INFLUENCE OF PRE-TREATMENT ON YIELD AND QUALITY OF MANDARIN JUICES

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ABSTRACT: Croatia in the Neretva valley produces remarkable quantities of mandarins but, as always, one part of yield, according to fruit characteristics, are not suitable for consumption as fresh. At the same time those fruit are suitable for processing industry e.g. for juice production. In Croatia, mandarin juice is not industrially produced, and generally is rare on market. The current study is contribution to mandarin juice processing with the aim to investigate influence of different pre-treatment on yield, quality, biological and sensorial attributes of obtained juices.
Mandarin from Neretva valley was used to produce cloudy juices on small scale equipment. Before pressing, fruits were peeled and treated (i) with maceration enzymes at various temperature and duration (EPT) (ii) by ultrasound (USPT) with different amplitude and duration. Control juices were produced without enzymes or USPT. In juices biologically active compounds (BAS) total carotenoids (TC), total phenolics (TP), and vitamin C were determined. All juices were sensory evaluated (SE) by quantitative descriptive method. The results were statistically analyzed. Considering the investigated parameters there are no remarkable differences among juices produced by enzyme treatments independent of enzyme concentration, temperature (20 or 50 ^{o}C) and duration of 30 or 60 min. regarding to BAS and SE. Also, different US treatment conditions did not show any remarkable influence on investigated parameters. But in comparison of these two types of treatment better yield and SE were obtained by US and higher content of TC was obtained by enzyme treatment. Treatment by US could be recommended to produce mandarin juice due to high yield and SE but also further investigations are needed to optimize US conditions due to better preservation of BAS.
Key words: *mandarin juice, ultrasound, enzyme maceration, biologically active compounds*

INTRODUCTION

Croatia in the Neretva valley produces remarkable quantities of mandarins but, as usual, one part of yield, are not suitable for consumption as fresh. At the same time those fruit are suitable for processing industry e.g. for juice production. In Croatia, mandarin juice is not industrially manufactured, and generally is rare on the market. Looking on juices from citrus fruit, on Croatian market, as well as on world market, orange juice is predominant. In comparison to orange juice mandarin juice has higher intensity of orange colour and may be added to orange juice to improve its colour (Perez-Lopez et al., 2006). Citrus juice production usually is based on extraction from unpeeled fruit and peel constituents which may pass into the juice later in technological process it should be removed (Bates et al., 2012). Since, mandarin is easy-to-peel fruit, mandarin juice could be produced with previous peeling (Tietel et al., 2010) what is convenient for SMEs (Bates et al., 2012) because peel constituents are not present in fresh juice after extraction which technologically simplifies finalization of juice. Yield of juices obtained by pressing is in dependence on pectin structure and content therefore pectolitic enzymes usually are used. To obtain higher yield certain enzyme treatment before pressing is desirable for many fruit (Chang et al., 1994, Levaj et al, 2010, Levaj et al., 2011). Recently in juice production, ultrasound technology has been investigated mostly as an substitution to common thermal treatment e.g. pasteurization in order to inactivate enzymes and microorganisms at mild temperature conditions (Tiwari et al., 2008; Dubrović et al., 2011; Rawson et al., 2011; Chandrapala et al., 2012; Fonteles et al., 2012) but in laboratory ultrasound is well-established as method for cell disintegration to extract intracellular components (Skauen, 1976, Valero et al., 2007). High-intensity cavitation punctures the cell walls and releases the cell content. Mandarin as other citrus fruit contain high valuable nutrients such as vitamin C, folate, dietary fiber, minerals (potassium) and biological active comounds (BACs), the terpenes and phenolic compounds, which synergistic action with vitamin C contribute to citrus antioxidant capacity (Codoner-Franch and Valls-Belles, 2010; Unno et al., 2011). Mandarin is rich in hesperidin, flavanon specific for citrus fruit, and with carotenoid, β-chryptoxanthin, which possess anti-inflammatory and anti-carcinogenic activity (Codoner-Franch and Valls-Belles, 2010; Kohno et al., 2001). Flavonoids also have a role in cardiovascular protection, (Codoner-Franch and Valls-Belles, 2010). Consquently, better releasing of hesperidin and β- chriptoxanthin, generally speaking, phenolics and carotenoids, in the processing of juice are desirable in order to better quality as well as health reasons. The current study is contribution to mandarin juice processing with the aim to investigate influence of ultrasound in comparison to thermal and enzyme treatment on yield, quality, biological and sensorial attributes of obtained juices.

MATERIAL AND METHODS

Mandarin fruit (*Citrus unshiu* Markovich), were harvested, twice with an interval of two weeks, in the Neretva Valley in 2010 and transported at 4 ℃ to Faculty of Food Technology and Biotechnology in Zagreb, hand peeled and frozen at -18 ℃ till juice manufacturing.

Juice manufacturing

After defrosting fruit was chopped by knife or homogenized in blender (Mixy Zepter, International) and was used to produce cloudy juices on small scale equipment according to scheme (Figure 1).

Ultrasonic pre-treatment (USPT)

Homogenized fruit (round 400 mL) was placed in a glass beaker (1000 mL), which served as the treatment chamber. An ultrasonic processor (S-4000, Misonix Sonicators, Newtown, CT, USA), set at 600 W, 20 kHz and 12–260 µm with a 19-mm diameter probe, was introduced into the vessel. Ultrasonication was carried out with 60 and 120 µm amplitude. Samples were treated ultrasonically for 5 and 10 min at 25 ℃ (table 1). Each experiment was conducted at least in duplicate.

Thermal (TPT) and enzyme pre- treatment (EPT)

Chopped fruit for was treated at 20 or 50 ^{o}C for 30 minutes (TPT) and for EPT with 50 or 150 ppm maceration enzymes (Endozym Pectofruit PR, AEB group, Italy) at 20 or 50 ^{o}C for 30 or 60 minutes (table 2). These concentrations and temperatures were selected according to the manufacturer's instructions.

Pressing was done on hydraulic pack press (Euclid Ltd., Croatia). Juices were kept in refrigeration at 8^{o}C until analysis. Juices were produced separately with the mandarin from the first harvest and with the mandarin from the second harvest. Each juice was separately analyzed.

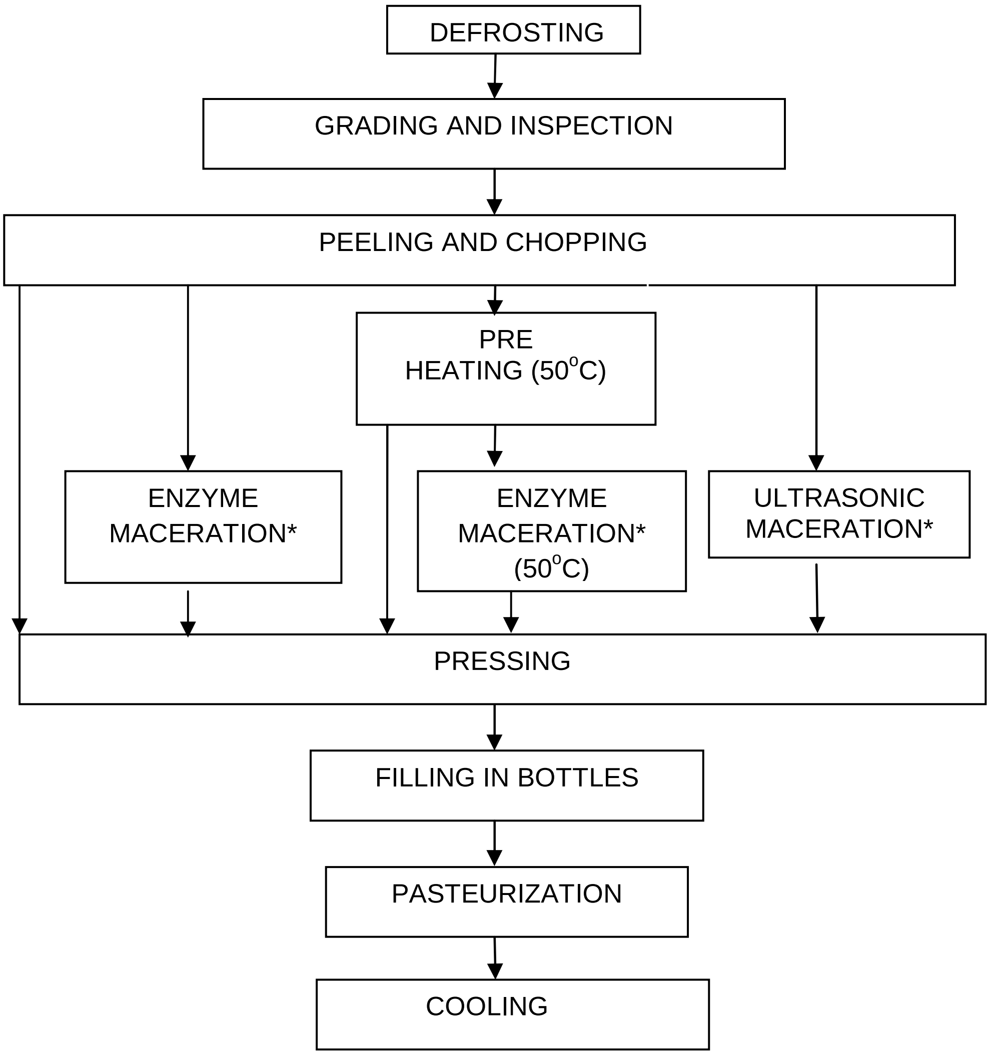
Table 1. Conditions of USPT and the code of produced juices

Code of sample	Amplitude/µm	Duration (min)
US/60/5	60	5
US/60/10	60	10
US/120/5	120	5
US/120/10	120	10

Table 2. Conditions of TPT and EPT and the code of produced juices

Code of sample	T (℃)	Enzyme (ppm)	Duration (min)
20/0/30	20	-	30
20/50/30	20	50	30
20/150/30	20	150	30
20/50/60	20	50	60
20/150/60	20	150	60
50/0/30	50	-	30
50/50/30	50	50	30
50/150/30	50	150	30

After pressing juice was filled in glass bottles of 200 mL and closed. Pasteurization of juices was done after filling at 85 ^{o}C/15 min. Cooling was done by immersing in cold water.
Yields were calculated on fruit pulp, after peeling.



*-see table 1 and 2

Figure 1. Scheme of juice manufacturing

Biologically active compounds determination and sensory evaluation

Total phenolics (TP) were extracted according to Coseteng and Lee (1987) and determined by spectrophotometer using Folin-Ciocalteu reagent (Ough and Amerine, 1988) based on gallic acid calibration so results are presented as equivalent of gallic acid (GAE).

Carotenoids were extracted by petroleum ether (V_u=total volume of extract (mL)), absorbance (λ=450 nm) ($A_{\lambda max}$) was measured by spectrophotometer and concentration was calculated by using extinction coefficient $E^{\%}_{1\ cm}$ (extinction coefficient in petroleum ether for β-carotene=2500) and formula (Vuelleumier, 1967):

$$\text{Total carotenoids (as β-carotene) (mg/100 g)} = \frac{A_{\lambda max} \cdot 1000 \cdot V_u}{m(g) \cdot E^{\%}_{1\ cm}}$$

Vitamin C were determined by titration with 2,6-diclorphenolindophenol (AOAC, 2002).

All analyses were done in duplicates.

Obtained juices were conducted to sensory evaluation by Quantitative descriptive analysis which was very comprehensive. The panellists were requested to list the terms appropriate to describe the colour, odour, taste, consistency and overall sensory impression whereas a total of 10 descriptive terms for all major sensory attribute categories were generated. The panellists scored the samples using a suitable line intensity scale, with scores awarded on a scale of 0 – 10 in which 10 indicated the highest intensity of evaluated sensory attribute. Sensory analysis was carried out by a trained panel consisting of fifteen members per session. The procedure was performed according to ISO 6564, ISO 8587 and ISO 11036 (in a sensory laboratory equipped according to ISO 8589) and in consistence with method from literature (Bursać et al., 2007; Bursać Kovačević et al., 2008).

Statistical data analysis

Statistical analysis was performed via analysis of variance (ANOVA) by Kruskal-Wallis test using SPSS (ver. 17) in order to investigate influence of pre-treatment (temperature, enzyme – temperature, and ultrasonic) on each determined parameter. Differences were considered significant at $p \leq 0.05$.

RESULTS AND DISCUSSION

All treatments were grouped as follow: (i) thermal pre-treatment (TPT) in order to investigate influence of temperature, (ii) enzyme pre-treatment (EPT) in order to investigate influence of enzyme concentration, temperature and duration of maceration, and (iii) ultrasonic pre-treatment (USPT) in order to investigate amplitude and duration of ultrasonic treatment.

Within each treatment there was not significant influence of temperature or enzyme concentration, temperature and duration of maceration or amplitude and duration of ultrasonic treatment on juice yield. However, USPT ranked highest yield followed by EPT and TPR (χ^2=11.60, p<0.01). Average values of juice yield are shown in table 3. Lieu and Le (2010) investigated US in grape mash treatment and also concluded that sonication treatment increased extraction yield.

Table 3. Average values of each treatment for yield, TP, TC and vitamin C

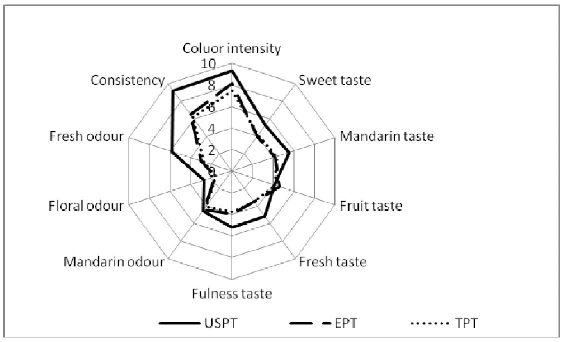
Treatment	Yield (%)	TP (mg/100 g)	TC (mg/100g)	Vitamin C (mg/100 mL)
TPT	65.27±6.09	76.45 ±26.98	0.75±0.26	17.18±4.16
EPT	75.55±6.75	73.36±37.55	0.75±0.11	17.81±2.11
USPT	83.16±5.97	71.07±11.38	0.44± 0.11	19.66±2.83

Influence of all treatments on BACs (TC, TP and vitamin C) was investigated. Content of TP were a little lower than previously determined in fresh mandarin (114.09 mg/100g) (Levaj et al., 2008) which may be due to a difference in the mandarin cultivars and the processing. According to average values for each treatment (table 3) the highest TP content was obtained by TPT and retention in USPT was 93 %. With prolonged time and higher amplitude loss of BACs was higher although the differences were not significant (table 4). Rawson et al. (2011) had similar observation for watermelon. TP content was not much affected at lower processing times of 0 to 6 min in comparison to higher processing times of 10 min where significant degradation of TP content was observed. Fonteles et al. (2012) also conclude that ultrasound caused reduction of TP content. In our study significant influence of pre-treatment was observed only for TC content (χ^2= 14.14, p=0.01). The average value for USPT was 40 % lower when compared to the highest average value of TC content determined for EPT (table 3). Although between all treatments there was no significant influence on vitamin C content, its average value of USPTs slight increased in comparison to other two treatments (table 4). Rawson et al. (2011) reported about degradation of vitamin C as processing time was increased from 0 to 10 min for an amplitude level of 24.1 μm at 25 ℃. In our study at lower amplitude was achieved higher vitamin C content which decreased with the higher amplitude but the differences were not significant (table 4).

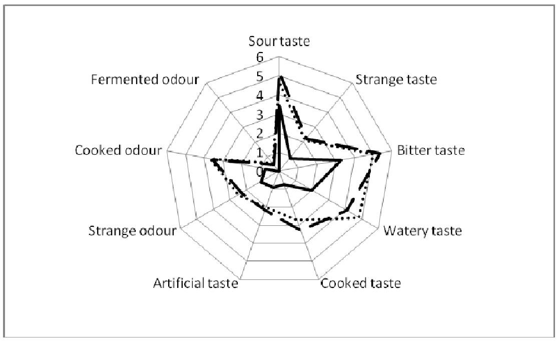
Table 4. Average values of each USPT for TP, TC and vitamin C

USPT	TP (mg/100 g)	TC (mg/100g)	Vitamin C (mg/100 mL)
US/60/5	76.66±4.72	0.42±0.01	22.26±0.49
US/60/10	72.78±18.07	0.45±0.19	21.92±0.98
US/120/5	75.37±1.84	0.44±0.16	16.70±2.45
US/120/10	59.45±13.36	0.47±0.13	17.74±0.0

Additionally, within each treatment there was no significant influence of temperature or enzyme concentration, temperature and duration of maceration or amplitude and duration of ultrasonic treatment on TC, TP or vitamin C content.



(a)



(b)

Figure 2. Results of sensory evaluation: (a) desirable attributes; (b) undesirable attributes

All sensory attributes are divided in two main evaluation blocs: first one include desirable attributes (see Fig. 1a) and the second one undesirable attributes (see Fig. 2b). Total sum of scores for desirable attributes and total sum of scores for undesirable attributes were calculated and mathematically adjusted that both blocks had the same share in total sensory score regardless of number of grades in blocs and than were subtracted. For desirable attributes (χ^2= 37.84, p<0.01) as for TSG (χ^2=54.53, p<0.01) the best ranking treatment was USPT, while TPT has the largest rank for undesirable attributes (χ^2=53.78, p<0.01).

CONCLUSIONS

Ultrasound is promising method in juice processing as a pre-treatment method to ensure alternative of thermal or enzymatic method according to results of sensory evaluation and yield. Additionally, better yield in this study was obtained in much shorter time when compared to conventional methods. Further investigations are needed to improve content of BAC in juices.

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