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## Summary

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## Impact of UV-C and thermal pasteurization on bioactive compounds, sensory characteristics and aroma profile of traditionally produced koruk vinegar

Auswirkungen von UV-C und thermischer Pasteurisierung auf bioaktive Verbindungen, sensorische Eigenschaften und das Aromaprofil von traditionell hergestelltem Koruk-Essig

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The aim of the study was to examine the effects of UV-C light applied by a designed reactor on bioactive components of traditionally produced koruk vinegar. Furthermore, microbial load, 5-HMF content, the volatile compounds, sensorial attributes and colour of vinegar were assessed. A reduction of 1.29-log CFUmL<sup>-1</sup> was achieved in total aerobic bacteria by applying a dose of 262.4 mJcm<sup>-2</sup>. A total of 22 volatile compounds, only total phenolic content changed significantly during UV-C treatment while antioxidant activity, total flavonoid and ascorbic acid content was not affected. Although the characteristic pungent sensation, aromatic intensity and richness in aroma of pasteurized vinegar significantly decreased, a non-significant difference in the sensorial properties was determined in all UV-C irradiated vinegar. UV-C treatment has potential for non-thermal pasteurization of koruk vinegar compared to thermal one due to the more preservative of its fresh-like characteristics.

Keywords: Koruk vinegar, UV-C light, bioactive component, aroma profile, sensory attributes

## Introduction

Vinegar has been used since ancient times and is an important element in European, Asian, and other cuisines. Vinegars are commonly used for pickling of fruits and vegetables and in the preparation of salad dressings, marinades, mayonnaise, mustard, canned foods and bakerv products (Hailu et al. 2012; Bhat et al. 2014; Budak et al. 2014). Vinegar is an acidic liquid produced by a twostage bioprocess. In the first stage, fermentable sugars are converted into ethanol by the action of yeasts, normally strains of Saccharomyces cerevisiae, while in the second stage, bacteria of the genus Acetobacter oxidize the ethanol to acetic acid (Bhat et. al. 2014). Vinegar could be produced from any alcoholic material from alcohol-water mixtures to various fruit wines (Hailu et al. 2012; Bhat et al. 2014). The classification of many different types of vinegar is usually based on the raw material used for its production. Traditional vinegar is produced from fruit juices such as grape, apple, plum, rice, and potato (Budak et al. 2014). Depending on the raw material, the alcoholic and acetification processes play a key role in vinegar production (Bhat et al. 2014).

Vinegar has various aminoacids, organic acids, phenolic compounds, melanoidins, vitamins and antioxidants which has many functional therapeutic properties including antimicrobial and antioxidant activities, reduction in blood pressure and the effects of diabetes (Johnston et al. 2006; Budak et al. 2014; Xia et al. 2020), anticarcinogenic, anti-infectious and antitumor effects (Chen et al. 2016; Karabiyikli and Sengun 2017), prevention of cardiovascular diseases and yeast infections (Leeman et al.2005; Ostman et al. 2005; Bhat et al. 2014). The number of studies in which characterization of traditionally produced vinegars and/or vinegars using different raw materials is fairly limited. However, phenolic components, which strongly vary depending on the production process and the raw material used, affect the antimicrobial and antioxidant potential of vinegar (Karabiyikli and Sengun 2017; Yıkmış et al. 2020).

The popularity of the fresh-like food products is getting increased by promoting the regular consumption of bioactive substances by many nutritional researchers. Although the thermal pasteurization is addressed to provide microbial reduction/inactivation and extend the shelf life of food products, reduction in organoleptic and nutritional properties are normally results related to this process (Petruzzi et al. 2017; Kaya and Ünlütürk 2019; Mieszczakowska-Frac et al. 2021). There is an increasing consumer demand for nonthermal or minimally processed products because of the consequences mentioned previously. Among the novel and alternative technologies, UV-C irradiation has been used in the food industry for different purposes including surface treatments of foods (US FDA 2001) like minimally fresh processed or ready-to-eat fruits/vegetables, reduction in pathogens in fish, meat and poultry process (Unluturk et al. 2008), dairy products (Cilliers et al. 2014; Barut Gök et al. 2021) various fruit and juices (Erkan et al. 2008; Pala and Toklucu 2013; Islam et al. 2016; Bhat et al. 2011; Barut Gök 2021). DNA has the ability to efficiently absorb UV-C light at a wavelength of 253.7 nm (Hoyer 1998), and inactivates most of the microorganisms (Allende et al. 2006), such as bacteria, moulds, yeasts and viruses. The primary mechanism of the UV-C light on inactivation of microorganisms (Price 1965; Unluturk et al. 2008; Gabriel and Nakano 2009) is due to the formation of pyrimidine dimers which prevent replication and provide inactivation of microorganisms (Abdul Karim Shah et al., 2016). Recent studies reported that UV-C irradiation can be used as an alternative to thermal pasteurization without any change in the physicochemical, nutritional, and sensorial quality of the juices (Chia, Rosnah, Noranizan, & Ramli, 2012; Kaya, Yildiz, & Unluturk, 2015; Müller, Noack, Greiner, Stahl, & Posten, 2014; Santhirasegaram et al. 2015). However, the application of UV-C irradiation is restricted for certain fruit juices due to the presence of high amount of color compounds and soluble and/or suspended particles, which reduce the penetration ability of UV light (Koutchma, Keller, Chirtel, & Parisi, 2004).

In the study, a designed reactor (Barut Gök 2018; Barut Gök 2021) was used to evaluate the efficacy of UV-C light on bioactive components (total phenolic content, total flavonoid content, total monomeric anthocyanin and ascorbic acid content, antioxidant activity) of traditionally produced koruk vinegar. Moreover, the efficacy of UV-C on the aroma profile of koruk vinegar, 5-HMF content, microbial load, sensorial attributes and colour were assessed compared to pasteurized one. There is no study regarding efficacy of UV-C treatment on the quality attributes of koruk vinegar or other traditional vinegar produced from different raw materials.

## **Materials and methods**

## Preparation of koruk vinegar

The traditional process was used for the vinegar production (Y1km1ş et al. 2020). Unripe grape (koruk) was pressed and filtered in order to produce the juice. The juice was inoculated by *Saccharomyces cerevisiae* (Laffort, Bordeaux, France) (3%) (Y1km1ş et al. 2021) for ethanol fermentation for 30 days at  $25\pm1$  °C and the wine was fermented for 60 days at  $28\pm1$ °C until the ethanol content was 0.5-1% after addition the acetic acid culture. Traditionally produced koruk vinegar (TV) was used as the control. Pasteurized vinegar (PV) was performed at  $65\pm1$ °C for 30 min. Ultraviolet-C treated vinegar (UVV) was obtained at three different UV-C doses as UVV1 (262.4 mJcm<sup>-2</sup>), UVV2 (65.9 mJcm<sup>-2</sup>) and UVV3 (32.9 mJcm<sup>-2</sup>).

## UV-C treatment and dosimetry

The modified UV-C reactor, used for vinegar treatment consisted of a grooved stainless steel semi-circle flow path positioned around a quartz glass tube containing a UV-C lamp (UV-C output of 28.1 W) (Barut Gök 2018; Barut Gök 2021). An irregular flow obtained with a secondary flow vortices formed in Dean flow reactors enables the irregular stirring of liquid which is intended to ensure that all liquid passed through the UV-C light providing an efficient disinfection. A peristaltic pump adjusted three different flow rates through the reactor.

The UV dose (D) was calculated as given by equation (2). t is the irradiation time and  $I_{avg}$  is the UV radiation intensity.

The average intensity  $(I_{avg})$  was quantified as given by equation (1) by using the incident intensity (I<sub>o</sub>, 22.3 mW cm<sup>-2</sup>) detected at the surface of quartz glass using a UVradiometer (UVP, USA) (Unluturk et al. 2008).  $A_e$  (1/cm) and L (cm) are the absorption coefficient detected at 254 nm wavelength and the path length of the cuvette, respectively. The slope of the curve of the absorption coefficient tion factor was used to estimate the absorption coefficient

of vinegar.  $A_e$  was estimated as 7.43 cm<sup>-1</sup>. UV-C doses were quantified as 262.4, 65.9 and 32.9 mJcm<sup>-2</sup> for vinegar, respectively.

$$I_{ave} (mW/cm^2) = I_0 x (1 - exp (-A_e x L))/(A_e x L)$$
(1)

$$D = I_{avg} x t \tag{2}$$

## **Microbial analysis**

Vinegars were evaluated for total aerobic bacteria, total yeast/mould and total *Enterobacteriaceae* counts after serial dilution in maximum recovery diluent (Merck, Germany). Total aerobic bacteria, yeast/mould count and total *Enterobacteriaceae* were inoculated on PCA (plate count agar), PDA (potato dextrose agar) and VRBG (violet red bile glucose) agar (Merck, Germany), respectively. The aerobic bacteria, total *Enterobacteriaceae* yeast/mould counts were incubated at 37±1°C for 48h, 37±1°C for 24h (Harrigan 1998) and 25±1°C for 5 days, respectively. All experiments were carried out in parallel and triplicates. The microbial population was reported as log CFUmL<sup>-1</sup>.

## Measurement of bioactive components

Total phenolic content (TPC) was analysed by the Folin-Ciocalteu reagent method according to the method described by Franke et al. (2004). The absorbance of the vinegar samples was measured at 720 nm wavelength after an incubation for one hour in darkness at room temperature (Singleton 1985). TPC was expressed as gallic acid equivalent in mg L<sup>-1</sup> (mg GAE L-1).

The total flavonoid content (TFC) was detected by the colorimetric method according to Zhishen et al. (1999). TFC was calculated and shown as mg of catechin equivalents/mL (mg CE mL-1). Antioxidant capacity of vinegars was performed by using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical with some modifications (Grajeda-Iglesias et al. 2016). CUPRAC (Cu (II) ion reducing antioxidant capacity) method was performed according to Apak et al. (2006). The absorption was read at 720 nm wavelength by a spectrophotometer. Determination of ascorbic acid (AA) was carried out with the ascorbic acid 2,6-dichlorophenolindophenol-titrimetric method (Ordóñez-Santos and Vázquez-Riascos, 2010). The results of AA were calculated as mg 100 mL<sup>-1</sup>. The pH-differential method was performed to calculate the total monomeric anthocyanin content of vinegars (Zhu et al., 2017). Absorbance measurements were performed by a spectrophotometer (SP-UV-VIS-300SRB, Spectrum Ins., Australia).

## HMF (Hydroxymethylfurfural)

A method described by LeBlanc et al. (2009) was applied for the 5-hydroxymethyl-2-furfural determination. The measurement is based on HMF with barbituric acid and *p*-toluidine to form a red-colored compound. The absorbance of the vinegars was analysed at 550 nm when the intensity of colour has reached a maximum level (LeBlanc et al., 2009).

#### **Colour measurements**

Colour analysis was performed using a colourimeter (Color meter, PCE-CSM 5, Germany). The colour components of vinegars were determined as  $L^*$  (brightness/darkness),  $a^*$  (redness/greenness),  $b^*$  (yellowness/blueness). The total colour difference (TCD), hue angle (h) and chroma (C) were calculated according to the following Equations 3–5 (Ordóñez-Santos et al., 2017).

The total colour difference  

$$TCD = ((\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2)^{1/2}$$
(3)

Chroma, 
$$C = (a^2 + b^2)^{1/2}$$
 (4)

hue angle, 
$$h = \tan^{-1}(b/a)$$
 (5)

# Total soluble solids (TSS, Brix), titratable acidity (TA) and pH

The TSS of vinegars was analysed using a refractometer (RX-7000 $\alpha$ , ATAGO Co., Japan) and was given in °Bx (Brix). pH values were determined using pH/mV Meter (Hannah, Hi, 2002, USA). The TA was calculated by potentiometric titration of vinegars with NaOH (0.1 N), and was stated as gram tartaric acid in 100 mL of vinegar (AOAC, 1990). All measurements were performed at 20°C ±1°C.

#### Sensory attributes

Thirty trained panellist evaluated the certain parameters of koruk vinegars such as aromatic intensity, richness in aroma, pungent sensation, general impression, ethyl acetate odour, taste, using a 9-point structured hedonic scale which was expressed from extremely dislike (1) to extremely like (9). Acceptance of sub-points was accepted as 6. The results were the mean of three replications (Callejón et al. 2008).

## Volatile compounds

Volatile compounds of vinegar were obtained by SPME with a Divinylbenzene/Carboxen/Polydimethylsiloxane (50/30 µm coating thickness; 2 cm length; Supelco, USA) fiber. After homogenization, 3 mL of the vinegar was immediately transferred into SPME vials (volume of 15 mL, Supelco, USA) within 2 min, followed by 10 µL of internal standard containing 81 mg kg<sup>-1</sup> of 2-methyl-3-heptanone (all organic volatile compounds except acids) and 2-methyl-pentanoic acid (for organic volatile acids; Sigma-Aldrich Co., USA) in methanol as internal standard. Vials were placed on a heater at 40 °C for 30 min to accumulate the volatiles up to headspace. Subsequently, fiber was injected in vial to absorb volatile compounds for 30 min. Desorption temperature was 250 °C in the MS sampler. Desorption of the extracted volatiles was carried out on a gas chromatography-mass spectrometry system (Shimadzu GC-2010, QP-2010, Shimadzu Co., Kyoto, Japan). Separation was achieved with the DB-Wax column (60m×0.25mm×0.25mm; J&W Scientific, USA). The volatile content of vinegar was identified by retention index (RI), using an n-alkane series  $(C_{10}-C_{26})$  under the same conditions. Mass spectral libraries of WILEY8 and NIST05 were used for identification.

#### Statistical analysis

All analyses were performed in triplicate and shown as mean  $\pm$  standard deviation (SD). Results were evaluated by performing one-way analysis of variance. Tukey's HSD test with a level of significance of p<0.05 was used to evaluate the differences among means. Statistical evaluation was performed using SPSS 22.0 software (SPSS Inc., USA). Cluster analysis (Ward method and hierarchical) and principal component analysis (PCA) were carried out with the JMP (12.2.0 SAS Institute, Inc., USA). Pearson correlation coefficient was analysed with OriginPro version 2017 (OriginLab, Northampton, USA.).

## **Results and discussion**

## **Microbial analysis**

Vinegar samples had an initial bacterial and fungal load of 2.92  $\pm 0.20$  and 1.31 ±0.07-log CFUmL<sup>-1</sup>, respectively (Table 1). Significant reductions have been achieved in all treated samples. However, higher bacterial reduction has been achieved by increasing the UV-C dose. The highest reduction for the bacterial population occurred at the highest UV-C dose of 262.4 mJcm<sup>-2</sup>. The number of total aerobic bacteria was reduced by 1.29,

0.83 and 0.52-log CFUmL<sup>-1</sup> at UV-C doses of 262.4, 65.9 and 32.9 mJcm<sup>-2</sup>, respectively. Although yeast and moulds were more resistant to ultraviolet treatment compared to bacterial load, an efficient reduction has been achieved in all UV-C doses in terms of fungal load. No Enterobacteriaceae growth was detected for treated and untreated samples. The findings are in accordance with the results of Feng et. al. (2012), who reported a 2.7 JmL<sup>-1</sup> UV-C treatment resulted in 0.6 and 0.53 log-CFUmL<sup>-1</sup> reductions in total aerobic bacteria (TAB) and yeast and mould (YM) count, respectively. Kaya and Ünlütürk (2019) reported that the log-reductions of S. cerevisiae after UV-C treatment were 0.40  $\pm$  0.04-log CFUmL $^{\text{--1}}$  and 0.54  $\pm 0.02$ -log CFUmL<sup>-1</sup> for high (6.36 ±0.04-log CFUmL<sup>-1</sup>) and low  $(4.55 \pm 0.09$ -log CFU mL<sup>-1</sup>) initial loads of verjuice samples, respectively. A 1607 JL<sup>-1</sup> UV-C exposure of orange and tropical juices resulted in 0.89-log and 0.30-log CFUmL<sup>-1</sup> reductions and 0.50 and 0.72-log CFUmL<sup>-1</sup> reductions in TAB and YM, respectively (Keyser et al. 2008). The findings obtained from the study performed with the current reactor, a 441 mJcm<sup>-2</sup> UV-C dose of grape juice inactivated 5.0 and 4.71-log CFUmL<sup>-1</sup> in TAB and YM, respectively (Barut Gök 2018). Microbial inactivation in treated samples was mostly dependent on the dose of the UV-C; however, this UV-C dose could be increased or decreased depending on the flow rate and transmittance or UV energy exposure time (Barut Gök 2021), type of reactor, the properties related to product and methods used for defining the UV-C dose.

## Evaluation of bioactive components

The antioxidant activity of vinegar is shown in Table 2. The results of antioxidant activity of vinegar samples by using the DPPH method did not significantly change after UV-C treatment. A significant decrease in the antioxidant activity of pasteurized vinegar samples observed compared to untreated and UV-C treated ones, regardless of the

**TABLE 1:** Effects of pasteurization and UV-C treatment on microbial load of koruk
 vinegar.

Samples	Total aerobic bacteria count (log CFU/mL)	Total <i>Enterobacteriaceae</i> count (log CFU/mL)	Yeast and mould count (log CFU/mL)
TV	2.92 ± 0.20 <sup>a</sup>	n. d.	1.31 ± 0.07 <sup>a</sup>
PV	1.76 ± 0.07 <sup>c</sup>	n. d.	n. d.
UVV1	1.63 ± 0.19 <sup>c</sup>	n. d.	n. d.
UVV2	2.09 ± 0.22 <sup>bc</sup>	n. d.	<1
UVV3	$2.40 \pm 0.14^{b}$	n. d.	<1

a, b, c Different letters in each row indicate significant difference at (p<0.05) (n = 3 ± SD). n. d., not detected; CFU, colony-forming unit; TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>).

> herein used methods. It has been confirmed in Fig 1 that there is a positive correlation (r=0.975) between DPPH and CUPRAC methods. However, a UV-C dose of 32.9 mJcm<sup>-2</sup> significantly induced the antioxidant activity of UV-C treated vinegars compared to fresh one in terms of CUPRAC. This result could be explained by the activity of phenylalanine ammonia-lyase. It was reported that either the enzyme activity might increase or decrease depending on the UV-C dose delivered. Nigro et al. (2000) reported an increasing phenylalanine ammonia-lyase activity with a low dose of UV-C (0.5 kJm<sup>-2</sup>), while high dose  $(2.5 \text{ kJm}^{-2})$  caused a lower enzyme activity. In a similar way, the results are in accordance with the findings of Bhat et. al. (2011) who reported a non-significant increase in the antioxidant capacity of star fruit juice after a UV-C dose of 2.158 Jm<sup>-2</sup> and antioxidant capacity of organic and conventional grapes after a UV-C dose of 65.6 Jm<sup>-2</sup> (Pinto et al. 2016). It has been reported that a non-significant change in the antioxidant capacity of UV-C treated apple and cranberry juice (Caminiti el al. 2011). Kaya et al. (2020) reported a similar antioxidant activity of untreated, combined treatment of pulsed light-mild heat and pasteurized verjuice samples. Islam et al. (2016) and Erkan et al. (2008) reported a statistically significant difference in antioxidant capacity of UV-C treated apple juice and significant decrease in strawberries.

> The total phenolic content of vinegar was shown in Table 2. There was a significant decrease in the phenolic content of samples after the pasteurization treatment. The UV-C dose of 32.9 mJcm<sup>-2</sup> had no significant effect on the phenolic content but a significant decrease was observed when the dose increased. Similarly, a significant decrease in the phenolic and antioxidant content of carrots (Gamboa-Santos et al. 2013) and red wine pomace (Pino-Garcia et al. 2017) was reported after heat treatment of samples. Various outputs in the literature concerning the relation between the UV-C light and phenolic content of products

**TABLE 2:** Effects of pasteurization and UV-C treatment on some bioactive components and physicocemical properties
 of koruk vinegar.

	Total phenolic compound (mg GAE/L)	Total flavonoids (mg CE/L)	DPPH (mg TEAC/mL)	CUPRAC (mg TEAC/mL)	Ascorbic acid (mg/ 100 mL)	HMF (mg/L)	рН	TA (acetic acid g/L )	oBrix
TV	443.63 ± 2.69°	67.71 ± 1.58 <sup>bc</sup>	$0.68 \pm 0.01^{b}$	$0.79 \pm 0.01^{b}$	2.19 ± 0.08ª	$0.09 \pm 0.01^{a}$	3.41 ± 0.01 <sup>a</sup>	$3.92 \pm 0.06^{a}$	$4.20 \pm 0.00^{a}$
PV	338.98 ± 3.20ª	59.23 ± 0.34ª	$0.63 \pm 0.00^{a}$	$0.76 \pm 0.00^{a}$	1.95 ± 0.17 <sup>a</sup>	$0.12 \pm 0.01^{a}$	3.41 ± 0.01 <sup>a</sup>	3.93 ± 0.03 <sup>a</sup>	$4.23 \pm 0.06^{\circ}$
UVV1	430.39 ± 2.68 <sup>b</sup>	65.77 ± 1.45 <sup>b</sup>	$0.67 \pm 0.01^{b}$	0.77 ± 0.01 <sup>ab</sup>	$2.12 \pm 0.08^{a}$	$0.10 \pm 0.01^{a}$	3.43 ± 0.03 <sup>a</sup>	3.93 ± 0.03 <sup>a</sup>	4.27 ± 0.06 <sup>a</sup>
UVV2	443.63 ± 3.03 <sup>cd</sup>	67.68 ±1.10 <sup>bc</sup>	$0.68 \pm 0.00^{b}$	$0.79 \pm 0.01^{ab}$	$2.17 \pm 0.06^{a}$	$0.10 \pm 0.01^{a}$	3.42 ± 0.00 <sup>a</sup>	$3.92 \pm 0.06^{a}$	$4.20 \pm 0.00^{a}$
UVV3	443.63 ± 2.16 <sup>d</sup>	70.37 ± 0.68°	$0.69 \pm 0.01^{b}$	0.81 ± 0.01°	2.16 ± 0.03 <sup>a</sup>	$0.10 \pm 0.01^{a}$	3.44 ± 0.02 <sup>a</sup>	3.92 ± 0.06 <sup>a</sup>	$4.20 \pm 0.00^{a}$

a. b. c Different letters in each row indicate significant difference at (p<0.05) (n = 3 ± SD). TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2. UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>): UVV3. UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>): GAE. Gallic acid equivalent: DDPH. Radical scavenging activity: CUPRAC. Cupric Reducing Antioxidant Capacity.

are available. The procured results from vinegar demonstrated parallelism with the studies (Noci et al., 2008; Islam et al., 2016) which a significant decrease and a non-significant change were reported in UV-C irradiated apple juices. Unlikely, Barut Gök (2018) reported a significant increase in UV-C treated white grape juice between doses of 239 and 1598 mJcm<sup>-2</sup> and apple juice (1668 mJcm<sup>-2</sup>) (Barut Gök et al. 2021), but not a significant change observed when the red grape juice (1232 mJcm<sup>-2</sup>) was used. Alothman et al. (2009) reported that exposure of UV-C could either increase or decrease the antioxidants, which are strongly dependent on the dose delivered, exposure time and the raw material. However, an induced phenolic content of vinegar content of vinegar samples was detected. This result is in agreement with the findings of Pala and Toklucu (2013) for UV-C treatment of orange juice. Their results showed that UV-C doses of 12.03, 24.06 and 48.12 kJL<sup>-1</sup> caused no significant differences between untreated (fresh) and UV-C treated samples in terms of ascorbic acid content. Unlikely, Bhat et al. (2011), Lemonie et al. (2010) and Vicente et al. (2006) reported a decreasing trend in ascorbic acid content of UV-C treated UV-C-heat treated broccoli, star fruit juice, and heat-treated strawberry samples, respectively. Likewise, Chia et al. (2012) found that the ascorbic acid content of UV-C treated and thermally pasteurised pineapple juices decreased significantly. Previously, Lemo-



FIGURE 1: Pearson's correlation coefficients of phytochemical and physicochemical properties of koruk vinegar.

samples were observed after UV-C doses of 65.9 and 32.9 mJcm<sup>-2</sup>, but not observed when the thermal treatment was used. Moreover, a positive correlation ( $r_{\rm DPPH}$ =0.996;  $r_{\rm CU-PRAC}$ =0.951) was determined between antioxidant capacity and total phenolic content of vinegars.

Grapes are rich sources of phenolic compounds such as flavonols. A non-significant increase in the total flavonoids of UV-C treated vinegars was recorded (Table 2). However, a significant reduction was detected in the total flavonoids of pasteurized vinegar samples compared to untreated and UV-C treated one. Our results are on par with previous reports on samples exposed to UV-C radiation (Gonzalez-Aguilar et al. 2007). Likewise, Bhat et al. (2011) reported an induced flavonoid content of starfruit juice after UV-C dose of 2.158 Jm<sup>-2</sup>. This could be explained with the fact that UV-C has been shown to have positive interactions, indicating an increase in the enzymes responsible for flavonoid biosynthesis (Alothman et al. 2009).

Fresh vegetables and fruits are important sources of dietary ascorbic acid (Lemonie et al. 2010). It is a significant nutrient that affects antioxidant capacity of product (Esteve et al. 2005; Chia et al. 2012). Table 2 shows Vitamin C content in treated vinegar following exposure to UV-C of different doses. Non-significant change in the vitamin C nie et al. (2007) reported an increase in ascorbic acid content of broccoli at a UV-C dose of 8 kJm<sup>-2</sup>. However, positive correlation was found between total ascorbic acid content and total phenolic content (r=0.998) as well as total ascorbic acid content and antioxidant activity ( $r_{DPPH}$ =0.995;  $r_{CUPRAC}$ =0.956).

## **HMF content**

UV-C light did not cause a statistically significant change in the HMF content of samples (Table 2). HMF is considered as a quality attribute due to its health concern in the fruit juices. Any process which produces as low as possible HMF is preferable for the product. Similarly, the results of the HMF analysis are consistent with the findings of Ros-Polski et al. (2016) who found that UV-C light is a confident novel method to thermal pasteurisation as it does not contribute to HMF formation. Unlikely, Aguilar et al. (2016) reported that the absorbed dose of irradiation increases when the HMF concentration was also increased. UV-C

radiation could be concluded as an alternative to thermal methods in order to reduce microorganisms as well as to eliminate HMF generation.

## **Colour analysis**

Significant differences in the colour parameters were recorded in vinegars after UV treatments (Table 3). A significant decrease in the L\* value was detected corresponding to increased UV-C dose. This enhancement might be based on the degradation of colour compounds, making the vinegar darker. However, parameters  $a^*$  and  $b^*$  showed an increasing and decreasing trend corresponding to UV-C dose, respectively. Previously, similar observations were determined in grape juice according to previous research studied with the current UV-reactor (Barut Gök 2021) and verjuice samples treated with pulsed light and mild heat treatment combination (Kaya et al. 2020). Similar to the findings, a decrease in the  $L^*$  value was detected after UV-C irradiation of grape juice (Müller et al. 2014). A lower  $b^*$  and a higher  $a^*$  value causes the intensity of red colour to increase. This observation is consistent with the results of other researchers such as Ibarz et al. (2005), Bhat et al. (2011) and Falguera et al. (2011) who have reported a decrease in  $b^*$  value of fruit juices. Guer-

rero-Beltrán and Barbosa-Cánovas (2005) found a decrease in  $b^*$ , reporting the photodestruction of the pigments of the UV-C treated apple juice. Additionally,  $\Delta E$  representing the magnitude of colour change showed a significant increase after all treatments. Similarly, Falguera et al. (2013) and Kaya et al. (2020) reported an increase in  $\Delta E$  of UV-C treated watermelon juice with the doses of 2.7 and 3.75 J mL<sup>-1</sup> and verjuices after a combination treatment

vinegar. L\* h\* a\* С н ΔE TV 30.00 ± 0.15<sup>d</sup> 12.99 ± 0.09<sup>a</sup> 17.90 ± 0.24<sup>d</sup> 22.12 ± 0.25<sup>cd</sup> 54.04 ± 0.196 ΡV 29.61 ± 0.14<sup>d</sup> 13.62 ± 0.02° 17.89 ± 0.08<sup>d</sup> 52.71 ± 0.12<sup>d</sup>  $0.79 \pm 0.09^{a}$  $22.49 \pm 0.07^{d}$ UVV1 28.46 ± 0.27<sup>a</sup> 13.21 ± 0.04<sup>b</sup>  $14.47 \pm 0.24^{\circ}$  $19.59 \pm 0.20^{\circ}$ 47.59 ± 0.39<sup>a</sup>  $3.78 \pm 0.60^{\circ}$ UVV2 28.72 ± 0.07<sup>b</sup> 13.94 ± 0.06<sup>d</sup> 16.19 ± 0.11<sup>b</sup> 21.36 ± 0.12<sup>b</sup> 49.27 ± 0.09b 2.35 ± 0.31<sup>b</sup> UVV3  $29.19 \pm 0.04$ 13.86 ± 0.05<sup>d</sup>  $17.01 \pm 0.06^{\circ}$  $21.95 \pm 0.08$  $50.83 \pm 0.02^{\circ}$ 1.51 ± 0.20<sup>at</sup>

**TABLE 3:** Effects of pasteurization and UV-C treatment on colour values of koruk

a, b, c, d, e Different letters in each row indicate significant difference at (p<0.05) (n = 3 ± SD). TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>).

of pulsed UV light and mild heat treatment, respectively. The UV-C treatment induces significant alterations in the chroma and hue values which is the angle between  $a^*$  and  $b^*$ , indicating the colour saturation of the analyzed object and the value is the ratio between  $a^*$  and  $b^*$  (Harder et al. 2009), respectively. Juices become less yellow and redder when the hue decreases (Esteve and Frigola, 2007; Chia et al. 2012). The reduction in chroma indicates that the colour of juices became less saturated with applications.

## Brix, pH and titratable acidity (TA)

The acidity and pH are important parameters that could be used as one of the most reliable indicators to evaluate the overall qualities for juice processing (Bhat et al. 2011). No significant effects were detected in the °Brix, pH and TA after UV-C treatments (Table 2). The results showed a parallelism with the studies (Noci et al. 2008; Caminiti et al. 2011; Syed et al. 2019; Kaya et al. 2020; Barut Gök 2021) regarding the data obtained that UV-C light did not induce significant changes in different juices and food



FIGURE 2: Sensory analysis chart for koruk vinegar. TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-Ctreated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>).

products. Unlikely, Bhat et al. (2011) reported that UV-C irradiation caused significant reduction in the acidity, which ranged between 6.24% and 6.73% in UV-C treated samples.

## Sensory analysis

The efficacy of UV-C on sensorial attributes of vinegar are shown in Fig 2 and Table 4. The sensorial analysis showed non-significant differences for doses used. The findings are parallel to Riganakos et al. (2017) and they were in correspondence with previous results (Manzocco et al. 2011), that did not report significant change in UV-C treated carrot juice and melon cubes, when compared with untreated ones (Falguera et al., 2011) Sensory analysis showed that the UV-C irradiated vinegar had better sensorial attributes, as compared to the conventional pasteurized ones. Significant reductions were observed in the sensorial attributes in terms of pungent sensation, richness in aroma and aromatic intensity of heat-treated samples. Similarly, Falguera et al. (2011) reported that thermally pasteurized

> apple juice tends to change colour and lose most of its flavour and vitamins during processing, unlike with the UV-C treated juice.

## Volatile compounds

A total of 22 and 19 volatile compounds were determined in the traditionally produced (TV) and pasteurized vinegars (PV), respectively (Table 5). Volatile compounds in the UV-C treated vinegars varied between 17 and 23. Six classes of free volatile complunds were characterized in the vinegar: (1) aldehydes (three compounds); (2) alcohols (five compounds); (3)terpenes (five compounds); (4)esters (seven compounds); (5)acids (five compounds). Vinegar flavour was especially affected by the raw ethanolic product from which it was made (Bhat et. al. 2014). Esters were the predominant aroma volatiles in UVV, PV and TV especially; ethyl acetate which has a fruity odour was the most abundant volatile compound, being major constituents of all samples. Untreated vinegars showed higher ester values. Esters are related to odour descriptors that represent a pleasant fruit-like smell. Treatments did not induce a significant change in esters like ethyl acetate, ethyl octanoate. However, pasteurization and the highest dose of UV-C did a significant decrease in ethyl heptanoate. Among the three aldehydes identified in the vinegar, benzaldehyde represented the largest portion of total quantified aldehydes.

**TABLE 4:** Colour, pungent sensation, richness in aroma, impression general, taste, aromatic intensity and ethyl acetate odour scores of koruk vinegar.

	Colour	Pungent sensation	Richness in aroma	Impression general	Taste	Aromatic intensity	Ethyl acetate odour
TV	7.58 ± 0.51ª	7.50 ± 1.17 <sup>b</sup>	7.33 ± 0.78 <sup>b</sup>	7.58 ± 1.16ª	7.42 ± 0.51 <sup>bc</sup>	7.58 ± 0.79 <sup>b</sup>	3.50 ± 0.67 <sup>a</sup>
PV	$7.00 \pm 0.60^{a}$	6.17 ± 0.72 <sup>a</sup>	6.33 ± 0.89ª	6.83 ± 0.72 <sup>a</sup>	6.25 ± 0.62 <sup>bc</sup>	5.67 ± 0.98°	2.92± 0.67 <sup>a</sup>
UVV1	7.42 ± 0.51 <sup>a</sup>	$6.75 \pm 0.87^{ab}$	7.33 ± 0.65 <sup>b</sup>	7.25 ± 0.97 <sup>a</sup>	6.58 ± 1.08 <sup>bc</sup>	$6.67 \pm 1.61^{ab}$	3.00 ± 0.60 <sup>a</sup>
UVV2	7.50 ± 0.52 <sup>a</sup>	7.78 ± 0.67 <sup>b</sup>	7.67 ± 0.78 <sup>b</sup>	7.58 ± 1.08 <sup>a</sup>	7.17 ± 0.94 <sup>bc</sup>	7.33 ± 0.49 <sup>b</sup>	2.92 ± 0.79 <sup>a</sup>
UVV3	$7.67 \pm 0.78^{a}$	7.50± 0.52 <sup>b</sup>	$7.25 \pm 1.06^{ab}$	7.67 ± 0.89 <sup>a</sup>	7.92 ± 1.00 <sup>bc</sup>	7.75 ± 0.62 <sup>b</sup>	3.67± 0.49 <sup>a</sup>

a.b.c.d.e Different letters in each row indicate significant difference at (p<0.05) (n = 3 ± SD). TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>).

The amount of this benzenoid decreased in pasteurization and UV-C treatment compared to untreated vinegar. Unlikely, it was reported that some treatments such as heat pasteurization might inactivate enzymatic transformations or induce chemical reactions affecting organoleptic properties of grape juice (Martinez et al. 2017). The identified  are in accordance with Yuan and Qian (2015); Slegers et al. (2015) and
 Martinez et al. (2017) who have reported the aroma profile before in grape and grape-derived
 products.
 All treatments caused

components of traditio-

nally produced vinegar

All treatments caused a reduction in the total amount of aldehydes, ke-

tones, acids and terpenes. However, an increase has been observed in alcohols of all treated samples. Although the highest increase was detected in PV, herbaceous odorant C6 alcohols such as (3-hexanol, 2-hexanol) were increased in all treated samples. The findings of the results are correndance with the result of Martinez et al. (2017) who reported an increase in the concen-

**TABLE 5:** Determination of volatile profiles of koruk vinegar.

Organic volatile compounds	RI	TV	PV	UVV1	UVV2	UVV3
Acetic acid	1457	31.14±5.65 <sup>b</sup>	19.86±0.91ª	18.55±1.36ª	22.86±2.79 <sup>ab</sup>	23.18±2.48 <sup>ab</sup>
Butanoic acid	1632	1.61±0.36°	n. d.	0.56±0.08 <sup>ab</sup>	1.03±0.32 <sup>bc</sup>	0.97±0.40 <sup>bc</sup>
Hexanoic acid	1851	2.85±0.24ª	2.17±0.45ª	2.46±0.44 <sup>a</sup>	3.14±0.77ª	3.08±0.61ª
Octanoic acid	2064	4.13±0.80 <sup>ab</sup>	2.70±0.65ª	3.55±0.57 <sup>ab</sup>	3.49±0.50 <sup>ab</sup>	4.45±0.65 <sup>b</sup>
Decanoic acid	2254	1.09±0.32 <sup>b</sup>	n. d.	n. d.	0.21±0.18ª	n. d.
Phenethyl alcohol	1926	21.16±3.16 <sup>a</sup>	19.78±2.98ª	28.71±4.26 <sup>ab</sup>	30.85±3.03 <sup>b</sup>	33.10±4.24 <sup>b</sup>
1-hexanol	1346	1.88±074ª	3.17±0.66 <sup>ab</sup>	1.93±0.56ª	1.89±0.20ª	4.03±1.23 <sup>b</sup>
2-hexanol	1228	n. d.	1.14±0.15ª	1.93±0.34 <sup>ab</sup>	1.89±0.35 <sup>ab</sup>	2.47±0.67 <sup>b</sup>
3-hexanol	1209	2.21±0.58ª	5.10±0.82 <sup>b</sup>	3.85±0.31 <sup>ab</sup>	3.91±0.57 <sup>ab</sup>	4.52±1.41 <sup>b</sup>
1-pentanol	1247	3.66±0.67 <sup>b</sup>	2.95±0.86 <sup>b</sup>	n. d.	3.57±0.79 <sup>b</sup>	4.13±0.96 <sup>b</sup>
Benzaldehyde	1542	4.89±1.12 <sup>b</sup>	1.88±0.42ª	2.84±0.69ª	3.01±0.48 <sup>a</sup>	3.16±0.35 <sup>ab</sup>
Hexanal	1082	2.58±0.73 <sup>b</sup>	1.19±0.62 <sup>ab</sup>	n. d.	1.81±0.63 <sup>b</sup>	1.75±0.18 <sup>b</sup>
Beta-lonone	1964	6.11±1.61ª	3.18±0.86ª	4.35±0.33ª	4.47±1.31ª	5.27±0.97ª
2-heptenal	1325	n. d.	n. d.	n. d.	n. d.	0.11±0.10 <sup>a</sup>
Ethyl acetate	867	54.81±9.51 <sup>a</sup>	35.74±7.94ª	34.10±5.78ª	39.77±15.31ª	42.17±6.22ª
Ethyl heptanoate	1343	8.76±1.29 <sup>b</sup>	3.84±1.01ª	4.01±0.98 <sup>a</sup>	5.60±1.81 <sup>ab</sup>	5.49±1.84 <sup>ab</sup>
Ethyl octanoate	1441	20.03±4.11ª	13.79±2.69ª	15.43±4.08ª	17.48±4.72ª	18.92±1.35ª
Isoamyl acetate	1125	4.85±1.36 <sup>d</sup>	n. d.	1.03±0.47 <sup>ab</sup>	2.45±0.58 <sup>bc</sup>	3.36±0.87 <sup>cd</sup>
Phenethyl acetate	1825	9.88±1.27 <sup>b</sup>	6.41±2.08 <sup>ab</sup>	4.49±0.45°	7.03±1.28 <sup>ab</sup>	6.98±1.12 <sup>ab</sup>
Ethyl laurate	1855	12.25±1.53 <sup>b</sup>	6.77±1.11ª	4.21±0.97 <sup>a</sup>	4.15±0.80 <sup>a</sup>	6.19±1.34ª
Ethyl palmitate	2280	n. d.	n. d.	n. d.	$0.14 \pm 0.14^{ab}$	0.22±0.05 <sup>b</sup>
para-cymene	1276	5.95±1.91 <sup>b</sup>	2.48±0.61ª	n. d.	1.10±0.35ª	1.55±0.39ª
Limonene	1198	19.81±2.48 <sup>b</sup>	11.47±1.97ª	9.44±1.58ª	11.36±1.45ª	13.55±2.52ª
Styrene	1248	2.07±0.66 <sup>b</sup>	0.47±028ª	n. d.	n. d.	0.55±0.24ª
Gamma-terpinene	1239	0.19±0.17 <sup>a</sup>	n. d.	n. d.	n. d.	n. d.
Total (mg/kg)		22	19	17	22	23
esters		110.58	66.55	63.27	76.62	83.33
alcohols		28.91	32.14	36.95	42.98	48.25
aldehydes		7.47	3.07	2.84	4.82	5.02
ketones		6.11	3.18	4.35	4.47	5.27
acids		40.82	24.73	25.12	30.73	31.68
terpenes		28.02	14.42	9.44	12.46	15.65

n. d., not detected; TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (35.9, mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>); RI, Retention index.

tration of herbaceous odorant C6 alcohols after hot-press juice processing. Although pasteurization induced a non-significant reduction in phenethyl alcohol, UV-C treatment induced a significant increase in this aroma compound. Decanoic acid which is a sour and citrus odour descriptor has lost in pasteurization and UV-C treatments except the dose of 65.9 mJcm<sup>-2</sup>.

Quantitation data are necessary to verify the changes in the aroma profile of vinegar to better understand the response of odour components in it. Factor analysis is shown in Fig 3 A. Detected volatile compounds in vinegar enabled good discrimination between all treatments and untreated samples (Fig 3A). When all of the aroma constituents were included, the PC1 (first principal component) explained 63.2% of the total variation, and PC2 explained 22.7% of the total variation. Samples pasteurized and UV-C treated of 65.9 and 262.4 mJcm<sup>-2</sup> doses were separated in the plot by PC1. The samples treated of 65.9 and 262.4 mJcm<sup>-2</sup> UV-C doses differ from others by a higher score on PC2, while samples UV-C treated at the highest dose, untreated and pasteurized samples are differentiated by their negative PC2 scores. Pasteurized vinegar was located at the bottom, showing a negative score for PC2, while treatment with a UV-C dose of 32.9 mJcm<sup>-2</sup> has an opposite score. UV-C dose of 262.4 mJcm<sup>-2</sup> and pasteurization are located to the left, showing a negative score for PC1, while untreated vinegar is located at the left side. Ethyl acetate and acetic acid have higher positive weight scores, while the compounds 3-hexanol and 2- hexanol which is



FIGURE 3: A: Principal component analysis (PCA) of volatile compounds in koruk vinegar. B: Dendrogram of hierarchical cluster analysis of koruk vinegar samples. TV, Traditionally produced koruk vinegar; PV, Pasteurized koruk vinegar; UVV1, UV-C-treated koruk vinegar (262.4 mJcm<sup>-2</sup>); UVV2, UV-C-treated koruk vinegar (65.9 mJcm<sup>-2</sup>); UVV3, UV-C-treated koruk vinegar (32.9 mJcm<sup>-2</sup>).

green and floral odour descriptor (Yuan and Qian 2015) had negative weight scores on PC1. Ethyl palmitate and phenethyl alcohol, which is a floral and sweet odour descriptor (Martinez et al. 2017) had higher positive scores, while the compound para-cymene and ethyl laurate had negative weight scores on PC2. When classified according to distances in cluster analysis, X (Acetic acid, phenethyl alcohol, ethyl octanoate, limonene), Y (ethyl acetate) and Z (butanoic acid, styrene and so on.) were clustered groups (Fig 3B). In clustering, the most distant (9.49 unit) aroma components were detected between acetic acid and butanoic acid, while the aroma components were detected 2-heptenal at the closest distance (0.01 unit) with ethyl palmitate (Data was not shown). Similarities were found when the graphs were analyzed despite cluster analysis according to distance compared to PCA.

## Conclusion

Thermal pasteurization is usually applied for assuring microbial safety that maintains the product safety during the shelf life period. However, organoleptic and nutritional quality losses may possibly occur depending on the thermal process. The findings of the current work demonstrated that UV-C light is a promising preservation technique with certain advantages. It is capable of inactivating microbial load in vinegar without change in bioactive compounds, sensorial attributes and aroma profile. UV-C light that is applied at relatively low doses might be a suitable alternative to increase vinegar bioactive compounds such as flavonoids and antioxidant capacity.

There is a lack of knowledge on the effects of the UV-C treatment in vinegar samples like traditionally produced from koruk juice. Furthermore, studies considering how the UV-C light influences not only the microbial count but also bioactive compounds, sensory and aroma profile of the product will be worthwhile to evaluate as a food preservation technique. Hence, UV-C could be effectively applied as a non-thermal technique to assure microbial safety while

carrying on the bioactive compounds and sensorial qualities of this vinegar product.

#### Data availability

The authors confirm that the data supporting the findings of this study are available within the article and its supplementary material.

## **Conflict of interest**

The authors declare that; there is no financial/personal interest or belief that could affect our objectivity.

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