Effects of low frequency ultrasonic treatment on the maturation of steeped greengage wine

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Abstract: To accelerate wine maturation, low frequency ultrasonic waves of 28 kHz and 45 kHz were used to treat the steeped greengage wine. The contents of total acid, total ester, fusel oil and the wine chromaticity were determined before and after the ultrasonic treatment. The volatile compounds were analyzed by GC-MS method, and the sensory quality was evaluated by panelist. The results indicated that ultrasonic treatment of the steeped greengage wine at 45 kHz 360 W for 30 min was effective to accelerate the aging process, where the fusel oil and alcohol compounds were significantly reduced and acid and ester compounds were significantly increased.

Keywords: Steeped greengage wine; Low frequency ultrasonic wave; Maturation

Introduction

Greengage wine is a popular alcoholic beverage in Southeast Asia, especially China, because of its unique fruit flavor and potential health benefits such as antioxidant and anti-cancer properties (Jo et al., 2006; Chen, Wu, Liu, & Li, 2008; Jeong, Moon, Park, & Shin, 2006; Adachi et al., 2007). The Chinese well-known story of “Discussing the heroes of the country while drinking greengage wine” happened more than 1,800 years ago in Three Kingdoms Period has granted it a historical culture. Greengage wine can be made by two different processes: fermentation and steeping (Li & Zhou, 2005). Several researchers have demonstrated that greengage wine made by steeping greengage fruit in rice wine could be more effective in maintaining the fruit flavor than that of the fermentation method (Yang, Wu, Peng, & Wang, 2005; Gao, Zeng, & Xiao, 2009). The steeping process is also a common operation in most industry practice. After the steeping process, the wine forms the basic flavor and body, but it is widely recognized that freshly steeped wine is undrinkable due to the harsh taste, pungent smell and too high concentration of fusel oil. The fusel oils are one group of the main flavor components of greengage wine, but a high concentration of fusel oils may cause dizziness, headache, thirst, and other uncomfortable symptoms (Watson & Preedy, 2003；Hori, Fujii, Hatanaka, & Suwa, 2003). Therefore, it is essential for the freshly steeped wine to be aged until its sensory properties become pleasurable. Generally, fresh wine is aged by natural maturation which requires a long time (more than half a year) and huge space, and consequently, is a low efficiency method in wine industry (Tao et al., 2012). To solve this problem, scientists have worked out a number of chemical and physical accelerating aging methods, such as oak wood bucket storage, micro-oxygenation, ultrasonic wave, and ultra-high pressure treatments (Chang & Chen, 2002; Nevares & Alamo, 2008; Alamo, Nevares, Gallego, Simon, & Cadahia, 2010; Van Jaarsveld & Hattingh, 2012; Madrera, Hevia, & Valles, 2013). However, there is little information on accelerating of steeped fruit wines including steeped greengage wine.

Ultrasonic wave, especially low frequency ultrasonic wave, can significantly accelerate some types of chemical reaction rates, and is reported to be a promising technique in shortening the wine aging process (Leonhardt & Morabito, 2007; Chang, 2005). Saterlay and Compton (2000) proposed that ultrasonic wave can create an acoustic cavitation of microbubbles. The violent implosion of these microbubbles leads to energy accumulations in hot spots, and generates extreme temperatures and pressures, which produce very high shear energy waves and turbulence (Hemwimol, Pavasant, & Shotipruk, 2006; Luque de Castro & Priego-Capote, 2007). Under this extreme micro-environment, chemical polymers are accelerated to be broken into numerous particles and recombined as new polymers with good flavor and body. In a recent critical review of ultrasonic wave on food properties and bioactivities, Soria and Villamiel (2010) concluded that the ultrasonic wave in a frequency range of 16 to 60 kHz is able to accelerate oxidation, polymerization and condensation of alcohol, aldehydes, esters and olefins in wines. A number of reports demonstrated that ultrasonic wave below 100 kHz could shorten the aging process of wine maturation (Leonhardt & Morabito, 2007; Chang, 2004; Chang, 2005). However, research on accelerating the aging of steeped greengage wine is still lack of information.

The objective of this study was to develop an accelerating aging method on steeped greengage wine by applying 28 kHz and 45 kHz ultrasonic treatment. The changes of flavor components as well as other substances in wine affecting the mouthfeel and quality, such as esters, acids and fusel oils, were evaluated after 15 days storage. This study may provide useful practical information to greengage wine industry in application of ultrasonic treatment in accelerating the wine aging.

2. Materials and Methods

2.1. Materials

The fresh steeped greengage wine was kindly provided by Galore Food Co. Ltd, Zhongshan city, China. The fresh wine was made by steeping fresh greengage fruit in rice alcohols for one year. This is the conventional method of greengage wine manufacturing in Asia. Most of un-dissolved particles were removed from the wine by a series of filtering steps. The steeped greengage wine was filled in separate polyethylene terephthalate (PET) containers (2 L/each), and kept in a dark and dry environment (15°C). After arrival of our laboratory, the fresh wine was stored less than 1 month before the experiment treatment. The alcohol concentration of the fresh wine was 17.5% (v/v). All solvents and chemicals (Sinopharm Ltd., China) used in this study were of analytical grade.

2.2. Ultrasonic treatment

The fresh greengage wine was treated by an ultrasonic KQ-600VDV bath

(Ultrasonic Instrument, Kunshan, China) with two separate frequencies: 28 kHz and 45 kHz. The ultrasonic power was adjusted at 240 W, 300 W and 360 W respectively. The equipment was filled with water as a medium for ultrasonic vibration transmitting, and the water was replaced after each treatment to keep the same ultrasonic heating effect. About 300 mL of fresh steeped greengage wine was filled in a 500 mL erlenmeyer flask with lid to reduce evaporation of volatile components. Then, the flask was placed in the center of the ultrasonic bath to assure the consistent of ultrasonic treatment. Samples were collected after 10, 20, 30, 40, and 50 min of treatment and stored in sealed glass containers to prevent evaporation loss. Ultrasonic untreated fresh greengage wine was also prepared in the same way as control. The wines were analysed after 15 days of storage at a dark and dry environment (15 °C) when the major flavor components in the wines were relatively stable after the ultrasonic treatment.

2.3 Total acid and ester determination

According to China Food Industry Standard Collection (2000), the total acid of steeped greengage wine was measured by the neutralization titration method and expressed as citric acid (g/L), and the total ester content was measured by the saponification reaction method and expressed as ethyl acetate (g/L).

2.4 Chromaticity determination

The chromaticity is one of the most typical sensory characteristics of fruit wines, which reflects the shade and intensity of the wine products (Rentzsch, 2009). According to Glories (1984), the chromaticity (I) of fruit wine was the sum of absorbance at 420, 520, 620 nm (I=A_{420}+A_{520}+A_{620}). The chromaticity of wines were determined on an UV2600 spectrophotometer (Techcomp, Shanghai, China), using de-ionized water as reference.

2.5 Fusel alcohol determination

Fusel oils are by-products of wine-making industry, and are mainly composed of n-propanol, n-butanol, isobutyl alcohol and isoamyl alcohol (Lachenmeier, Haupt, & Schulz, 2008). In the present study, fusel oils refer to isobutyl alcohol and isoamyl alcohol because they are the main fusel oil components in Chinese traditional rice wine (Shen, 1998), which was used in the steeping of greengage fruit. Steeped greengage wine was distilled to eliminate the effect of wine colour and the distillation was used for the analysis. Total contents of fusel oils were determined by PDAB (p-Dimethylaminobenzaldehyde) colorimetry on the UV2600 spectrophotometer at 520 nm against reagent blank as reference (AOAC, 1984). The concentrations of individual fusel oils of isobutyl alcohol and isoamyl alcohol were determined by gas chromatography (GC 2010, Shimadzu, Japan) using a DB-WAX column (60.0 m×250 µm I.D., 0.32 mm film thickness, Supelco, USA). Oven temperature program was: from holding at 40°C for 5 min, to 180°C with an increase of 10 °C /min, keeping for 5 min. The injection temperature was 250 °C. Flow rate were: N_2, 1.2 mL/min; H_2, 47 mL/min; Air, 400 mL/min.

2.6 Volatile compounds determination

The volatile compounds of steeped greengage wine were extracted by headspace solid phase micro-extraction. The optimal ultrasonic-treated wine and untreated wine

(8 mL each) were placed in 15 mL vials with 2.4 g NaCl respectively. The vials was sealed and preheated at 25°C for 10 min. A CAR-PDMS extraction fiber (Supelco, USA) was inserted into the vials and fractionated from the sample matrix at 45°C in a thermal block for 30 min until the equilibration of volatiles. Then, the fiber was removed and inserted immediately into an injection port of a gas chromatograph (GC 6890, Agilent, USA) and desorbed for 3 min at 250 °C.

The qualitative analyses of volatile compounds were carried out on a gas chromatography mass spectrophotometer (GC 6890/MS 5975, Agilent, USA) using a DB-WAX column (30.0 m×250 μm I.D., 0.25 μm film thickness, Supelco, USA). Nitrogen was used as carrier gas with a flow rate of 1.2 mL/min. Oven temperature program was: from holding at 40°C for 4 min, to 60°C with an increase of 6 °C /min, then increasing by 10 °C/min until oven temperature reached 230 °C (8 min) and the injection temperature was 250 °C. The parameters of the mass spectrophotometer were: interface temperature, 250 °C; ion source temperature, 200 °C; electron impact (EI) spectra obtained at 70 eV; filament current, 200 uA; electrode stem source temperature, 350 °C; scanning mass range of 33-450 m/z.

The identification of flavor compounds was achieved by comparing the Kovats index (KI) of a series of n-alkane (C_7-C_{21}) with the mass spectra library of NIST98 (National Institute of Standards of Technology, Hewlett-Packard, MD, USA). The integration reports were accepted if matching degree was above 800. The relative contents of flavor compounds were determined by comparing the percentage of peak areas.

2.7 Sensory evaluation

Ultrasonic treated and untreated wine samples were sensory evaluated by 10 qualified and experienced panelists in the School of Food Science and Technology, Jiangnan University. Each panelist was in good health condition and has been trained before the evaluation. The blind tasting and centesimal score system (O.I.V., 1990) was applied to evaluate the wine's quality. Based on the distribution of appearance (20 scores), aroma (30 scores), taste (40 scores) and typicality (10 scores), all the samples were presented to the panelists separately and randomly in a sensory evaluation room at 21±1°C. After consultation with sales representatives of the wine manufacturing company of Galore Food Co. Ltd and from a market point of view, samples with a total score of over 80 were considered as good and acceptable, over 85 were excellent, between 70 and 80 were common, and below 70 were unacceptable.

2.8 Statistical analyses

Every determination was repeated three times and two replications of one treatment were performed. All the data were statistically analyzed by the software of SPSS 17.0 (SPSS Inc., Chicago, IL, USA). The significant differences were determined at the 95% level.

3. Results and discussion

3.1 Total acid content

The contents of total acid in steeped greengage wine increased significantly after ultrasound treatments (Table 1). Under the ultrasound frequency of 28 kHz and power of 240 W, the highest concentration of total acid was 14.22±0.07 g/L at 30 min, whereas the highest level of 14.21±0.07 g/L and 14.25±0.09 g/L were achieved under 300 W and 360 W power at 40 min respectively. No significant differences were observed for the total acid contents among the ultrasonic treated wines, which suggested that the ultrasound may have quickly (about 10 min) promoted the formation of acids in the greengage wine. In addition, the total acid under 45 kHz treatment were higher than those of 28 kHz, with the highest concentration of 14.14±0.07 g/L, 14.24±0.04 g/L and 14.36 ± 0.07 g/L at 50 min for the three powers respectively (Table 1).

Citric acid, malic acid, and tannic acid are the main organic acids in steeped greengage wine that contribute to the wine’s quality (Gao, Zeng, Xiao, 2009). The increase of total acid could be explained by the oxidation of unsaturated alcohols and aldehydes under ultrasonic conditions. The cavitation and mechanical effect of ultrasound is able to create an extreme micro-environment of high temperature and pressure, which in turn facilitate the activity of reactive molecules. Moreover, it was favorable to the formation of acids in wines because of the dissociation of oxygen that caused by cavitation bubble collapse (Petrier, Combet, & Mason, 2007).

3.2 Total ester content

The concentration of total ester also increased in the ultrasonic treated samples (Table 2). At the ultrasonic frequency of 28 kHz, the highest concentrations were 1.45±0.04 g/L, 1.66±0.04 g/L, and 1.63±0.06 g/L at 10 min under the three powers respectively, which also suggested a very quick ultrasonic effect on esterification. At the 45 kHz ultrasound and 240 W power, the highest concentration of total ester was 1.55±0.03 g/L at 20 min. Both the 300 W and 360 W power treatments had the highest level of 1.71±0.02 g/L at 10 min and 30 min, respectively (Table 2). It showed that the frequency of 45 kHz was more effective than 28 kHz in promoting the esterification in the wines, which may be caused by the more intense interaction between alcohols and acids under the higher frequency. The enhanced esterification effect between alcohols and acids under ultrasonic treatment has also been observed by Ince, Tezcanli, Belen, & Apikyan (2001). However, in the present study, ultrasonic treatment of longer than 30 min was not favorable to the greengage wine maturation as the total ester contents were decreased (Table 2), possibly because the heating effect of ultrasonic energy would have accelerated the evaporation of esters. Therefore, 10-30 min ultrasonic treatment might be appropriate for a significant esterification of the steeped greengage wine.

3.3 Chromaticity

The chromaticity of steeped greengage wine after ultrasonic treatments changed slightly as shown in Table 3. Although there had some differences among the ultrasonic treated and untreated samples, the chromaticity values were in a small variation range of 1.18-1.25, which suggested that the ultrasonic treatment has no negative effect on the color of the wines. It was reported that the mechanical effect of ultrasonic treatment accelerated the condensation of pigment compounds and increased the chromaticity, and ultrasound processing was able to affect the anthocyanins degration and the color of grape juice (Tiwari, Patras, Brunton, Cullen, & O̍Donnell, 2009; Tiwari, Patras, Brunton, Cullen, & O̍Donnell, 2010). However, the effect of ultrasonic wave on wine chromaticity was not extensively investigated and the mechanism is not fully understood.

3.4 Fusel oil content

The results indicated that ultrasonic wave was able to reduce the concentration of fusel oil in steeped greengage wine significantly when compared with untreated sample (Table 4). At the frequency of 28 kHz, the lowest concentrations of fusel oil were determined in 10-30 min under all the three powers, with the values of 394.33±7.54 mg/L, 374.33±14.71 mg/L, 392.00±5.51 mg/L, respectively. However, after 30 min, the concentration of fusel oil increased slightly. This might be caused by the release of fusel oils from the degradation of associated-alcohols in the wine (Lin, Zeng, & Yu, 2013).

For the 45 kHz treatment, the variations of fusel oil content were different for the three powers. Under the 240 W power, the lowest concentration of fusel oil was 377.00±5.29 mg/L at 10 min, whereas for the 300 w and 360 W was 400.67±2.03 mg/L and 358.00±2.00 mg/L at 30 min, respectively (Table 4). Generally, the ultrasonic of 28 kHz, 300 W and 45 kHz, 360 W were suitable for reducing the concentration of fusel oils. The concentration of individual fusel oil component was analyzed by GC and the results in the treatment of 45 kHz and 360 W sample were showed in Table 5. It indicated that the lowest concentration of isobutyl alcohol and isoamyl alcohol was 117.77 mg/L and 224.62 mg/L under 45 kHz, 360 W and 30 min ultrasonic treatment, which was consistent with the lowest total fusel oil content using colorimetry determination, although the sum of isobutyl alcohol and isoamyl alcohol was 4.36% lower than that of the total fusel oils, suggesting some other minor fusel oil components may also exist in the wine. The sonochemical effect on reducing fusel oils in greengage wine could be related to advanced oxidative processes with the production of hydroxyl radical (Mason, 2003). The ultrasound activates the surface hydroxyl of fusel oil to free radical which is in favour of oxidation and esterification, and therefore increases the acids and esters with the sacrifice of fusel oils, as discussed in sections *3.1* and *3.2*.

3.5 Flavor content

According to our preliminary determination, the optimal ultrasonic-accelerated wine was the sample that was treated by 45 kHz, 360 W ultrasonic for 30 min, and therefore was used for volatile compound analysis. The GC-MS total ion chromatogram of aroma components in treated wine and untreated wines were shown in Fig. 1 and the contents were present in Table 6. About 38 and 39 volatile compounds were determined in the treated and untreated wine respectively, which included esters, alcohols, aldehydes, ketones and other compounds. Compared with the untreated wine, 3 more esters (ethyl isovalerate, isoamyl acetate and ethyl heptanoate) were determined and 1 ester (ethyl furoate), 1 aldehydes (decanal), 1 ketone (3-hydroxy-2-butanone) were not determined (Fig. 1 and Table 6) in the ultrasonic treated sample.

The results showed that the esters in steeped greengage wine increased by 7.74%

after ultrasonic irradiation, which represented a significant flavor variation (*p*<0.05). Esters are one of the main flavor contributors in alcoholic beverages. The content of ethyl acetate and ethyl benzoate was increased by 1.57% and 2.42% in treated wine, suggesting the acceleration of esterification. However, another major flavor contributor to the greengage wine, alcohols decreased 6.75% after the ultrasonic treatment, especially with the decrease of ethyl alcohol by 7.40%. It was suggested that the increase of ester compounds in greengage wine could be the conversion of alcoholic compounds, as discussed by Ince, Tezcanli, Belen, & Apikyan (2001), and above sections of *3.2* and *3.4*. It was reported that benzaldehyde was a characteristic volatile compound in steeped greengage wine (Yang, Wu, Peng, & Wang, 2005), but no significant change was detected for benzaldehyde in the present work. The content of benzaldehyde decreased from 19.41% to 19.21% after ultrasonic treatment. Meanwhile, other volatile conpounds changes slightly without significant variation.

3.6 Sensory evaluation

Steeped greengage wine has its characteristic taste and flavor which are contributed from several compounds such as organic acids and volatile materials. Sensory evaluation is a very important tool to assess its quality and consumer acceptability. Evaluated by the 10 panelists, the sensory scores of wine samples were presented in Table 7. Sensory scores of the 28 kHz treated wines increased as the ultrasonic power and time increasing. The highest scores of 28 kHz under three powers were 82.10±3.64, 83.03±3.59, 83.40±2.77 at 50 min, 40 min and 40 min, respectively. In the first 30 min of 45 kHz, the score changes were similar to those of

28 kHz treatments, but the increase was more remarkable. However, for the 50 min treated wine at both 28 and 45 kHz frequencies and powers, their scores declined when compared to 30 and 40 min treated ones. The sensory evaluation was relevant to the chemical indices such as total esters. For example, the highest sensory evaluation score was 84.40±2.85 which suggested an excellent wine quality after ultrasonic treatment at the frequency of 45 kHz, 360 W for 30min (Table 7), and the concentration of total esters of this sample was 1.71±0.02 g/L, also the highest in the treated wines (Table 2). However, after 30 min treatment, the sensory scores of the greengage wines decreased as the ultrasonic power increased (Table 7). Generally, the results of sensory evaluation were highly in agreement with the results of chemical analysis. Suitable ultrasonic frequency and power treatments were able to accelerate the aging process by reducing the fusel oil and alcohol compounds and increasing acid and ester compounds, and therefore, improve the sensory quality.

4. Conclusion

In the present work, changes in total acid, total ester, fusel oil, chromaticity, volatile compounds and sensory quality of steeped greengage wine by ultrasonic treatment were investigated. The results showed that low frequency ultrasonic treatment had a positive effect on the aging process of steeped greengage wine according to the chemical analysis and sensory evaluation. After ultrasonic treatment, the concentrations of total acids and esters were increased which was the accelerated oxidation reactions of fusel oils and alcohols. The optimal ultrasonic treatment conditions for accelerating the aging of steeped greengage wine were 45 kHz frequency, 360 W and 30 min.

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References

Adachi, M., et al. (2007). The “*Prunus mume* Sieb. et Zucc.” (UME) is a rich natural source of novel anti-cancer substance. *International Journal of Food Properties, 10,* 375－384.

AOAC. (1984). *AOAC 959.05*, Fusel oil in distilled liquors-spectrophotometric method. In “Official Methods of Analysis,” 14th Ed. Association of Official Analytical Chemists. Washington, D.C., USA.

Chang, A. C., & Chen, A. C. (2002). The application of 20 kHz ultrasonic waves to accelerate the aging of different wines. *Food Chemistry, 79,* 501－506.

Chang, A. C. (2005). Study of ultrasonic wave treatments for accelerating the aging process in a rice alcoholic beverage. *Food Chemistry, 92,* 337－342.

Chang, A. C. (2004). The effects of different accelerating techniques on maize wine maturation. *Food Chemistry, 86,* 61－68.

Chen, Z. Y., Wu, J. J., Liu, X. M., & Li, S. F. (2009). Antioxidation and HPLC determination of Lyoniresnol in plum fermented wine. *Food Science, 30,* 82 – 85.

China Food Industry Standards Collection. (2000). *Beverage and Alcohol.* National Food Fermentation Standardization Center. Beijing, China.

Del Alamo, M., Nevares, I., Gallego, L., De Simon, B. F., & Cadahia, E. (2010). Micro-oxygenation strategy depends on origin and size of oak chips or staves during accelerated red wine aging. *Analytical Chimica Acta, 660,* 92 – 101.

Gao, M., Zeng, X. A., & Xiao, L. M. (2009). Determination of benzaldehyde content in plum fruit wine by high performance liquid chromatography. *Liquor Making Science & Technology, 5,* 110 – 112.

Glories, Y. (1984). La coleur des vins rouges. 2e partie: mesure, origine et interpretation. *Connaiss Vigne vin, 18,* 253 – 271.

Hemwimol, S., Pavasant, P., & Shotipruk. (2006). Ultrasound-assisted extraction of anthraquinones from roots of Morinda citrifolia. *Ultrason Sonochemistry, 13,* 543 – 548.

Hori, H., Fujii, W., Hatanaka, Y., & Suwa, Y. (2003). Effects of fusel oil on animal hangover models. *Alcoholism: Clinical and Experimental Research, 27,* 37S – 41S.

Ince, N. H., Tezcanli, G., Belen, R. K., & Apikyan, I. G. (2001). Ultrasound as a catalyzer of aqueous reaction systems: the state of the art and environmental applications. *Applied Catalysis B: Environmental, 29,* 167 – 176.

Jo, S. C., et al. (2006). Antioxidant activity of *Prunus mume* extract in cooked chicken breast meat. *International Journal of Food Science and Technology, 41,* 15 – 19.

Jeong, J. T., Moon, J. H., Park, K. H., & Shin, C. S. (2006). Isolation and characterization of a new compound from *Prunus mume* fruit that inhibits cancer cells. *Journal of Agricultural and Food Chemistry, 54,* 2123 – 2128.

Lachenmeier, D. W., Haupt, S., & Schulz, K. (2008). Defining maximum levels of higher alcohols in alcoholic beverages and surrogate alcohol products. *Regulatory Toxicology and Pharmacology, 50,* 313 – 321.

Lee, P. R., Yu, B., Curran, P., & Liu, S. H. (2011). Effect of fusel oil addition on volatile compounds in papaya wine fermented with *Williopsis saturnus* var. *mrakii* NCYC 2251. *Food Research International, 44,* 1292 – 1298.

Leonhardt, C. G., & Morabito, J. A. (2007). Wine aging method and system. United State Patent, US 7220439 B2.

Li, H. L., & Zhou, J. J. (2005). Production technology and healthy function of plum wine. *China Brewing, 2,* 46 – 48.

Lin, Z. R., Zeng, X. A., & Yu, S. J. (2012). Enhancement of ethanol-acetic acid esterification under room temperature and non-catalytic condition via pulsed electric field application. *Food Bioprocess Technology, 5(7),* 2637 – 2645.

Luque de Castro, M. D., & Priego-Capote, F. (2007). Ultrasound-assisted preparation of liquid samples. *Talanta, 72,* 321 – 334.

Madrera, R. R., Hevia, A. G., & Valles, B. S. (2013). Comparative study of two aging systems for cider brandy making. Changes in chemical composition. *LWT-Food Science and Technology, 54,* 513 – 520.

Mason, T. J. (2003). Sonochemistry and sonoprocessing: the link, the trends and (probably) the future. *Ultrasonics Sonochemistry, 10,* 175 – 179.

Matsuura, K., Hirotsune, M., Nunokawa, Y., Satoh, M., & Honda, K. (1994). Acceleration of cell growth and ester formation by ultrasonicwave irradiation. *Journal of Fermentation and Bioengineering, 77,* 36 – 40.

Nevares, I., & Del Alamo, M. (2008). Measurement of dissolved oxygen during red wines tank aging with chips and micro-oxygenation. *Analytical Chimica Acta, 621,* 68 – 78.

O. I. V. (1990). Recueil des methodes internationales d’analyse des vins et des mouts. Office International de la Vigne et du Vin. Paris, France.

Perez-Magarino, S., & Gonzalez-SanJose, M. L. M. (2002). Physico-chemical parameters justifying the vintage qualification in wines from Spanish Protected Designation of Origin. *European Food Research and Technology, 214,* 444 – 448.

Rentzsch, M., Weber, F., Durner, D., Fischer, U., & Winterhalter, P. (2009). Variation of pyranoanthocyanins in red wines of different varieties and vintages and the impact of pinotin A addition on their color parameters. *European Food Research and technology, 229,* 689 – 696.

Petrier, C., Combet, E., & Mason, T. (2007). Oxygen-induced concurrent ultrasonic degradation of volatile and non-volatile aromatic compounds. *Ultrasonics Sonochemistry, 14,* 117 – 121.

Saterlay, A. J., & Compton, R. G. (2000). Sonoelectroanalysis-an overview. *Fresenius’ Journal of Analytical Chemistry, 367,* 308 – 313.

Shen, Y. F. (1998). Liquor production technology encyclopedia. Beijing, China.

Sun, J. C., Yu, B., Curran, P., & Liu, S. Q. (2012). Lipase-catalysed transesterification of coconut oil with fusel alcohols in a solvent-free system. *Food Chemistry, 134*, 89 – 94.

Soria, A. C., & Villamiel, M. (2010). Effect of ultrasound on the technological properties and bioactivity of food: a review. *Trends in Food Science & Technology, 21,* 323 – 331.

Tiwari, B. K., Patras, A., Brunton, N., Cullen, P. J., O'Donnell, C. P. (2010). Effect of ultrasound processing on anthocyanins and color of red wine grape juice. *Ultrasonics Sonochemistry, 17,* 598 – 604.

Tiwari, B. K., O'Donnell, C. P., Patras, A., Brunton, N., & Cullen, P. J. (2009). Anthocyanins and color degradation in ozonated grape juice. *Food and Chemical Toxicology, 47,* 2824 – 2829.

Tao, Y., et al. (2012). Effects of high hydrostatic pressure processing on the physicochemical and sensorial properties of a red wine. *Innovative Food Science & Emerging Technologies, 16,* 409 – 416.

Van Jaarsveld, F. P., & Hattingh, S. (2012). Rapid induction of ageing character in brandy products. Ageing and general overview. *South African Journal of Enology & Viticulture, 33,* 225 – 252.

Watson, R. R., & Preedy, V. R. (2003). Nutrition and alcohol: Linking nutrient interactions and dietary intake. Washington, D. C: CRC Press.

Yang, Y. H., Wu, S. H., Wang, X. H., & Peng, Q. (2005). Analysis of flavoring compositions in green plum fruit fermenting wine and green plum fruit steeping wine by GC-MS. *Liquor Making Science & Technology, 9,* 80 – 83.

Zeng, X. A., Yu, S. J., Zhang, L., & Chen, X. D. (2008). The effects of AC electric field on wine maturation. *Innovative Food Science & Emerging Technologies, 9,* 463 – 468.

Table 1 Changes of total acid (g/L) in steeped greengage wine after 28 kHz and 45 kHz ultrasonic treatment

Frequency (kHz)	Power (W)	Treating time (min)					
		0	10	20	30	40	50
28	240	[a]12.82±0.03a	13.93±0.18b	13.98±0.05bc	14.22±0.07d	14.10±0.04cd	14.17±0.04d
	300	12.82±0.03a	14.06±0.06b	14.09±0.04b	14.14±0.04bc	14.21±0.07c	14.14±0.07bc
	360	12.82±0.03a	14.16±0.05bc	14.15±0.09bc	14.06±0.07b	14.25±0.09c	14.11±0.05b
45	240	12.82±0.03a	14.07±0.07b	14.11±0.04b	14.08±0.07b	14.08±0.09b	14.14±0.07b
	300	12.82±0.03a	14.13±0.07b	14.17±0.04bc	14.22±0.06bc	14.13±0.07b	14.24±0.04c
	360	12.82±0.03a	14.17±0.05b	14.16±0.04b	14.19±0.09b	14.22±0.09b	14.36±0.07c

[a] Different letters in the same row indicate significant different ($p \le 0.05$).

Table 2 Changes of total ester (g/L) in steeped greengage wine after 28 kHz and 45 kHz ultrasonic treatment

Frequency	Power	Treating time (min)					
(kHz)	(W)	0	10	20	30	40	50
28	240	[a]1.22±0.03a	1.45±0.04d	1.42±0.04cd	1.30±0.06ab	1.26±0.03a	1.36±0.05bc
	300	1.22±0.03a	1.66±0.04d	1.56±0.04c	1.56±0.02c	1.26±0.06a	1.45±0.05b
	360	1.22±0.03a	1.63±0.06d	1.48±0.04c	1.37±0.07b	1.48±0.02c	1.52±0.07c
45	240	1.22±0.03a	1.54±0.03c	1.55±0.03c	1.50±0.05c	1.43±0.04b	1.55±0.03c
	300	1.22±0.03a	1.71±0.02e	1.63±0.03de	1.56±0.02cd	1.47±0.03b	1.53±0.04bc
	360	1.22±0.03a	1.55±0.03d	1.47±0.05bc	1.71±0.02e	1.41±0.06b	1.52±0.03cd

[a] Different letters in the same row indicate significant different ($p \leq 0.05$).

Table 3 Changes of chromaticity in steeped greengage wine after 28 kHz and 45 kHz ultrasonic treatment

Frequency (kHz)	Power (W)	Treating time (min)					
		0	10	20	30	40	50
28	240	[a] 1.21±0.00b	1.18±0.00a	1.19±0.01a	1.20±0.01b	1.19±0.01a	1.20±0.02b
	300	1.21±0.00a	1.21±0.01a	1.20±0.01a	1.22±0.01b	1.23±0.01c	1.24±0.01c
	360	1.21±0.00a	1.21±0.01a	1.22±0.01a	1.23±0.01ab	1.25±0.02b	1.25±0.01b
45	240	1.21±0.00b	1.22±0.01b	1.21±0.01b	1.22±0.01b	1.21±0.01b	1.19±0.01a
	300	1.21±0.00ab	1.20±0.01a	1.22±0.01bc	1.23±0.01c	1.23±0.01c	1.22±0.01bc
	360	1.21±0.00a	1.22±0.01ab	1.23±0.01b	1.24±0.01c	1.22±0.01ab	1.22±0.02ab

[a] Different letters in the same row indicate significant different ($p \le 0.05$).

Table 4 Changes of fusel oil (mg/L) in steeped greengage wine after 28 kHz and 45 kHz ultrasonic treatment

Frequency (kHz)	Power (W)	Treating time (min)					
		0	10	20	30	40	50
28	240	[a]445.45±3.32c	394.33±7.54a	425.67±4.06b	397.67±7.22a	405.00±5.69a	419.33±3.96b
	300	445.45±3.32e	390.00±4.04b	412.33±5.04d	374.33±14.71a	412.33±5.78d	400.02±7.57c
	360	445.45±3.32b	403.67±9.95a	393.67±8.41a	392.00±5.51a	400.56±3.93a	393.33±3.53a
45	240	445.45±3.32d	377.00±5.29a	422.33±5.04c	397.33±7.36b	403.00±1.53b	409.00±4.58bc
	300	445.45±3.32c	410.67±0.89ab	423.33±4.33b	400.67±2.03a	411.33±2.03ab	422.33±4.70b
	360	445.45±3.32e	400.67±5.54cd	380.67±5.69b	358.00±2.00a	411.00±5.77d	391.67±4.98bc

[a] Different letters in the same row indicate significant different ($p \leq 0.05$).

Table 5 Changes of individual fusel oil components (mg/L) after 45 kHz, 360 W ultrasonic treatment.

Component	Treating time (min)					
	0	10	20	30	40	50
Isobutyl alcohol	141.21	126.94	123.32	117.77	121.65	119.74
Isoamyl alcohol	296.59	262.14	243.51	224.62	267.05	244.98

Table 6 Volatile compounds in steeped greengage wine with and without 45 kHz ultrasonic treatment

R.T/min	Compounds	Normalized content/%	
		Untreated	treated
Esters		8.94	16.68
3.44	Ethyl acetate	6.31	7.88
6.82	Ethyl butyrate	0.03	0.06
7.27	Ethyl 2-methylbutyrate	0.02	0.12
7.71	Ethyl isovalerate	-	0.05
9.13	Isoamyl acetate	-	0.19
14.05	Ethyl heptanoate	-	0.06
14.41	Ethyl lactate	0.05	0.06
15.75	Ethyl caprylate	0.21	2.99
18.39	Ethyl furoate	0.02	-

18.57	Ethyl caprate	0.02	0.48
19.02	Ethyl benzoate	1.79	4.21
20.38	Ethyl phenylacetate	0.06	0.15
20.72	Ethyl salicylate	0.14	0.13
24.77	Methyl hexadecanoate	0.03	0.05
26.27	Diethyl phthalate	0.15	0.13
27.16	Triethyl citrate	0.08	0.07
Alcohols		67.42	60.67
5.07	Ethyl alcohol	56.56	49.16
8.93	Isobutyl alcohol	1.90	1.67
10.60	n-Butyl alcohol	0.02	0.05
12.02	Isoamyl alcohol	6.01	7.09
14.58	Hexyl alcohol	0.15	0.21

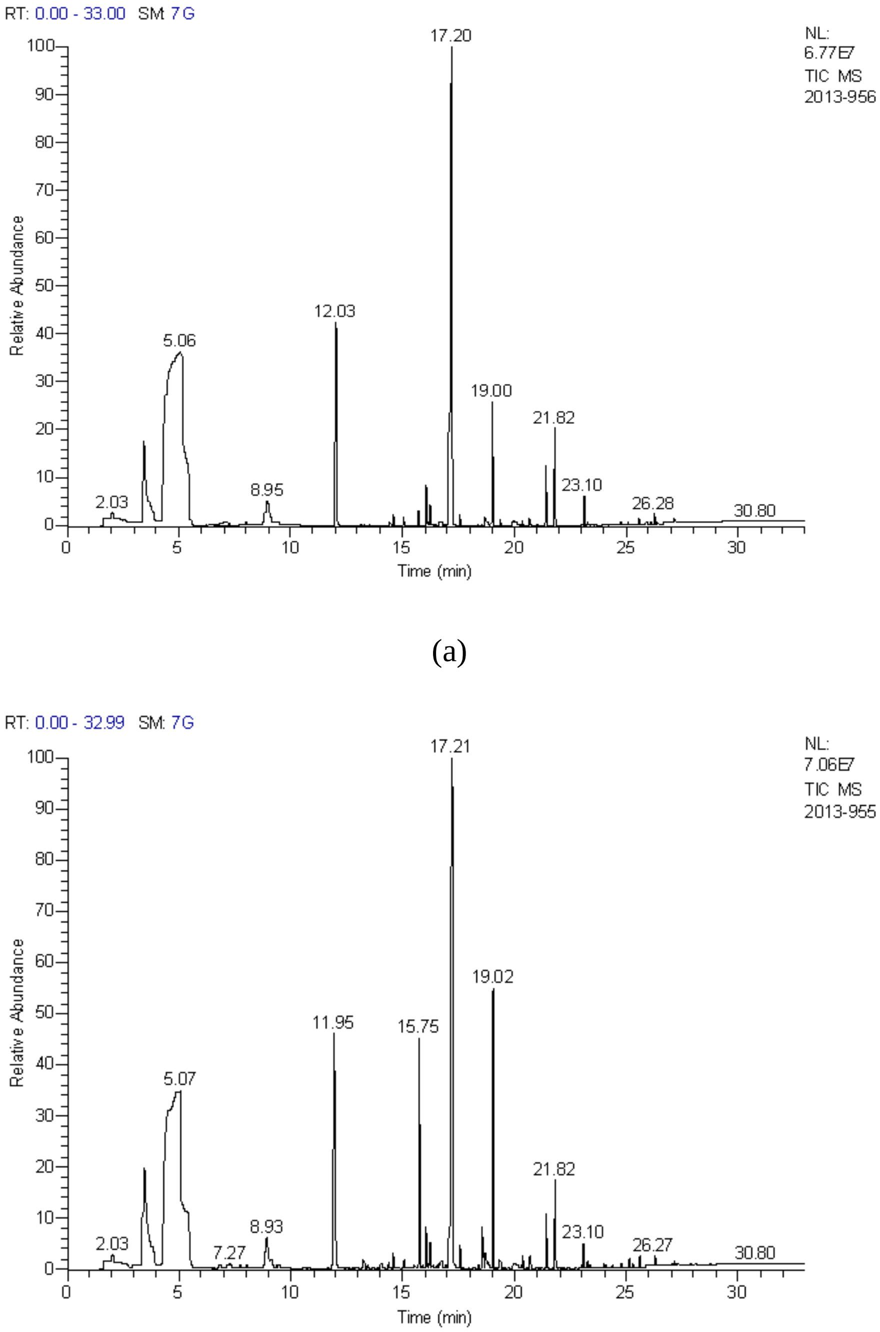
16.17	n-heptanol	0.05	0.05
17.56	1-Octanol	0.15	0.30
19.36	alpha-Terpineol	0.07	0.12
21.41	Benzyl alcohol	0.89	0.71
21.82	Phenethyl alcohol	1.62	1.31
Aldehydes&Ketones		20.39	20.38
8.00	Hexanal	0.07	0.07
13.26	Octanal	0.03	0.11
15.09	Nonanal	0.16	0.14
16.25	Furfural	0.32	0.35
16.67	Decanal	0.06	-
16.75	trans ,trans-2, 4- Heptadienal	0.08	0.16
17.21	Benzaldehyde	19.41	19.21

13.52	3-Hydroxy-2-butanone	0.02	-
18.82	Acetophenone	0.10	0.19
20.68	2-Hydroxyacetophenone	0.14	0.15
Others		0.92	0.96
16.03	Acetic acid	0.71	0.64
21.01	Caproic acid	0.02	-
23.02	4-ethyl-2-methoxyphenol	0.02	-
23.24	Octanoic acid	0.06	0.10
25.29	Decanoic acid	-	0.05
25.59	2,4-Di-tert-butylphenol	0.08	0.12
25.89	tert-Butylhydroquinone	0.03	0.05

Table 7 Sensory evaluation scores of steeped greengage wine after 28 kHz and 45 kHz ultrasonic treatment

Frequency (kHz)	Power (W)	Treating time (min)					
		0	10	20	30	40	50
28	240	[a]76.93±7.56a	79.58±5.82ab	80.95±3.85ab	81.20±4.54b	81.45±4.57b	82.10±3.64b
	300	76.93±7.56a	80.73±3.96ab	81.55±3.89b	82.45±3.88b	83.03±3.59b	82.95±3.34b
	360	76.93±7.56a	81.28±3.87b	81.90±3.59b	83.05±3.52b	83.40±2.77b	81.95±3.49b
45	240	76.93±7.56a	80.48±5.06ab	81.45±4.66ab	82.50±3.68b	83.05±3.42b	82.18±3.28b
	300	76.93±7.56a	80.90±4.57ab	82.23±3.75b	83.28±2.99b	81.88±4.23b	80.93±4.78ab
	360	76.93±7.56a	81.48±4.06b	83.35±2.82bc	84.40±2.85c	81.00±4.84b	79.15±5.86ab

[a] Different letters in the same row indicate significant different ($p \leq 0.05$).



(a)

(b)

Fig. 1 Total ion chromatogram of aroma components in steeped greengage wines analysed by GC-MS: (a) untreated, and (b) 45 kHz 360 w ultrasonic wave treatment for 30 min. Peak identification refers to Table 6.