstock (5.75˚Brix). Shivam grafted on to *Solanum capsicoides* rootstock showed the lowest TSS value (5.50˚ Brix). Graft combination of TNAU tomato hybrid CO3 on to *Solanum sisymbrifolium* rootstock showed high percentage of titrable acidity (0.79%) followed by the combination, Shivam grafted onto *Solanum sisymbrifolium* rootstock (0.69%). Three combinations include TNAU tomato hybrid CO3 and Shivam as a scion with *Solanum capsicoides* as a rootstock and Shivam with *Solanum torvum* as a rootstock showed least percentage of titrable acidity (0.65) than other grafted plants. Significant variations among the total chlorophyll content of different treatments were observed and increase in chlorophyll content was observed in grafted plants than non-grafted plants. Shivam plants and TNAU tomato hybrid CO 3 grafted on *Solanum sisymbrifolium* the showed higher chlorophyll content of 2.81 mg/g and 2.78 mg/g. While, the non-grafted plants *viz.*, Shivam (2.23 mg/g) and TNAU tomato hybrid CO 3 (2.31 mg/g) showed lowest chlorophyll content when compared with the grafted plants (Table.1 and Fig.1).

Most important factor in vegetable production is its nutritional quality. The pH, TSS, titrable acidity, lycopene and ascorbic acid is highly considered for value addition. Fruit size, yield and quality of tomato fruits were affected by scion, but it may be altered by rootstocks.

Titrable acidity was more in TNAU tomato hybrid CO 3 grafted onto *Solanum sisymbrifolium* rootstock. Tomato grafted with suitable rootstocks improved the titrable acidity. This is in accordance with the results observed by (Flores *et al.*, 2010; Turhan *et al.*, 2011) [3, 24]. TSS content were also varies among the grafted plants significantly. TNAU tomato hybrid CO 3 grafted on *Solanum sisymbrifolium* rootstock showed higher TSS content among other grafted plants. Turhan *et al.* (2011) [24] also reported that rootstock influences the TSS content in grafted plants. Similarly, total soluble solids were more in grafted plants of Cecilia on to Beaufort rootstock (Mohammed *et al.*, 2009) [10].In the present study, though there is a significant difference among the treatments, it is noticed that the genetic parameter influence is more pronounced between two varieties when compared to the rootstock characters.

Effect of grafting on fruit quality parameters of grafted and non-grafted plants

Fruits of *Solanum torvum* recorded higher amount of ascorbic acid (66.66 mg/100g) followed by *Solanum sisymbrifolium* (56.23 mg/100g) and *Solanum capsicoides* rootstocks (55.66 mg/100g). Lower amount of ascorbic acid was recorded in Shivam (33.19 mg/100g) and TNAU tomato hybrid CO 3 (33.65 mg/100g). Scion has highly influenced the lycopene content of grafted plants as all the graft combinations showed results on par to that of scion. Highest lycopene content was recorded in Shivam grafted on to *Solanum sisymbrifolium* rootstock (5.69 mg/100g) followed by TNAU tomato hybrid CO 3 on to *Solanum sisymbrifolium* rootstock (5.65 mg/100g). The β carotene (1.07 mg/100g) was also higher in TNAU tomato hybrid CO 3 on to *Solanum sisymbrifolium* rootstock whereas Shivam grafted on to *Solanum sisymrifolium* and *Solanum capsicoides* recorded the least β carotene (1.00 mg/100g) among graft combinations. Among the non-grafted plants, the highest (1.20 mg/100g) and the

lowest (0.98 mg/100g) β carotene content was observed in *Solanum torvum* and *Solanum capsicoides* rootstocks. Out of the different grafted and non-grafted plants, Shivam and TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* rootstock showed higher total carotenoids (6.45 mg/100g and 6.43 mg/100g) and TNAU tomato hybrid CO 3 grafted on to *Solanum capsicoides* rootstock showed least of 6.22 mg/100g of total carotenoids. Then non-grafted *Solanum sisymbrifolium* rootstock (5.72 mg/100g) recorded the highest total carotenoids and *Solanum torvum* rootstock recorded the lowest (4.90 mg/100g) total carotenoids. All the graft combination possessed increased total phenol content comparing non-grafted plants between which, TNAU tomato hybrid CO 3 and Shivam grafted on *Solanum torvum* contain higher amount of total phenol (3.73 mg/100g and 3.72 mg/100g) and both the hybrid plants grafted on *Solanum capsicoides* contain less total phenol (3.13mg/100g each). Among the rootstocks the highest total phenol content (7.74 mg/g) was registered by *Solanum sisymbrifolium* and the lowest (6.02 mg/g) by *Solanum capsicoides* (Table 2 and Fig.2). Increased lycopene content was noticed in grafted watermelon (Salam *et al*., 2002) [18]. The highest ascorbic acid content was noticed in Shivam and TNAU tomato hybrid CO 3 grafted on *Solanum sisymbrifolium*. Zhu *et al.* (2006) [27] and Zhang *et al.* (2019) [26] revealed that grafting with suitable rootstock increased the ascorbic acid content. In addition, 10.59% increase in ascorbic acid content was noticed in grafted brinjal than non-grafted brinjal (Na *et al.*, 2012) [11]. But, it was reduced by grafting as per the study conducted by Turhan *et al.* (2011) [24] in tomato. In this study, mild increase in ascorbic acid content was noticed in tomato plants grafted with all the three rootstocks than the non-grafted plants.

Lycopene content is the most important factor that determines the colour of the fruit, which plays a major role in market value of the commodity. Total carotenoid content of the tomato fruit contains mostly lycopene and β carotene. In most of the cases the lycopene content remains the same in grafted and non-grafted plants. In agreement with this result, (Romano and Paratore (2000) [16]; Khah *et al.* (2006) [7]; Turhan *et al.* (2011) [24] also found that no significant differences among the grafted and non-grafted plants for lycopene content. Decrease in lycopene content was also found in grafted plants by (Mohammed *et al.*, 2009) [10]. Slight difference in pH value was found among the grafted plants. This was in contrary to the results given by Khah *et al.* (2006) and Turhan *et al.* (2011) [7, 24] where no difference in pH was identified in grafted plants. From these observations, it could be concluded that significant differences were not noticed among the grafted and non-grafted plants in terms of lycopene, β carotene and total carotenoids content. However, it could be clearly visualized that, though the rootstocks have significant level of ascorbic acid and total phenols, when compared to non-grafted tomato plants, it does not have pronounced effect on ascorbic acid and total phenol content of tomato grafted plants. Hence, the fruit quality parameters of grafted plants cannot be altered by rootstocks through translocation while grafting. It indicates that the quality is determined by scion not by the rootstock. These results are in line with Sherly (2011) [19] in brinjal and Dhivya (2013) [1] in Tomato.

Effect of grafting on the antioxidant enzyme properties

The immature and mature fruits of Shivam (0.84 g/min and 0.53 g/min) and TNAU tomato hybrid CO 3 (0.77 g/min and 0.46 g/min) showed less peroxidase activity than the grafted combination. This indicates the influence of rootstock on peroxidase activity of grafted plants. Three wild rootstocks were analyzed for peroxidase activity and results revealed that *Solanum sisymbrifolium* rootsotck had higher peroxidase activity (5.32 g/min) followed by *Solanum torvum* rootsotck (4.41 g/min). Out of three rootstocks *Solanum sisymbrifolium* showed increase in catalase activity in both immature and mature rootstocks (8.19 µg of H_2O_2/g/min and 7.60 µg of H_2O_2/g/min) followed by *Solanum torvum* (7.83 µg of H_2O_2/g/min and 6.40 µg of H_2O_2/g/min). The least activity was observed in *Solanum capsicoides* (5.65 µg of H_2O_2/g/min and 4.26 µg of H_2O_2/g/min). However, *Solanum capsicoides* (0.1 µg of H_2O_2/g/min) showed less activity of catalase in leaf comparing the three rootstocks. Non-grafted TNAU tomato hybrid CO 3 showed catalase activity of 1.93 µg of H_2O_2/g/min, which is lesser than Shivam (2.12 µg of H_2O_2/g/min) (Table 3).

The least Polyphenol oxidase (PPO) activity was recorded in TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* rootstock (0.37 g/min). Comparing non-grafted hybrids, Shivam recorded higher (0.32 g/min) than TNAU tomato hybrid CO 3 (0.34 g/min). Catalase and peroxidase activity was more in grafted plants than the non-grafted hybrid. At ten days after grafting, catalase activity was more in Shivam grafted on to *Solanum sisymbrifolium* rootstock, whereas peroxidase activity was more in TNAU tomato hybrid grafted on *Solanum torvum* rootstock. Present findings are in line with the findings of Tamilselvi *et al.* (2016) [22] and Tamilselvi and Pugalendhi (2017) [21].

Grafting increases the level of Reactive Oxygen Species (ROS) that stimulate defence antioxidant enzymes *viz.*, catalase and peroxides. Catalase and peroxidase eliminate ROS and helps in wound healing at graft union. More stress was produced in grafted plants due to grafting. Less activity of antioxidant enzymes was observed in non-grafted plants than the grafted plants (Xu *et al*., 2005) [25]. Similar results were reported by Gulen *et al.* (2002) [5]. Fernandez Garcia *et al.* (2004) [2], observed increase in catalase and peroxidase activity at 4th day of grafting during which the xylem differentiation at the graft union takes place by lignifications.

Fruit peroxidase activity helps in protecting the membrane integrity by inhibiting the breakdown of lipid and delaying the ethylene action (Olaiya, 2010) [12]. Peroxidase activity was increased in the fruits of grafted plants comparing then non-grafted plants. Presence of peroxidase activity helps in resistance to plant diseases. Solasodine content was significantly different among the fruits of grafted, non-grafted and rootstock species. The least solasodine content was found in TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock and Shivam grafted on to *Solanum sisymbrifolium* rootstock (0.019%). Solasodine content was higher in rootstock species compared to grafted and non-grafted plants. It indicates that the solasodine content was scion specific in grafted tomato plants. Similarly, Solasodine content in brinjal grafted with *Solanum viarum* revealed that the solasodine content in grafted plants was scion specific (Srinivas and Krishnan, 1996) [20]. Sherly (2011) [19] endorsed similar results in brinjal when grafting with *S.torvum* rootstock. Contrary to this study solasodine content in eggplant was increased when grafted with wild *Solanum* species which was due to the more adsorption efficiency of species used as rootstock than cultivated eggplant (Kumar *et al.*, 2017) [9].

Table 1: Performance of tomato grafted plants on fruit quality parameters

Treatments	pH	TSS (Brix)	Titrable acidity (%)	Total chlorophyll (mg/g)
TNAU tomato hybrid CO3 grafted on to *Solanum torvum* rootstock (T_1)	4.63	5.63	0.67	2.59
TNAU tomato hybrid CO3 grafted on to *Solanum sisymbrifolium* rootstock (T_2)	4.77	5.79	0.79	2.78
TNAU tomato hybrid CO3 grafted on to *Solanum capsicoides* rootstock (T_3)	4.49	5.54	0.65	2.50
Shivam grafted on to *Solanum torvum* rootstock (T_4)	5.33	5.61	0.65	2.62
Shivam grafted on to *Solanum sisymbrifolium* rootstock (T_5)	5.63	5.75	0.69	2.81
Shivam grafted on to *Solanum capsicoides* rootstock (T_6)	5.27	5.50	0.65	2.47
TNAU tomato hybrid CO 3 (T_7)	4.38	5.51	0.66	2.31
Shivam(T8)	5.19	5.46	0.64	2.23
S. Ed	0.13	0.14	0.01	0.06
CD	0.29	0.31	0.04	0.14

Table 2: Effect of grafting on fruit quality parameters of grafted and non-grafted plants

Treatments	Ascorbic acid (mg/100g)	Lycopene (mg/100g)	β carotene (mg/100g)	Total carotenoids (mg/100g)	Total phenols (mg/g)
TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock (T_1)	35.44	5.51	1.07	6.37	3.73
TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* rootstock (T_2)	37.66	5.65	1.01	6.43	3.67
TNAU tomato hybrid CO 3 grafted on to *Solanum capsicoides* rootstock (T_3)	36.14	5.55	1.03	6.22	3.13
Shivam grafted on to *Solanum torvum* rootstock (T_4)	36.35	5.53	1.02	6.34	3.72
Shivam grafted on to *Solanum sisymbrifolium* rootstock (T_5)	37.92	5.69	1.00	6.45	3.54
Shivam grafted on to *Solanum capsicoides* rootstock (T_6)	36.34	5.57	1.00	6.37	3.13
TNAU tomato hybrid CO 3 (T_7)	33.65	5.51	0.99	5.85	2.45
Shivam(T8)	33.19	5.54	1.08	5.91	2.23
Rootstock					
Solanum torvum	66.66	0.43	1.20	4.90	6.49
Solanum sisymbrifolium	56.23	5.63	0.98	5.72	7.74
Solanum capsicoides	55.66	5.11	1.04	5.53	6.02
S. Ed	1.14	0.14	0.02	0.16	0.13
CD	2.41	0.29	0.05	0.34	0.27

Table 3: Effect of grafting on the antioxidant enzyme properties

Treatments	Peroxidase (absorbance at 430 nm /g/min)			Catalase (µg of H_2O_2 /g/min)			Polyphenol oxidase (OD /g/ min)		Solasodine of fruits (%)
	Leaf (90 DAG)	Immature	Mature	Leaf (90 DAG)	Immature	Mature	Fruits	Leaf	
TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock (T_1)	1.32	1.34	0.80	2.31	1.67	0.96	0.29	0.45	0.019
TNAU tomato hybrid CO 3 grafted on to *Solanum sisymbrifolium* rootstock (T_2)	1.76	1.23	0.75	2.45	1.89	1.32	0.21	0.37	0.020
TNAU tomato hybrid CO 3 grafted on to *Solanum capsicoides* rootstock (T_3)	1.23	1.20	0.64	1.96	1.45	0.86	0.28	0.44	0.022
Shivam grafted on to *Solanum torvum* rootstock (T_4)	1.66	1.32	0.91	2.57	1.92	1.26	0.26	0.47	0.028
Shivam grafted on to *Solanum sisymbrifolium* rootstock (T_5)	1.71	1.26	0.87	2.62	1.77	1.19	0.20	0.39	0.019
Shivam grafted on to *Solanum capsicoides* rootstock (T_6)	1.11	1.17	0.78	2.22	1.56	0.92	0.24	0.40	0.024
TNAU tomato hybrid CO 3 (T_7)	1.76	0.77	0.46	1.93	1.26	1.63	0.10	0.32	0.020
Shivam (T_8)	1.84	0.84	0.53	2.12	1.34	1.12	0.17	0.34	0.023
Rootstocks									
Solanum torvum	4.41	5.67	4.82	6.87	7.83	6.40	2.54	4.38	1.740
Solanum sisymbrifolium	5.32	5.59	4.79	6.77	8.19	7.60	2.21	3.12	1.230
Solanum capsicoides	3.12	4.98	4.12	4.89	5.65	4.26	2.16	2.20	1.321
S. Ed	0.08	0.08	0.39	0.11	0.12	0.10	0.03	0.05	0.021
CD	0.16	0.18	0.83	0.23	0.25	0.22	0.07	0.10	0.044

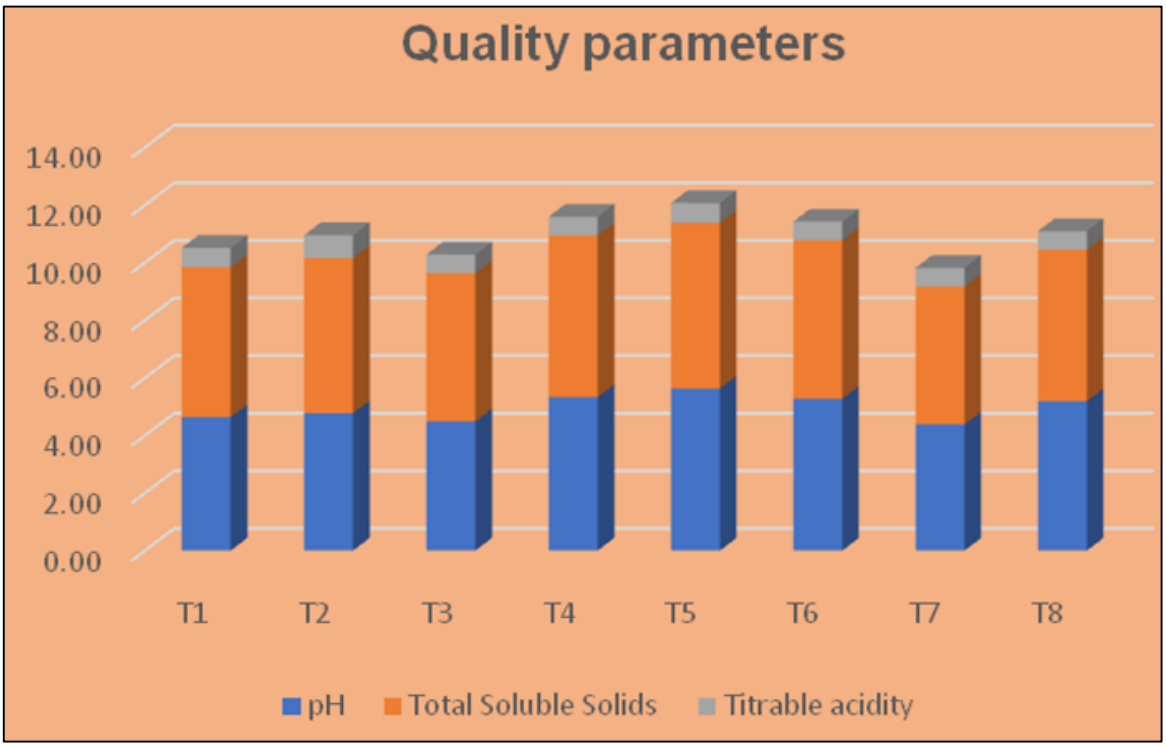

Fig 1: Performance of tomato grafted plants on fruit quality parameters

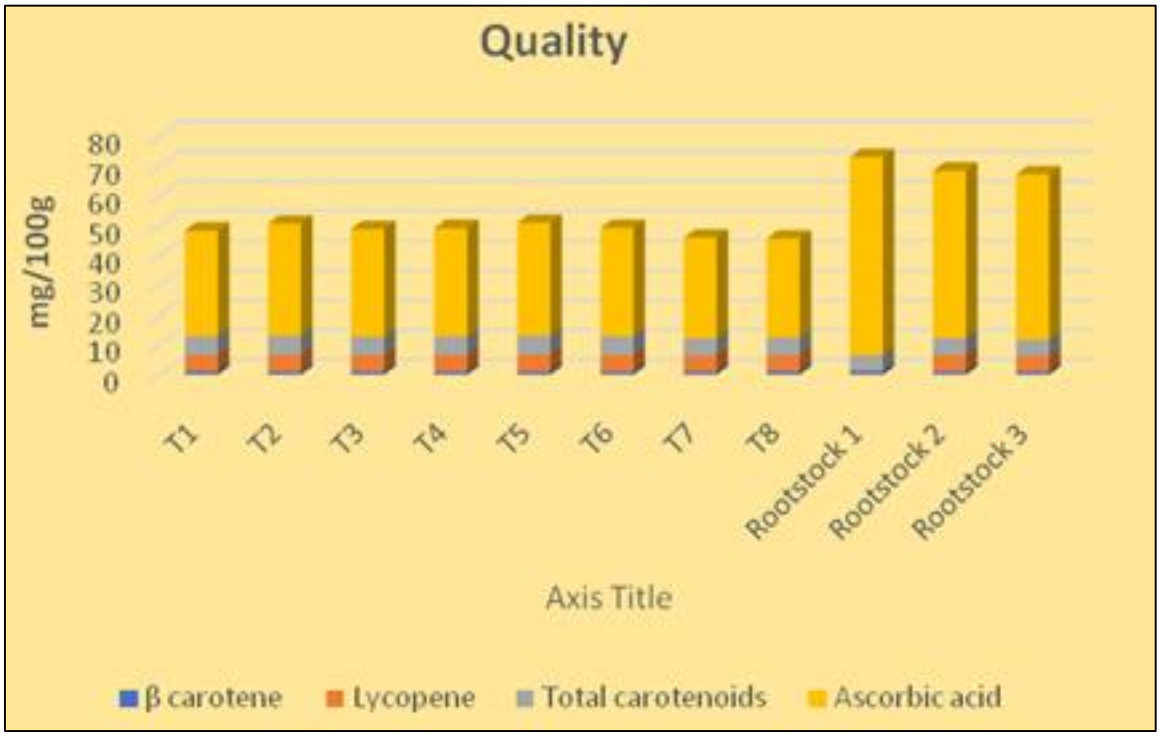

Fig 2: Effect of grafting on fruit quality parameters of grafted and non-grafted plants

Conclusion

The results of the experiment revealed that TSS (5.79 ℃Brix) and titrable acidity (0.79%) was superior in TNAU tomato hybrid CO 3 grafted on *Solanum sisymbrifolium* rootstock. The ascorbic acid (37.92 mg/100g), total chlorophyll (2.81 mg/g), pH (5.63) and lycopene (5.69 mg/100g) was high in Shivam grafted on to *Solanum sisymbrifolium* rootstock. Similarly, total phenol content was high in TNAU tomato hybrid CO 3 grafted on to *Solanum torvum* rootstock (3.73 mg/g). Catalase activity was more in immature fruits of Shivam grafted on *Solanum torvum* rootstocks (1.92 g/min) and in mature fruits of TNAU tomato hybrid CO 3 grafted on *Solanum sisymbrifolium* (1.32 g/min) rootstock. Among the graft combinations solasodine content was observed more in Shivam grafted on to *Solanum torvum* rootstock (0.028%). In the present study it could be concluded that though there is a marginal improvement in the different parameters studied the ultimate quality is determined by the scion only not by the rootstocks.

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