

## Research Article

# Evaluation of Tomato Waste Extract as Natural Source of Antioxidant in Cookies

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### ABSTRACT

Oxidation cause deterioration of food color, flavor, texture and also results in nutrient degradation. Tomato peel and seeds are the major bio-waste of tomato processing industry, these are natural rich sources of bioactive substances that play a significant role in prevention of diseases. The proposed work was to utilize tomato waste (peel and seeds) extract (TWE) as natural source of antioxidant in cookies at three levels (0.1, 0.12 and 0.14%) and evaluated, rancidity, physical and organoleptic properties of cookies at storage interval of 0,15, 30 and 45 days. The results indicated that TWE was high in phenols ( $48.35 \pm 1.25$  mg GAE/100 g), flavonoids ( $46.88 \pm 1.20$  mg RE/100 g), lycopene content ( $46.16 \pm 0.64$  mg/100 g) and also had high DPPH value ( $92.3 \pm 0.3\%$ ). Also, the data demonstrated that TWE had significant effect on, rancidity, physical and organoleptic properties of cookies from  $T_0$  to  $T_3$  as function of treatment. During storage, TWE had non-significant effect on physical properties of cookies. The results also showed that TWE had good inhibition effect on rancidity of the cookies. The inhibition effect of TWE at level of 0.14% was equivalent to the effect of BHT at 0.01%. Organoleptic performance exhibited that the cookies with 0.12% TWE were the most acceptable. Finally, addition of TWE as source of natural antioxidant can be recommended to improve shelf life of food products.

**Key words:** Tomato waste extract, oxidation, lycopene, cookies, rancidity properties

### INTRODUCTION

Degradation and deterioration of food products results in lipid oxidation and decreases the shelf life of foods. Both natural and synthetic antioxidants are used to prevent oxidation. Food industries widely used synthetic antioxidants such as Tetra Butyl Hydro Quinine (TBHQ), Butylated Hydroxyl Anisole (BHA) and Butylated Hydroxyl Toluene (BHT) in food products but now consumers are aware about harmful effects of synthetic antioxidants and they prefer natural sources of antioxidants because they are safe and non-toxic (Nanditha and Prabhasankar, 2009). By-products obtained after the processing of fruit and vegetable are rich source of bioactive compounds and can be

used as a source of natural antioxidant to prevent oxidation (Umbreen *et al.*, 2014). Tomato processing industries generates large amount of waste that contained carotenoid rich skin and seeds that are not properly utilized (Strati and Oreopoulou, 2014). The main carotenoid in tomato waste is lycopene (70-80%) that gives them red color while others include phytoene (5.3%), phytofluene (2.8%),  $\beta$ -carotene (3.7%),  $\xi$ -carotene (0.9%),  $\gamma$ -carotene (1.2%) and lutein (2.0%). Both lycopene and  $\beta$ -carotene are three times higher in skin and peels of tomato and have ability to act as antioxidants and/or singlet oxygen quenchers. The quenching constant of lycopene *in vitro* has been found to be more than double that of  $\beta$ -carotene and 100-fold that of  $\alpha$ -tocopherol (Vagi *et al.*, 2007). The essential oils and extracts of fruit and vegetable waste also contained these active compounds and can be used to increase the shelf life of food products (Bagamboula *et al.*, 2004). Tomato waste (peel and seeds) extract contained considerable amounts of carotenoids (lycopene and beta-carotene) and exhibit strong antioxidant activity and can be used in cosmetics, pharmaceutical applications and also to produce high value-added food products like butter, ice cream, mayonnaise, baked goods, breakfast cereals, spreads, bottled water, carbonated beverages, fruit and vegetable drinks, soybean beverages, candy, soups and salad dressings in order to extend their shelf life due to presence of natural antioxidants (Stajcic *et al.*, 2015; Kaur *et al.*, 2011; Yang *et al.*, 2006). Tomato peel extracts also improved storage stability of crude cotton and sunflower oil (Elbadrawy and Sello, 2011; Nawal *et al.*, 2008).

Bakery products, specifically cookies are considered as the most suitable vehicle for the supplementation (Dhingra *et al.*, 2012) but they are more susceptible to oxidative degradation because of more oil used in their preparation that limited their shelf life (Lean and Mohamed, 1999). In food industries synthetic antioxidants are mostly used to prevent the food products from oxidation. But the consumers demand green food products with high safety, quality and nutritional values such as essential oils and extracts from plant origin (Nielsen and Rios, 2000). As tomato waste extract also has ability to prevent oxidation due to presence of higher amounts of phenolics and lycopene contents. Therefore in the present project, we aimed to utilize tomato waste extract as source of natural antioxidant in cookies and compare their effect with BHT antioxidant.

## MATERIALS AND METHODS

**Procurement of raw material:** Fully ripe fresh tomatoes (*Lycopersicon esculentum* Mill) (surface area with more than 90% red color), without any visual defects, were collected from Agricultural farm of Faisalabad, Pakistan. Tomatoes were washed by water to remove dust and unwanted particles from the surface, boiled the tomatoes at 85 °C temperature for 10 min. After boiling, skin and seeds of tomatoes were removed (Hasanuzzaman *et al.*, 2014). Skin and seeds of tomatoes were dried in oven at 60 °C up to 5% moisture content. After drying, all material was milled into flour and stored in desiccators for later use (Kumar *et al.*, 2011). Analytical grade chemicals were purchased from Sigma-Aldrich Chemie GmbH, Germany.

**Preparation of tomato waste extract:** Ethanolic extract of tomato waste (peel and seeds) powder TWP was prepared by adding 120 mL ethanol, 30 mL distilled water in 15 g dried tomato peel and seeds powder (TWP). Then samples were placed in orbital shaker for 6-8 h at 240 rpm at room temperature (AOCS, 1998). The supernatant from each flask was filtered with what man No. 1 filter paper. The solvent from the supernatant was separated at 50 °C in a rotary vacuum evaporator (EYELA, N-N series, Japan) leaving behind crude extract. The extract of each sample was weighted to determine the yield of antioxidant extract and stored at 4°C until use.

**Total phenolic contents:** Total phenolic contents in TWE were carried out through using Folin-Ciocalteu method as described by Chan *et al.* (2008).

**Total flavonoid contents:** Total flavonoid contents in TWE were determined using colorimetric method described by Dewanto *et al.* (2002) with slight modification.

**Lycopene contents:** Extraction and quantitative determinations of lycopene were conducted according to Fish *et al.* (2002) using a mixture of hexane:ethanol:acetone (v/v/v 2:1:1) containing 0.05% of BHT.

**DPPH free radical scavenging assay:** The scavenging activity of the extracts was estimated by using 1, 1-diphenyl-2-picrylhydrazyl (DPPH) as a free radical model and a method adapted from Magalhaes *et al.* (2006).

**Development and characterization of cookies:** Cookies in these experiments were prepared from wheat flour and TWE (tomato peel and seeds extract) in three different concentrations 0.01% BHT (T<sub>1</sub>), 0.1% tomato waste (peel and seeds) extract (T<sub>2</sub>), 0.12% tomato waste (peel and seeds) extract (T<sub>3</sub>) and 0.14% tomato waste (peel and seeds) extract (T<sub>4</sub>). Cookies were prepared according to the method given in AACC (2000) with slight modifications. The basic ingredients used were 200 g of composite flour, 100 g shortening, 100 g of granulated cane sugar, 1 g of beaten whole egg and 3 g of baking powder. The dry ingredients were weighed and mixed thoroughly in a bowl by hand for 3-5 min. Shortening was added and rubbed in until uniform. The egg was added and dough was thoroughly kneaded in a mixer for 5 min. The dough was rolled thinly on a sheeting board to a uniform thickness of 3 inches having 1 inch diameter and cut with the help of biscuit cutter. The cookies were baked on greased pans at 425 °F for 10 min in a baking oven. The prepared cookies were cooled to room temperature (30±2 °C) and packed in high density polyethylene bags.

**Rancidity tests:** Rancidity tests like free fatty acid, peroxide value and saponification value were analysed through following the methodology of AACC (2000) method No. 940.28, 965.33 and 920.160, respectively at 0 day interval up to a storage period of 45 days at room temperature.

**Physical parameters:** The physical parameters like diameter, thickness, breaking strength and spread factor were analysed through following the methodology of AACC (2000) at 0 day interval up to a storage period of 45 days at room temperature. Diameter (mm) of six cookies was determined by placing the cookies next to each other horizontally and the total diameter was measured. The thickness (mm) was measured by placing six cookies on one another and the total height was measured. The tests were repeated thrice to bring meticulousness. The spread factor was calculated according to the formula i.e.,  $SF = D/T$  and the evaluation of texture expressed as breaking strength was measured by using the three point bend rig technique by a texture analyzer (TA-TX2i Plus, Stable Micro System Surrey, UK) (Piga *et al.*, 2005).

**Color measurement:** The color of both sides of carrot pomace powder (CCP) cookies was measured using a Hunter's Lab colour analyzer. In the Hunter's lab colorimeter, the color of a sample is denoted by the three dimensions, L\*, a\* and b\* that gives measurement of the lightness, redness/greenness and yellowness/blueness of the product respectively and storage study of 45 days were also done (Kumar *et al.*, 2011).

**Organoleptic evaluation:** Organoleptic evaluation of cookies was done for various attributes like color, flavor, taste, texture and overall acceptability. Trained panelists were asked to list their preference on a 9-point Hedonic scale (where 1 = dislike extremely and 9 = like extremely) on fortnightly basis for consecutive 45 days (Meilgaard *et al.*, 2007).

**Data analysis:** Data obtained was statistically analyzed and interpreted by analysis of variance (ANOVA) with M-Stat C software<sup>®</sup> package. The Least Significance Difference (LSD) test at 5% probability level was applied to compare the treatment means (Steel *et al.*, 1997).

## RESULTS AND DISCUSSION

### Phyto-chemical test

**Total phenolic and flavonoid contents:** Total phenolic and flavonoid contents in TWE were  $48.35 \pm 1.25$  mg GAE/100 g and  $46.88 \pm 1.20$  mg RE/100 g, respectively (Table 1). It is reported that the total phenolic contents present in peels, pulp and seeds were  $36.9 \pm 0.8$ ,  $33.3 \pm 0.5$  and  $17.6 \pm 0.9$  mg GAE/100 g, respectively (Fuentes *et al.*, 2013). However, according to Toor *et al.* (2005) tomato peel and seeds of three cultivars on average contained 52% flavonoid contents.

**Lycopene content:** Data regarding lycopene extraction has been presented in Table 1. According to results tomato waste (peel and seeds) extract (TWE) contained  $44.16 \pm 0.64$  mg/100 g lycopene content. This result is in accordance with Choudhari and Ananthanarayan (2007) who demonstrated that tomato peel and seeds had higher concentration of lycopene than the whole tomato pulp. Similar outcomes were also observed by Toor and Savage (2005).

**DPPH free radical scavenging assay:** According to result higher DPPH value ( $92.3 \pm 0.3\%$ ) of TWE was observed (Table 1). Kalogeropoulos *et al.* (2012) reported that the by-products (seed plus peel) obtained after processing of tomato has a similar amount of antioxidant activity measured according to DPPH and FRAP methodologies, than the raw whole tomato. As tomato waste extract is rich source of lycopene and phenolic compounds so they are responsible for higher scavenging activity. It has been documented that antiradical scavenging activity is related to substitution of hydroxyl groups in the aromatic rings of phenolics, thus contributing to their hydrogen-donating ability (Yen *et al.*, 2005).

### Product analysis (cookies)

#### Rancidity analysis of cookies

**Measuring Free Fatty Acids (FFA), Peroxide Value (PV) and Saponification Value (SV):** In the present study, oxidation degree on cookies samples were determined by measuring free fatty acid, peroxide value, saponification value in the presence of BHT and tomato waste (peel and seeds) extract (TWE) for 45 days of storage. Data thus presented in Fig. 1-3 revealed that treatments and storage periods and their interactions altered free fatty acid, peroxide value and saponification value significantly.

Among the treatments the values of free fatty acid, peroxide and saponification value of  $T_0$  cookies samples with 0.01% BHT (control) in TWE cookies were  $0.07 \pm 0.008$  g/100 g,  $0.12 \pm 0.01$  meq/100 g and  $170 \pm 2.05$  mg KOH/100 g). While the values of FFA, PV and SV ranged from  $0.10 \pm 0.01$  to  $0.07 \pm 0.008$  g/100 g,  $0.13 \pm 0.009$  to  $0.09 \pm 0.01$  meq/100 g and  $174 \pm 1.63$  to  $166 \pm 1.24$  mg KOH/100 g in TWE cookies, respectively. In TWE cookies minimum FFA, PV and SP No. was observed in  $T_3$  (0.14% tomato peel and seeds extract) and this value was less than values of FFA, PV and SV of  $T_0$  (control) because of addition of more TWE extract that contained abundant amount of antioxidants specially lycopene contents. Lycopene exhibits higher singlet oxygen ( $O_2$ ) quenching ability as compared to other antioxidants that may helpful in prevention of oxidation (Rizk *et al.*, 2014).

During 45 days of storage FFA, PV and SV of TWP cookies increased significantly (Fig. 1-3) but that change was less than control. This indicates that shortening in cookies become rancid due to the oxidation of unsaturated glycerides leading to development of peroxides and/or due to hydrolysis of glycerides resulting in increased levels of Free Fatty Acids (FFA).

Table 1: Phytochemical tests of tomato waste extract

Phenolic contents (mg GAE/100 g)	48.35±1.25
Flavonoid contents (mg RE/100 g)	46.88±1.20
Lycopene content (mg/100 g)	44.16±0.64
DPPH (%)	92.3±0.3

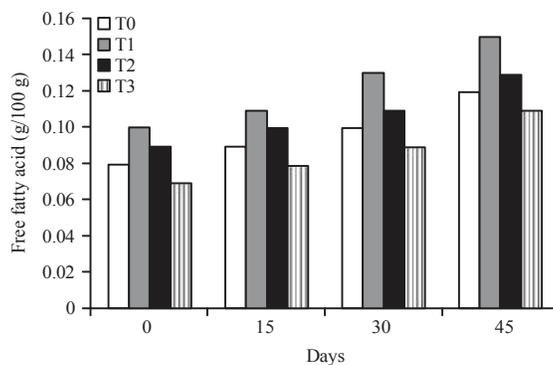


Fig. 1: Effect of storage on free fatty acid of TWE cookies

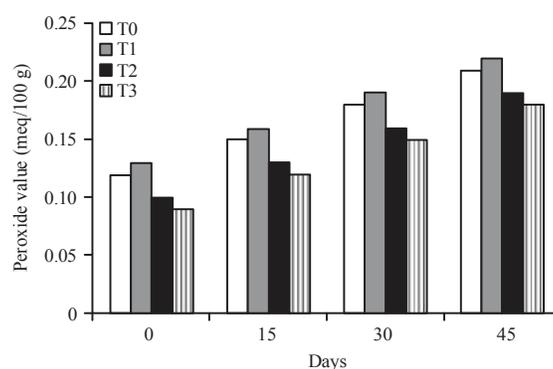


Fig. 2: Effect of storage on peroxide value of TWE cookies

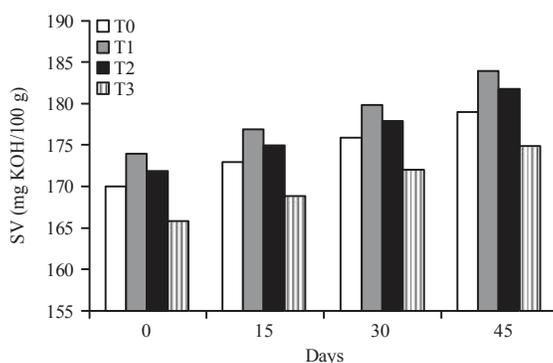


Fig. 3: Effect of storage on saponification value of TWE cookies

These results are comparable with Bhangar *et al.* (2008) and Noorolahi *et al.* (2012) who found that addition of natural plant extracts in cookies may increase their shelf life due to presence of antioxidants.

**Physical analysis:** The study of mechanical properties of cookies is important to evaluate the quality parameters from consumer point of view. These properties of cookies are dependent on properties of their dough matrix and baking behaviour. Physical parameters studied include diameter, spread ratio, breaking strength, thickness and color value. Mean of treatments presented in Table 2 indicates that diameter, thickness, spread ratio and breaking strength of TWE cookies were significantly affected as a function of their ingredients. However, storage periods and interactions showed non-significant changes in these parameters.

Table 2: Influence of TWE with different levels on the physical parameters of cookies

Treatments	Diameter (mm)	Thickness (mm)	Breaking strength (F, N)	Spread ratio (mm)
T <sub>0</sub>	81.76±0.004A	8.94±0.008D	9.06±0.008D	9.14±0.004A
T <sub>1</sub>	81.58±0.008B	8.96±0.009C	9.19±0.008C	9.10±0.01B
T <sub>2</sub>	81.55±0.008C	8.97±0.01B	9.23±0.01B	9.09±0.008C
T <sub>3</sub>	81.45±0.01D	8.98±0.004A	9.27±0.008A	9.08±0.004D

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference ( $p < 0.05$ ), T<sub>0</sub>: Cookies sample with 0.01% BHT, T<sub>1</sub>: Cookies sample with 0.1% TWE, T<sub>2</sub>: Cookies sample with 0.12% TWE, T<sub>3</sub>: Cookies sample with 0.14% TWE

Progressive decrease in diameter and increase in thickness of cookies was observed with the addition of tomato waste extract in cookies (Table 2). Maximum diameter (81.76±0.004 mm) and minimum thickness (8.94±0.008 mm) was observed in T<sub>0</sub> (control) cookies with 0.01% BHT. While minimum diameter and maximum thickness was recorded in T<sub>3</sub> (0.14% TWE) and their values were (81.45±0.01 mm and 8.98±0.004 mm), respectively. Diameter decreased and thickness of cookies increased due to presence of soluble dietary fiber present in tomato waste powder that may solubilise in the solvent. The fiber that is oligosaccharide has ability to attract the water more, due to which the dough viscosity increased that leads the thickness increased and thickness decreased (Pasha *et al.*, 2008).

According to results maximum spread factor (9.14±0.004 mm) was observed in T<sub>0</sub> (control) and minimum spread factor (9.08±0.004 mm) was recorded in T<sub>3</sub> of TWE cookies. T<sub>0</sub> cookies with 0.01% BHT were least hard than all other cookies. Hardness increased from (9.19±0.008 to 9.27±0.008mm) in TWE cookies from T<sub>0</sub> to T<sub>3</sub> (Table 2) due to presence of soluble dietary fiber that has higher water holding capacity which results in increased dough viscosity that leads the spread factor decreased. The hardness of cookies was as a result of development of gluten network. Gluten promotes the network development by attracting the water molecules (Srivastava *et al.*, 2014).

The influence of storage on the diameter, thickness, spread ratio and breaking strength of cookies prepared from waste powder are given in the Table 3. During storage physical parameters of cookies decreased but that change was non-significant. The result was nearly matched with the findings of Mushtaq *et al.* (2010) and Rehman *et al.* (2013) who reported that the change in physical parameters of rusks and cookies were due to increase in moisture contents with the passage of time.

**Color:** The color of cookies which is one of the characteristics affects the acceptability of end product by the consumer. The surface color, L\* (brightness), a\* (redness) and b\* (yellowness) values of cookies samples were measured. The values obtained for color of tomato waste (peel and seeds) extract (TWE) cookies are shown in Table 4.

These values signify the lightness/darkness of the samples. Lesser color values show a darker surface of the cookies. In general, tomato waste extract addition in cookie samples considerably decreased the 'L' and 'b' values which indicate the brightness and yellowness while the 'a' value showing the redness increased with supplementation.

The dark color of cookies was due to presence of red colored carotenoids like lycopene and β-carotene present in TWE (George *et al.*, 2004). Therefore, when tomato waste extract was added to the flour, cookies became darker and the creamy-yellow color of the cookies turned into orange-yellow.

During 45 days of storage L\*, a\* and b\* values decreased from T<sub>0</sub> to T<sub>3</sub> but that change was non-significant (Mushtaq *et al.*, 2010; Aslam *et al.*, 2014) as shown in Table 5.

**Organoleptic evaluation:** The organoleptic evaluation is very important criterion to evaluate the response of judges towards the end product and they rate the liking on a scale. The cookies prepared from different level of Tomato waste (peel and seeds) extract (TWE) were subjected to sensory evaluation for color, taste, flavor, texture and overall acceptability at 0, 15, 30 and 45 days interval of storage. All the quality attributes of cookies assessed organoleptically showed significant change among treatment and storage (Table 6 and 7).

Table 3: Effect of storage on the means of physical parameters of TWE cookies

Storage	Diameter (mm)	Thickness (mm)	Breaking strength (F/N)	Spread ratio (mm)
0	81.58±0.11	8.96±0.01	9.18±0.01	9.10±0.02
15	81.58±0.11	8.96±0.01	9.18±0.01	9.10±0.02
30	81.57±0.11	8.95±0.01	9.17±0.01	9.09±0.02
45	81.57±0.11	8.95±0.01	9.17±0.01	9.09±0.02

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference (p<0.05)

Table 4: Influence of TWE with different levels on the color of cookies

Treatments	L*	a*	b*
T <sub>0</sub>	70.04±0.008A	3.22±0.01D	23.93±0.008A
T <sub>1</sub>	69.07±0.004B	5.75 ±0.01C	22.78±0.008B
T <sub>2</sub>	67.22±0.004C	5.85±0.005B	21.21±0.008C
T <sub>3</sub>	65.43±0.004D	5.95 ±0.02A	20.36±0.008D

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference (p<0.05), T<sub>0</sub>: Cookies sample with 0.01% BHT, T<sub>1</sub>: Cookies sample with 0.1% TWE, T<sub>2</sub>: Cookies Sample with 0.12% TWE, T<sub>3</sub>: Cookies sample with 0.14% TWE

Table 5: Effect of storage on the means of color of TWE cookies

Storage	L*	a*	b*
0	67.94±1.76	5.19±1.14	22.07±1.38
15	67.93±1.76	5.19±1.13	22.07±1.38
30	67.93±1.76	5.18±1.14	22.06±1.38
45	67.92±1.76	5.18±1.13	22.06±1.38

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference (p<0.05)

Table 6: Influence of TWE with different levels on the organoleptic evaluation of cookies

Treatments	Color	Taste	Texture	Flavor	Overall acceptability
T <sub>0</sub>	7.12±0.01A	7.00±0.02A	8.00±0.04A	7.40±0.03A	7.30±0.08A
T <sub>1</sub>	6.90±0.02C	6.77 ±0.02C	7.60 ±0.02C	7.18±0.02C	7.15 ±0.04C
T <sub>2</sub>	7.04±0.04B	6.80 ±0.02B	7.77 ±0.01B	7.32±0.02B	7.22 ±0.04B
T <sub>3</sub>	6.70±0.04D	6.50±0.02D	7.40±0.01D	7.12±0.02D	7.07 ±0.04D

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference (p<0.05), T<sub>0</sub>: Cookies sample with 0.01% BHT, T<sub>1</sub>: Cookies sample with 0.1% TWE, T<sub>2</sub>: Cookies sample with 0.12% TWE, T<sub>3</sub>: Cookies sample with 0.14% TWE

Table 7: Effect of storage on the means of organoleptic evaluation of TWE cookies

Storage	Color	Taste	Texture	Flavor	Overall acceptability
0	6.94±0.15A	6.76 ±0.17A	7.69±0.22A	7.25±0.11A	7.18±0.08A
15	6.76±0.14B	6.57±0.16B	7.49±0.21B	7.08±0.13B	7.00±0.11B
30	6.59±0.17C	6.36±0.16C	7.29±0.23C	6.87±0.13C	6.82±0.12C
45	6.29±0.19D	6.16±0.16D	7.04±0.23D	6.62±0.15D	6.62±0.12D

Values are mean ±standard error of mean of three samples of each sample, analyzed individually in triplicate. Mean followed by different superscript letters in the same column represent significant difference (p<0.05)

The scores for all the parameters were lower than that of the control cookies. In general, up to about 0.12% substitution level of TWE yield higher scores which were close to the control for all the parameters studied. Substitution levels of 0.14% produced TWE cookies with relatively lower values for all the parameters. The results are in close agreement with those of Arshad *et al.* (2014) who observed that color, flavor, taste, texture and overall acceptability scores of multigrain cookies decreased due to presence of more fiber that had higher water holding capacity.

During storage color, taste, texture, flavor and overall acceptability of TWE cookies decreased from T<sub>0</sub> to T<sub>3</sub> (Table 7). These results are in accordance with Sharif *et al.* (2005) who have associated the changes in organoleptic evaluation parameters with the absorption of moisture, generation of free fatty acids and increase in peroxide value in cookies during storage.

## CONCLUSION

From above discussion it can be concluded that every year fruit processing industry is wasting a considerable amount of bio-active material that can play a vital role to cure and prevent many diseases. In present study tomato waste (peel and seeds) extract cookies were analyzed by replacing for different levels (0.1, 0.12 and 0.14%) on proximate, physical, sensory and rancidity properties. Results obtained showed that tomato waste extract had better effect on physical, sensory and rancidity properties of cookies. So it can be concluded from results that tomato waste extract improved the quality of food products and also increased their shelf life.

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