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RESEARCH ARTICLE

Grafting onto Different Rootstocks Influences Yield, Quality, Chemical Composition, and Bioactivities of Tomato Fruits under Greenhouse Conditions

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Abstract

Objective: Grafting is a possible chemical free solution that shows several changes in plants, with the aid of vigorous rootstock. Methods: The influence of grafting on the yield and fruit quality of tomato cultivars grown under greenhouse condition was conducted during two successive seasons of 2016 and 2017. The metabolic composition of fruits of tomato *Solanum lycopersicum* L. Nefret hybrid (T1) grafted onto four rootstocks were identified by gas chromatography coupled to mass spectrometry (GC-MS). The levels of oxidative stress markers; malondialdehyde (MDA), superoxide dismutase (SOD), and glutathione (GSH) were determined in the rat's liver tissue. The levels of aspartate and alanine aminotransferases and alkaline phosphatase (AST, ALT and ALP) were analyzed for the determination of liver functions. Kidney function parameters of serum urea and creatinine levels were also measured. Histopathological examinations of the liver and kidney tissues were also performed. Results: The effects of grafting on the vegetative growth, yield, and fruit quality characteristics, *i.e.*, fruit weight, length, diameter, total soluble solids content and titratable acidity of T1, grafted onto four rootstocks have showed great variability. The vegetative growth, *i.e.*, plant height of T2, was increased when *S. lycopersicum* L. var. VFN (T2) was used as a rootstock. T2 has showed no adverse effects in all biochemical parameters under investigation compared by the other treatments. Aldehydes, ketones, esters, fatty acids, terpenoids, carotenoids, phenolics and alkaloids were the main identified components and the major compound was ß-ionone (13.37%) in T2. Conclusion: The nutritional content of T2 and that it has showed no alteration in oxidative stress markers, liver and kidney function tests and their histopathological patters support its safety. While, un-grafting or grafting onto *Solanum melongena* L. cv. Balady (T3) may be a tool to improve the crop quality.

Keywords: *Greenhouse-grown tomato, Grafting, GC-MS, Carotenoids, Lycopene, Polyphenols, Liver, Kidney, Histopathology.*

Introduction

Grafting onto resistant rootstocks was first introduced to vegetable production in Japan and Korea in the late 1920s as a strategy against Fusarium wilt and other diseases [1]. In North America, innovations in Tomato grafting occurred during the 1930s and 1940s when tomato was grafted onto jimson weed (*Datura stramonium* L.) as a method for root-knot nematode control. Grafting commercial cultivars onto desirable rootstocks represents fast, easy and successful strategy to cope such drastic effects as well as improve yield and fruit quality [2]. Grafting was reported to enhance tolerance of plants to heavy metals [3], improving nutrient uptakes so that the plants becomes more efficient uptake and use of water and plant nutrients and subsequently an increase in plant strength [4].Grafting is agronomically important because one can combine desirable aboveground characteristics (such as fruit size) and underground characteristics (such as resistance to soil-borne diseases).Grafting increases the economic harvesting period and

a parallel increase in yield and a reduction in the use of agricultural chemicals [5]. Also grafting has other advantages as it can solve many agricultural problems much faster than plant breeding programs which cost much money and need long time [6], beside enhancing nutrient uptake [4], increases synthesis of endogenous hormones [7] and improves water use efficiency [8]. The main purpose of grafted seedlings is to increase the yield and quality of fruits by combining a disease resistant rootstock with a genetically superior scion [9].

The quality characteristics might be affected by grafting as a result of the translocation of metabolites associated with fruit quality to the scion through the xylem and/or modification of physiological processes of the scion [8]. Rootstock/scion combination must be carefully chosen for optimal fruit quality [10]. The crop type, growers experience and preference, available facilities, grafts number, grafting cost and the purpose of grafting are a critical issue for grafting technique must be used [11].

The success in grafting technique mainly depends on the appropriate choice for scion/ rootstock combinations, using of proper grafting method and grafts maintaining [12]. Now the use of grafted seedlings has been increasingly popular in the production of many fruit-bearing vegetables such as watermelon, cucumber, oriental melon, muskmelon, tomato, eggplant, and red pepper.

Recently, Egypt had a strong competition in grafting industry to provide grafted Solanaceous crops with high quality and better performance to growers [13]. The tomato (*Solanum lycopersicum*) and its wild relatives (genus Solanum, section Lycopersicon) originated in western South America. The name *Lycopersicon esculentum* breaches the International Code of Botanical Nomenclature (ICBN) was widely used until recently.

Today, the tomato is one of the major vegetable crop plants in the world. Currently, more than 120 million tons of tomatoes are produced annually worldwide. The biological activities of Solanum species are due to the presence of different chemical constituents as alkaloids, polyphenols and carotenoids. Also, tomatoes are the main source of lycopene, in addition to vitamin C, vitamin E, pro-vitamin A carotenoids and ß -carotene. Pharmacological evaluation of bioactive substances which could be moved from the rootstocks to scion and hence to fruits and their relation to human health has recently recorded a growing interest amongst researcher's worldwide [14].However, while voluminous pharmacological studies have been conducted to ascertain the uses of various plants, very few plants have been thoroughly evaluated for their detrimental effect. Reports of efficacy are, by far, more numerous than those on toxicity [15].

Therefore, a need to further investigation of plants to incorporate the observations of short and long-term toxicity especially in the most important organs in the body like liver and kidney must be taken into consideration. This study aimed to evaluate the effect of rootstocks on the growth, yield, quality and chemical constituents of tomato plants grown under greenhouse condition and moving of bioactive substances from scion to fruits through biochemical investigation in a rat model.

Materials and Methods

Plant Materials

Two field experiments were carried out during two successive seasons of 2016 and 2017 at Kaha Research Station, Horticulture Research Institute, Agricultural Research Centre, Ministry of Agriculture, Egypt. Four rootstocks were examined in this experiment beside a tomato hybrid was used as scion (*Solanum lycopersicum* L. Nefret hybrid) (T1).

The tested rootstocks were: tomato (*Solanum lycopersicum* L. var. VFN) (T2), eggplant (*Solanum melongena* L. cv. Balady) (T3), Tomato (*Solanum lycopersicum* L. var. VF) (T4), and Datura plant (*Datura stramonium* L.) (T5). Seeds of rootstocks *Datura stramonium* L. and *Solanum melongena* L. *cv*. Balady were sown two weeks earlier in July to get seedlings. While seeds of tomato rootstock VFN, VF and scion were sown at the same time in the last week of July.

Seeds of all rootstocks, and scion were sown in seedling trays (84 cells). After 45 days from sowing the stems of the scion and rootstocks were cut at right angles leaving 2-3 leaves depending on temperature and

tapered stems of the scion were placed into the cleft of the cut-end of the rootstocks, followed by clipping [16]. Seedlings were placed under a plastic tunnel. After 4-5 days, plastic of the tunnel was gradually opened for adaptation. The agricultural operations were applied according to the recommendation of the Ministry of agriculture and Land Reclamation of Egypt. The plants were arranged in completely randomized design with three replicates.

Agricultural Determinations

Vegetative Growth

Physical Parameters

Vegetative growth parameters were determined using five plants randomly chosen from each experimental plot as follow: plant height (cm) was determined after 60 and 180 days from transplanting, stem thickness of union zone (cm), and plant dry weight (%).

NPK Content and Total Chlorophyll in Leaves

Mature non-senescent leaf samples from 5th node from apex were taken to determine nitrogen, phosphorus and potassium contents. Plant leaves samples were oven dried at 60°C. After drying, samples were ground using a pestle and a mortar for determination of mineral composition. Ash of the plant samples was digested using the H_2SO_4 and H_2O_2 [17].

The total nitrogen concentration (%) was determined using the modified micro-Kjeldahl method [18]. The total concentrations (%) of phosphorus and potassium using spectrophotometer with the technique of the ascorbic acid method and flame photometer, respectively were carried out [19, 20]. The total chlorophyll content in the 5th leaves using Minolta Chlorophyll Meter SPAD- 501 as SPAD unit was also determined [21].

Fruit Characteristics

Physical Parameters

A random sample of 10 fruits from each plot was randomly chosen to determine the average fruit weight (g), fruit length (cm), diameter (cm), fruit firmness (kg/cm^2) of each individual tomato fruit was measured at two points of the equatorial region by using a pressure (Digital force-Gouge Model FGV-0.5A to FGV-100A. Shimpo instruments).

Yield Parameters

Early yield per plant (kg) was calculated as the total fresh weight of fruits harvested from the first fourth pickings. Total yield per plant: All tomato fruits reached to the pink stage after six weeks from transplanting were picked weekly through the harvesting period and weighted.

Chemical Parameters

Total soluble solids percentage (TSS %) was determined using hand Refractometer [22]. Total titratable acidity (g citric/100 g fresh weight) was determined by titration with 0.01 N Na OH using phenolphthalein as indicator [22]. Vitamin C (ascorbic acid) (mg/100 g f.w.) was estimated by titration with 2, 6-dichlorophenolindophenol dye [22].

Quantitative Determination of Bioactive Metabolites

Preparation of Samples for Analysis

The fresh fruits of each grafted plant and the un-grafted (200 g) were extracted separately with CH_2Cl_2 for 3 hrs, on an orbital shaker in the dark at room temperature. Each extract was separated by centrifugation (13,000 ×g, 10 minutes). The supernatant was taken, the residue was re-suspended in 50 mL of the same solvent, and the mixture was again separated by centrifugation.

The two resulting supernatants were then combined and concentrated under reduced pressure at 40°C. Each residue was dried over anhydrous sodium sulfate filtered, and stored at 4 °C in a sealed vial for further analysis till dryness to get 0.39, 0.42, 0.38, 0.40, and 0.51 g of the crude extracts of the four rootstocks (T2-T5) and the control (T1), respectively [23].

Gas Chromatography Coupled to Mass Spectrometry (GC-MS)

The chemical constituents of the four rootstocks (T2 -T5) and the control (T1) were analyzed by GC-MS. GC-MS was carried out using total ion monitoring mode on a Finnigan mass Spectrometer model SSQ 7000 equipped with library software Wiley 138 and NBS 75. Capillary DB-5 (methyl polysiloxane containing 5% phenyl groups) column 25 m x 0.25 mm i.d. was used.

The initial column temperature was started at 60°C for 2 min., programmed at 60-100°C (2°C/min) and 100-250°C (5°C/min). Helium was used as the carrier gas at a constant flow rate of 1.0 mL/min. Injection voltage 70 eV was used. Molecular ions (scan mass range: 40-450 mz^{-1}) were monitored for identification. The identification of the separated compounds was based on their retention indices, relative to a homologous series of *n*-alkane (C8-C20) on the DB-5 capillary column under the same operating conditions and computer matching with GC/MS spectra from the library software data and those reported in literature [24].

Chemicals

All chemicals were of high analytical grade, products of Merck, Germany and Sigma, USA.

Biological Determinations

Animals and Ethics

Male Wistar albino rats (100-120 g) were selected for this study. They were obtained from the Animal House, National Research Centre in Egypt. All animals were housed in environmentally controlled condition with free access of water and diet. They were kept one week for acclimatization before starting the experiment. Anesthetic procedures and handling with animals complied with the ethical guidelines of Medical Ethical Committee of the National Research Centre in Egypt to ensure that animals do not suffer at any stage throughout the experiment.

Acute Toxicity

Ninety male rats were divided into 15 groups (6 rats each). Each group was orally administered with one kind of tomato (100, 250 and 500 mg/kg bode weight) and observed after 24 hours. No dead animals were observed along the acute toxicity test for different concentrations. Therefore, the recommended dose was 250 mg/kg for studding the chronic effects and for further biological determinations.

Chronic Toxicity

Sixty rats were divided into six groups (10 rats each). Group T0: rats were given daily oral dose of 0.5 mL distilled water for two months and served as control group. Groups T1-T5: rats were given daily oral dose of 0.5 mL of each kind of tomato with a dose of 250 mg/kg for two months.

Sample Preparation

Blood was collected from each animal by puncture of sublingual vein in clean and dry test tubes, left 10 minutes at room temperature to clot, and centrifuged at 3000 rpm for serum separation. The separated serum was stored at -80°C for further determinations of liver and kidney function tests. Liver tissue was homogenized in cold 0.9 N NaCl (1: 9 w/v) solution, centrifuged at 3000 rpm for10 minutes, separated from the supernatant and stored at−80°C for further antioxidant determinations.

Biochemical Assays

The hepatic oxidative stress markers; malondialdehyde (MDA), superoxide dismutase (SOD), and glutathione (GSH) were estimated [25]. Serum aspartate and alanine aminotransferases and alkaline phosphatase [26] were estimated by biodiagnostic kit (Biogamma, Stanbio, West Germany). Serum urea and the creatinine were also measured [27].

(f)- Histopathological Analysis

Liver and kidney tissues were fixed in 10 % formalin. Paraffin embedded samples were prepared for sectioning at 4-µm thickness. Slides were stained with hematoxylin and eosin and examined by light microscope [25, 28].

Statistical Analysis

Data of the agricultural study were subjected to standard analysis of variance procedure [29]. The least significance difference (LSD) between groups was calculated whenever F values were significant at 5% level. Data of the biological study were done by using one-way analysis of variance (ANOVA), CoStat software Computer Program accompanied with LSD between groups at $P < 0.05$.

Results and Discussion

Agricultural Determinations Study

Vegetative Growth

Physical Parameters

According to the obtained results, in Table (1) it could be illustrating that grafting of tomato plants had significantly increased the physical parameters of vegetative growth

(*i.e.*, plant height, stem thickness, and dry weight) as compared to control (un-grafted) ascribable which gave the lowest data. Tallest plants pointed clearly with the grafted tomato Nefret hybrid (T1) onto VFN (T2) rootstock in both the periods, i.e. 60 and 180 days after transplanted. While grafting tomato Nefret hybrid onto eggplant (T3) rootstock showed significant increment and recorded the highest results of both stem thickness and dry weight. This trend is true for both seasons of the study as shown in Table (1). These results are supported by the findings of [30] who suggested that superior growth of grafting may be due to promote the movement of water and nutrients from rootstock to scion as a result of the better development of vascular bundles which depends on the good adhesion between rootstock and scion.

Table 1: Effect of different rootstocks onto vegetative growth of tomato plants

Rootstocks	Plant height (cm) after 60 days	Plant height (cm) after 180 days	Stem thickness (cm)	Dry weight (%)
First season				
T1	83.30	206.67	2.20	25.17
T2	92.87	275.67	2.40	25.23
T3	88.27	235.67	2.63	25.83
T4	90.90	258.00	2.23	25.27
T5	89.23	251.67	2.57	25.50
LSD 5%	1.12	8.50	0.06	0.10
Second season				
T1	89.97	238.08	2.24	22.01
T2	94.72	281.18	2.45	30.97
T3	81.71	256.20	2.68	29.22
T4	92.63	262.90	2.28	28.07
T5	90.95	210.63	2.62	25.48
LSD 5%	0.99	5.35	0.04	0.08

T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. Var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4: Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.)

NPK Content and Total Chlorophyll in Leaves

Regarding to the effect of rootstocks on NPK contents, data stated that positive effect was detected among rootstocks however, all grafted plants contained high percentage of NPK in their leaves especially when the plants were grafted onto Datura (T5) rootstocks which produced the highest results followed in descending order by VFN (T2), VF (T4) and eggplant (T3) compared to those grown without un-grafted (T1) which gave the worst results as shown in Table (2) The effect of the rootstock on the mineral composition of plant leaves was principally explained with physical characteristics of the root system, such as lateral and vertical development, which resulted in enhanced uptake of water and minerals [30].

Suggested that the uptake and/or utilization efficiency of macronutrients by plants may be enhanced by grafting onto some rootstocks. This is ascribed mainly to the root characteristics of these rootstocks, which are more vigorous than those of the highly productive cultivated varieties. Many studies revealed that some graft combinations were significantly more efficient in absorbing and indeed, transporting nutrients to the shoot, such as phosphorus, nitrogen, potassium, magnesium, calcium, iron, or other micronutrients, in comparison with un-grafted plants [31].

Concerning the total chlorophyll content in leaves, data in Table (2) reveal that, tomato plants grafted onto different rootstocks were significantly sufficient to encourage the capability of these plants to enrich the content of total chlorophyll in their leaves. On the other side, Datura (T5) rootstock was significantly the superior one rootstock which gave the highest content as compared with other rootstocks or those of un-grafted plants that showed the lowest results which were in agreement with other study [32].

Grafting can improve net photosynthesis rate and enhance assimilate accumulation and thus enhancing growth potential and dry

matter accumulation in roots, stems and leaves. Grafting, which improves stomata conductance and intercellular CO_2 concentration, will strengthen the transfer capability of photosynthetic substrates and the supply capability of photosynthetic materials to ensure increased photosynthesis efficiency [5].

Table 2: Effect of different rootstocks onto NPK nutrients and total chlorophyll content of tomato plants

Rootstocks	Nitrogen (N)%	Phosphorus (P)%	Potassium (K)%	Total Chlorophyll (Spad)
First season				
T1	3.72	1.87	3.84	42.60
T2	4.36	2.33	4.31	49.45
T3	4.11	2.09	3.76	47.50
T4	4.34	2.24	4.24	49.40
T5	4.73	2.84	4.80	49.97
LSD 5%	0.04	0.08	0.08	0.03
Second season				
T1	3.79	1.88	4.39	50.33
T2	4.45	2.11	3.92	50.97
T3	4.18	2.29	4.32	47.40
T4	4.44	2.18	3.83	48.40
T5	4.82	2.80	4.89	43.42
LSD 5%	0.15	0.08	0.17	0.60

T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4: Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.)

Fruit Characteristics

Physical and Yield Parameters

As regard to the effect of different rootstocks on yield and its components, data presented in Table (3) clear that grafted tomato plants onto different rootstocks under investigation significantly increased both early and total yield more than un-grafted plants especially when using VFN as a rootstock which showed superiority results[33] Our results in grafted and un-grafted tomato plants, were matched with another study suggested that the higher yield of fruit from grafted tomato plants was most likely an effect of the vigorous root system of the rootstock [34]. The increased yield of grafted plants is also believed to be due to enhanced water and mineral uptake. The increment in the early and total yield may be attributed to improvement of the increase of nutrient uptake and water use efficiency [35]. Moreover, the differences in yield between grafted and un-grafted (control) plants may be explained by the increment of the fruit weight. On the other side, there were no significant differences among rootstocks with respect to fruit length, fruit diameter and fruit firmness in both seasons under investigation.

Table 3: Effect of different rootstocks onto yield and fruit quality of tomato fruits

Treatments	Early yield (kg/plant)	Total yield (kg/plant)	Fruit weight (g)	Fruit length (cm)	Fruit diameter (cm)	fruit firmness (cm)
First season						
T1	2.55	4.70	156.76	6.00	7.00	9.60
T2	2.97	6.07	179.87	6.17	7.63	8.77
T3	2.28	5.82	173.97	5.50	7.20	9.83
T4	2.89	5.97	174.33	7.13	6.53	8.90
T5	2.80	6.02	162.14	6.00	6.73	9.70
LSD 5%	0.13	0.32	0.94	-	-	-
Second season						
T1	2.59	5.93	159.7	6.11	7.13	9.78
T2	3.03	6.19	183.47	6.29	7.79	8.94

T3	2.32	4.78	177.10	5.60	7.33	10.0
T4	2.94	6.09	178.65	7.27	6.66	9.0
T5	2.95	6.14	166.59	6.12	6.86	9.89
LSD 5%	0.06	0.12	1.12	-	-	-

T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4: Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.)

Chemical Parameters

The obtained results indicate that total soluble solids (TSS) and vitamin C were significantly influenced by different rootstocks. The greatest TSS content in tomato fruits was recorded in grafted tomato Nefret onto VF (T4) meanwhile, the highest vitamin C values were observed in grafted tomato Nefret onto VFN (T2) rootstock as presented in Table (4). A similar mention was recorded that the type of rootstock significantly influenced the characteristics defining fruit quality [36]. In our current study, there were no significant differences between grafted tomato plants onto different rootstocks and those un-grafted without grafting in the terms of titratable total acidity in fruits in the two growing seasons (Table 4).

Table 4: Effect of different rootstocks onto TSS, vitamin C and titratable total acidity of tomato fruits

Rootstocks	TSS %	Vitamin C.%	Acidity%
First season			
T1	5.00	21.20	0.62
T2	5.60	29.77	0.62
T3	5.43	28.20	0.75
T4	5.63	27.03	0.63
T5	4.90	24.53	0.66
LSD 5%	0.62	1.82	-
Second season			
T1	5.09	21.60	0.63
T2	5.71	30.36	0.64
T3	5.53	28.71	0.68
T4	5.74	27.55	0.64
T5	4.99	25.00	0.67
LSD 5%	0.07	0.69	-

T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4: Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.)

The Effect of Grafting on the Four Rootstocks on Chemical Composition of Grafted and un-grafted Plants

The chemical constituents of the total ethanolic extracts of different rootstocks of tomato (T2-T5) and un-grafted (T1) plants were identified by GC/MS technique. The results (Tables 5-9) revealed that forty-seven compounds were identified from the different rootstocks and the un-grafted plants of tomato. GC/MS analysis allowed the identification of 98.53 % (Table 6), 98.24% (Table 7), 98.63% (Table 8), 98.85% (Table 9) of the total crude extract of T2, T3, T4 and T5 compared with 97.03% of the un-grafted plants (T1) (Table 5).

Table 5: GC/MS analysis of the total ethanolic extract of T1 (*Solanum lycopersicon* L. Nefret hybrid) as the control group

RT	Compound Name	Area %	MW	Formula
7.17	*cis*-3-Hexenal	1.04	98	$C_6H_{10}O$
9.66	*cis*-2-Hexenal	1.98	98	$C_6H_{10}O$
11.07	*trans*-2-Hexen-1-al	1.93	100	$C_6H_{12}O$
15.29	Heptanal	1.83	100	$C_6H_{12}O$
18.50	*trans*-2-Heptenal	1.34	112	$C_7H_{12}O$
27.62	Linalool	0.90	154	$C_{10}H_{18}O$
29.93	Camphor	0.88	152	$C_{10}H_{16}O$
30.34	O,N-Diacetyl tomatidine	0.92	499	$C_{31}H_{49}NO_4$
31.95	α-Terpineol	0.97	154	$C_{10}H_{18}O$
32.85	Cantaxanthin	0.97	564	$C_{40}H_{52}O_2$
33.02	Decanal	0.91	156	$C_{10}H_{20}O$

33.77	*p*-Menth-1-en-9-al	1.02	156	$C_{10}H_{20}O$
34.00	Oleyloleate	1.03	532	$C_{36}H_{68}O_2$
35.74	Methyl palmitate	20.56	270	$C_{17}H_{34}O2$
36.56	2-Decenal	3.67	154	$C_{10}H_{18}O$
37.03	7-(Hydroxymethyl)-2-methoxy-xanthone	2.02	256	$C_{15}H_{12}O_4$
37.22	*iso*-Propyl-14-methyl pentadecanoate	19.22	298	$C_{19}H_{38}O_2$
38.97	Ricinoleic acid, methyl ester, acetate	5.76	354	$C_{21}H_{38}O_4$
39.10	Linolenic acid, methyl ester	3.64	402	$C_{24}H_{34}O_5$
39.53	Methyl stearate	2.63	298	$C_{19}H_{38}O_2$
40.21	9, 12-Octadecadienoic acid (Z, Z).	7.44	280	$C_{18}H_{32}O_2$
41.04	Eugenol	0.84	164	$C_{10}H_{12}O_2$
41.75	Lycopene	1.02	536	$C_{40}H_{56}$
42.46	9-Octadecenoic acid -2-(octadecyloxy) ethyl ester	1.10	578	$C_{38}H_{74}O_3$
42.52	Zeaxanthin	0.98	568	$C_{40}H_{56}O_2$
46.42	ß-Ionone	8.39	192	$C_{13}H_{20}O$
49.94	ß-Methylionone	1.21	206	$C_{14}H_{22}O$
50.79	Benzophenone	1.06	182	$C_{14}H_8O_2$
53.16	Lycoxanthin	0.99	552	$C_{40}H_{56}O$

Methyl palmitate represented the major component, 20.56% followed by *iso*-propyl-14-methyl pentadecanoate (19.22%). Camphor (0.88%) was the minor compound (Table 5). β-Ionone represented the major component 12.34% of the rootstock T2 where O, N-diacetyl tomatidine (0.82%) was the minor compound (Table 6).

Table 6: GC/MS analysis of the total ethanolic extract of T2 (*Solanum lycopersicon* L. Nefert hybrid onto *Solanum lycopersicon* var. VFN)

RT	Compound Name	Area %	MW	Formula
7.30	*cis*-3-Hexenal	3.92	98	$C_6H_{10}O$
9.84	*cis*-2-Hexenal	4.81	98	$C_6H_{10}O$
11.30	*trans*-2-Hexen-1-al	4.39	100	$C_6H_{12}O$
19.92	Isoterpinolene	6.30	136	$C_{10}H_{16}$
26.86	Linalool	4.89	154	$C_{10}H_{18}O$
29.32	Camphor	5.75	152	$C_{10}H_{16}O$
32.89	α-Terpineol	3.85	154	$C_{10}H_{18}O$
34.63	Oleyloleate	3.65	532	$C_{36}H_{68}O_2$.
35.07	Carvone	3.10	150	$C_{10}H_{14}O$
35.72	Pentadecanoic acid,14-methyl, methyl ester	3.80	270	$C_{17}H_{34}O_2$
36.81	Glycerol 1,3- dihexadecanoate	8.21	568	$C_{35}H_{68}O_5$
37.57	O,N-Diacetyl tomatidine	0.82	314	$C_{19}H_{14}N_4O$
38.96	6,9-Octadecadienoic acid, methyl ester	7.06	294	$C_{19}H_{34}O_2$
36.81	Glycerol 1,3- dihexadecanoate	8.21	568	$C_{35}H_{68}O_5$
39.09	Methyl-2-hydroxyoctadeca-9,12,15-trienoate	5.74	308	$C_{19}H_{32}O_3$
39.59	Olein, 3-palmito2-stearo-1	3.62	860	$C_{55}H_{104}O_6$
41.85	Lycopene	5.67	536	$C_{40}H_{56}$
46.41	ß-Ionone	12.34	240	$C_{15}H_{28}O_2$,
48.72	ß-Methylionone	3.91	206	$C_{14}H_{22}O$
53.04	Lycoxanthin	6.70	552	$C_{40}H_{56}O$

The major compound of the rootstock T3 was palmitic acid (14.38%) and 2-methyl-2-octen-4-one (1.63%) was the minor compound (Table 7).

Table 7: GC/MS analysis of the total ethanolic extract of T3 [*Solanum lycopersicon* L. Nefert hybrid onto Eggplant (*Solanum melongena* L. CV. **Balady)].**

Rt	Compound name	Area %	MW	Formula
12.12	*trans*-2-Hexen-1-al	2.03	100	$C_6H_{12}O$
15.32	1-Tertbutyl-1,3-dihydro-2-himidazole-2-one	1.80	140	$C_7H_{12}N_2O$
18.70	*trans*-2-Heptenal	2.07	112	$C_7H_{12}O$
24.26	Pyrrolidin-2-one,5-heptyl	3.22	183	$C_{11}H_{21}NO$
25.02	Methyl-24-methylhexacosanoate	2.09	424	$C_{28}H_{56}O_2$
28.72	Linalool	3.56	154	$C_{10}H_{18}O$
29.43	Camphor	3.83	152	$C_{10}H_{16}O$
30.34	2,6-Nonadienal (E,Z)	3.39	138	$C_9H_{14}O$
34.62	ß-Ionone	3.22	192	$C_{13}H_2O$
35.06	2-Methyl-2-octen-4-one	1.63	140	$C_9H_{16}O$
35.72	O,N-Diacetyltomatidine	2.43	499	$C_{31}H_{49}NO_4$
37.06	Oleyloleate	2.32	532	$C_{36}H_{68}O_2$
37.77	Methyl palmitate	10.44	270	$C_{17}H_{34}O_2$
38.98	Palmitic acid	14.38	256	$C_{16}H_{32}O_2$
39.11	Propyl-14-methyl pentadecanoate	2.84	270	$C_{17}H_{34}O_2$

40.14	12,15-Octadecadienoic acid, methyl ester	11.37	294	$C_{19}H_{34}O_2$
41.87	Linolenic acid, methyl ester	6.02	292	$C_{17}H_{32}O_2$
42.00	Methyl stearate	7.62	282	$C_{19}H_{38}O$
42.43	Lycopene	3.20	536	$C_{40}H_{56}$
42.50	9-Octadecenoic acid -2-(octadecyloxy) ethyl ester	3.79	578	$C_{38}H_{74}O_3$
43.74	Zeaxanthin	3.44	568	$C_{40}H_{56}O_2$
46.40	9,19-Cyclolanostane-3,7-diol	1.98	444	$C_{30}H_{52}O_2$
52.28	Lycoxanthin	2.22	552	$C_{40}H_{56}O$

2-Decenal represented the major component (11.85 %) of T4 where Lycopene (1.22 %) was the minor compound (Table 8).

Table 8: GC/MS analysis of the total ethanolic extract of T4 (*Solanum lycopersicon* onto *Solanum lycopersicon* L var. VF)

Rt	Compound name	Area %	MW	Formula
6.18	5,10-*bis*(3-aminophenyl)15,20-diphenylporphyrin	0.33	644	$C_{44}H_{32}N6$
6.49	*cis*-3-Hexenal	5.39	93	$C_6H_{10}O$
10.07	*cis*-2-Hexenal	3.47	98	$C_6H_{10}O$
12.62	*trans*-2-Hexen-1-al	3.16	100	$C_6H_{12}O$
13.50	Heptanal	2.65	114	$C_7H_{14}O$
30.34	Camphor	5.39	152	$C_{10}H_{16}O$
30.39	*α*-Terpineol	4.47	154	$C_{10}H_{18}O$
32.67	Cantaxanthin	4.06	564	$C_{40}H_{52}O_2$
32.91	Methyl palmitate	10.02	270	$C_{17}H_{34}O_2$
35.72	2-Decenal	11.85	334	$C_{20}H_{30}O_4$
36.56	Palmitic acid, ethyl ester	5.58	284	$C_{18}H_{36}O_2$
37.03	*iso*-Propyl-14-methyl pentadecanoate	3.48	270	$C_{17}H_{34}O_2$
37.38	Ricinoleic acid, methyl ester, acetate	5.80	354	$C_{21}H_{38}O_4$
38.68	Methyl stearate	3.27	616	$C_{36}H_{56}O_8$
39.53	Lycopene	1.22	536	$C_{40}H_{56}$
41.76	9-Octadecenoic acid -2-(octadecyloxy) ethyl ester	4.08	578	$C_{38}H_{74}O_3$
42.41	Zeaxanthin	5.20	568	$C_{40}H_{56}O_2$
42.76	Phytofluene	4.47	542	$C_{40}H_{62}$
45.66	O,N-Diacetyltomatidine	4.44	499	$C_{31}H_{49}NO_4$

The major compound of T5 was methyl palmitate (15.52%) where hexadecadien-7, 11-ol-1 (1.06%) was the minor compound (Table 9).

Table 9: GC/MS analysis of the total ethanolic extract of T5 [*Solanum lycopersicon* L. Nefert hybrid onto *datura* (*Datura stramonium* L.)]

Rt	Compound name	Area %	MW	Formula
8.65	*cis*-2-Hexenal	2.01	98	$C_6H_{10}O$
12.64	*trans*-2-Hexen-1-al	2.86	100	$C_6H_{12}O$
16.22	Heptanal	2.94	114	$C_7H_{14}O$
17.73	*trans*-2-Heptenal	2.00	112	$C_7H_{12}O$
20.17	Isoterpinolene	2.94	136	$C_{10}H_{16}O$
25.79	(E)-4-(2-Methylphenyl)1-dimethylamino-1-butene	2.35	189	C13H19N
28.73	Linalool	3.85	154	$C_{10}H_{18}O$
29.93	Camphor	3.75	152	$C_{10}H_{16}O$
26.86	*α*-Terpineol	4.08	154	$C_{10}H_{18}O$
27.62	Cantaxanthin	2.62	564	$C_{40}H_{52}O_2$
33.77	*p*-Menth-1-en-9-al	3.11	152	$C_{10}H_{16}O$
33.81	O, N-Diacetyl tomatidine	2.70	499	$C_{31}H_{49}NO_4$
33.99	ß-Ionone	6.92	192	$C_{13}H_2O$
34.00	Benzophenone	3.38	182	$C_{14}H_8O_2$
34.41	2,2-Dipropyl-N-ethylpiperidine	1.82	197	$C_{13}H_{27}N$
35.71	Methyl palmitate	15.52	270	$C_{17}H_{34}O_2$
36.33	Pyrrolo[1,2-a] pyrazine1,4-dione, hexahydro-3(2-methyl propyl)	3.20	210	$C_{11}H_{18}N_2O_2$
37.02	2-Decenal	8.40	284	$C_{18}H_{36}O_2$
37.06	Palmitic acid	2.80	256	$C_{16}H_{32}O_2$
38.95	(Z, Z)-7,15-Tetracosadiene-1,24-diol	8.27	366	$C_{24}H_{46}O_2$
38.97	Ricinoleic acid, methyl ester, acetate	2.21	354	$C_{21}H_{38}O_4$
39.10	Linolenic acid, methyl ester	2.03	292	$C_{19}H_{32}O_2$
40.14	Hexadecadien-7,11-Ol-1	1.06	238	$C_{16}H_{30}O_2$
44.06	Oleyloleate	3.50	532	$C_{36}H_{68}O_2$
44.36	Lycopene	2.82	592	$C_{41}H_{84}O$
46.42	1-Hentetracontanol	2.03	593	$C_{41}H_{84}O$
49.79	2-Hydroxymethyl-5,10,15,20-tetraphenylporphyrin	1.04	644	$C_{45}H_{32}N_4O$

The percentage of esters in T1, and the rootstocks T2, T3 and T5 was in high percentage. Aldehydes were the highest ratio in T4 (24.52%), followed by T1 (13.72%) but was the lowest one in T3 (7.79%). Fatty acids were higher in the control T1 (13.20%) followed by the rootstock T3 (12.73%), where it's lowest percentage in T5 (2.80%). Terpene hydrocarbons and terpenoids have the higher percentage in the rootstock T2 (23.89%) followed by T5 (14.73 %) where the lowest percentage was in T1 (3.75%). Alkaloids have the higher percentage in the rootstock T5 (12.11%). T2 -T4 showed very low percentage as well as the control group. It was noticed that alcohols were found only in T5 by the ratio 9.33% (Table 9). Also, phenolics (0.84%) were found only in T1 where steroids (0.90%) were present only in T3 (Table 5 and 7, respectively. Plants have the capacity to synthesize, accumulate secondary metabolites that may act as molecules of different bioactivities due to interactions with human receptors. These low molecular-weight substances derived from the fatty acid, amino acid and carbohydrate pools constitute a heterogenous group of molecules with saturated and unsaturated, straight-chain, branched-chain and cyclic structures bearing various functional groups, e.g. alcohols, aldehydes, ketones and esters [8].

In the present study, all the rootstocks of tomato fruits showed the presence of antioxidant metabolites such as carotenoids, phenolic compounds, and phenolic acids. Many studies have reported the ability of these compounds to provide effective protection by neutralizing free radicals, which are unstable molecules linked to the development of a number of degenerative diseases and conditions [37].

Other antioxidants as chlorogenic acid, and flavonoids (luteolin, quercitrin, and quercetin) were high in ripe tomato [38]. It is well-established that free radicals are associated with the process that leads to cell degeneration, especially in organs such as liver and kidney.

Intrahepatic accumulation of reactive oxygen species (ROS) is thought to be an important cause for many diseases related to liver and kidney. Carotenoids have been well described that are able to scavenge ROS [37]. Lycopene, a carotenoid identified in the investigated tomatoes, has been suggested to has antioxidant activity, due to the long-chain conjugated double bonds, so may play a role in certain diseases related to the oxidative stress [39].

Lycopene could protect cells against oxidative damage and thus decrease the effect of chronic diseases. The *cis* isomers of lycopene play beneficial role in the body biological process than all-trans-lycopene [39]. In our present study lycopene has the highest ratio in the T2 group (5.67 %, Table 6) followed by T4 (3.22%, Table 8) compared with the lowest value (1.02%, Table 5) in the control T1. This result is in agreement with what was reported that the grafted crop onto the commercial hybrid rootstock tending to increase the lycopene concentration by 40% than the fruits from un-grafted plants. Carotenoid cleavage enzymes (carotene oxygenase derivatives) were reported to metabolize lycopene to biologically active metabolites, which can ameliorate nonalcoholic fatty liver disease [40].

Carotenoids were reported to be highly efficient in mitigating the hepatotoxic impacts of tramadol by preventing lipid peroxidation and initiating modifications in the expression and activity of antioxidant pathways [39]. The concentration of lycopene was related to the high K concentration in the fruit in tomato. The antioxidant activity of carotenoids (alpha-carotene, beta-carotene, cryptoxanthin, lutein and lycopene), taken with the diet or through nutritional supplements, was reported to benefit human health [37]. The administration of tomato is beneficial in reducing heavy metal accumulation in the liver. Tomato extract was reported to inhibit nonalcoholic steatohepatitis-promoted hepatocarcinogenesis mainly as a result of reduced oxidative stress [41].

The identification and contents of aldehydes, ketones, esters, fatty acids, terpenoids, carotenoids, alkaloids and phenolic compounds were determined by GC-MS in fruits on grafting between the three rootstocks (T2-T4), compared with un-grafted plants (T1).

The results show how grafting can influence the phenolic content of tomato fruits.

Several defense-related secondary compounds like phenols could be synthesized by the plant as phenolic compounds are

important components in tomato for their beneficial effects on human health. The presence of such compounds could partially explain the pharmacological properties of this plant and demonstrates its importance in alimentation and daily intake [42].

Several dietary phytochemicals potentially regulate the equilibrium between oxidant and antioxidant species. Phenolic compounds, display a remarkable array of biological and pharmacological activities [43]. The level of expression of phenolics and biochemical components, *viz*. peroxidase (POD), polyphenoloxidase (PPO), acid phosphatase, total phenol and *ortho*-dihydroxy phenol and other biochemical changes is the general response associated with plant disease resistance.

Various elicitors' molecules like Chitosan induced a significant increase in the activities of PPO and POD, and increased the phenolic compounds in tomato fruits and thus providing protection against the ingress of pathogens [44].

The antioxidant capacity of tomato could be related to the content of phytochemicals such as phenolics and flavonoids. Varieties of tomato plant are subject to oxidative stress, showing a response to antioxidant enzymes [45]. The fruits of tomato synthesize metabolites such as phenolic compounds and pigments, such as chlorophyll and carotenoids, and other nutrients that benefit human health [37].

The current study showed the presence of steroidal alkaloids in fruit extracts of the four rootstocks (T2 -T5) and the control (T1) where it recorded the highest ratio in T5 (12.11%) and it was the lowest in T2 (0.82%) This could represent a synergistic effect with phenolic compounds against oxidative stress [45]. The effect of grafting on the accumulation of major phenolic constituents in tomato fruit was reported [8]. Health compounds might be affected by grafting as a result of the translocation of metabolites associated with fruit quality to the scion through the xylem and/or modification of the physiological processes of the scion [8].

According to the results of this study, T2 and T3 extracts significantly recovered the parameters of liver functions in plasma, reduced malondialdehyde and enhanced glutathione levels, as well as enhanced all antioxidant enzyme activity in all tissues. Polyphenolic compounds showed anti-oxidant and anti-inflammatory properties, able to synthesize metal chelating proteins and exert important functions in reducing the risk of human diseases [46].

In the present work, phenolics were noticed in all rootstocks while steroids (2.98%) were present only in T3 (Table 7). Phenolic compounds are strong antioxidants capable of removing free radicals, chelate metal catalysts, activate antioxidant enzymes, reduce α-tocopherol radicals and inhibit oxidases. The root exudate profile emitted from plants grafted onto rootstock was distinctly different than from un-grafted roots providing details about specific constituents having some role in suppression of the pathogen [44].

The current study, tomato fruits of grafted plants were considered to be safe for consumption. In the present work, tomato scion of tomato T1 onto tomato T2 had a positive effect on performance, chemical composition and bioactivity. The rootstock T2 was reported to be safe with low alkaloids content (0.82%). Therefore, in countries like Egypt, where vegetable cultivation is basically carried out by traditional methods, the grafting technique defined in this study could increase tomato yield and performance and provide higher profit to the farmers.

Since rootstock has an impact on plant cultivation performance as well as on fruit yield and quality, various tomato cultivars can be utilized in grafting experiments to find out the best combinations. Differences of chemical composition of tomato fruits, regarding to phenolics, carotenoids, esters, alkaloids content, were also found due to grafting.

In general, the quality parameters were higher in the grafted plants (Table 4). Figure 1 is the diagram illustrated the total percentages of different phytochemical identified by GC/MS in the total ethanolic extracts of the five tomato treatments (T1-T5).

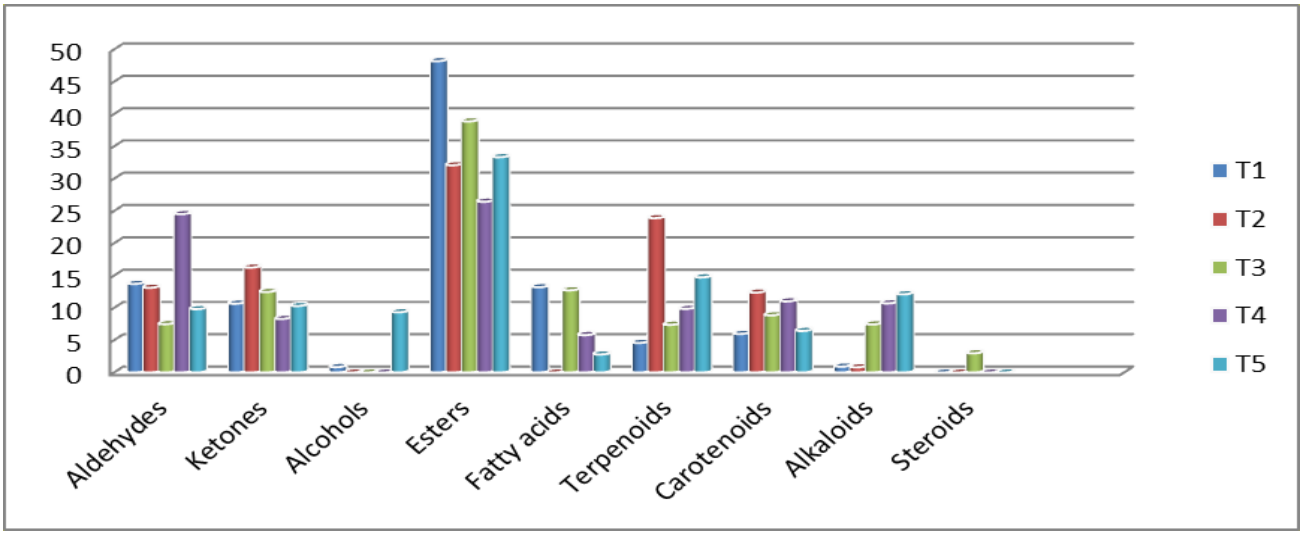

Figure 1: The total percentages of different phytochemical identified by GC/MS in the total ethanolic extracts of the five tomato treatments

The effect of tomato-derived lycopene consumption on markers of inflammation and oxidative stress was reported [47]. The anti-inflammatory effects derived by tomato products consumption were superior to that of lycopene delivered as a single compound. The components of tomato such as soluble solids, ascorbic acid and total soluble sugar content were increased in un-grafted plants than those in grafted ones [48]. Concentration of the main components in tomato fruits such as aldehydes, ketones, esters, fatty acids, terpenoids, carotenoids, phenolics and alkaloids were variable.

In Vivo Biological Estimations

Oxidative Stress Markers

Acute toxicity study revealed extracts safety after 24 hours of administrations. No animals died due to all the tested concentrations. Chronic toxicity study showed 25% mortality of animals upon administration with 250 mg/kg body weight of T5 tomato only, revealed its toxicity effect. Additionally, rats in this group showed nasal bleeding. Liver and kidney are target organs for toxic chemicals due to their essential functions in detoxification and excretion processes. Thus, they are considered highly useful in toxicity studies because of their sensitivity to harmful compounds and their potential to predict toxicity [15].

The exposure to stress leads to the formation of reactive oxygen species (ROS) favoring the oxidative stress, inducing physiological and behavioral changes are interfering with the maintenance of homeostasis of an organism, which can cause cell damage and damage to lipids, DNA, proteins, mitochondria, and cell membranes [49].

MDA is the most commonly used test for lipid peroxidation in biomedical sciences since MDA is one of the major aldehydes formed after breakdown of lipid hydroperoxides. Thus, it is considered as a good biomarker of oxidative damage caused by free radicals [28, 50]. The most significant disturbance in the antioxidant defense is a decrease in GSH concentration and SOD levels and increment of lipid peroxidation [51]. These observations are in line with our results through reduction in GSH, SOD and increment of MDA levels in all treated groups. SOD is considered a front line of defense against the potentially cytotoxic O2− free radicals that cause oxidative stress [51].

Superoxide dismutase transforms O2 − to the more stable hydrogen peroxide (H_2O_2), which converted enzymatically into H2O by catalase and glutathione peroxidase [52]. As SOD is a glutathione-level-dependent enzyme, its activity was decreased by the depletion of glutathione level [53]. In the present study and regarding to the oxidative stress markers, SOD enzyme level recorded significant decrease after administration of normal rats with different types of tomato extracts as compared with the control group.

It decreased by 33.00, 58.00, 49.40 and 57.40% after administration with T1, 3, 4 and 5, respectively (Table 10). Insignificant decrease in SOD level was noticed after administration with T2. With respect to GSH, significant decrease in its level after administration of normal rats with T4 (30.60%) and 5 (33.40%) was recorded.

Normal rats administered by T1, 2 and 3, recorded insignificant decrease as compared with control group.

Contradictory, insignificant increase in MDA level after administration of normal rats with T1-5 as compared with control group (Table 10). Nutrients with antioxidant properties can neutralize free radicals preventing the loss of cellular integrity [49]. Therefore, dietary antioxidants are effective means to limit lipid peroxidation *in vivo*. Recent investigations have been focused on natural molecules to identify consumer concerns about safety and toxicity [55]. The significant changes in oxidative stress markers under investigation revealed elevation of free radicals after administration of certain kinds on tomato especially in T5. T2 recorded insignificant changes in oxidative stress indices revealed that it did not initiated free radicals.

Table 10: Effect of different tomato's treatments on hepatic antioxidant levels

Groups	SOD (µg /mg protein)	% change	GSH (µg/gm tissue)	% change	MDA (µmol/mg protein)	% change
Control rats (T0)	5.00^{a} ± 0.43	---	8.87^{a}± 1.76	---	0.22 ^{ab}± 0.02	---
T1	3.35 ^{b}± 0.23	(-33.00)	8.42 ^{a}± 0.29	(-5.07)	0.23 ^{ab}± 0.009	(+4.50)
T2	4.77 ^{a}± 0.78	(-4.60)	8.30 ^{a}± 1.66	(-6.40)	0.23 ^{ab}± 0.023	(+4.50)
T3	2.10 ^{c}± 0.49	(-58.00)	7.74 ^{a}± 2.48	(-12.73)	0.24^{ab}± 0.005	(+9.09)
T4	2.53 ^{bc}± 0.19	(-49.40)	6.15 ^{b}± 2.98	(-30.60)	0.25 ^{a}± 0.02	(+13.60)
T5	2.13 ^{c}± 0.89	(-57.40)	5.90 ^{b}± 0.19	(-33.40)	0.24 ^{a}± 0.012	(+9.09)

Data are expressed as mean ± SD of six rats in each group. T1: Tomato (*Solanum lycopersicum* L.) var. Nefret. T2: Tomato (*Solanum lycopersicum* L.) var. VFN. T3: Eggplant (*Solanum melongena* L.) cv. Balady. T4*:* Tomato (*Solanum lycopersicum* L.) var. VF. T5: Datura (*Datura stramonium* L.). Groups having the same letters are insignificant while those having different letters are significantly different at $P < 0.05$. Values between brackets are % changes versus control group

Liver Function Enzymes

The liver function indices revealed significant increase in AST enzyme after administration of normal rats with T1 and T4 by 29.40 and 46.10%, respectively as compared with the control group (Table 11), while significant increase in ALT enzyme reached to 38.15% was recorded in T4, as compared to the control group. Insignificant increase in ALP enzyme was noticed after administration of normal rats with all rootstocks under investigation (Table 11). T2 was not able to change the liver function enzymes that revealed its safety. The significant increment of AST, ALT and ALP enzymes in certain groups under investigation was in accordance with previous studies [56] who explained this elevation to the increase in hepatic cell membrane fragility that led to enzyme release into circulation.

Table 11: Effect of different tomato's treatments on liver function enzymes level

Groups	AST		ALT		ALP	
	Value (unit/L)	% change	Value (unit/L)	% change	Value (unit/L)	% change
Control rats (T0)	0.78 ^{b}±0.035	---	1.52 ^{b} ±0.06	---	257.44 ^{a}± 1.86	---
T1	1.01 ^{a}± 0.12	(+29.40)	1.64 ^{b} ± 0.48	(+7.80)	277.64 ^{a}±5.06	(+7.80)
T2	0.80 ^{b}± 0.14	(+2.50)	1.53 ^{b} ± 0.18	(+0.60)	259.34 ^{a}± 5.80	(+0.70)
T3	0.81 ^{b}± 0.04	(+3.80)	1.58 ^{b}± 0.06	(+3.90)	257.76 ^{a}± 7.22	(+0.10)
T4	1.14 ^{a}± 0.09	(+46.10)	2.10 ^{a}± 0.16	(+38.15)	266.86 ^{a}± 1.66	(+3.60)
T5	0.89 ^{b}± 0.13	(+14.10)	1.58 ^{b}± 0.04	(+3.90)	270.66 ^{a}± 21.7	(+5.10)

Data are expressed as mean ± SD of six rats in each group. T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. Var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4*:* Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.). Groups having the same letters are insignificant while those having different letters are significantly different at $P < 0.05$. Values between brackets are % changes versus control group

Kidney Function Tests

Concerning kidney function parameters, significant increase in urea level was recorded in T4 and 5 as compared to control group. It reached to 16.98 and 45.16 %, respectively, while insignificant increases were recorded in T1, 2 and 3 (Table 12). Creatinine level showed insignificant increase after administration of normal rats with different tomato extracts (Table 12). Similarly, T2 did not affect the kidney function parameters under investigation. The elevated levels of both urea and creatinine in certain groups revealed renal injury [49, 53]. This was in agreement with [54] who reported that renal injuries were associated with elevation of urea and creatinine levels, where the serum creatinine level does not

rise until at least half of the kidney nephrons are destroyed. Group T2 recorded insignificant effects on liver and kidney indices, revealed its safety.

Table 12: Effect of different tomato's treatments on kidney function parameters of serum urea and creatinine levels

Groups	Urea		Creatinine	
	(mg/dl)	% change	(mg/dl)	% change
Control rats (T0)	63.76 ^{bc} ± 8.40	---	15.68 ^{a}± 1.48	---
T1	65.43 ^{bc}± 18.50	(+2.65)	17.44 ^{a}± 0.42	(+11.22)
T2	62.49 ^{bc} ± 12.62	(+1.99)	16.20 ^{a}± 2.53	(+3.31)
T3	92.56 ^{a} ± 21.44	(+45.16)	17.15 ^{a}± 0.47	(+9.30)
T4	52.93 ^{d}± 3.60	(+16.98)	17.51 ^{a}± 0.65	(+11.60)
T5	70.21 ^{b} ± 8.93	(+10.11)	17.62 ^{a}± 0.46	(+12.30)

Data are expressed as mean ± SD of six rats in each group. T1: Tomato (*Solanum lycopersicum* L. Nefret hybrid). T2: Tomato (*Solanum lycopersicum* L. var. VFN). T3: Eggplant (*Solanum melongena* L. cv. Balady). T4*:* Tomato (*Solanum lycopersicum* L. var. VF). T5: Datura (*Datura stramonium* L.). Groups having the same letters are insignificant while those having different letters are significantly different at $P < 0.05$. Values between brackets are % changes versus control group.

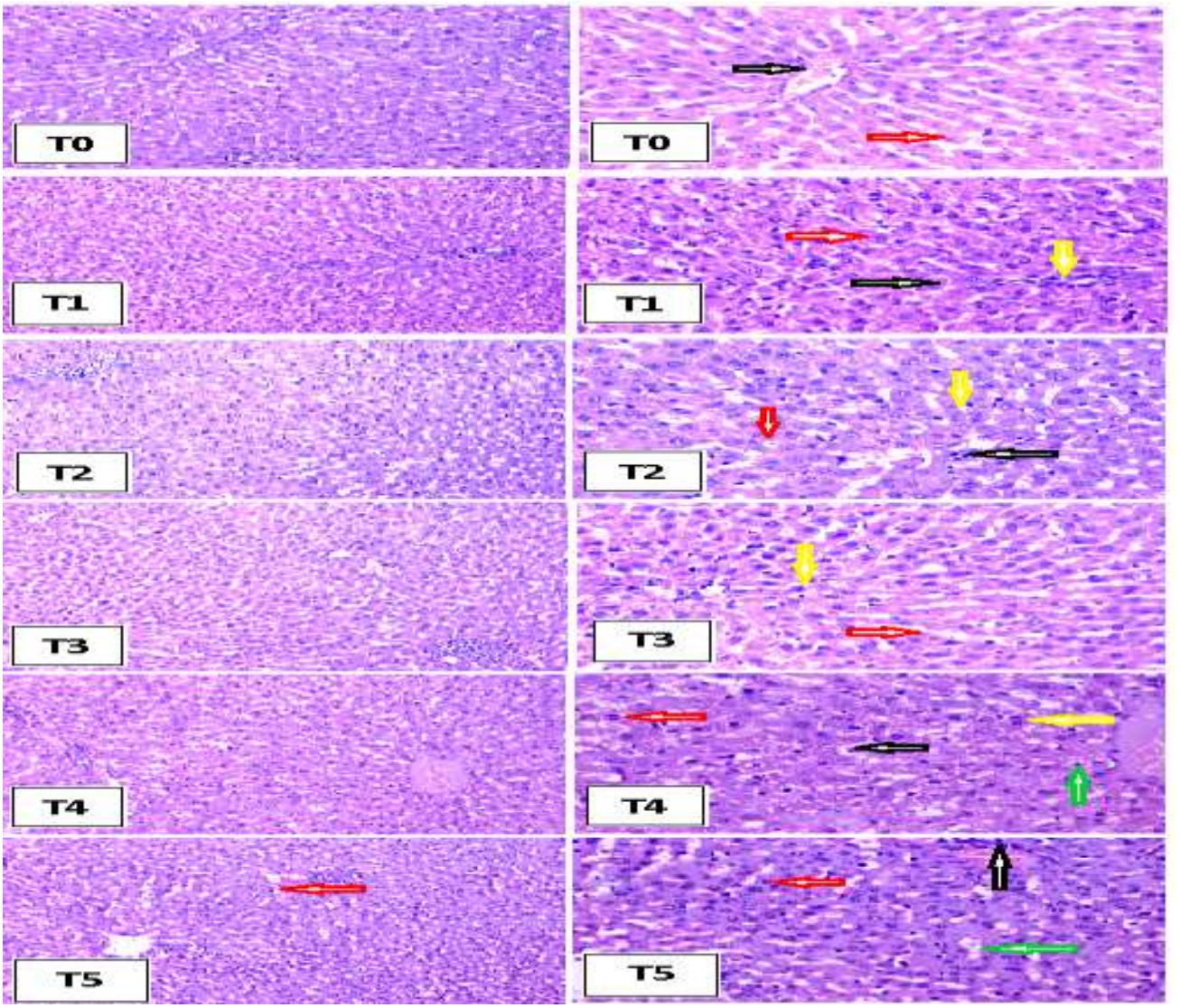

Fig. 2: Photomicrographs of liver section from control group (T0) showed hepatic tissue with normal structure and architecture, normal hepatocytes arranged in thin plates (red arrow), congested central vein (black arrow) and normal portal tract (yellow arrow). Liver section of T1 group showed preserved (intact) lobular hepatic architecture (red arrow) and scattered lymphocytes at portal tracts (black arrow (arrow) and in between hepatocytes (yellows arrow). Liver section from T2& T3 groups showed preserved lobular hepatic architecture (red arrow) with aggregate of lymphocytes in between hepatocytes (black arrow), congested and dilated sinusoids (yellow arrow). Liver section from T4 group showed intact lobular hepatic architecture, moderate hepatocyte with ballooning (black arrow) and binucleated hepatocytes (yellow arrow), central vein congestion (green arrow), congested and dilated sinusoids (red arrow). Liver section from T5 group showed preserved (intact) lobular hepatic architecture (red arrow) with aggregate of lymphocytes in between hepatocytes (black arrow), congested central vein (yellow arrow), congested and dilated sinusoids (green arrow) (H&E, x200, x400)

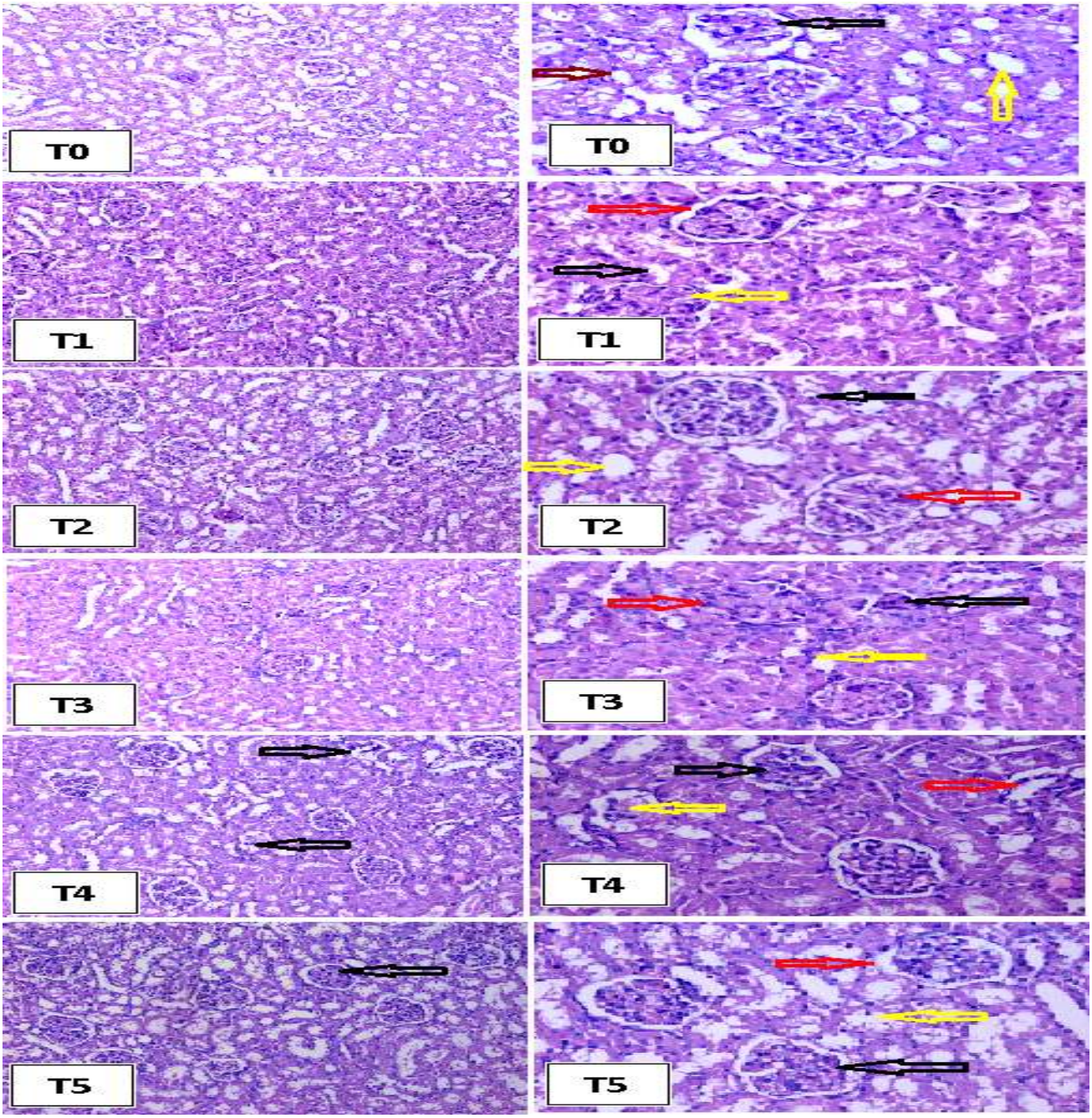

Fig.3: Photomicrographs of kidney section from control group (T0) showed renal cortex with normal renal corpuscle, normal glomerulus (red arrow), normal juxtaglomerular apparatus, normal proximal convoluted (black arrow) and distal convoluted (yellow arrow) tubules. Kidney section from T1&T2 groups showed renal cortex with almost normal renal corpuscle, normal glomerulus (red arrow), normal juxtaglomerular apparatus, normal pattern of proximal convoluted (black arrow) and distal convoluted (yellow arrow) tubules. Kidney section from T3 group showed few of the glomeruli corpuscles with obliterated cells (hyperplasia of epithelial cells lining the partial layer of Bowman's capsule) (red arrow). Proximal convoluted tubules show destructed epithelial cells (yellow arrow), destructed epithelial of distal convoluted tubules (yellow arrow) and congested blood vessels (blue arrow). Kidney section from T4 group showed few of the glomeruli corpuscles with obliterated and destructed cells (black arrow). Proximal convoluted tubules show destructed epithelial cells (yellow arrow) and destructed epithelial lining of distal convoluted tubules (red arrow). Kidney section from T5 group showed renal cortex with renal corpuscle of almost normal glomerulus (black arrow), normal juxtaglomerular apparatus, normal pattern of proximal convoluted (red arrow) and distal convoluted (yellow arrow) tubules (H&E, x200, x400)

Regarding to liver and kidney architectures, they showed certain degree of histopathological changes as illustrated in Figures 2 and 3, respectively. Administration of rats with T2 tomato recorded the most normal architectures of liver and kidney. These observations give an additional support that T2 stabilized cell membrane structure and function, led to preserve liver function enzymes and antioxidant levels to their normal levels. Therefore, the activity exhibited by this rootstock of ripe tomato could be attributed to their phenolic compounds and the mechanism through which they possibly do this, could be by their radical scavenging abilities and reducing power [38]. The worst observations in liver and kidney architectures were recorded in T5.

Conclusion

It could be concluded that tomato grafting onto different rootstocks under investigation encouraged both vegetable growth beside riches total chlorophyll content and NPK percentage in their leaves, Moreover, it increased early and total yield as well as improved fruit quality. T2 did not record any side effects revealing its safety. The activity of T2 may be attributed to the valuable components such as aldehydes, ketones, esters, fatty acids, terpenoids, carotenoids, phenolics and alkaloids have identified by GC-MS. The rootstock datura, unfortunately, showed the worst effect on oxidative stress markers as well as liver and kidney functions.

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