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The effect of ultrasonic treatments on cloudy quality-related quality parameters in apple juice

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ABSTRACT

The effects of ultrasonic treatment on the cloudiness (cloud level and cloud stability), sedimentation and storage of apple juice were studied at various amplitude levels (50 and 100 μm), pulse durations (50% and 100%), and temperatures (40, 50 and 60 °C). Ultrasound at high amplitude, temperature, and time minimized the coarse particles in the apple juice and increased its cloudiness quality. The ultrasonic treatment increased the cloudiness level up to 16.9 times and the cloud stability up to 9.8 times. Up to 58% of solid particles causing sedimentation were suspended after the treatment. Even after storage for four months, the apple juice remained cloudy without any additional processing. Total yeasts and molds were completely inactivated (<1 log cfu/mL) by the ultrasonic treatment. These findings suggest that thermosonication could be considered a good alternative to conventional treatment in order to achieve the desired cloudiness in cloud apple juice processing.

Industrial relevance: The article demonstrates some interesting results in using ultrasonic treatments on cloud quality parameters in apple juice. The ultrasonic treatments applied at high temperature improve dramatically cloud value and cloud stability, and decrease sedimentation level of the juice. The treatments also completely inactivate yeasts and molds in the juice. Thus thermosonication could be considered as a good alternative to achieve the desired cloudiness in apple juice processing.

1. Introduction

Apple (*Malus domestica*) is the most widely cultivated fruit in many regions of the world. It is a rich source of bioactive compounds, such as flavonoids, phenolic acids, carotenoids, minerals and vitamins, with various health benefits (Abid et al., 2014; Pfannhauser, Fenwick, & Khokhar, 2001).

Cloudy apple juice is rich in bioactive compounds, whereas clear apple juice, produced by removing the bioactive compounds, is more popular among consumers. Cloudy apple juice is produced by direct pressing of the mash without the addition of enzymes. It has pulp particles dispersed in a serum constituted by macro-molecules such as pectin and proteins (Kolniak-Ostek, Oszmianski, & Wojdylo, 2013; Wucherpfennig, Dietrich, Kanzler, & Will, 1987).

The major quality parameters of cloud apple juices are their cloudiness and microbial safety. The cloudy properties of apple juices are a decisive factor in consumer acceptance of apple juice and the prevention of yeast and mold growth in acidic juice is important on health grounds. The cloud stability of apple juice is generally related to the diameters of the particles in the juice. Particle sizes above 0.5 μm to 0.65 μm in apple juice are unstable and settle out, whereas those below 0.5 μm are held in suspension by Brownian motion and effectively do not settle (Beveridge, 2002; Stahle-Hamatschek, 1989; Zimmer, Pecoroni, Dietrich, & Gierschner, 1994).

Over the past few years, it is observed that modifications in the food processing techniques depend on increased demands. The novel techniques change chemical, physical and organoleptic properties of food. Ultrasonic treatment is a major novel technique used in the food industry for numerous processes, such as cooking, cutting, emulsification/homogenization, crystallization, extraction and microbial inactivation. (Pingret et al., 2012). Cheng, Soh, Liew, & Teh (2007) reported that ultrasonic treatment increased the cloud stability of guava juice. Tiwari, Muthukumarappan, O'Donnell, & Cullen (2009) reported that the cloud value and stability of sonicated orange juice were better than those of a control juice. In addition to structural modification, cavitation might produce chemical effects, such as the release of free radicals, and it might reduce the content or bioavailability of some nutrients (Pingret, Fabiano-Tixier, & Chemat, 2013). However, in our previous study (Başlar & Ertugay, 2013), it was determined that total phenolic component loss by ultrasound was lower than heat treatment under similar conditions. The differences at reported results might be due to ultrasonic treatment conditions such as temperature, exposure time and equipment, and also structure of foods and bioactive compounds profile.

The objective of this study was to determine the effect of sonication on the cloud quality of apple juice under different treatment temperatures,

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amplitudes, and pulse durations. The cloud quality was determined using the cloud value, cloud stability, and sedimentation. The effect of ultrasonic treatments on the particle size and yeast and mold activity in the apple juice was also studied.

2. Material and methods

2.1. Material

Apples (*Golden delicious*) were purchased from a local fruit distribution center in Erzurum, Turkey. The apples were washed and the juice was extracted using a home juice extractor (Philips HR1861-XXL, China). The apple juice was filtered using a filter (approximately 1 mm pore) and ultrasound was immediately applied to the unclarified apple juice.

2.2. Ultrasound and heat treatments

An ultrasonic processor (UP400S, Hielscher, Germany) with a 22 mm diameter probe was used for sonication (Başlar & Ertugay, 2013; Karaman, Yilmaz, Ertugay, Başlar, & Kayacier, 2012). Fresh apple juice was sonicated at various amplitude levels (50 and 100 μm), ultrasonic pulse durations (50% and 100%), treatment temperatures (40, 50, and 60 °C), and exposure times (5 and 10 min) at a constant frequency of 24 kHz. The ultrasound probe was immersed in the juices at a depth of approximately 2.5 cm within 200 mL of apple juice in a glass reactor of 250 mL with a cooling jacket. The cooling water temperature was experimentally determined before every treatment, because of temperature rise during sonication. The temperature of the samples was kept constant using a water bath (Memmert, Germany), combined with a peristaltic pump working at 7 L/min. Pulsed ultrasound (P-US) was used with a mean of pulsed duration of 0.5 s on and 0.5 s off (50%). The pasteurization of the apple juice in a glass tube was carried out at 90 °C for 1 min. Untreated samples of the same apple juice served as controls. The juices were stored in jars at 25 °C for 4 months.

2.3. Cloud value and stability

The apple juice samples were centrifuged at 760 ×g for 10 min and the absorbance of the supernatant was measured at 660 nm using a spectrophotometer (T60V Spectrophotometer, PG Instruments, UK) to determine the cloud values (Versteeg, Rombouts, Spaansen, & Pilnik, 1980; Tiwari, Muthukumarappan, O'Donnell, & Cullen, 2008). Cloud stability was determined using the method of Stahle-Hamatschek (1989). The samples were centrifuged at 4200 ×g for 15 min, and the absorbance of the supernatant was measured at 625 nm. Cloud stability was calculated according to the following equation (Genovese, Elustondo, & Lozano, 1997):

$$CS\% = \frac{C_A}{C_0} \times 100 \quad (1)$$

where C_A is the absorbance after centrifuging and C_0 is the absorbance before centrifuging.

2.4. Sedimentation

The apple juice (100 mL) samples were placed in measuring cylinders and stored at +4 °C for 24 h. After the storage, separation was determined volumetrically.

2.5. Microscopic imaging

Three drops of apple juice were dropped on the lam and dried in an oven at 60 °C. After drying, methylene blue (1%) was dropped, and the dye was removed by immersing in distilled water. Different regions were randomly selected and imaged with a microscope with 10× lens (Nikon 50i).

2.6. Total yeast and mold count

The apple juice samples were serially diluted with 0.1% sterile peptone water, and 0.1 mL of the diluted (or non-diluted) samples was inoculated potato dextrose agar (pH: 3.5) using the spread-plating method. After the inoculation, the plates were incubated at 25 °C for five days.

2.7. Sensorial analysis

Sensorial properties of the untreated, pasteurized, and ultrasound-treated apple juice samples were determined by a panel of 12 laboratory staff members. For the sensorial analysis, the ultrasound treatment was applied to apple juice samples at amplitudes of 50 and 100 μm at 40, 50, and 60 °C for 10 min, and a pulse was not used. The samples were cooled by refrigeration and were served with a glass of water. The samples were graded for five sensory attributes: color & appearance, odor, flavor, cloudiness, and general acceptability of samples. These were evaluated with a point scale of 1 (poor) to 9 (excellent). Also, panelists were asked, "Did you detect a metallic smell?"

2.8. Statistical analysis

Statistical analysis was performed using SPSS 15.0 (SPSS Inc., Chicago, USA). Variance analysis (ANOVA) was applied to determine the differences between the experimental runs.

3. Results and discussion

3.1. Cloud value and stability

The cloud value and stability of the cloud apple juice are major quality parameters. The cloud stability indicates how much apple juice is stable after centrifugation at 4200 ×g for 15 min. This level of centrifugation is considered equivalent to one year of storage (Beveridge, 2002; Stahle-Hamatschek, 1989).

The ultrasonic treatment enhanced the cloud value and stability of the apple juice, with the treatment temperature ($p < 0.01$), amplitude ($p < 0.01$) and treatment time ($p < 0.01$) significantly influencing the results. The best results were obtained at the highest processing time (10 min), temperature (60 °C), amplitude (100 μm), and pulse cycle (100%) of ultrasonic treatment. The cloud stability was determined as 21.8% at the condition. The treatment conditions caused a 16.9-fold increase in the cloud value and a 9.8-fold rise in the cloud stability in comparison with controls. These dramatic changes were even observed with the naked eye by the researchers.

In this study, the treatment temperature played a very important role in the cloud level and stability. The ultrasonic treatments of apple juice at 60 °C for 10 min increased the cloud level and cloud stability between 13.4 and 16.9 times and 8.6 and 9.8 times compared to control samples, respectively. The same level of ultrasonic treatment applied at 50 °C increased the cloud level and cloud stability between 5.0 and 8.6 times and between 1.7 and 3.5 times, respectively. Ultrasonic treatment applied at 40 °C generally did not change the cloud level or the stability. Although the cloud value of the apple juice increased significantly after the treatments at 50 °C, the cloud stability was insufficient. Figs. 1 and 2 show that the highest cloud quality parameters were gained by ultrasound applied at 60 °C and an amplitude of 100 μm. At a temperature of 50 °C, pulsed ultrasound produced higher cloud value and greater stability. Fig. 4 shows that the ultrasound applied at high temperatures (thermosonication) fragmented a greater number of large particles in apple juice. Increasing the treatment time (from 5 min to 10 min) at 60 °C slightly increased the cloud level (except for pulse of