Arch Lebensmittelhyg 73,
180–186 (2022)
DOI 10.53194/0003-925X-73-180

ISSN 0003-925X

Korrespondenzadresse:
leylanatheghi@iauvaramin.ac.ir

[1])Department of Food Science and Technology, Faculty of Agriculture, Varamin-Pishva Branch, Islamic Azad University, Varamin, Iran; [2])Associate Professor, Department of Food Science and Technology, Faculty of Agriculture, Varamin-Pishva Branch, Islamic Azad University, Varamin, Iran; [3])Halal Research Center of IRI, Ministry of Health and Medical Education, Tehran, Iran

Comparison of antioxidant potency, pectin methyl esterase activity, and microbial contamination in red grape juice samples pasteurized by ultrasonication and thermal process

Vergleich der antioxidativen Wirkung, der Pektin-Methylesterase-Aktivität und der mikrobiellen Kontamination in roten Traubensaftproben, die mit Ultraschall und thermischen Verfahren pasteurisiert wurden

Ghazaleh Abdullahi Moghaddam Masouleh[1]), Leila Nateghi[2]), Masoumeh Moslemi[3])

Summary

Today, people tend to consume products with high nutritional value. Ultrasound-assisted pasteurization is one of alternative methods for thermal pasteurization. In this study, red grape juice samples were treated by ultrasonication at different powers (10, 105, and 200 W), temperatures (0, 30, and 60 °C), and times (2, 6, and 10 min). After optimization by response surface methodology (Box-Behnken method), antioxidant potency, pectin methyl esterase activity, and microbial contamination (total count of microorganisms, coliforms, molds, and yeasts) of the red grape juice treated by the optimized ultrasonication were compared to those achieved by thermal pasteurization (at 90 °C for 30 sec) as control. By increasing power from 10 to 200 W, temperature from 0 to 60 °C, and time from 2 to 10 min, antioxidant potency, activity of pectin methyl esterase, and microbial population decreased in the samples, significantly. The optimum condition of ultrasonication to achieve the least microbial growth (non-detectable), and the highest antioxidant potency (IC_{50} of 62.3062 µl/ml) and pectin methyl esterase activity (0.0568 PEU/ml) was predicted at 180 W, 42 °C, and 8 min. After optimization, there was no significant difference between microbial contamination in the sample treated by ultrasonication and the thermally pasteurized red grape juice, but higher antioxidant potency and pectin methyl esterase activity was observed in the ultrasound-treated sample than control. Therefore, grape juice can be pasteurized by ultrasonication instead of thermal process, through which bioactive compounds are better preserved without safety concern in term of microbial contamination.

Keywords: red grape juice, thermal pasteurization, ultrasonication, antioxidant activity, pectin methyl esterase, microbial contamination

Introduction

Ultrasonication is of fast, non-thermal and non-destructive processes commonly used in food industries that protects quality and organoleptic properties of the products (Wambura et al., 2008; Irkilmez et al., 2017). In addition, it has high efficiency and includes low energy and maintenance costs (Zinoviadou,et al., 2015).

Red grape with scientific name of *Vitis vinifera* is a genus of *Vitaceae* family consisted of 11 genera and more than 600 species. Grapes are divided into seeded and seedless, which are found in red, black, yellow, and green. Red grape has fascinating effect in improvement of memory performance and control of Alzheimer's disease (Patrice et al., 2006). Therefore, protection of its bioactive compounds under industrial processing to benefit from the highest functional properties is of concern.

Thermal pasteurization is the common processing method used in juice industry. Despite its high potential in inactivation of microorganisms, high temperatures adversely affect nutrients and bioactive compounds of the products and may lead to loss of vitamins, non-enzymatic browning, protein denaturation, loss of flavor, discoloration, etc. (Vikram et al., 2005; Irkilmez et al., 2017). For example, vitamin C is susceptible to high temperatures and is significantly reduced during thermal pasteurization. Moreover, physicochemical properties of juices such as acidity, pH, Brix, and color may change after browning (Khandpur et al., 2015; Samani et al., 2015). On the other hand, unprocessed fruit juices might be contaminated by microorganisms especially molds and yeasts, that their growth reduces the nutritional value other than production of microbial toxins, leading to unpleasant sensory attributes (Elvira et al., 2014; Ortuño et al., 2015).

Several studies have been done about effect of ultrasound waves on quality attributes of foods. For example, antioxidant activity of red raspberry puree at frequencies of 20–1000 kHz for 30 min (Golmohamadi et al., 2013), total phenols, flavonoids, and antioxidant capacity of pear juice at power of 750 W, frequency of 20 kHz, and amplitude of 70% (Saeeduddin et al., 2015), structural and physical properties and lycopene content of guava juice at power of 1000 W, frequency of 20 kHz, at 25 ºC for 3–9 min (Campoli et al., 2018), and microstructure of pectin and carbohydrates, and rheological properties of kiwi juice at power of 400 W and frequency of 20 kHz for 4–16 min (Wang et al., 2020) have been investigated. We studied total phenol content and anthocyanin level of red grape juice after treatment by ultrasonication in our previous work for the first time (Abdollahi Moghaddam Masouleh et al., 2022). In the current work, antioxidant potency by DPPH inactivation method, pectin methyl esterase (PME) activity, and microbial contamination (total microorganisms, coliforms, yeasts and molds) of the samples were evaluated in the laboratory. For this purpose, different temperatures (0, 30, and 60 °C), ultrasound powers (10, 105, and 200 W), and times (2, 6, and 10 min) were studied for pasteurization of red grape juice by ultrasound waves and the optimum condition was determined by Response Surface Methodology (RSM) and Box-Behnken method. Results of the optimized ultrasonication (i.e., the least microbial contamination and the most antioxidant and PME activity) were compared to the results of thermal pasteurization at 90 °C for 30 sec.

Materials and methods

Materials

Red grape was purchased from local market (Tehran, Iran). Phenolphthalein, sodium hydroxide, and plate count agar were purchased from Merck company (Germany). Lauryl sulfate tryptose broth, brilliant green lactose bile broth, and Dichloran Rose-Bengal Chloramphenicol Agar were prepared from Sigma-Aldrich company (USA).

Methods

Sample preparation, design of experiments, and pasteurization

Red grape juice was produced by mechanical force after washing of fruits. An ultrasonicator instrument model AMMM (Switzerland) with output of 1000 W under frequency of 20 ±0.5 kHz equipped with cylinder probe made of titanium with a diameter of 20 mm was used. Design of experiments was done by three variables of temperature (0, 30, and 60°C), time (2, 6, and 10 min), and power (10, 105, and 200 W) (Table 1). Three levels of the variables were determined based on a preliminary study. Results of the ultrasonication method were optimized by RSM (Mini-Tab software version 16). The optimal process was the condition leading to the least microbial contamination (total count, coliform, mold, and yeast) and the highest antioxidant and PME activity. In parallel, red grape juice was thermally pasteurized at 90 °C for 30 sec as a common processing method in food industry. At the end, the results of the optimized ultrasonication and the thermal pasteurization were compared.

PME activity

PME activity was measured by using the method described by Kimball (1999). At first, 20 ml of pectin solution (10 g pectin and 15.3 g NaCl in 1000 ml distilled water) was added to 5 ml of red grape juice in a 100-ml beaker. Then, pH of the solution was adjusted to 7 with NaOH (2 N). Then the pH of solution was readjusted to pH 7.7 with NaOH (0.05 N). Then 0.1 ml of NaOH with 0.05 normality was added to the sample at once and the time taken to regain

TABLE 1: *Treatments developed by RSM (Box-Behnken method) for ultrasonication of red grape juice samples.*

Treatment	Power (W)	Temperature (°C)	Time (min)
1	105	30	6
2	10	60	6
3	10	30	2
4	200	0	6
5	10	0	6
6	105	60	10
7	105	60	2
8	105	30	6
9	200	30	10
10	105	30	6
11	105	0	10
12	200	60	6
13	10	30	10
14	105	0	2
15	200	30	2

Journal of Food Safety and Food Quality 73, Heft 6 (2022), Seiten 169–194

pH 7.7 was recorded. The enzyme activity unit (PEU) was calculated according Equation 1 (Kimball, 1999).

$$\text{PEU}\left(\frac{\text{unit}}{\text{ml}}\right) = \frac{(0.05\ \text{N NaOH})\times(0.1\ \text{ml of NaOH})}{(5\ \text{ml sample})\times(\text{time (min)})} \qquad \text{Equation 1}$$

Antioxidant activity

Antioxidant potency of the samples was measured by 2,2-diphenyl-1-picrylhydrazyl (DPPH) method. In this method, 0.1 ml of DPPH solution (0.1 mM in methanol) was added to 3.9 ml of red grape juice at different concentrations (100, 200, 300, 400, and 500 mg/kg). The mixture was placed in darkness for 30 min. Then, absorbance of the mixture was read at 517 nm by spectrophotometer (NanoDrop ND-1000, USA). Distilled water was used as a control. Antioxidant potency was calculated according to Equation 2 (Mousavi et al., 2022).

$$\text{DPPH inhibition (\%)} = \frac{AB-AS}{AB}\times 100 \qquad \text{Equation 2}$$

Where, AS is sample absorbance, and AB is blank absorbance. In our study, antioxidant activity is expressed as IC50, that is the concentration able to scavenge 50% of DPPH radicals.

Microbial tests

Total microorganisms were enumerated in plate count agar after incubation at 30 °C for 72 h (ISIRI 1-5272, 2015). For coliforms, 10 ml of lauryl sulfate tryptose broth was inoculated with 1 ml of red grape juice and incubated at 37 °C for 24–48 h. Then, 1 ml of the incubated media in which the bacteria were grown (the turbid media or those containing gas bubbles) was transferred to 10 ml of brilliant green lactose bile broth and incubated again at 37 °C for 24–48 h. Number of coliforms per ml of the sample was determined based on the tubes containing gas bubbles (ISIRI 11166, 2008a). For mold and yeast, Dichloran Rose-Bengal Chloramphenicol Agar was used and the inoculated medium was incubated at 25 °C for 3–5 days (ISIRI 1-10899, 2008b).

Results and Discussion

Analysis of variances (ANOVA) and predicted models developed by RSM (Box-Behnken method)

As seen in Table 2, power, temperature, time, and squared power (a^2) had significant effect on PME activity and antioxidant potency in red grape juice pasteurized by ultrasonication. Square of temperature (b^2) and time (c^2) and interactions of power, temperature, and time had no significant effect on PME and antioxidant activity of red grape juice treated by ultrasonication. Similar results were observed for single effect of each variable and also squared power (a^2) on total microorganisms, coliforms, mold, and yeast in the ultrasonicated samples. In addition, interaction of power and temperature showed significant effect on microbial growth. Predicted models developed by RSM for PME and antioxidant activity and microbial growth in the ultrasound-treated samples are presented in Table 3.

TABLE 2: *Results of ANOVA developed by RSM (Box-Behnken method) for red grape juice treated by ultrasonication at different levels of power, temperature, and time.*

Source of changes	Mold and yeast (cfu/ml)		Coliforms (cfu/ml)		Total micro-organisms (cfu/ml)		IC_{50} (µl/ml)		PME activity (PEU/ml)	
	P-value	F-value	P-value	F-value	P-value	F-value	P-value	F-value	P-value	F-value
Regression constant	0.000*	180.26	0.001*	31.26	0.000*	131.98	0.000*	168.82	0.000*	69.93
Linear effects	0.000*	459.52	0.000*	74.61	0.000*	325.28	0.000*	471.90	0.000*	195.50
Power (a)	0.000*	1209.68	0.000*	181.7	0.000*	848.15	0.000*	1120.58	0.000*	452.71
Temperature (b)	0.000*	156.77	0.002*	31.95	0.000*	187.83	0.000*	266.10	0.000*	112.80
Time (c)	0.018*	12.10	0.024*	10.17	0.006*	20.87	0.003*	29.03	0.006*	20.99
Square effects	0.000*	72.56	0.007*	13.84	0.001*	39.13	0.001*	33.06	0.008*	13.05
Power × Power (a^2)	0.000*	210.02	0.001*	40.97	0.000*	116.59	0.000*	99.11	0.002*	31.89
Temperature × Temperature (b^2)	0.176	2.48	0.416	0.79	0.951	0.00	0.600	0.31	0.158	2.76
Time × Time (c^2)	0.765	0.10	0.925	0.01	0.509	0.51	0.570	0.37	0.222	1.95
Interactions	0.020*	8.71	0.051*	5.34	0.069*	4.51	0.322	1.50	0.389	1.23
Power × Temperature (a × b)	0.004*	24.19	0.011*	15.17	0.015*	13.21	0.107	3.85	0.223	1.93
Power × Time (a × c)	0.37	0.97	0.45	0.67	0.703	0.16	0.980	0.00	0.249	1.70
Temperature × Time (b × c)	0.37	0.97	0.699	0.17	0.703	0.16	0.459	0.64	0.797	0.07
Lack of fit	0.156	14.84	0.211	15.17	0.315	14.87	0.417	10.11	0.563	9.86

TABLE 3: *Predicted models developed by RSM (Box-Behnken method) for red grape juice treated by ultrasonication at different levels of power, temperature, and time.*

Source	Model	R^2	R^2-adjusted
PME activity (PEU/ml)	$Y = 0.071885 - 0.015838a - 0.007906b - 0.003410c + 0.006188a^2 - 0.001819b^2 - 0.001528c^2 - 0.001463ab - 0.001372ac + 0.000286bc$	99.21%	97.79%
IC_{50} (µl/ml)	$Y = 56.5819 + 7.4825a + 3.6462b + 1.2044c - 3.2755a^2 - 0.1839b^2 - 0.1997c^2 - 0.6204ab + 0.0085ac - 0.2537bc$	99.67%	99.08%
Total microorganisms (cfu/ml)	$Y = 8.333 - 12.75a - 6b - 2c + 6.958a^2 - 0.0417b^2 + 0.4583c^2 + 2.25ab - 0.25ac - 0.25bc$	99.58%	98.83%
Coliforms (cfu/ml)	$Y = 5.0000 - 11.6250a - 4.8750b - 2.7500c + 8.1250a^2 + 1.1250b^2 - 0.1250c^2 + 4.7500ab + 1.0000ac - 0.5000bc$	98.25%	95.11%
Mold and yeast (cfu/ml)	$Y = 6.6667 - 12.5000a - 4.5000b + 0.1667c + 7.6667a^2 - 0.8333b^2 + 0.1667c^2 + 2.5000ab - 0.5000ac + 0.5000bc$	99.69%	99.14%

a: power, b: temperature, c: time

Antioxidant potency and PME activity of red grape juice samples

The tested and the predicted IC_{50} (µl/ml) and PME activity (PEU/ml) in red grape juice samples treated by ultrasound waves are presented in Table 4.

According to the results, IC_{50} (µl/ml) increased by increasing power from 10 to 200 W, temperature from 0 to 60 °C, and time from 2 to 10 min. The highest IC_{50} (63.335 µl/ml) was found at 200 W, 60 °C, and 6 min, while the lowest IC_{50} (41.669% µl/ml) was found at 10 W, 0 °C, and 6 min (Table 4).

Antioxidants are secondary metabolites of plants that prevent oxidation reactions stimulated by environmental factors such as light, oxygen, and microbiological attack (Hidalgo et al., 2017). Colorful fruits are full of antioxidants playing vital role in human health (Rawson et al., 2011). Antioxidants are susceptible to environmental factors and may be degraded during conventional processing. Use of ultrasound waves in combination with mild heating can significantly reduce processing time, energy consumption, and production costs. Therefore, other than cost-effectiveness, biologically active compounds such as phenols and anthocyanins are better preserved (Sulaiman et al., 2016; Zou et al., 2016). Although, use of high time and temperature during sonication may reduce antioxidant potency due to degradation of antioxidant compounds (Sulaiman et al., 2017). It is in agreement to our results, so that antioxidant potency decreased by increasing time and temperature and also power of ultrasonication (Table 4). Similar results were observed in our previous study, through which anthocyanin and total phenol content of red grape juice decreased by increasing power, time, and temperature of ultrasonication (Abdollahi Moghaddam Masouleh et al., 2022). Interesting results were observed in study of Aadil et al. (2013), who investigated the effects of ultrasound treatment on quality of grapefruit juice. In their study, sonication of grapefruit for 30, 60, and 90 min at 28 kHz and temperature of 20 °C led to significant improvement in free radical scavenging activity, total antioxidant capacity, total phenols, flavonoids, and flavonols.

As seen in Table 4, PME activity of red grape juice decreased significantly by increasing power from 10 to 200 W, temperature from 0 to 60 °C, and time from 2 to 10 min. The highest PME activity (0.1 PEU/ml) was measured in the sample treated at 10 W and 0 °C for 2 min, and the lowest PME activity (0.050 PEU/ml) was found in the red grape juice treated at 200 W and 60 °C for 6 min (Table 4). PME activity reduces turbidity of fruit juice and causes a clear texture by separation of methyl group from pectin structure (Vasantha Rupasinghe and Yu, 2012). Enzyme inactivation by ultrasound treatment can be attributed to the bubbles produced by ultrasound waves, which further burst in the environment. The bursting bubbles trigger physical and chemical reactions. Sonolysis of water molecules to hydroxy free radicals is of these reactions. These radicals easily react with biological molecules such as enzymes, through which chemical structure of enzymes is altered leading to their reduced biological activity (Vercet et al., 2001; Kadkhodaee and Povey, 2008; Bora et al., 2017;). Such deteriorative effect is accelerated when the product is processed at high temperature as a result of hydrogen bonds dissociation and thermal deamidation of amino acids such as asparagine and glycine (Tanaka and Hoshino, 2003).

TABLE 4: *Comparison of tested and predicted IC_{50} (µl/ml) and PME activity (PEU/ml) in red grape juice samples pasteurized by ultrasonication.*

Treatment	Tested IC_{50} (µl/ml)	Predicted IC_{50} (µl/ml)	Tested PME activity (PEU/ml)	Predicted PME activity (PEU/ml)
1	56.681	56.582	0.072	0.072
2	49.368	49.907	0.086	0.086
3	44.317	44.428	0.095	0.094
4	58.118	57.579	0.070	0.070
5	41.669	41.373	0.100	0.099
6	60.980	60.795	0.060	0.058
7	59.544	58.894	0.063	0.064
8	56.676	56.582	0.072	0.072
9	61.913	61.802	0.055	0.056
10	56.389	56.582	0.072	0.072
11	53.360	54.010	0.074	0.073
12	63.335	63.631	0.050	0.051
13	47.174	46.820	0.088	0.090
14	50.910	51.094	0.078	0.080
15	59.022	59.376	0.068	0.065

Microbial growth in red grape juice samples

Microbial population in red grape juice samples treated by ultrasonication under different conditions of power, temperature, and time was enumerated in the laboratory and compared to those predicted by RSM (Table 5). According to the results, power, temperature, and time had significant effect in suppression of microorganisms in the samples, so that microbial population decreased by increasing power from 10 to 200 W, temperature from 0 to 60 °C, and time

TABLE 5: *Comparison of tested and predicted microbial growth in red grape juice samples treated by ultrasonication.*

Treatment	Tested count (cfu/ml)	Predicted total count (cfu/ml)	Tested mold and yeast (cfu/ml)	Predicted mold and yeast (cfu/ml)	Tested coliform (cfu/ml)	Predicted coliform (cfu/ml)
1	8.000	8.333	7	6.667	6.000	5.667
2	20.000	19.750	19	19	17.000	16.750
3	31.000	30.250	28	27.75	26.000	25.375
4	6.000	6.250	3	3	4.000	4.250
5	35.000	36.250	32	33	30.000	31.000
6	1.000	1.300	1	1.10	1.000	1.375
7	4.000	5.000	2	2.25	2.000	2.875
8	8.000	8.333	6	6.667	5.000	5.667
9	ND*	ND	ND	ND	ND	ND
10	9.000	8.333	7	6.667	6.000	5.667
11	14.000	13.000	9	8.75	11.000	10.125
12	ND	ND	ND	ND	ND	ND
13	27.000	26.750	27	26.25	23.000	22.875
14	17.000	16.500	13	12.25	13.000	12.625
15	5.000	5.250	3	3.75	3.000	3.125

*ND: non-detectable

from 2 to 10 min. In this regard, the highest microbial growth was found in the sample treated at 10 W and 0 °C for 6 min, and the lowest growth was observed in the samples treated at 200 W and 30 °C for 10 min, and 200 W and 60 °C for 6 min.

According to Iran regulation, total microorganisms, coliforms, and mold and yeast should be less than 1 cfu/ml in fruit juice (ISIRI 3414, 2019). Our study revealed that there was no significant difference between microbial growth in the ultrasound-treated sample under optimum condition (200 W, 60 °C, 10 min) and the thermally pasteurized red grape juice (90 ºC for 30 sec). Similar results were reported by Valero et al. (2007) in study of orange juice, and Saeeduddin et al. (2015) in study of pear juice for suppression of microorganisms by ultrasound treatment.

Thermal ultrasound can inactivate microbes in fruit juice. However, microorganisms do not respond to ultrasonication in same way and effectiveness of sonication depends on amplitude of ultrasound waves, processing time and temperature, volume of processed juice, and juice composition. Microbial destruction is mainly due to changes of cell membrane, local heat generation, physical pressure induced by bursting bubbles, and formation of free radicals and hydrogen peroxide in the environment under sonication (Lee et al., 2009; Bhat et al., 2011; Chandrasekhar et al., 2012; Mohideen et al., 2015).

Tomadoni et al. investigated the protective effect of ultrasonication alone or in combination with pomegranate extract in kiwi juice. In their study, ultrasound treatment alone was able to significantly reduce the number of yeasts and molds possibly by bubble cavitation process (Tomadoni et al., 2015). In addition, ultrasound treatment could extend shelf life of blueberry juice (Mohideen et al., 2015), orange juice (Guerrouj et al., 2016), carrot juice (Adiamo et al., 2018), grapefruit juice (Aadil et al., 2017), and strawberry juice (Bhat and Goh, 2017) by similar mechanisms.

Single optimization for effect of ultrasonication on antioxidant potency, PME activity, and microbial growth in red grape juice

Result of single optimization for antioxidant activity in red grape juice intended for ultrasound treatment is shown in Figure 1a. Accordingly, the best result of IC50 (39.7242 µl/ml) was predicted at 10 W, 0 °C, and 2 min with 100% desirability.

Result of single optimization for PME activity (PEU/ml) in red grape juice intended for ultrasonication is depicted in Figure 1b. As seen in the figure, the optimum condition to achieve the maximum PME activity (0.0993 PEU/ml) was predicted at 10 W and 0 °C for 2 min with 99.261% desirability.

Single optimization of microbial growth in ultrasound-treated products with 100% desirability was achieved at 179 W, 60 °C, and 10 min for total count of microorganisms (Figure 1c) and 200 W, 60 °C, and 10 min for coliforms (Figure 1d), and yeasts and mold (Figure 1e).

Simultaneous optimization for effect of ultrasonication on antioxidant potency, PME activity, and microbial growth in red grape juice

According to Figure 2, the optimum condition to achieve the highest PME activity (0.0568 PEU/ml) and the least

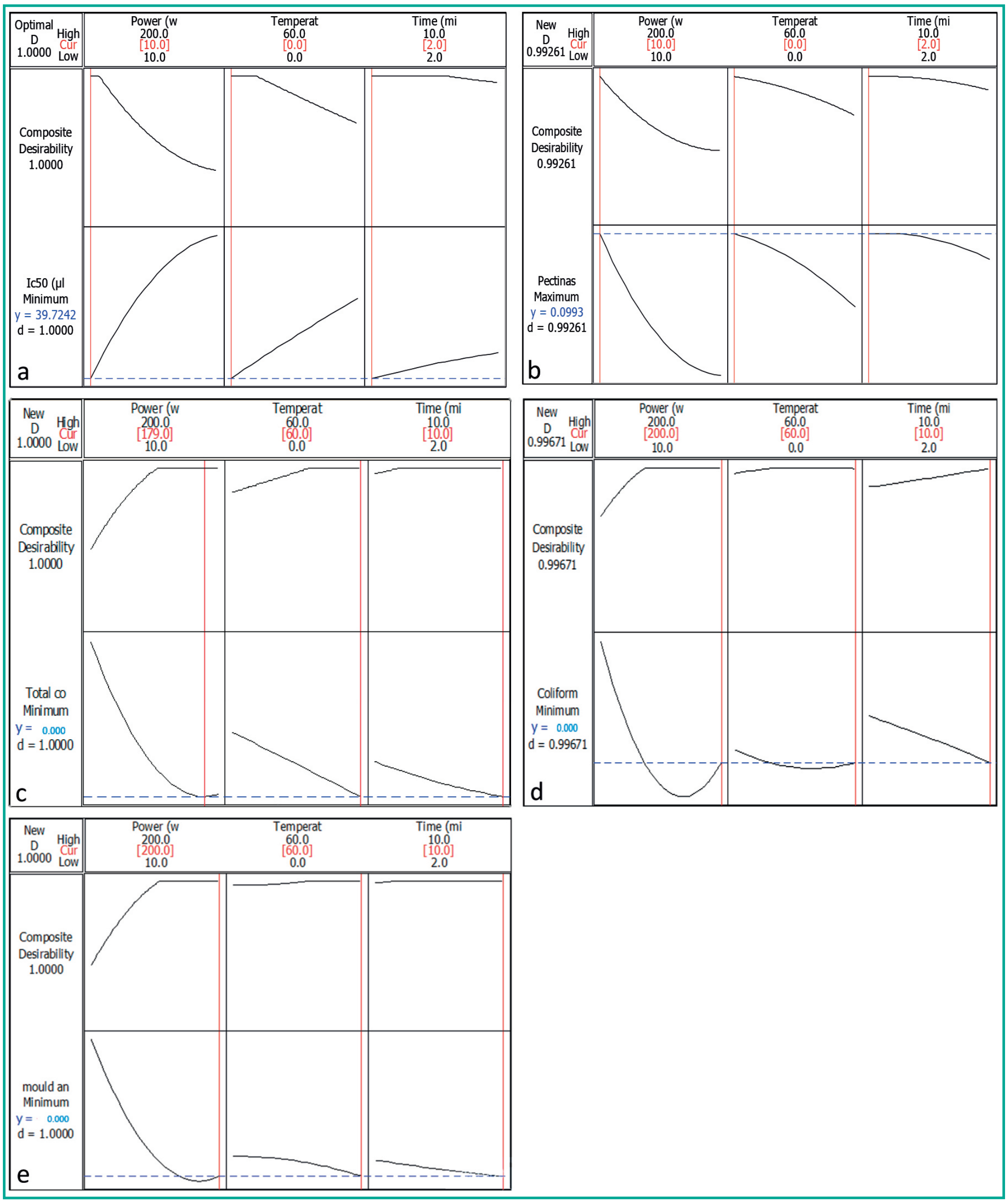

FIGURE 1: *Single optimization of a) IC_{50} (µl/ml), b) PME activity (PEU/ml), c) total count of microorganisms (cfu/ml), d) coliforms (cfu/ml), e) mold and yeast (cfu/ml) in red grape juice samples for ultrasound treatment.*

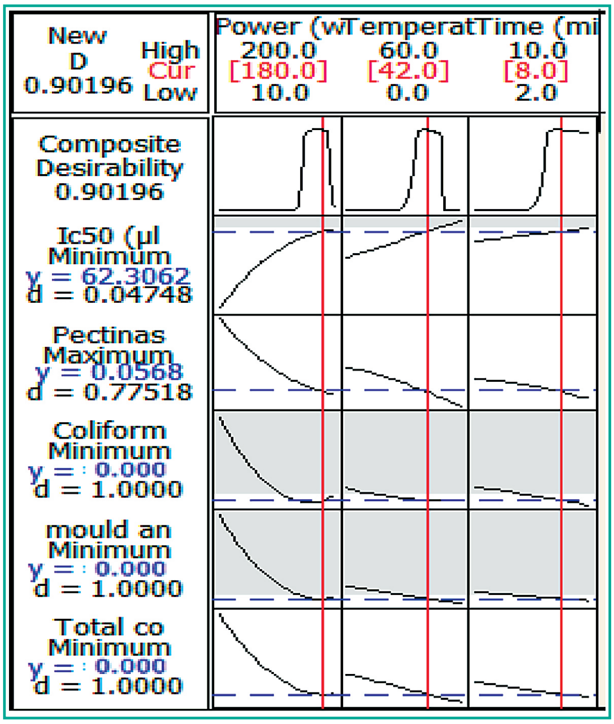

FIGURE 2: *Simultaneous optimization of PME activity, antioxidant potency, and microbial growth in red grape juice intended for ultrasound-assisted pasteurization.*

IC_{50} (62.3062 µl/ml) and microbial growth (non-detectable) in red grape juice after ultrasonication was predicted at 180 W, 42 °C, and 8 min with 90.196% desirability.

Comparison of the ultrasound-treated sample under optimum condition and the thermal pasteurized sample

Table 6 shows the results of antioxidant potency, PME activity, and microbial growth in both thermally pasteurized (90 °C for 30 sec) and optimized ultrasonicated (180 W, 42 °C, 8 min) samples.

According to the table, both methods were efficient enough in suppression of microorganisms in the red grape juice samples. Nonetheless, the bioactive compounds (e.g., antioxidants) were better preserved in the ultrasonicated sample. On the other hand, the higher activity of PME in the sample treated by ultrasonication is of interest in food industry due to a clear matrix required for grape juice. Therefore, ultrasonication under optimized conditions could be considered as efficient alternative for thermal pasteurization in juice industry.

TABLE 6: *Comparison of the pasteurized sample at 90 °C for 30 sec with the ultrasonicated sample under optimum condition.*

Test	Optimum ultrasonication	Thermal pasteurization
IC_{50} (µl/ml)	60.132 ± 0.018^{A}	67.13 ± 0.008^{B}
PME activity (PEU/ml)	0.052 ± 0.001^{A}	0.038 ± 0.002^{B}
Total microorganisms (cfu/ml)	ND	ND
Coliforms (cfu/ml)	ND	ND
Molds and yeasts (cfu/ml)	ND	ND

*Different uppercase letters indicate significant difference ($p \leq 0.05$) in rows. **ND: non-detectable

Conclusion

This study investigated the effects of ultrasonication under optimized condition on bioactive compounds and in suppression of microbial population in red grape juice compared to thermal pasteurization. The results indicated that by increasing power from 10 to 200 W, temperature from 0 to 60 °C, and time from 2 to 10 min, PME activity and microbial population decreased, and IC50 increased, significantly. Simultaneous optimization of the variables for ultrasonication of red grape juice to achieve the least microbial growth, and the highest PME and antioxidant activity suggested a processing at 180 W, 42 °C, and 8 min with 90.196% desirability. Both the thermal pasteurization and the optimized ultrasonication could suppress the microorganisms up to non-detectable level. Nonetheless, higher PME activity and antioxidant potency were detected in the sample treated by ultrasonication under optimized condition than the thermally pasteurized sample. In conclusion, ultrasonication can be considered as efficient alternative for thermal pasteurization in fruit juice processing. Although, our results were achieved in the laboratory. For scaling up, its cost-effectiveness should be investigated.

Conflict of interest

None of the authors has conflict of interest. The research was done by the authors themselves and no external body supported the research financially.

References

Aadil RM, Zeng X, Han Z, Sun D (2013): Effects of ultrasound treatments on quality of grapefruit juice. Food Chem. 141: 3201–3206.

Aadil RM, Zeng X, Han Z, Sahar A, Ahmed Khalil A, Ur Rahman U, Khan M, Mehmood T (2017): Combined effects of pulsed electric field and ultrasound on bioactive compounds and microbial quality of grapefruit juice. J Food Process Preserv 1–6. https://doi.org/10.1111/jfpp.

Abdollahi Moghaddam Masouleh Gh, Nateghi L, Moslemi M (2022): Comparison of Microbial Loads and Bioactive Compounds of the Grape Juice Samples Treated by Ultrasonication and Thermal Pasteurization. Appl Food Biotechnol. 9(3): 217–225. http://dx.doi.org/10.22037/afb.v9i3.37919

Adiamo OQ, Ghafoor K, Al-Juhaimi F, Babiker EE, Ahmed IAM (2018): Thermosonication process for optimal functional properties in carrot juice containing orange peel and pulp extracts. Food Chem 245: 79–88.

Bhat R, Kamaruddin NSBC, Min-Tez L, Karim AA (2011): Sonication improves Kasturi lime (Citrus microcarpa) juice quality. Ultrason Sonochem 18(6): 1295–300.

Bhat R, Goh KM (2017): Sonication treatment convalescent the overall quality of hand-pressed strawberry juice. Food Chem 215: 470–476.

Bora SJ, Handique J, Sit N (2017): Effect of ultrasound and enzymatic pretreatment on yield and properties of banana juice. Ultrason Sonochem (37): 445–451.

Campoli SS, Rojas ML, do Amaral JEPG, Canniatti-Brazaca SG, Augusto PED (2018): Ultrasound processing of guava juice: Effect on structure, physical properties, and lycopene in vitro accessibility. Food Chem 268: 594–601.

Chandrasekhar J, Madhusudhan M, Raghavarao K (2012): Extraction of anthocyanins from red cabbage and purification using adsorption. Food Bioprod Process 90(4): 615–623.

Elvira L, Durán CM, Urréjola J, de Espinosa FRM (2014): Detection of microbial contamination in fruit juices using non-invasive ultrasound. Food Control 40: 145–150.

Journal of Food Safety and Food Quality 73, Heft 6 (2022), Seiten 169–194

Golmohamadi A, Möller G, Powers J, Nindo C (2013): Effect of ultrasound frequency on antioxidant activity, total phenolic and anthocyanin content of red raspberry puree. Ultrason Sonochem 20(5): 1316–1323.

Guerrouj K, Sánchez-Rubio M, Taboada-Rodríguez A, Cava-Roda RM, Marín-Iniesta F (2016): Sonication at mild temperatures enhances bioactive compounds and microbiological quality of orange juice. Food Bioprod Process 99 (Supplement C): 20–28.

Hidalgo G, Pilar Almajano M (2017): Red fruits: extraction of antioxidants, phenolic content and radical scavenging determination: a review. Antioxidants 6(1): 1–27.

Iranian National Standardization Organization (2008a): Microbiology of food and animal feeding stuff – Horizontal method for the detection and enumeration of coliforms – Most probable number technique. Institute of Standards and Industrial Research of Iran, Standard No. 11166. 1st. edition.

Iranian National Standardization Organization (2008b): Microbiology of food and animal feeding stuff – Horizontal method for the enumeration of yeasts and molds – Part 1: Colony count technique in products with water activity greater than 0.95. Institute of Standards and Industrial Research of Iran, Standard No. 10899-1. 1st. edition.

Iranian National Standardization Organization (2015): Microbiology of the food chain Horizontal method for the enumeration of microorganisms – Part 1: Colony count at 30 °C by the pour plate technique. Institute of Standards and Industrial Research of Iran, Standard 454 No. 5272-1. 1st. edition.

Iranian National Standardization Organization (2019): Microbiology of Soft Drinks-Fruit juice, Vegetable juice and products –Specifications and test methods. Institute of Standards and Industrial Research of Iran, Standard No. 3414. 2nd Revision.

Irkilmez MU, Başlar M, Sağdiç O, Arici M, Ertugay MF (2017): The effect of ultrasonic treatments on turbidity, microbial load, and polyphenol oxidase (PPO) activity of plum nectar. J Food Meas Charact 11(2): 380–387.

Kadkhodaee R, Povey M (2008): Ultrasonic inactivation of Bacillus alpha-amylase: I. effect of gas content and emitting face of the probe. Ultrason Sonochem 15: 133–142.

Khandpur P, Gogate PR (2015): Understanding the effect of novel approaches based on ultrasound on the sensory profile of orange juice. Ultrason Sonochem 27(Supplement C): 87–95.

Kimball DA (1999): Citrus Processing. A complete Guide, 2nd Ed. Kluwer Academic/Plenum Publishers, New York.

Lee H, Zhou B, Liang W, Feng H, Martin SE (2009): Inactivation of Escherichia coli cells with sonication, manosonication, thermosonication, and manothermosonication: microbial responses and kinetics modeling. J Food Eng 93 (3): 354–364.

Mohideen FW, Mis Solval K, Li J, Zhang J, Chouljenko A, Chotiko A (2015): Effect of continuous ultra-sonication on microbial counts and physicochemical properties of blueberry (Vaccinium corymbosum) juice. LWT- Food Sci Technol 60(1): 563–570.

Mousavi SA, Nateghi L, Javanmard Dakheli M, Ramezan Y, Piravi Vanak Z (2022): Maceration and ultrasound-assisted methods used for extraction of phenolic compounds and antioxidant activity from Ferulago angulate. J Food Process Preserv 46(3):e16356.

Ortuño C, Balaban M, Benedito J (2015): Modelling of the inactivation kinetics of Escherichia coli, Saccharomyces cerevisiae and pectin methylesterase in orange juice treated with ultrasonic-assisted supercritical carbon dioxide. J Supercrit Fluids 90(Supplement C): 18–26.

Patrice L, Thierry T, Mark R (2006): Historical Origins and Genetic Diversity of Wine Grapes. Trends Genet 22 (9): 511–519.

Rawson A, Patras A, Tiwari BK, Noci F (2011): Effect of thermal and non-thermal processing technologies on the bioactive content of exotic fruits and their products: Review of recent advances. Food Res Int 44(7): 1875–1887.

Saeeduddin M, Abid M, Jabbar S, Wu T, Hashim MM, Awad FN, Zeng X (2015): Quality assessment of pear juice under ultrasound and commercial pasteurization processing conditions. LWT- Food Sci Technol 64(1): 452–458.

Samani BH, Khoshtaghaza MH, Lorigooini Z, Minaei S, Zareiforoush H (2015): Analysis of the combinative effect of ultrasound and microwave power on Saccharomyces cerevisiae in orange juice processing. Innov Food Sci Emerg Tech 32(Supplement C): 110–115.

Sulaiman A, Farid M, Silva FVM (2016): Strawberry puree processed by thermal, high pressure, or power ultrasound: Process energy requirements and quality modeling during storage. Food Sci Tech Int 23(4): 293–309.

Sulaiman A, Farid M, Silva FVM (2017): Quality stability and sensory attributes of apple juice processed by thermosonication pulsed electric field and thermal processing. Food Sci Tech Int 23 (3), 64–71.

Tanaka A, Hoshino E (2003): Similarities between the thermal inactivation kinetics of Bacillus amyloliquefaciens α-amylase in an aqueous solution of sodium dodecyl sulfate and the kinetics in the solution of anionic phospholipid vesicles. Biotechnol Appl Biochem 38: 175–181.

Tomadoni B, Moreira MDR, Espinosa JP, Ponce A (2015): Individual and Combined Effects of Pomegranate Extract and Ultrasonic Treatments on Kiwifruit Juice Quality Parameters. J Food Process Eng. Wiley Online, doi10.1111/JFPE.12339

Valero M, Recrosio N, Saura D, Muñoz N, Martí N, Lizama V (2007): Effects of ultrasonic treatments in orange juice processing. J Food Eng 80: 509–516.

Vasantha Rupasinghe H, Yu L (2012): Emerging preservation methods for fruit juices and beverages. Food additive 10: 32–48.

Vercet A, Burgos J, Crelier S (2001): Inactivation of Protease and Lipase by Ultrasound. Innov Food Sci Emerg Tech 2(2): 139–150.

Vikram VB, Ramesh MN, Prapulla SG (2005): Thermal degradation kinetics of nutrients in orange juice heated by electromagnetic and conventional methods. J Food Eng 69(1): 31–40.

Wambura P, Yang W, Wang Y (2008): Power Ultrasound Enhanced One-Step Soaking and Gelatinization for Rough Rice Parboiling. Int J Food Eng 4(4): 1556–3758.

Wang J, Wang J, Kranthi Vanga S, Raghavan V (2020): High-intensity ultrasound processing of kiwifruit juice: Effects on the microstructure, pectin, carbohydrates, and rheological properties. Food Chem 270: 126–121.

Zinoviadou KG, Galanakis CM, Brnčić M, Grimi N, Boussetta N, Mota MJ, Barba FJ (2015): Fruit juice sonication: Implications on food safety and physicochemical and nutritional properties. Food Res Int 77: 743–752.

Zou Z, Xi W, Hu Y, Nie C, Zhou Z (2016): Antioxidant activity of citrus fruits. Food Chem 196: 885–896.

Address of corresponding author:
Assoc. Prof. Leila Nateghi
Department of Food Science and Technology
Faculty of Agriculture
Varamin-Pishva Branch
Islamic Azad University, Varamin
Iran
leylanatheghi@iauvaramin.ac.ir