

Influence of power ultrasound on the quality parameters of grapefruit juice during storage

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Abstract

Fresh grapefruit juice samples were subjected to ultrasound treatment at various treatment times (30, 60 and 90 min) in a bath type sonicator at a frequency of 28 KHz, radiating power of 70% (420 W) and 20 °C. Grapefruit juice samples were stored in sterilized polypropylene tubes at 4±1 °C for a period of 28 days. This study was initiated with the objective of calculating the effect of ultrasound treatment and storage time on acidity, pH, °Brix, electrical conductivity (EC), hunter color values (L*, a* and b*), non-enzymatic browning (NEB), cloud value (CV) and bioactive compounds of grapefruit juice. Results showed that there was no change in pH, °Brix and acidity, while a significant decrease in all the color values (L*, a* and b*) and significant increase in EC, cloud value, total antioxidant capacity (TAC), DPPH free radical scavenging activity, total phenolics (TP), total flavonoids (TF) and total flavonols was observed in all the juice samples sonicated for 30, 60 and 90 min as compared to control. During storage, NEB and hunter color values (L*, a* and b*) were increased, while CV, DPPH (2,2-diphenyl-1-picrylhydrazyl) free radical scavenging activity, TAC, TP, TF and total flavonols were decreased significantly in all the ultrasound treatments, but more stability was determined in sonication treatment of 90 min as compared to control during storage period of 28 days. The findings of this study suggested that sonication for 90 min may be employed successfully in the processing of grapefruit juice at industrial scale.

Keywords: Ultrasound processing; grapefruit juice; storage; cloud value; polyphenols; antioxidant capacity.

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Introduction

Citrus fruits are generally prominent because of their higher contents of bioactive and polyphenolic compounds which play a significant role in controlling the risk of many diseases such as mortality rate of cancer and cardiovascular diseases [1]. The health benefits of citrus fruit have mainly been attributed to the presence of bioactive compounds [2] which are the main reason for the health benefits of these natural products are phytochemicals, especially phenolics [3]. Among them, grapefruit is a very common variety that can significantly contribute to a healthy human diet and is commonly used as a breakfast fruit. Mostly used as chilled, cut in half, loosened from the peel and skin membranes with a special curved grapefruit knife [4]. It is also an excellent source of many nutrients and phytochemicals, which play an important part in the human health and a good source of sugar organic acid and phenolic compounds. Grapefruit and grapefruit juice, which are rich in calories, have been shown to help speed up the metabolism. Its concentration and nature mostly affects the taste and organoleptic properties [5]. The NEB is a key point for the quality deterioration factor which caused the degradation of ascorbic acid, formation of brown pigments and the reactions between amino acids and reducing sugars in the citrus juices [6]. Previously, researchers have done some work on phenolic compounds obtained from grapefruit, and few studies have been published which

suggests that they could play a key role in the total antioxidant capacity of grapefruit juice [1]. Antioxidant compounds which are extracted from plants were acknowledged as active oxygen scavengers [7].

Nowadays, consumers have become more conscious about health and diet. There is an increase in demand by consumers for healthy, fresh like products with increased shelf life and juices with minimum flavor and vitamin losses which stimulate the way for new food processing and preservation techniques [8]. Thermal processing technique is the most familiar technique for improving the shelf life of juices by reducing the activity of microbes and enzymes. However, losses in terms of color, taste, flavor, sensory and nutritional qualities were occurred when food was treated with heat [9]. According to the demands of consumer, researchers have more interest in non-thermal technologies which can improve the nutritional value, retain the original properties and maintain the quality of food products [10]. Among these non-thermal technologies, some technologies have got considerable attention of the researchers due to specific actions in biological materials and undesirable changes in food [11]. Ultrasound is considered to be a promising and emerging alternative technique for the heat treatment of food processing industry [12]. Moreover, ultrasound has identified as a prospective technology to meet the FDA requirements

of 5 log reduction in relevant microorganisms found in fruit juices [13]. Ultrasound was seen to be valuable for minimal processing, less energy input and reduced processing time [14]. In liquids, application of high power sonication creates the cavitation bubbles due to pressure changes that result in the increase of localized temperature up to 5000K and 50,000 kPa [15]. The overall goal of this study was to evaluate the storage effects of ultrasound on the different quality aspects of grapefruit juice. Few reports are available on the storage effect of ultrasound processing, but no report is available related to grapefruit juice on selected parameters.

Materials and Methods

Chemicals

The chemical reagents 2,2-diphenyl-1-picrylhydrazyl (DPPH), catechin hydrate, sulfuric acid, quercetin, ammonium molybdate, gallic acid were obtained from Aladdin Industries Company Limited (Shanghai, China) and sodium hydroxide was purchased from Nanjing Chemical Reagent Company Limited (Nanjing, China). Ethanol, Methanol, sodium nitrite and aluminium chloride were attained from Sinopharm Chemical Reagent (Shanghai, China). Folin-Ciocalteu reagent was bought from Guoyao Reagent Company Limited (Shanghai, China). Sodium acetate anhydrous, sodium carbonate and sodium phosphate were gained from Guanghua Chemical Factory (Guangdong, China). Ascorbic acid was bought from Accu Standard Inc. (New Heaven, CT 06,513, USA). All chemicals and reagents used in the study were of analytical (AR) grade.

Preparation of grapefruit juice

Freshly grapefruits were obtained from a local supermarket (Guangzhou, China) to produce fresh grapefruit juice. The grapefruits were screened, washed and crushed using a domestic juice extractor (JM352, Guangdong Midea Group Co. Ltd., China). The juice was then filtered through sterilized double layer muslin cloth to remove the impurities and then used for ultrasound treatments.

Ultrasound treatment

Ultrasound treatment was performed immediately after fresh juice was extracted. The juice was vortex-mixed and divided into four parts as control (0 min) and sonicated for 30, 60 and 90 min. The ultrasound was carried out at 28 kHz frequency, power radiation 70% (420 W) and temperature 20 °C by using an ultrasonic cleaner (SB-600DTY, Ningbo Scientz Biotechnology Company Limited, Ningbo, China). All

the sonication treatments were carried out in the dark to avoid any possible interference of light.

Packaging and storage

Juice samples were stored at 4±1 °C in sterilized polypropylene tubes (Guangzhou Jie Sheng Kang Biotechnology Co. Ltd., Guangzhou, China). All sonicated treated and control samples were stored at 4±1 °C. The samples were analyzed at 0 days (immediately after treatment), 7, 14, 21 and 28 days during storage.

Determination of pH, °Brix, acidity, CV, EC, color values and NEB

The pH was determined by a digital pH meter, °Brix was measured by Abbe refractometer and acidity was investigated by the method of Redd et al. [16]. The CV was measured by using the method described by Versteeg et al. [17] while EC was determined by conductivity meter and the color of the juice samples was measured using a colorimeter (WSF-J, Shanghai Precision & Scientific Instrument Company Limited, China) based on 3 color co-ordinates, namely L*, a*, b*. The color values were expressed as L* (whiteness/darkness), a* (redness/greenness) and b* (yellowness/blueness). To determine the NEB, 10 ml of grapefruit juice sample was centrifuged (12,500 g, 10 min and 25 °C). The supernatant was collected and clarified by utilizing a 0.45 µm filter. The browning index was determined as the absorbance at 420 nm using spectrophotometer at the room temperature [18].

Determination of contents of total phenolics, flavonoids and flavonols

Total phenolics of grapefruit juice were determined by spectrophotometer method using Folin-Ciocalteu reagent with some minor amendments [19]. Briefly 1 ml of 10 % Folin-Ciocalteu reagent was added to a 0.5 ml of a known concentration of the sample. The mixture was mixed well and left it for 6 min, and then 2 ml of a 20% sodium carbonate solution was added to the above mixture. The total phenolics were measured at 760 nm using the spectrophotometer after reaction for 60 min at 30 °C. A calibration curve was prepared by using a standard solution of gallic acid and the results of total phenolics were expressed as µg of gallic acid equivalents (GAE) per gram of sample.

Total flavonoids contents were determined by a method described by Kim et al. [20] with minor modification. In short, 0.25 ml of the known sample was mixed with 1.25 ml of de-ionized water in a plastic tube and then 5% sodium nitrite solution (75

μL) was added. After 6 min, 150 μL of a 10% aluminum chloride solution was added and then after 5 min, 0.5 ml of 1 M sodium hydroxide was added. The final volume was made up to 2.5 ml with distilled water and mixed well. The absorbance was measured at 415 nm by using a spectrophotometer. The results were expressed as μg of (+)-catechin equivalents per gram of sample.

Total flavonols of grapefruit juice samples were measured by using the method of Kumaran et al. [21]. A known aliquot of sample (2 ml) or standard was mixed with 2 ml of 2% AlCl_3 solution, and then 3.0 ml (50 g/L) sodium acetate solution was added. The mixture was placed at 20 °C for 150 min. The absorption was measured at 440 nm by using the spectrophotometer and the results were described as μg of quercetin equivalents per gram of sample.

Determination of DPPH free radical scavenging activity and total antioxidant capacity (TAC)

The DPPH free radical scavenging activity and total antioxidant capacity (TAC) were measured by using the modified method of Aadil et al. [22].

Statistical analysis

All the measurements were performed in triplicate. For statistical analysis, a completely randomized design was used with a two-way factorial experiment design and significant differences between mean values were determined by LSD pair-wise comparison test at a significance level of $p < 0.05$. Statistical analyses were conducted by using Statistix 9.0 software (Analytical Software, Tallahassee FL, USA).

Results and Discussion

Effect of sonication and storage on quality parameters of grapefruit juice

In juices, pH and acidity are the important quality parameters to assess and maintain the shelf life. The °Brix is one of the most important factors for grading the quality and used to specify the percentage of soluble sugar in the juice [23]. Table 1 shows the effect of sonication and storage of grapefruit juice on °Brix, pH and acidity. In the present study, we observed that all the sonicated treatments at 0 days showed the non-significant effect as compared to control treatment, whereas an increase in pH and °Brix of grapefruit juice with the increase of storage time was observed in all the treatments. While a significant decrease in acidity was observed in all treatments on the 28th day of storage. A significant increase in pH was in agreement with a storage study of sonicated

orange juice [24] which indicates the initiation of fermentation or spoilage.

Generally, liquid foods are electrical conductors due to the presence of nutrients such as proteins, fatty acids, minerals and vitamins [25]. The EC of the sonicated treated grapefruit juice was measured during the storage time. The observed changes in EC are depicted in Table 1. Just after processing, EC was increased at different sonication treatment times as compared to control while during storage time of 28 days. EC was significantly decreased in all the treatments. The decrease in electrical conductivity of grapefruit juice during the storage period might be due to the loss of nutrients responsible for electrical conductivity.

Cloud value

Cloud value plays a significant role in improving the color and flavor of fruit juices, which is interconnected to the particles composed of a mixture of lipids, pectin, cellulose and protein [26, 27]. In fruit juices, cloud stability is visual quality parameters for conclusive acceptance of consumer [28]. The effects of ultrasound treatments on the CV of grapefruit juice in relation to storage time of 28 days are depicted in Fig. 1. Our study showed that the cloud values of all the sonication treatments were increased after processing (day 0) while during storage time of 28 days cloud values were decreased in all the sonication treatments, but showed more stability in sonication treatment (90 min) as compared to control. Our results were in agreement with Tiwari et al., [29] who observed the same behavior in the sonicated orange juice during storage. During ultrasound treatments, the possible reason of increase in the cloud value might be attributed to the breakdown of bigger molecules into smaller ones due to high pressure gradient exerted by cavitation [30]. Another reason is that the application of ultrasound breaks the linear pectin molecule, reducing its molecular weight and resulting in weaker network [31].

Color Values

Color is the most important quality parameter on which consumer's preferences is based and also plays an important role in the appearance and acceptability of food [32]. It can accentuate the ultimate acceptance, mean for rapid identification and serve as an indicator of organoleptic and nutritional quality of food during processing and subsequent storage because it is linked with the perception of some characteristics that appear to be representative of the quality of a fruit juice [34]. Color differences between sonicated and different

Table 1 Effect of sonication and storage period on the quality parameters of grapefruit juice.

Treatments	Time (Days)	pH	TA (%)	TSS (°Brix)	NEB	EC (ms/cm)	Color		
							L*	a	B
Control	0	4.70fghi±0.01	0.20a±0.01	9.60a±0.05	0.215j±0.02	2.78de±0.02	7.16h±0.05	5.25g±0.07	-11.23p±0.04
	7	4.69jkla±0.01	0.20ab±0.01	9.60a±0.10	0.261i±0.07	2.69f±0.03	7.45e±0.04	5.65de±0.05	-10.97n±0.06
	15	4.71f±0.01	0.19bcd±0.01	9.10bc±0.05	0.286h±0.05	2.47hi±0.04	7.59d±0.08	5.79c±0.07	-10.19l±0.08
	21	4.73de±0.01	0.18cde±0.01	9.00c±0.10	0.298g±0.06	2.32k±0.06	7.81b±0.08	5.97b±0.06	-9.39h±0.07
	28	4.74bc±0.01	0.16g±0.01	8.80d±0.10	0.315f±0.05	2.08l±0.06	7.98a±0.06	6.35a±0.06	-8.11d±0.08
US (30 min)	0	4.70fghi±0.01	0.20ab±0.01	9.60a±0.10	0.261i±0.07	2.85cd±0.05	7.01i±0.07	4.67m±0.04	-10.20l±0.06
	7	4.69kl±0.01	0.20a±0.01	9.55a±0.05	0.282h±0.06	2.73ef±0.04	7.33f±0.06	4.79kl±0.08	-9.89k±0.04
	14	4.71fg±0.01	0.18cde±0.01	9.12bc±0.10	0.299g±0.09	2.52gh±0.05	7.47e±0.06	4.93ij±0.03	-9.36h±0.08
	21	4.73cde±0.01	0.18de±0.01	9.03bc±0.06	0.313f±0.05	2.41ij±0.04	7.72c±0.04	5.26g±0.09	-8.21e±0.02
	28	4.75ab±0.01	0.15g±0.01	8.75d±0.05	0.359c±0.07	2.11l±0.07	7.85b±0.07	5.59de±0.08	-7.36a±0.03
US (60 min)	0	4.70ghij±0.01	0.20a±0.01	9.60a±0.06	0.303g±0.03	2.94b±0.04	6.88j±0.07	4.76l±0.05	-10.45m±0.05
	7	4.70ijkl±0.01	0.19abc±0.01	9.50a±0.10	0.325e±0.05	2.78de±0.06	7.20gh±0.04	4.88jk±0.09	-9.68j±0.07
	14	4.70fgh±0.01	0.18de±0.01	9.10bc±0.10	0.347d±0.05	2.56g±0.04	7.36f±0.07	5.06h±0.06	-9.16g±0.07
	21	4.74bcd±0.01	0.16fg±0.01	9.05bc±0.09	0.369c±0.07	2.41ij±0.06	7.60d±0.08	5.39f±0.07	-8.03c±0.06
	28	4.76a±0.01	0.16g±0.01	8.70d±0.10	0.391ab±0.07	2.13l±0.06	7.72c±0.06	5.68d±0.05	-7.47b±0.05
US (90 min)	0	4.70hijk±0.01	0.20ab±0.01	9.50a±0.10	0.321ef±0.07	3.08a±0.08	6.75k±0.05	4.88jk±0.07	-11.95q±0.05
	7	4.70ijkl±0.01	0.19bcd±0.01	9.50a±0.05	0.344d±0.06	2.87bc±0.04	7.07i±0.07	5.02hi±0.07	-11.09o±0.07
	14	4.70ghij±0.01	0.17ef±0.01	9.15b±0.09	0.369c±0.04	2.67f±0.03	7.24g±0.05	5.21g±0.07	-10.45m±0.03
	21	4.73e±0.01	0.16g±0.01	9.05bc±0.09	0.388b±0.04	2.51gh±0.05	7.46e±0.05	5.56e±0.03	-9.56i±0.03
	28	4.76a±0.01	0.16g±0.01	8.73d±0.06	0.401a±0.02	2.16l±0.05	7.61d±0.07	5.85c±0.03	-8.59f±0.08

Means followed by the same letter within each column are not significantly different ($p < 0.05$); US: ultrasonic technique, TA: Titratable acidity, EC: electrical conductivity, NEB: Non-enzymatic browning

treatment time intervals (30, 60 and 90 min) and color values (L^* , a^* , b^*) are expressed in Table 1. During sonication treatment, it was found that all the color values L^* (whiteness), a^* (redness) and b^* (yellowness) were decreased at 0 days, but a significant increase was observed during storage period of 28 days in all the treatments. These results are in agreement with the observations of Tiwari et al. [29]. Changes in color during sonication might be due to cavitation, which induced several physical, chemical and biological reactions, such as breakdown of susceptible particles and increased diffusion rates [33].

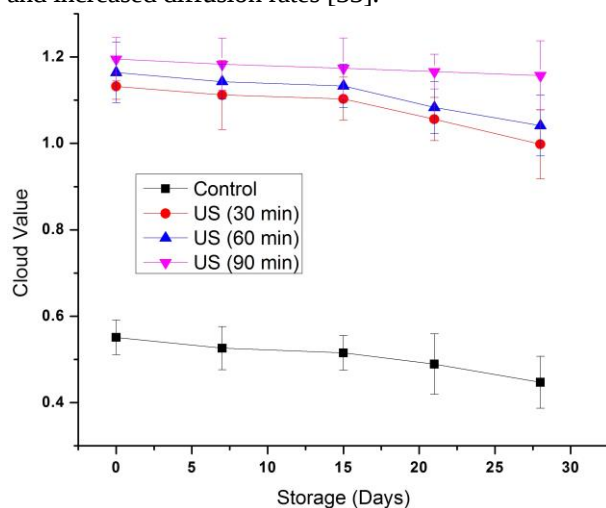


Fig. 1: Effect of sonication and storage period on the cloud value of grapefruit juice.

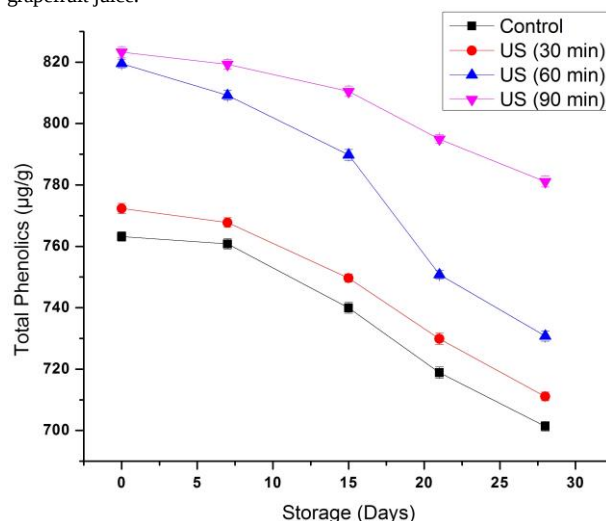


Fig. 2: Effect of sonication and storage period on the total phenolic compounds of grapefruit juice.

Non-enzymatic browning (NEB)

Browning is a complex process, which involves several factors, including enzyme activity, reducing sugar, presence of ascorbic acid, and other promoters influencing the browning reaction and an imperative

restraint which is commonly used to indicate the formation of brown color in juices [35, 36]. Table 1 shows the effect of ultrasound and storage time on NEB of grapefruit juice during storage time of 28 days. During storage time of 28 days, significant increase in NEB was observed in all the treatments of grapefruit juice which was in agreement with the observations of sonicated orange juice [29]. Increase in NEB could be due to the breakage of color pigments due to ultrasound treatment. Similar changes were reported in a sonicated apple cider [34]. In citrus juices, the ascorbic acid degradation during storage is related to be a key chemical reaction responsible for NEB [37].

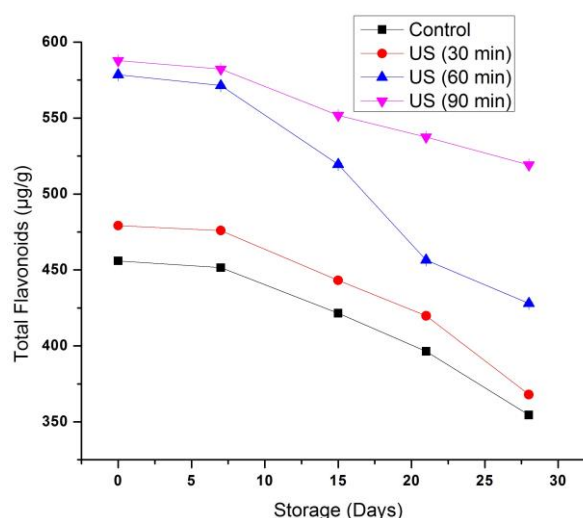


Fig. 3: Effect of sonication and storage period on the total flavonoid compounds of grapefruit juice.

Total phenolic, total flavonoid and total flavanol, total antioxidant capacity and DPPH free radical scavenging activity

Grapefruit is a rich source of health promoting flavonoids, especially flavanones, which are shown to possess several physiological properties [38]. A large number of epidemiological studies are associated with the consumption of fruits and vegetables with decreased risks of development of diseases such as cancer and coronary heart disease [39] due to the presence of health promoting phytochemicals such as carotenoids, flavonoids, phenolic compounds and vitamins [40, 41]. Antioxidant capacity has been used to evaluate the antioxidant potential status of tissue, which is a function of the bioactive compounds. The effect of ultrasound treatment and storage time on TP, TF, total flavonols, DPPH free radical scavenging activity and TAC of grapefruit juice during storage period of 28 days are shown in Fig. 2, 3, 4, 5 and 6, respectively. At processing time (day 0), significant

increase in TP, TF, total flavonols, DPPH free radical scavenging activity and TAC were observed in all the ultrasound treatment as compared to control while at day 28, significant decrease was observed in all the sonicated treatments but more stability was observed in sonication treatment (US 90 min) as compared to control treatment. In many plant species, there is a good relationship between antioxidant activity and total phenols and was extensively investigated in different fruits and vegetables [42]. A significant increase in the total antioxidant activity is related to the presence of high concentration of total polyphenol content in vegetables and fruits. Previous study of Gao et al. [43] observed significant decrease in antioxidant capacity of sonicated apple-carrot juice in a storage study of 28 days.

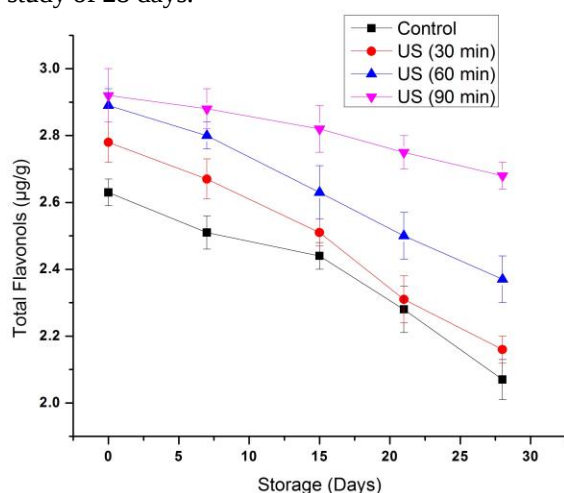


Fig. 4: Effect of sonication and storage period on the total flavonol compounds of grapefruit juice.

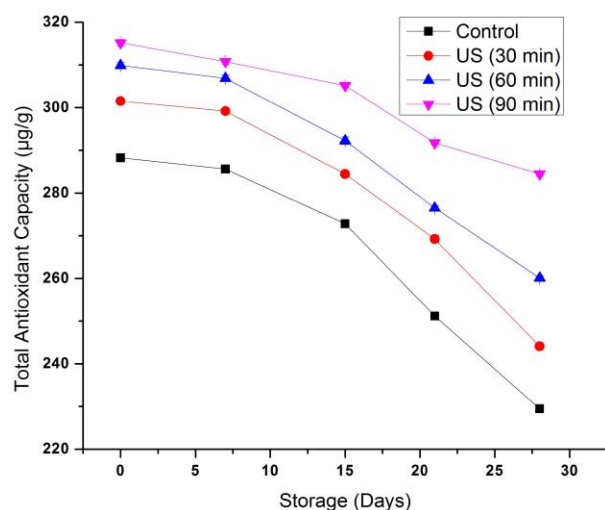


Fig. 5: Effect of sonication and storage period on the total antioxidant capacity of grapefruit juice.

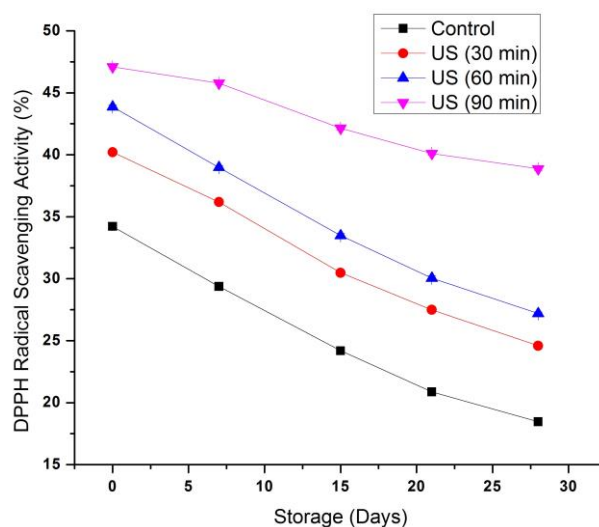


Fig. 6: Effect of sonication and storage period on the DPPH radical scavenging activity of grapefruit juice.

Conclusions

The present study was conducted to evaluate the effects of the sonication technique on selected quality attributes of grapefruit juice during storage period of 28 days. The pH, acidity, NEB, color and cloud values, TP, TF, flavonols, DPPH free radical scavenging activity and TAC were significantly affected during storage period. While sonication treatment (90 min) improved and preserved more TP, TF, flavonols, DPPH free radical scavenging activity, TAC and cloud values of grapefruit juice as compared to control during storage period. However, further research needs to be conducted for the optimization of processes by changing variables such as power, frequency, time and temperature of treatment. Based on the results obtained, sonication seems a promising non-thermal technique to enhance the quality of juice as shown by its effects on selected parameters investigated during refrigerated storage. Therefore, it is suggested that the technique may potentially be implemented for grapefruit juice processing to preserve its nutritional and sensory quality for consumer's satisfaction.

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