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EFFECT OF UV-C LIGHT ON THE INACTIVATION OF *ZYGOSACCHAROMYCES ROUXII* IN APPLE JUICE: EVALUATION OF PHENOLIC AND ANTIOXIDANT CONTENTS

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ABSTRACT
Improved quality and safety can be achieved by UV-C light which is a non-thermal method for fruit juices. *Zygosaccharomyces rouxii* causes spoilage problems in sugar foods, fruit juices and concentrates. In this research, the effect of UV-C light in different doses (4, 8, 12 J/cm^2) was investigated for inactivation of *Z.rouxii* in apple juice and also some quality properties were evaluated such as color, total phenolic and antioxidant contents. As a result, applying 4 J/cm^2 irradiation gave the highest phenolic and antioxidant contents as 229.89±2.19 (mgGAE. mL^{-1}) and 0.032±0.05 (EC_{50}, µg/mL) respectively. Color values significantly affected from the increase in doses of irradiation (P<0.05), The reduction UV-C exposure in *Zygosaccharomyces rouxii* was achieved as 1.45 log CFU ml^{-1} for 4 J/cm^2; 2.09 log CFU ml^{-1} for 8 J/cm^2 and lastly, 1.94 CFU/ml for 12 J/cm^2 treatment. These findings indicate that low doses of UV-C irradiation can effectively inactivate organisms while preserving quality characteristics of apple juice.
Keywords: Apple juice, ultraviolet light, irradiation, *Zygosaccharomyces rouxii*, phenolic content, quality

UV-C IŞIĞININ ELMA SUYUNDAKİ *ZYGOSACCHAROMYCES ROUXII*'NİN İNAKTIVASYONUNA ETKİSİ: FENOLİK VE ANTİOKSİDAN İÇERİKLERİNİN DEĞERLENDİRİLMESİ

ÖZ
Meyve sularında kalite ve güvenlik, termal olmayan bir yöntem olan UV-C ışığı ile artırılabilir. *Zygosaccharomyces rouxii,* şekerli gıdalarda, meyve sularında ve konsantrelerde bozulmaya neden olan bir mikroorganizmadır. Bu araştırmada, farklı dozlarda (4, 8, 12 J/cm^2) uygulanan UV-C ışığının elma suyundaki *Z. rouxii*'nin inaktivasyonu üzerindeki etkisi incelenmiş ve ayrıca renk, toplam fenolik madde içeriği ve antioksidan kapasite gibi bazı kalite özellikleri değerlendirilmiştir. Sonuçlar, 4 J/cm^2 ışınlamanın en yüksek fenolik ve antioksidan içerik değerlerini sırasıyla 229.89±2.19 mg GAE/mL ve 0.032±0.05 EC50 (µg/mL) olarak sağladığını göstermiştir. Renk değerleri, ışınlama dozlarının artışıyla belirgin şekilde değişmiştir (P< 0.05).UV-C ışığına maruz bırakılan *Z. rouxii*'nin azalması sırasıyla 4 J/cm^2 için 1.45 log CFU/mL, 8 J/cm^2 için 2.09 log CFU/mL, 12 J/cm^2 için 1.94 log CFU/mL olarak saptanmıştır. Bu bulgular, düşük dozda UV-C ışığı uygulamasının bozulmaya neden olan mikroorganizmaları etkili bir şekilde inaktive ederken elma suyunun kalite özelliklerini koruyabileceğini göstermektedir.
Anahtar kelimeler: Elma suyu, ultraviyole ışık, ışınlama, *Zygosaccharomyces rouxii*, fenolik madde, kalite

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INTRODUCTION

Apple (*Malus domestica* Borkh.; Rosaceae) is considered to be one of the most economically and culturally significant fruits, which is grown in all temperature zones (Patocka *et al.,* 2020; Spengler, 2019). Apples are a rich source of various nutrients, such as dietary fibers, vitamins, minerals, phenolic and bioactive compounds (Müller et al., 2021; Hyun and Jang, 2016). Its high nutritional value increases its preference among consumers. It is known that it has anti-cancer, anti-diabetic, anti-inflammation and anti-obesity effects (Hyun and Jang, 2016). Thus, apple juice is a mostly preferred fruit juice by the consumers worldwide (Müller et al., 2021).

Fruit juices have a low pH value and are typically abundant in carbohydrates and complex nitrogen sources, in that way offering optimal growth substrates for the proliferation of spoilage yeasts, molds, and certain acid-tolerant bacteria (Aneja *et al*., 2014). Microbial spoilage has the potential to cause unfavorable alterations in the nutritional and sensory characteristics of fruit juices and beverages. Yeasts can be tolerable to the high acidity found in fruit juices and can easily thrive in anaerobical conditions (Saloma, 2018). However, certain strains of yeasts can cause alteration in different food and beverages such as wine and fruit juices (Hernandez *et al*., 2018). In fruit juices generally *Zygosaccharomyces, Candida, Saccharomyces, Rhodotorula, Issatchenkia, Hanseniaspora, and Pichia* causes spoilage (Xiang *et al*., 2020; Hernández *et al.,* 2018). However, yeasts belonging to the *Zygosaccharomyces* genus are regarded as the most common spoilage yeasts in sugary food and beverage products, and are the reason for significant financial losses within these sectors (Hernández *et al*., 2018, Rojo *et al.,* 2015, 2017). Hereby, it is clear that this microbiological degradation in the fruit juice industry has major economic consequences (Marvig *et al*., 2015).

Zygosaccharomyces rouxii is a facultative anaerobic yeast that can tolerate ethanol, sulfur dioxide and acetic acid and grow to 1.8-8 pH and 0.62 water activity (Karaman, 2020; Sperber and Doyle, 2009). The spoilage caused by *Zygosaccharomyces rouxii* strains, comprehended by consumers with the formation of non-desired odors affecting the products. These odor compounds effect quality and may cause many waste in this type of products (Escott *et al*., 2018).

Thermal treatments are used in food industry to protect the last product by inhibiting the growth of microorganisms and enzymes (Riganakos *et al*., 2017). Nevertheless, thermal processing may cause some undesirable changes such as nutrient loss, color alteration and changes in sensory attributes (Riganakos *et al.,* 2017). It especially affects the color quality of anthocyanin containing fruit juices (such as apple and grapefruit), since anthocyanins degrade and form colorless or unwanted brown-colored pigments (Pala and Toklucu, 2011; La Cava and Sgroppo, 2019). Due to the growing consumer preference for healthy and minimally processed, preservative-free products, there is an effort to create innovative non-thermal technologies that can increase the quality and extend the shelf life of such products (La Cava and Sgroppo, 2019). For this reason, new non-thermal processes has been devoloped and combined, such as UV-C, pulsed electric field, and high hydrostatic pressure achieving equivalent or even higher degrees of stability and safety in food processing (La Cava and Sgroppo, 2019). Non-thermal pasteurization methods can help to denature enzymes and eliminate microorganisms with less harmful effects on sensory and nutritional quality of foods (Xiang *et al*., 2018).

In recent decades, a range of non-thermal techniques has been developed, with ultraviolet light (UV) emerging as an outstandingly promising technology. United States Food and Drug Administration (USFDA) and the United States Department of Agriculture (USDA) specified the Ultraviolet (UV) irradiation as an alternative food processing technology for fruit juices in recent years (Feliciano *et al*., 2019). This is mainly beacuse of its user-friendly nature and its ability to efficaciously eliminate a wide array of microorganisms (Bintsis *et al*., 2000). Moreover, UV treatment does not generate any chemical residues, so its appeal as a reasonable method for microbial control enhances (Guerrero–Beltrán

and Barbosa–Cánovas, 2004). UV light is between 100 and 400 nm region of the electromagnetic spectrum. This UV range may be further divided and classified as UV-A (315–400 nm), UV-B (280–315 nm), UV-C (200–280 nm), and the vacuum UV range (100–200 nm) (Baysal *et al.*, 2013). The UV-C light has germicidal effect on microorganisms such as bacteria, yeasts, molds and viruses (Caminiti *et al.*, 2012).

Recent studies demonstrated that UV-C light technology is one of the technologies studied to preserve the fruit juices such as orange juice (Prado *et al.*, 2019; Feliciano et al., 2019; Hakguder Taze *et al.*, 2015), carrot juice (Riganakos *et al.*, 2017), grapefruit juice (La Cava and Sgroppo, 2019; Unluturk and Atilgan, 2015), mango juice (Santhirasegaram *et al.,* 2015), kale juice (Pierscianowski et al., 2021) and apple juice (Baysal e*t al.*, 2013; Gouma *et al,* 2015; Xiang *et al.*, 2020). On the other hand, studies have been conducted on trying novel treatments for inactivation of different yeasts or bacteria that cause food spoilage in apple juice with UV-C (Baysal *et al.,* 2013; Sauceda-Gálvez *et al.*, 2020; Gabriel, 2012; Gouma *et al.*, 2015; Akwu *et al.*, 2021), but limited study investigated the UV-C effect on the *Z. rouxii* inhibition in apple juice (Xiang *et al.*, 2020). Previous researches investigate the UV-C effect on different microorganisms in apple juice such as Sauceda-Gálvez et al. (2020), investigate the single and combined UV-C and ultra-high pressure homogenisation treatments on inactivation of *Alicyclobacillus acidoterrestris* spores in apple juice also the same researchers evaluate the ultraviolet light (uv-c) effectiveness in the inactivation of bacterial spores inoculated in cloudy apple juice. In another study, Akwu et al. (2022), investigate the effect of germicidal short wave-length ultraviolet light on the polyphenols, vitamins, and microbial inactivation in highly opaque apple juice. Akgün and Ünlütürk (2017), determine the effects of ultraviolet light emitting diodes (LEDs) on microbial and enzyme inactivation of apple juice. Gabriel et al. (2012), inactivation of Escherichia coli O157:H7 and spoilage yeasts in germicidal UV-C-irradiated and heat-treated clear apple juice. They evaluate different doses and also investigate the effect on the individual polyphenolic content and in-vitro total antioxidant activity of apple juice (Islam et al., 2016). By this point of view this study will also add valuable contribution to the literature. Also there is no study in specially evaluating the phenolics and antioxidants after this effect.

Therefore in this study, it was aimed to investigate the inactivation effects of UV-C technology on the *Zygosacchoremyces rouxii* in clear apple juice. Additionally comparing the quality effects such as phenolic and antioxidant contents and color between control (not UV-C light irradiated) and UV-C application groups.

MATERIAL AND METHODS

Materials

Commercially available pasteurized apple juice (Harvest 100% Apple Juice, Dimes, İzmir, Turkey) was provided from a local market in İzmir, Turkey. Chemicals were analytical grade and provided by Merck (Darmstadt, Germany) or Sigma-Aldrich Trading Co. Ltd. (Buchs, Switzerland).

Culture and Medium Preparation

This study was carried out with osmophilic yeast, *Zygosaccharomyces rouxii* (DSM 7,525, DSMZ GmbH, Germany), supplied as lyophilized. According to the DSMZ, yeast malt agar was created from different media components including yeast extract, 3 g/L; malt extract, 3 g/L; peptone, 5 g/L; dextrose, 10 g/L; and agar 20 g/L (Merck, Darmstadt, Germany). Yeast cells were grown on liquid medium and incubated at 30°C shaker incubator (Biosan, ES-20, Latvia), at 120 rpm for 18 hr (Xiang et al., 2018). 2-ml sterilized water was added for the suspension of the lyophilized culture then transferred to liquid medium and incubated at 25°C in static incubator (Nüve En 30, Turkey) at least 3 days (Chen and Tseng, 1997). The last concentration of the culture was determined as almost 10^7 CFU/ml. Cells were applied to centrifuge process (6000 rpm, 10 min, 4 °C) after growing by a centrifuge (Hettich mikro centrifuge, 200R, Germany). After this, supernatant was removed, and a tube was filled with sterile 0.1% peptone water (Xiang et al.,

2020). Uninoculated apple juices were used as the control samples.

Inoculation of Yeast and UV-C Treatment of Apple Juice

UV-C (UVP XX–15, UVP Inc., CA, USA) application was carried out using an mercury UV lamp with peak radiation at 254 nm wavelength. Flat black painted tube was used for the UV radiation which was in the same size with a Petri dish. The samples were placed in 6 cm diameter Petri dishes directly below the collimated UV beam and stirred continuously during the irradiation with a vortex mixer at 14000rpm (IKA, Yellowline TTS 2, IKA® Werke GmbH & Co. KG, Germany). The irradiance I0 of the lamp was measured by a UV–VIS radiometer supplied with UVX–25 sensor (UVX, UVP Inc., CA, USA) placed at the same distance from the UV lamp as the plates. The UV lamp was preheated for approximately 30 minutes before starting the UV treatment to ensure a consistent intensity level (Baysal *et al.*, 2013).

For UV-C application, 25 ml of apple juice inoculated with 0.25 ml *Z. rouxii* cells was transferred to sterilized plastic petri dishes (90 mm×15 mm).The distance of apple juices to the lamp was kept constant as 10 cm, from this distance, different doses (4, 8, 12 J/cm^2) were processed by taking into consideration of different studies and also equipment properties as reference (Hayes et al., 2012; Xiang et al., 2020; Sauceda-Gálvez et al., 2020). The UV intensity was kept constant at 0.0071 W/cm^2**.** Exposure times were equal to UV doses divided by UV intensities (J/cm^2). The exposure times of the samples to UV-C application were calculated as 9.38, 18 and 28 minutes, respectively.

Microbiological analysis

After UV-C treatment, for the determination of microbial counts 1 ml of each sample was taken and transferred to 9 ml peptone water (0.1%) dilutions were made. Counts were determined by plating the diluted samples onto yeast malt medium by taking 0.1 ml from the dilutions. After plating, the petri dishes were left to incubate at 25 ^{o}C for 3 days, and the microorganism counts in the petri dishes were performed two replications. Results are expressed as $\log_{10}$CFU/mL (Chen and Tseng, 1997).

Determination of physicochemical properties of apple juice

Total soluble solids content

Total soluble solids (TSS) were measured by a digital refractometer (Hanna, Romania) at 25°C. Results are expressed as °Brix (AOAC, 2000).

pH

The pH values of apple juices at 25°C were determined using a pH meter (Thermo Scientific Orion 3 Star, USA) (AOAC, 2000).

Total titratable acidity

Titratable acidity (TA) determination was carried out with using 0.1 M NaOH until pH 8.2 was reached and the results were expressed as "g malic acid/100 mL apple juice"(g TA·100^{-1} mL^{-1}) Titratable acidity was calculated as shown in Equation 1.

$$\text{Titratable Acidity \%} = V * F * E * 100 / m \quad (1)$$

where V is the amount of 0.1 N NaOH used (ml), F is the factor of the base solution used in the titration, E is 1 ml of 0.1 N NaOH equivalent of acid amount (g), m is the actual amount of sample titrated (ml) (Cemeroğlu, 2018).

Total phenolic content

The total phenolic compounds in the apple juice was determined by using the Folin-Ciocalteu method with a modification (Franke et al., 2014; Xiang et al., 2020). Gallic acid was used as a standard. Sample extract (500 µL) was mixed with Folin-Ciocalteu reagent (1.0 mL). A total of 2.0 mL of sodium carbonate solution (7.5%, w/v) and 1.5 mL of distilled water were added to the mixture. The solution was then vortexed thoroughly for 15 seconds and left to rest at room temperature for 60 minutes. The absorbance at 760 nm wavelength was measured using a Varian Cary 50 Scan (Australia) model spectrophotometer. The results were reported as milligrams of gallic acid equivalent per 100 mL of apple juice (mg GAE·100^{-1} mL^{-1}).

Determination of Total Antioxidant Activity

For the determination of antioxidant activity of apple juice, DPPH assay is used. After the necessary dilutions were made (first diluted 1:10 with 80% methanol, then diluted again 1:10 from the dilute sample); 20, 40, 60, 80 and 100 µl of the extract were transferred to 5 different tubes. 600 µl DPPH solution was added on each tube. The total volume in each tube was then made up to 6 mL with methanol. After the tubes were kept in a dark environment at 25ºC for 15 minutes, the absorbance of the samples were determined on a spectrophotometer at a wavelength of 517 nm using methanol as a blank (Cemeroğlu, 2018). Antioxidant activity was expressed as (EC_{50}, µg/mL) (Rydzak *et al.,* 2020).

Color parameters

Colorimetric measurement of the samples was carried out using a Chromameter (Konica Minolta CR- 600, Japonya). The color values were expressed as the L*, a* and b*. L* represents whiteness or brightness/darkness, a* is the red/green coordinate and b* the yellow/blue coordinate (Gök, 2021). The L* value represents lightness and darkness, with a higher L* value indicating a lighter-colored product. An increase in the b* value means a higher yellow intensity and an increase in the a* results in a higher red intensity. The instrument was calibrated before each analysis.

Statistical Analyses

The results were statistically analyzed using analysis of variance (ANOVA) in SPSS 13 software (SPSS Inc., Chicago, IL, USA), with the Duncan test applied to determine differences among treatments at a significance level of $P<0.05$. Each experiment was conducted at least three times, and the means and standard deviations were calculated for the results.

RESULTS AND DISCUSSION

Assesment of microbial inactivation

The initial microorganism numbers of *Zygosaccharomyces rouxii* were determined as 3.24 x10^7 CFU ml^{-1} for apple juice. Apple juice samples were exposed to UV-C radiation doses of 4 J/cm^2, 8 J/cm^2 and 12 J/cm^2. The reduction in *Zygosaccharomyces rouxii* was achieved as 1.45 log CFU ml^{-1} for 4 J/cm^2 radiation exposure; 2.09 log CFU mL^{-1} for 8 J/cm^2 and lastly, there was a reduction of 1.94 CFU mL^{-1} for a 12 J/cm^2 radiation treatment. It can be seen that the most reduction of microorganisms was obtained after a 8 J/cm^2 radiation treatment. Similar to this study, Galvez et al., (2020) explained that the increase in doses not provide an increase in lethality. Similarly, Taze *et al* (2015) studied with a UV dose of 108.42 mJ/cm^2, and found the maximum log reduction in yeast and mould count as 1.76 log_{10} after 20 min of UV exposure (I0=1.32mW/cm^2). In parallel with this, Bhat *et al.* (2011), remarked a reduction in yeasts and mould counts by 2-log cycle on UV treatments. Keyser et al.(2008), found a 1.32 log CFU/ml- 4.48 log CFU/ml reductions in, strawberry and guava-and-pineapple juice and mango nectar. At UV intensity levels of 1.31, 0.71, and 0.38 mW/cm^2, log reductions of 2.1±0.3, 1.6±0.1, and 0.8±0.1, respectively, were achieved in apple juice after 15 minutes of treatment (Baysal et al., 2013). UV-C treatments proved to be more effective, reaching a lethality of 5.5 log10 CFU/mL with a dose of 21.5 J/mL at 20°C. In contrast, UV-C treatments were less efficient at higher doses, achieving a maximum lethality of 4.07 log10 CFU/mL with a dose of 28.7 J/mL (Sauceda *et al*., 2020).

In another study, (Fenoglio *et al*., 2020*), L. plantarum, E. coli,* and *S. cerevisiae* microorganisms were cultivated in clear pear juice and the inactivation effect of UV-C application on these microorganisms was examined. 4.4-5.5 log reductions were found for *L. plantarum, E. coli and S. cerevisiae*. In the study conducted by Lapena *et al.* (2022), the inactivation of *Escherichia coli*, *Salmonella enterica* and *Listeria monocytogenes* was assessed under different ultraviolet C (UVC_{254nm}) treatments in apple juice. *E. coli and S. enterica* populations were significantly reduced ($P<0.001$) after UVC_{254nm} treatment with 904.0±1.0 mJ/cm^2 and a dose of 1200.0±1.0 mJ/cm^2 was needed to significantly reduce ($P<0.001$) of *L. monocytogenes*. After 20 minutes of UV-C exposure (16.8 kJ/m^2), the spores of *Alicyclobacillus acidoterrestris*, *Alicyclobacillus herbarius*, and *Alicyclobacillus cycloheptanicus* were significantly reduced by more

than 4 log CFU/mL, with counts falling below the detection limit of the method (<1.7 log CFU/mL). Meanwhile, the spores of *Alicyclobacillus acidocaldarius* were even more sensitive to UV-C, with a similar reduction observed after just 15 minutes of exposure (12.6 kJ/m²) (Baysal *et al*., 2013). The populations of *Z. rouxii* in apple juice were reduced by 4.86-log and 5.46-log values following UVC-LED irradiation at doses of 800 and 1200 mJ/cm², respectively (P<0.05) (Xiang et al., 2020). Additionally, a flow rate of 0.0078 L/min (frequency 30 Hz) with a UV-C dose of 13.75 mJ/cm² effectively reduced *S. typhimurium* by 5 log10 CFU/mL in pineapple juice, meeting the FDA standards (Mansor *et al*., 2014). In a previous research it was determined that the decreases in inactivation of yeasts *Z. rouxii, Z. bailii and S. cerevisiae* were significant (P<0.05). Compared to control group with a UV dosage of 3.36 mJ/cm², 3.8-5.0 log reductions found (Hayes et al., 2012). In a study, 10 mJ/cm² UV dose at 0.288645 mW/cm² average irradiance resulted in 3.4 $\log_{10}$ CFU/mL reduction of *E. coli* at a depth of 1.5 cm using UV-LEDs emitting light at 263 nm (Akwu *et al*., 2022). The inactivation of *S. aureus* followed a linear decrease with increasing exposure time, and after 30 minutes of UV exposure, a notable reduction in bacterial levels was observed in the apple juice. Xiang *et al.* (2020), who studied with *Z. rouxii* in apple juices, found similar results to this study. 4.86- and 5.46-log values after UVC-LEDs irradiation at 800 and 1200 mJ/cm² (P<0.05), respectively.

Total soluble solid content, pH and titratable acidity

Physical and chemical properties such as pH, total soluble solids, titratable acidity, total phenolic content, total antioxidant content and color values (L*,a*,b*) of the apple juice control samples were shown in Table 1. Total soluble solid content and titratable acidity were found in untreated (control) samples as 11.3±0.05 (ºBx) and 0.29±0.009 (%). According to Müller et al. (2014), untreated pH value of apple juice was found to be as 3.62±0.04 similar to this study (3.93±0.07).

Table 1. Physicochemical properties for control and treated samples

Quality parameters	Different doses of radiation (J/cm²)			
	Control samples	4	8	12
Total soluble solid content (ºBrix)	11.3±0.05^{a}	11.55±0.0^{a}	11.48±0.037^{b}	11.22±0.068^{b}
pH	3.93±0.007^{a}	4.09±0.068^{b}	4.07±0.072^{b}	4.06±0.10^{b}
Titratable acidity (g TA·100^{-1} mL^{-1})	0.29±0.009^{a}	0.08±0.016^{b}	0.09±0.016^{b}	0.08±0.026^{b}
Total phenolic content (mgGAE·100^{-1} mL^{-1})	240.46±1.20^{a}	229.89±2.10^{b}	218.36±.1.50^{c}	216.25±1.05^{c}
Antioxidant activity (EC_{50}, µg/mL)	0.046±0.05^{a}	0.032±0.05^{b}	0.018±.0.05^{c}	0.013±.0.05^{c}
L*	63.26±0.98^{a}	60.70±0.58^{b}	60.13±0.51^{b}	59.43±0.85^{b}
a*	-2.60±0.13^{a}	-2.84±0.44^{a}	-3.73±0.26^{b}	-3.89±0.16^{b}
b*	19.83±0.83^{a}	22.88±0.49^{b}	23.49±0.67^{b}	27.97±0.95^{c}

*Values in the raws with different superscripts (a, b, c) differ (P <0.05).

The results obtained from this study indicated that the content of total soluble solids differ significantly from the control group (P<0.05). pH value did not change within the treatment groups (P>0.05), but significantly differ from the control group (P<0.05). Additionally, titratable acidity changed significantly (P<0.05). The fact that the pH value remains constant throughout the process can be seen as one of the advantages of UV-C application. A noticable pH change can cause some undesired changes in sensory characteristics. pH and acidity are important

criteria that enhances the shelf life of fruit juice during the process control for maintaining the quality of the final product (Bhat *et al.,* 2011). Also, pH and ºBrix can impact the lethality rate in UV-treated apple juice (Gök, 2021; Koutchma *et al.*, 2004; Caminiti *et al.*, 2011). These present results are in parallel with earlier studies in the literature, no significant changes were indicated in pyhsicochemical parameters values such as pH, total soluble solids and total phenolic content in apple juice (Pala & Toklucu, 2013; Kyriakos et al., 2017; Noci *et al.*, 2017; Walkling-Ribeiro *et al.*, 2008; Falguera, Pagan and Ibarz, 2011; Caminiti *et al.*, 2012). Teja *et al.* (2017), studied ultraviolet treatment with treatment times of 5, 10, and 15 min for a distance of sample from lamp source (8.6,13.7, 18.6 and 22.8 cm) at 1 mm sample thickness. They also not found any significant effects on pH, TSS of apple and pineapple juice. In another study, Bhat *et al.* (2011), applied ultraviolet light (0, 30 and 60 min) to starfruit juice and detected significant decrease in the titratable acidity, but the decrease in ºBrix and pH were not significant. Conformably, Torkanmani *et al.* (2011), After UVC no significant alteration was observed in juice pH and color and also Unluturk and Atilgan (2015) who studied with grape juice, after UVexposure no changes were seen in the values of pH, total soluble solid, and titratable acidity. Comparably, Xiang et al. (2020) found no significant differences in other characteristics, such as pH, electrical conductivity, titratable acidity, total soluble solids, reducing sugar, and lightness (L*) value of apple juice exposed to 1200 mJ/cm² (P>0.05). It is known that treatments in food industry may cause some unwanted changes in quality characteristics (Riganakos *et al.*, 2017). On the other hand, UV-C treatment causes a little change in these characteristics as it can be seen in this studies results. This makes the UV-C treatment an ideal alternative for food industry.

Evaluation of Total Phenolic Content and Antioxidant Activity

Phenolics in apple and apple juice shows antioxidant activity against free radicals and they are thought to account for the beneficial and healthy effects of apple and apple juices on human nutrition (Pavun *et al.*, 2018). For this reason, it is desirable that the processes applied to apple juice for preservation purposes do not reduce the content of this phenolic substance. Total phenolic content of apple juice is considered as an important quality character (Gök, 2021). UV-C application can either increase or decrease the antioxidant values of the juice. These values are strictly dependent on the time of exposure, delivered dose and the raw material (Gök, 2021). In Table 1, it can be seen that after applying different doses of radiation, there was significant phenolic content change between the control, 4 (J/cm²) and the other groups ($P \leq 0.05$), while between the groups of 8 and 12 (J/cm²) doses there were no important changes found ($P>0.05$). In parallel to recent results, phenolic contents were decreased after UVC treatments (La Cava *et al.*, 2019; La Cava and Sgroppo 2015). According to Noci *et al.*, (2008) with an UV-C treatment (2.66-53.1 J/cm²) total phenol content of apple juice decreased significantly**.** While, according to Feng et al. (2013), treatment of watermelon juice with 37 J/mL doses did not result in a significant change ($P>0.05$) in total phenolic content. Inversely to these results, in mango juice there were significant increases after 15 and 30 min exposure of UV-C light in phenolics, carotenoids, flavonoids and thus antioxidant capacities (Santhirasegaram et al., 2015). According to Caminiti *et al.* (2011), UV-C treatment of apple juice didn't have a significant effect on the phenolic content ($P>0.05$). Islam *et al.*, (2016) discovered that the total phenolic content in the control and irradiated apple juice samples ranged from 9.79 to 9.48 mg GAE·100^{-1} mL^{-1}, indicating that UV irradiation did not cause any significant changes ($P>0.05$). Alternatively, it is well-established that heat treatments significantly decreases the polyphenols in apple juice ($P<0.05$). According to Aguilar *et al.*, (2007) there was a 32.2% reduction of polyphenols in thermally-threated apple juice.

Antioxidant activity decreased significantly after UV-C in all groups compared to untreated one. The minimum decrease was found in the 4 J/cm² sample group. Similarly, Teja et al. (2017), studied ultraviolet during 5, 10, and 15 min for a distance of sample from lamp source (8.6,13.7, 18.6 and

22.8 cm) at 1 mm sample thickness. They stated that the antioxidant activity of apple and pineapple juices showed a decreasing trends with respect to an increase in dosage level. The obtained results suggested that ultraviolet treatment conditions slightly affect the quality parameters of the both juices. Significant reductions were measured that UV-C irradiation in apple juices at in the concentrations of phenolic and antioxidant compunds (Islam *et al.,* 2016). In contrast to this, Bhat *et al.* (2011), applied ultraviolet light (0, 30 and 60 min) to starfruit juice and detected an improvement on the antioxidants, including % DPPH inhibition, total phenols, flavonols, flavonoids, and antioxidant capacity, were measured following UV treatment at 60 min application. Nonetheless, the UVC-LEDs irradiation caused reduction in total phenolic content and antioxidant activity ofapple juice (Xiang *et al.* 2020).

Significant decrease in total phenol content was established, while antioxidant capacity was not reduced significantly (Noci *et al.*, 2008). In the phenolics the highest content was found in UV-30 min. sample with an increase of 31%. However, for 60 min samples, degradation of flavonoids was obtained. This could be due to the prolonged UV-C exposure time, which generates excessive stress, thereby suppressing the flavonoid content. They also mentioned that DPPH inhibition increased by 91.2% by UV-C treated juice. The maximum increase in reducing capacity (12%) was found with control and 30 min sample. As mentioned earlier, the stress response induced by UV-C processing may enhance the extraction of antioxidant compounds, which is consistent with the findings of Bhat et al. (2011), where UV-C exposure increased the antioxidant capacity of starfruit juice. The differences between untreated and UV-C treated (48.12 kJ/L dose) orange juice were minimal in terms of organic acids, antioxidant capacity, and phenolic content. The total phenol content and antioxidant capacity of untreated orange juice were 1124.13 mg gallic acid/L and 4.71 mmol trolox equivalent/mL, respectively. These parameters showed no significant change following UV-C and heat treatments ($P>0.05$) (Pala and Toklucu, 2013).

Results of Color Measurement

Color parameters are very important for consumer preferences. In this study, the lightness, greenness and yellowness of control group was 63.26±0.98, -2.60±0.13 and 19.83±0.83 respectively. UV-C treatment caused changes in brightness (L*). Thus, the brightness of apple juice, which plays an important role in the consumer preference and acceptance, was protected better in the minumum dosage group than the others. a* and b* parameters showed changes after the UV-C exposure ($P\leq0.05$). The yellowness and greenness of the apple juice samples increased with the radiation application. Thus, after the UV-C treatment, apple juices became darker, greener and more yellow. In a different study (Müller *et al.*, 2014), untreated apple juice's color characteristics such as L*, a* and b* values are determined as 30.5±0.8, 1.0±1.0 and 18.6±1.7 respectively. Similar results were found, after the UV-C and UV-B treatment of apple and grape juice, the brightness value also decreased, and a^* and b^* values showed increase with the treatment (Müller *et al.*, 2014). Falguera *et al.* (2011), investigated UV irradiation on physicochemical properties of apple juice from different sources. After applying 400, 500, 600 and 700 nm wavelenght on four different apple juices, the brightness of each apple juice was decreased significantly. In another study, the results indicated that no significant changes ($P>0.05$) were observed in the L*, a*, and b* values during storage for the control (fresh), thermally treated, and UV-C treated samples. Also they indicated that laboratory observations using the human eye revealed no evident difference between the fresh, thermally treated, and UV-C treated samples (Riganakosa *et al.*, 2017). Results showed significant improvements for brightness in star fruit irradiation (Bhat *et al.*, 2011). Though, for orange juice slight changes were detected in a* and b* values whereas L* value remained almost constant after UV exposure (Taze *et al.*, 2015). Teja *et al.* (2017), studied with apple and pineapple juices, found that the color parameters (L*, a* and b*) were

slightly affected by ultraviolet treatment (Teja *et al*., 2017).

CONCLUSION

Apple juice, one of the most consumed fruit juices worldwide due to its taste and high nutritional values, provide a growing medium to *Z. rouxii* if not properly handled. This work aimed to address the impact of UV-C technologies on apple juice. Findings from the present study revealed that the logarithmic reduction in *Z. rouxii*. was significant, especially in the samples exposed to 8 J/cm². The content of total soluble solids, pH value and titratable acidity changed significantly (P<0.05), also the changes between the total phenolic contents have found to be significant (P<0.05). On the other hand, the color parameters (L^*, a^*, b^*) changed slightly with UV-C radiation treatment. The increases in the doses caused decreases in the quality values. Thus, mild ultraviolet treatment on apple juice is an ideal alternative to heat treatments. In further studies, UV-C technology, maybe combined with other processes at low dosages of irradiation.

CONFLICTS OF INTEREST

The author state that they have no conflicts of interest.

REFERENCES

Aguilar-Rosas SF, Ballinas-Casarrubias ML, Nevarez-Moorillon GV, Martin-Belloso O, Ortega-Rivas, E. (2007). Thermal and pulsed electric fields pasteurization of apple juice: Effects on physicochemical properties and flavour compounds. *Journal of Food Engineering*, 83(1), 41-46.

Akwu AS, Patras A, Pendyala B., Kurup A., Chen FC, Vergne, MJ (2022). Effect of germicidal short wave-length ultraviolet light on the polyphenols, vitamins, and microbial inactivation in highly opaque apple juice. *bioRxiv,* 2022-07.

Almeida ETC, Barbosa IM, Tavares JF, Barbosa Filho JM, Magnani M, de Souza, EL (2018), Inactivation of spoilage yeasts by Mentha spicata L. and M.×villosa Huds. essential oils in cashew, guava, mango, and pine apple juices. *Frontiers in Microbiology,* 9, 1111.

Aneja KR, Dhiman R, Aggarwal NK, Aneja, A. (2014). Emerging preservation techniques for controlling spoilage and pathogenic microorganisms in fruit juices. *International Journal of Microbiology,* 2014, 758942.

Akgün, M. P., Ünlütürk, S. (2017). Effects of ultraviolet light emitting diodes (LEDs) on microbial and enzyme inactivation of apple juice. *International Journal of Food Microbiology*, 260, 65-74.

Barut Gök S. (2021). UV-C treatment of apple and grape juices by modified UV-C reactor based on Dean vortex technology: Microbial, physicochemical and sensorial parameters evaluation. *Food and Bioprocess Technology*, 14, 1055-1066.

Baysal AH, Molva C, Unluturk, S. (2013).UV-C light inactivation and modeling kinetics of Alicyclobacillus acidoterrestris spores in white grape and apple juices. *International Journal of Food Microbiology,* 166(3), 494–498. DOI: 10.1016/j.ijfoodmicro.2013.08.015

Bhat, R., Ameran, S. B., Voon, H. C., Karim, A. A.,Tze, L. M. (2011). Quality attributes of starfruit (Averrhoa carambola L.) juice treated with ultraviolet radiation. *Food Chemistry*, 127(2), 641-644.

Bintsis T, Tzanetaki E.L, Robinson R.K, (2000). Existing and potential applications of ultraviolet light in the food industry — A critical review. *Journal of Food Science Application* 80, 637–645

Caminiti, IM., Noci, F., Munoz, A., Whyte, P., Morgan, DJ., Cronin, DA., et al. (2011). Impact of selected combinations of non-thermal processing technologies on the quality of an apple and cranberry juice blend. *Food Chemistry, 124*(4), 1387–1392.

Caminiti IM, Palgan, I, Muñoz A, Noci F, Whyte P, Morgan DJ, Cronin DA, Lyng JG, (2012) The effect of ultraviolet light on microbial inactivation and quality attributes of apple juice. *Food Bioprocess Technology*, 5, 680–686.

Cemeroğlu, B. (2018). Gıda analizi . Gıda Teknolojisi Derneği Yayınları.

Chen C, Tseng CW (1997) Effect of high hydrostatic pressure on the temperature

dependence of Saccharomyces cerevisiae and Zygosaccharomyces rouxii. *Process Biochemistry*, 32(4):337-343 pp

Escott C, Fresno, JMD, Loira I. (2018). *Zygosaccharomyces rouxii*: Control Strategies andApplications in Food and Winemaking. *Fermentation*, 4(3),69.

Falguera V, Pagán J, Ibarz A. (2011). Effect of UV irradiation on enzymatic activities and physicochemical properties of apple juices from different varieties. *LWT-Food Science and Technology,* 44(1), 115-119.

Feliciano RJ, Estilo EEC, Nakano H, Gabriel, AA, (2019), Ultraviolet-C resistance of selected spoilage yeasts in orange juice. *Food Microbiology*, 78(October 2018), 73–81. DOI: 10.1016/j.fm.2018.10.003

Fenoglio D, Ferrario M, Schenk M, Guerrero S. (2020). Effect of pilot-scale UV-C light treatment assisted by mild heat on E. coli, L. plantarum and S. cerevisiae inactivation in clear and turbid fruit juices. Storage study of surviving populations. *International Journal of Food Microbiology,* 108767. DOI: 10.1016/j.ijfoodmicro.2020.10

Franke, SIR (2004) Food Chemistry Study of antioxidant and mutagenic activity of different orange juices. *88*, 45–55. DOI: 10.1016/j.foodchem.2004.01.021

Gabriel, A. A. (2012). Inactivation of Escherichia coli O157: H7 and spoilage yeasts in germicidal UV-C-irradiated and heat-treated clear apple juice. *Food Control*, 25(2), 425-432.

Gouma M, Gayán E, Raso J, Condón S, Álvarez I (2015) Inactivation of spoilage yeasts in apple juice by UV-C light and in combination with mild heat. *Innovative Food Science and Emerging Technologies,* 32, 146–155. DOI: 10.1016/j.ifset.2015.09.008

Guerrero–Beltrán JA, Barbosa–Cánovas GV, (2004) Review: advantages and limitations on processing foods by UV light. *Food Sci. Technol. Int.* 10, 137–148.

Hakguder Taze B, Unluturk S, Buzrul S, Alpas H. (2015). The impact of UV-C irradiation on spoilage microorganisms and color of orange juice. J*ournal of Food Science and Technology*, *52*(2), 1000–1007. DOI: 10.1007/s13197-013-1095-7

Hayes JC, Laffey, JG, McNeil B, Rowan, N. J. (2012). Relationship between growth of food-spoilage yeast in high-sugar environments and sensitivity to high-intensity pulsed UV light irradiation. *International journal of food science & technology*, 47(9), 1925-1934.

Hernández, A, Pérez-Nevado F, Ruiz-Moyano S, Serradilla MJ, Villalobos MC, Martín A, Córdoba MG (2018). Spoilage yeasts: What are the sources of contamination of foods and beverages?. I*nternational journal of food microbiology*, 286, 98-110.

Huang Z, Hu H, Shen F, Wu B, Wang X, Zhang B, ... Zhang X. (2018). Relatively high acidity is an important breeding objective for fresh juice-specific apple cultivars. *Scientia Horticulturae,* 233, 29-37.

Hyun TK, Jang KI. (2016). Apple as a source of dietary phytonutrients: an update on the potential health benefits of apple. *EXCLI Journal*, 15, 565.

Islam MS, Patras A, Pokharel B, Wu Y, Vergne MJ, Shade L, ... Sasges, M. (2016). UV-C irradiation as an alternative disinfection technique: Study of its effect on polyphenols and antioxidant activity of apple juice. *Innovative Food Science & Emerging Technologies*, 34, 344-351.

Karaman K, Sagdic O, Yilmaz MT. (2020).Potential of natamycin to control growth of *Zygosaccharomyces* spp. in apple juice during storage. *International Journal of Food Microbiology*, 332(February), 108771. DOI: 10.1016/ j.ijfoodmicro.2020.108771

Keyser, M., Műller, I. A., Cilliers, F. P., Nel, W., Gouws, P. A. (2008). Ultraviolet radiation as a non-thermal treatment for the inactivation of microorganisms in fruit juice. *Innovative food science & emerging technologies*, 9(3), 348-354.

Koutchma T, Keller S Chirtel S, Parisi, B. (2004).Ultraviolet disinfection of juice products in laminar and turbulent flow reactors. *Innovative Food Science and Emerging Technologies*, 5, 179–189.

La Cava ELM, Sgroppo SC (2019).Combined Effect of UV-C Light and Mild Heat on Microbial

Quality and Antioxidant Capacity of Grapefruit Juice by Flow Continuous Reactor. *Food and Bioprocess Technology*, 12(4), 645–653. DOI: 10.1007/s11947-019-2239-1

Legras JL, Merdinoglu D, Cornuet JM, Karst F. (2007).Bread, beer and wine: Saccharomyces cerevisiae diversity reflects human history. *Molecular Ecology*, 16(10), 2091–2102.

Mansor, A., Shamsudin, R., Adzahan, N. M., Hamidon, M. N. (2014). Efficacy of ultraviolet radiation as non-thermal treatment for the inactivation of Salmonella typhimurium TISTR 292 in pineapple fruit juice. *Agriculture and Agricultural Science Procedia*, 2, 173-180.

Marvig CL, Kristiansen RM, Nielsen DS (2015). Growth/no growth models for Zygosaccharomyces rouxii associated with acidic, sweet intermediate moisture food products. *International Journal of Food Microbiology*, 192, 51–57. DOI: 10.1016/j.ijfoodmicro.2014.09.021

Müller WA, Valentina M, Pasin A, Sarkis, JR (2021), International Journal of Food Microbiology Effect of pasteurization on Aspergillus fumigatus in apple juice : Analysis of the thermal and electric effects. *International Journal of Food Microbiology*, 338(July 2020), 108993. DOI: 10.1016/j.ijfoodmicro.2020.108993.

Müller A, Noack L, Greiner R, Stahl MR, Posten C. (2014). Effect of UV-C and UV-B treatment on polyphenol oxidase activity and shelf life of apple and grape juices. *Innovative Food Science & Emerging Technologies,* 26, 498-504.

Nicolau-Lapeña, I., Colás-Medà, P., Vinas, I., Alegre, I. (2022). Inactivation of Escherichia coli, Salmonella enterica and Listeria monocytogenes on apple peel and apple juice by ultraviolet C light treatments with two irradiation devices. *International Journal of Food Microbiology,* 364, 109535.

Noci F, Riener, J, Walkling M, Cronin DA, Morgan DJ, Lying, JG. Ultraviolet irradiation and pulsed electric fields (PEF) in a hurdle strategy for the preservation of fresh apple juice. *Journal Food Engineering*. 2008, 85, 141–146.

Pala ÇU, Toklucu AK (2011). Effect of UV-C light on anthocyanin content and other quality parameters of pomegranate juice. *Journal of Food Composition and Analysis*, 24(6), 790–795. DOI: 10.1016/j.jfca.2011.01.003

Patocka J, Bhardwaj K, Klimova B, Nepovimova E, Wu Q, Landi M, Wu W. (2020). Malus domestica: A review on nutritional features, chemical composition, traditional and medicinal value. *Plants*, 9(11), 1408.

Pavun L, Uskokovic-Markovic, S, Dikanović D, Djurdjevic, P. (2018). Determination of flavonoids and total polyphenol contents in commercial apple juices. *Czech Journal of Food Science,* 36, 2018 (3): 233–238

Pierscianowski J, Popović V, Biancaniello M, Bissonnette S, Zhu Y, Koutchma T. (2021), Continuous-flow UV-C processing of kale juice for the inactivation of E. coli and assessment of quality parameters. *Food Research International*, 140(May 2020). DOI: 10.1016/ j.foodres.2020.110085

Prado DB do, Szczerepa MM dos A, Capeloto OA., Astrath NGC, Santos, N. C. A. dos, Previdelli, I. T. S., Nakamura, C. V., Mikcha, J. M. G., Abreu Filho, B. A. de. (2019). Effect of ultraviolet (UV-C)radiation on spores and biofilms of Alicyclobacillus spp. in industrialized orange juice. *International Journal of Food Microbiology*, 305(May). DOI: 10.1016/ j.ijfoodmicro.2019.108238

Riganakos KA, Karabagias IK, Gertzou I, Stahl M. (2017). Comparison of UV-C and thermal treatments for the preservation of carrot juice. *Innovative Food Science and Emerging Technologies*, 42(June), 165–172. DOI: 10.1016/ j.ifset.2017.06.015

Rojo MC, López FA, Lerena MC, Mercado L, Torres A, Combina M. (2015). Evaluation of different chemical preservatives to control *Zygosaccharomyces rouxii* growth in high sugar culture media. *Food Control*, 50, 349-355.

Rojo MC, Palazzolo CT, Cuello R, Gonzalez M, Guevara F, Ponsone ML, Combina, M. (2017). Incidence of osmophilic yeasts and Zygosaccharomyces rouxii during the production

of concentrate grape juices. *Food microbiology*, 64, 7-14.

Rydzak L, Kobus Z, Nadulski R, Wilczyński K, Pecyna A, Santoro F, Krzywicka, M. (2020) Analysis of selected physicochemical properties of commercial apple juices. *Processes*, 8(11), 1457.

Saloma M. (2018) 16.1 introduction. 291–308. DOI: 10.1016/B978-0-12-802230-6.00016-3

Santhirasegaram, V., Razali, Z., George, D. S., Somasundram, C. (2015). Comparison of UV-C treatment and thermal pasteurization on quality of Chokanan mango (Mangifera indica L.) juice. *Food and Bioproducts processing*, 94, 313-321.

Sauceda, G. J. N., Tió-Coma, M., Martinez-Garcia, M., Hernández-Herrero, M. M., Gervilla, R., Roig-Sagués, A. X. (2020). Effect of single and combined UV-C and ultra-high pressure homogenisation treatments on inactivation of Alicyclobacillus acidoterrestris spores in apple juice. *Innovative Food Science & Emerging Technologies*, 60, 102299.

Sperber WH, Doyle, M. P. (2009). Compendium of the Microbiological Spoilage of Foods and Beverages.

Spengler, RN. (2019). Origins of the apple: The role of megafaunal mutualism in the domestication of Malus and rosaceous trees. *Frontiers in Plant Science* 10, 617.

Singleton VL, Rossi JA (1965) Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American journal of Enology and Viticulture*, 16(3), 144-158.

Teja CK, Sanganamoni S, Prabhakar B, Rao, P. S. (2017) Effect of UV–C Light Treatment on Physicochemical and Bioactive Compounds in Apple and Pineapple Juices. *International Journal of Current Microbiology and Applied Sciences*. 6, 2321-2333.

Unluturk S, Atilgan MR (2015). Microbial Safety and Shelf Life of UV-C Treated Freshly Squeezed White Grape Juice. *Journal of Food Science*, 80(8), M1831–M1841. DOI: 10.1111/1750-3841.1295.

Xiang Q, Liu X, Li J, Liu S, Zhang H, Bai, Y. (2018). Effects of dielectric barrier discharge plasma on the inactivation of Zygosaccharomyces rouxii and quality of apple juice. *Food Chemistry*, 254(136), 201–207. DOI: 10.1016/j.foodchem.2018.02.008

Xiang Q, Fan L, Zhang R, Ma Y, Liu S, Bai Y. (2020). Effect of UVC light-emitting diodes on apple juice: Inactivation of Zygosaccharomyces rouxii and determination of quality. *Food Control*, 111, 107082.

Walkling-Ribeiro, M., Noci, F., Cronin, DA., Riener, J., Lyng, JG., Morgan, DJ. (2008). Reduction of Staphylococcus aureus and quality changes in apple juice processed by ultraviolet irradiation, pre- heating and pulsed electric fields. *Journal of Food Engineering*. 89: 267-273.