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The effects of the fermentation with immobilized yeast and different cherry varieties on the quality of cherry brandy

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Summary

The aim of this research was to investigate influence of different fermentation processes (by immobilized yeast cells and classical fermentation) and different cherry varieties (*Maraska*, *Montmorencys* and *Kelleris*) on the chemical and sensorial characteristics of cherry brandies. Cherry brandies were analyzed to determine chemical composition, aroma profile and sensory properties. Cherry brandies produced by immobilized yeast cells had a higher content of aldehydes, but lower content of total acids, total extract, higher alcohols and esters compared to the samples produced by classical fermentation process. Furfural was not detected in cherry distillates produced by immobilized yeast cells, while distillates produced by classical fermentation process had very low content. Cherry brandies produced by classical fermentation process had significantly higher content of benzaldehyde which has great influence on aroma of cherry brandies. Ethyl octanoate which is considered one of the most important contributors to the aroma of alcoholic distillates was found in the highest concentrations in *Maraska* distillates. The best evaluated sample was brandy produced from *Maraska* cherry variety fermented by immobilized yeast cells followed by brandy produced also from *Maraska* cherry variety, but by classical fermentation process.

Keywords: cherry brandy, fermentation, quality

Introduction

Cherries are the smallest members of the stone fruit family: Rosaceae, genus: *Prunus*. They comprise over a hundred species, classified in pomological terms into two distinct groups: corymb or racemosa. Among the most important species *Prunus avium* L., known as "sweet cherry", and *Prunus cerasus* L., known as "sour cherry" are the most important. The sour cherry varieties are divided in three main groups depending on tree habit and fruit characteristics: Kentish cherries, morellos and marasca (Ferretti et al., 2010). The main characteristics related to cherry fruit quality are sweetness, sourness, firmness and rich source of anthocyanins (pigments that are responsible for red colour of fruit) (Pedisić et al., 2009; Ferretti et al., 2010). Cherries are mainly consumed as fresh fruit. Food industry process them to different products: dried (with or without sugar), frozen or canned cherries, as juice or concentrate, and alcoholic beverages (Chaovanalikit et al., 2004; Ferretti et al., 2010).

Alcoholic beverages are complex mixtures mainly comprised of ethanol, water and a large number of minor compounds that may be present in the raw materials or formed during the distinct stages of the manufacturing process such as: alcohols, acids, esters, aldehydes, polyphenols, metals, aminoacids, etc. (Rodríguez Madrera et al., 2006). Quality of alcoholic

beverages is influenced by many factors during the production process (raw material, fermentation, distillation, etc.). Distilled spirits from cherry (Kirschwasser, and Cherry brandy) are produced in many regions of the world. Kirschwasser is mainly produced in Germany, France and Switzerland and has full fruit aroma and smooth finish. Maraska Kirschwasser is made by distilling the famous Maraska cherry which is grown only on the Adriatic coast. Alcoholic beverages produced from sour cherry fruits contain many volatile aroma compounds, including benzaldehyde, linalool, hexanal, phenylacetaldehyde and eugenol, contribute to the typical fruity flavour and aroma (Willemsens and Boelsens 1996).

Immobilized yeast technology has attracted continual attention in the fermentation processes over the last years because of faster fermentation, possibility of continuous operation, and increased volumetric productivity. Production with immobilized yeast cells indicate that different reactor and system designs have great influence on quality of final product (Virkejari and Kronolof, 1998; Miličević et al., 2012).

The aim of this research was to investigate influence of the fermentation process with immobilized yeast cells on the chemical and sensorial characteristics of cherry brandies produced from different cherry varieties (*Maraska*, *Montmorencys* and *Kelleris*).

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Materials and methods

Preparation of cherry brandies

Preparations of cherry brandies were carried out in Zvečevo d.d. food industry Požega.

The cherry brandies were produced from three cherry varieties: *Maraska* (MAR-CL and MAR-IYC), *Montmorencys* (MON-CL and MON-IYC) and *Kelleris* (KEL-CL and KEL-IYC).

Samples MAR-CL, MON-CL and KEL-CL were fermented by classical fermentation process: fermentation with selected yeast *Feromol-Bouquet 125* and controlled temperature during fermentation of 18-20 °C. The duration of fermentation was 21 days. Samples MAR-IYC, MON-IYC and KEL-IYC were fermented by yeast cells *Feromol-Bouquet 125* immobilized in Ca-alginate gel. Fermentation was carried out in *internal loop gas-lift fermentor* (Fig. 1) and controlled temperature during fermentation of 18-20 °C. The duration of fermentation was 3 days.

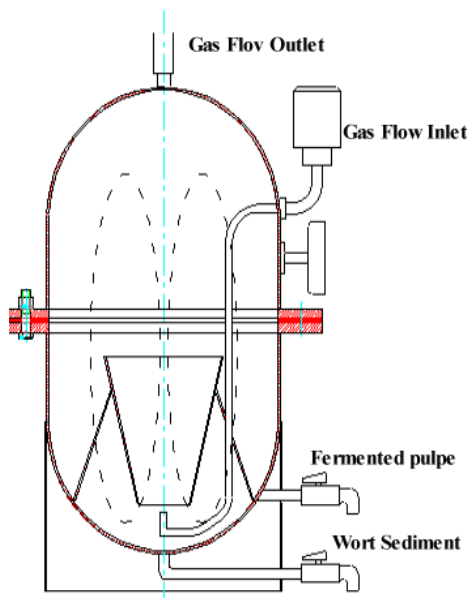


Fig. 1. Reactor for fermentation with immobilized yeast cells

Samples were distilled in industrial copper clip distillation device as shown in the scheme (Fig. 2). The samples containing approximately 70% vol alcohol were taken from the middle fraction, or with recommended alcohol concentration in distillates, while the first *head* and the last *tail* fraction were not used. All selected samples were distilled according to the same distillation protocol.

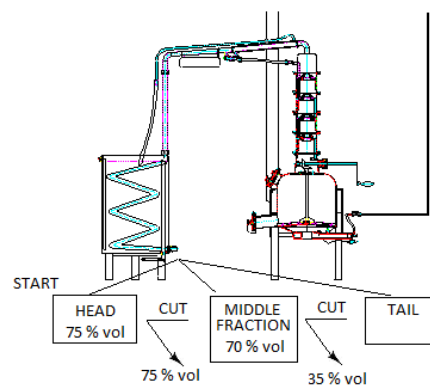


Fig. 2. Scheme of equipment used for distillation of cherry brandies

Chemical analysis

Chemical analysis of cherry brandies (ethanol, extract, SO₂, aldehydes, higher alcohols, total acids, esters, methanol and furfural) was conducted according to standard AOAC (1995) procedures.

Analyses of aroma substances

The major volatile components were analyzed according to European Community Reference Methods for the analysis of spirits using gas chromatography (EEC 2000, EEC 2008).

Gas chromatography analyses were performed by gas chromatograph (Chrompack 437A, USA) equipped by split/splitless injector and an FID detector. For analysis of distillates a Chrompack Poraplot capillary column (25 m x 0.25 µm i.d. 0.25 µm) was used. Initial oven temperature was kept at 35 °C for 7 min, then raised at 10 °C/min to 80 °C followed by 25 °C/min to 180 °C, and kept for 4 min at 180 °C. Qualitative analysis was done by comparing the standard retention times (analytical grade from Merck KGaA Darmstadt, Germany) with the corresponding peaks of samples. The quantification was carried out by comparing the peak areas to those of the Merck standards. All analyses were run in triplicates. Experimental data were analysed by analysis of variance (ANOVA).

Sensory analysis

A sensory analysis of samples was performed according to the method of positive scoring factor according to the German DLG model (Koch, 1986). This model was based on 4 sensorial experiences: colour, clearness, odour and taste, which are marked with grades 0 to 5, including 0, while the average grade is multiplied by the significance factor. Sensory assessment was conducted in two repetition cycles by ten sensory testing experts.

Results and discussion

Chemical composition of cherry brandies is presented in Table 1. Ethanol content of cherry distillates was in range from 68.01% (MAR-CL) to 69.98% (MON-CL) and total SO₂ from 3.85 (KEL-CL) to 9.20 mg/L (MON-IYC). Ethanol content is very important for the mouth-feel and flavour of alcoholic beverages, as such lower content of ethanol may cause the reduction of some aroma substances in distillates (Nykanen and Suomalainen, 1983; Conner, 1998). SO₂ was added during fermentation to protect the pulp from non-controlled fermentation process, such as browning and oxidation (Nikičević and Tešević, 2010; Miličević^b et al., 2012).

Distillates fermented by classical fermentation process had higher content of total extract where MAR-CL had the highest value of 0.064 g/L, followed by MON-CL (0.034 g/L), KEL-CL (0.024 g/L), MAR-IYC (0.018 g/L), KEL-IYC (0.017 g/L), and MON-IYC (0.011 g/L).

Cherry distillates produced by immobilized yeast cells had a higher content of aldehydes (265.40-290.00 mg/L; classical fermentation=105.00-180.00 mg/L), but lower content of total acids (247.00-353.85 mg/L; classical fermentation=447.60-485.00 mg/L) and esters (1013.46-1368.78 mg/L; classical fermentation=1528.23-2378.23 mg/L) compared to the samples produced by classical fermentation process. Total acids and esters are responsible for a so-called sweet floral-basic aroma of alcoholic beverages (Miličević et al., 2013). Esters are mostly formed from esterification of fatty acids with

alcohols during the fermentation and ageing process and can be influenced by many factors, such as fermentation temperature, oxygen availability and fermentation strains (Nikičević et al., 2011).

Cherry brandies produced by classical fermentation process had significantly higher content of higher alcohols. MAR-CL had the highest content of higher alcohols (1562.14 mg/L), followed by MON-CL (1330.71 mg/L), KEL-CL (845.27 mg/L), MAR-IYC (595.00 mg/L), MON-IYC (577.20 mg/L) and KEL-IYC (267.20 mg/L). Content of higher alcohols in the distillates depends on several factors such as raw material, yeast strains and fermentation conditions (Rodriguez Madrera et al., 2006). Aldehydes and higher alcohols may cause formation of acetal during maturation of distillates and development of pleasant aroma, without sharp alcoholic odour tones (Miličević^b et al., 2012).

Methanol is not a yeast fermentation product, and its concentration depends on raw material composition and ripening state of fruits (Suarez Valles et al., 2005). The results show that in all cherry distillates methanol was below the maximum permitted content (12 g/L) established by EU *Regulation*, 2008.

Furfural was not detected in cherry brandies produced by immobilized yeast cells, while distillates produced by classical fermentation process had very low furfural content (0.001-0.006 mg/L). Furfural formation is basically due to the presence of pentose residue in the mash as well as to a amadori decomposition product via 1,2-enolisation (Versini et al., 2009).

Table 1. Chemical composition of cherry brandies

| | MAR-CL | MON-CL | KEL-CL | MAR-IYC | MON-IYC | KEL-IYC |
|------------------------------|---------|---------|---------|---------|---------|---------|
| Ethanol (% vol.) | 68.01 | 68.98 | 69.27 | 69.84 | 69.60 | 69.01 |
| Total extract (g/L) | 0.064 | 0.034 | 0.024 | 0.018 | 0.011 | 0.017 |
| Total SO ₂ (mg/L) | 5.54 | 6.85 | 3.85 | 6.04 | 9.20 | 5.24 |
| Total acidity (mg/L) | 447.60 | 485.00 | 484.20 | 247.00 | 351.20 | 353.85 |
| Aldehydes (mg/L a.a.) | 180.00 | 105.00 | 174.00 | 290.00 | 283.00 | 265.40 |
| Esters (mg/L a.a.) | 2378.23 | 1528.23 | 1797.80 | 1033.10 | 1013.46 | 1368.78 |
| Higher alc. (mg/L a.a.) | 1562.14 | 1330.71 | 845.27 | 595.00 | 577.20 | 267.20 |
| Furfural (mg/L a.a.) | 0.006 | 0.001 | 0.002 | n.d. | n.d. | n.d. |
| Methanol (g/L a.a.) | 0.12 | 0.04 | 0.04 | 0.08 | n.d. | n.d. |

n.d. - not detected

MAR - *Maraska* variety; MON - *Montmorencys* variety; KEL - *Kelleris* variety

CL - samples fermented by classical fermentation process; IYC - samples fermented by immobilized yeast cells

Aroma compounds, their levels, odour attributes and thresholds are most important for quality and authenticity of distilled spirits and liqueurs (Nikičević et al., 2011). Several hundred compounds, such as alcohols, esters, aldehydes, ketones, volatile acids, terpenes contribute to the complex aroma profile of alcoholic beverages. Table 2 shows the average contents of the important aroma substances of cherry brandies.

The aroma of cherry distillates is significantly affected by the benzaldehyde, which originates from the enzymatic

degradation of amygdalin in the stones of the fruits, passing into the mash during fermentation and later into the distillate (Nikičević et al., 2011). Cherry distillates produced by classical fermentation process had significantly higher content of benzaldehyde. MAR-CL had the highest content (2.27 mg/L), followed by MON-CL (1.69 mg/L), KEL-CL and MAR-IYC (1.58 mg/L), MON-IYC (1.48 mg/L) and KEL-IYC (1.38 mg/L).

The most significant acetate esters present in cherry distillates are isoamyl acetate and 2-phenylethyl

acetate (Nikičević et al., 2011). They are mainly responsible for the floral and fruity aroma of the distillates (Schmid and Grosch, 1986; Tešević et al., 2005). Among cherry varieties, *Maraska* distillates had the highest, while *Kelleris* distillates had the lowest content of isoamyl acetate and 2-phenylethyl acetate. Isoamyl acetate content followed the order: MAR-CL (9.6 mg/L) > MON-CL (9.40 mg/L) > MAR-IYC (9.02 mg/L) > MON-IYC (8.35 mg/L) > KEL-CL (7.50 mg/L) > KEL-IYC (7.34 mg/L). Cherry distillates produced from *Montmorencys* and *Kelleris* by immobilized yeast cells had very low content of 2-phenylethyl acetate (in trace), while MAR-CL had 0.81 mg/L, MON-CL 0.61 mg/L, MAR-IYC 0.6 mg/L and KEL-CL 0.3 mg/L.

Results showed low concentration of terpenes (linalool, limonene, α -terpineol and nerolidol), where distilled samples produced by immobilized yeast cells had higher content of mentioned substances. Despite the low concentration of these compounds, their presence is relevant because they harmonically synergize to produce the characteristic cherry aroma (Nikičević et al., 2011).

Distillation enhanced the concentration of ethyl esters and many of the higher alcohols (Schreier et al., 1978). Ethyl acetate, formed by esterification of acetic acid with ethanol, was in range between 7.49 mg/L (MAR-IYC) to 4.20 mg/L (MON-CL). Other esters were in lower concentration: acetaldehyde 3.15-5.15 mg/L;

ethyl octanoate 2.56-5.92 mg/L; ethyl hexanoate 1.62-2.96 mg/L; ethylundecanoate 1.10-2.15 mg/L; ethyl dodecanoate 0.90-1.80 mg/L; ethyl decanoate 0-1.74 mg/L and ethyl lactate 0-0.41 mg/L. The ethyl esters of fatty acids from hexanoate to ethyl decanoate related to a ripe fruits aroma and were at medium to low level which is in accordance with Versini et al. (2009). Ethyl octanoate which is considered one of the most important contributors to the aroma of alcoholic distillates (Nikičević et al., 2011) was found in the highest concentrations in *Maraska* distillates, where distilled samples produced by immobilized yeast cells had lower content of mentioned substance.

Cherry distillates produced by classical fermentation process had significantly higher content of isoamyl alcohol and isobutyl alcohol. Isoamyl alcohol content followed the order: MON-CL (9.90 mg/L) > KEL-CL (8.80 mg/L) > MAR-CL (8.79 mg/L) > MON-IYC (6.39 mg/L) > KEL-IYC (6.16 mg/L) > MAR-IYC (5.39 mg/L); and isobutyl alcohol: KEL-CL (5.37 mg/L) > MON-CL (3.90 mg/L) > MAR-CL (3.62 mg/L) > MON-IYC (3.60 mg/L) > MAR-IYC (3.42 mg/L) > KEL-IYC (3.30 mg/L).

The levels of 2-phenylethanol detected in cherry brandies were in range 2.75 (MAR-IYC) - 4.52 mg/L (MON-CL). 2-phenylethanol was of fermentative origin and contributes to the aroma of alcoholic beverages with floral or rose notes (Rodriguez Madrera et al., 2006; Versini et al., 2009).

Table 2. Average contents (mg/L) of the major volatile components of cherry brandies

| | MAR-CL | MON-CL | KEL-CL | MAR-IYC | MON-IYC | KEL-IYC |
|-----------------------|-----------|-----------|-----------|-----------|------------|-----------|
| Benzaldehyde | 2.27±0.03 | 1.69±0.02 | 1.58±0.03 | 1.58±0.02 | 1.48±0.06 | 1.38±0.03 |
| Isoamyl acetate | 9.60±0.01 | 9.40±0.02 | 7.50±0.1 | 9.02±0.02 | 8.35±0.01 | 7.34±0.03 |
| 2-Phenylethyl acetate | 0.81±0.03 | 0.61±0.02 | 0.30±0.03 | 0.60±0.01 | tr. | tr. |
| α -Terpineol | 2.47±0.02 | 1.57±0.01 | n.d. | 2.84±0.01 | 2.14±0.01 | 1.84±0.01 |
| Linalool | 0.92±0.02 | 0.62±0.03 | 0.32±0.03 | 0.98±0.03 | 0.65±0.03 | 0.42±0.02 |
| Limonene | 0.16±0.03 | tr. | tr. | 0.19±0.02 | 0.11±0.01 | 0.14±0.02 |
| Nerolidol | 0.06±0.02 | tr. | tr. | 0.08±0.02 | 0.07±0.02 | 0.06±0.02 |
| Ethyl acetate | 6.57±0.01 | 4.20±0.01 | 4.31±0.01 | 7.49±0.02 | 4.52±0.01 | 4.36±0.02 |
| Acetaldehyde | 5.15±0.02 | 3.76±0.03 | 4.11±0.01 | 4.58±0.01 | 3.15±0.02 | 3.35±0.02 |
| Ethyl octanoate | 5.92±0.01 | 2.69±0.01 | 2.92±0.01 | 4.18±0.01 | 2.62±0.01 | 2.56±0.01 |
| Ethyl hexanoate | 2.96±0.02 | 2.11±0.02 | 1.92±0.03 | 2.66±0.02 | 2.01±0.02 | 1.62±0.04 |
| Ethyl dodecanoate | 1.80±0.01 | 1.62±0.02 | 1.11±0.01 | 1.20±0.02 | 1.01±0.02 | 0.90±0.02 |
| Ethylundecanoate | 2.15±0.02 | 2.10±0.02 | 1.55±0.03 | 1.95±0.02 | 1.15±0.02 | 1.10±0.03 |
| Ethyl decanoate | 1.74±0.02 | 0.91±0.02 | 1.18±0.01 | n.d. | n.d. | n.d. |
| Ethyl lactate | 0.07±0.02 | 0.41±0.02 | n.d. | n.d. | n.d. | n.d. |
| Isoamyl alcohol | 8.79±0.01 | 9.90±0.01 | 8.80±0.01 | 5.39±0.03 | 6.39±0.03 | 6.16±0.03 |
| Isobutyl alcohol | 3.62±0.01 | 3.90±0.03 | 5.37±0.01 | 3.42±0.02 | 3.60±0.032 | 3.30±0.03 |
| 2-phenylethanol | 2.97±0.01 | 4.52±0.01 | 3.45±0.01 | 2.75±0.01 | 3.45±0.01 | 3.55±0.02 |
| 1-propanol | 2.75±0.01 | 4.70±0.03 | 1.20±0.01 | 1.00±0.01 | 3.70±0.03 | 1.72±0.03 |
| Undecanoic acid | 2.96±0.02 | 2.83±0.02 | 2.62±0.02 | 2.93±0.02 | 2.88±0.02 | 2.60±0.02 |
| Dodecanoic acid | 0.28±0.02 | 0.20±0.02 | 0.23±0.02 | 0.25±0.02 | 0.16±0.01 | 0.14±0.01 |

n.d. - not detected; tr. - traces

MAR - *Maraska* variety; MON - *Montmorencys* variety; KEL - *Kelleris* variety

CL - samples fermented by classical fermentation process; IYC - samples fermented by immobilized yeast cells

Cherry brandies produced by classical fermentation process had 1-propanol content in range 1.20 (KEL-CL)-4.70 mg/L (MON-CL), while 1.00 (KEL-IYC)-3.70 mg/L (MON-IYC) was content range for distillates produced by immobilized yeast cells. 1-propanol is significant aroma substance of apricot, plum, cherry and other fruit brandies. Bandion (1972) reported a 1-propanol content of 3800

mg/100 mL of pure alcohol for a cherry brandy. Free fatty acids are usual components of alcoholic beverages which are produced by yeast metabolism of carbohydrates and have a small effect on the flavour of distillates (Tešević et al., 2009). Undecanoic acid (ranging between 2.6-2.96 mg/L) and dodecanoic acid (ranging between 0.14-0.28 mg/L) were identified in the analyzed samples.

Table 3. Results of sensory analyses of cherry brandies (*German DLG model*)

| Sample | Assessment characteristics | | | | TOTAL (max 100 points) |
|---------|----------------------------|------------------------------|--------------------------|--------------------------|---------------------------|
| | Color (max 15 points) | Clearness (max 15 points) | Odour (max 25 points) | Taste (max 45 points) | |
| MAR-CL | 15.00 | 15.00 | 23.50 | 45.00 | 98.50 |
| MON-CL | 14.90 | 14.90 | 22.50 | 44.10 | 96.40 |
| KEL-CL | 15.00 | 14.90 | 23.50 | 44.10 | 97.50 |
| MAR-IYC | 15.00 | 15.00 | 25.00 | 44.10 | 99.10 |
| MON-IYC | 15.00 | 15.00 | 23.00 | 40.05 | 93.05 |
| KEL-IYC | 15.00 | 15.00 | 23.00 | 38.70 | 91.70 |

MAR - *Maraska* variety; MON - *Montmorencys* variety; KEL - *Kelleris* variety

CL - samples fermented by classical fermentation process; IYC - samples fermented by immobilized yeast cells

Table 3 presents results of sensory analyses of cherry brandies according to German *DLG* model. Total sensory analyses points of cherry distillates were in range of 91.7-99.1.

The best evaluated sample was brandy produced from *Maraska* cherry variety which is fermented by immobilized yeast cells (99.1 total points of maximal 100 points) followed by brandy produced also from *Maraska* cherry variety, but by classical fermentation process (98.5 total points). As mentioned earlier, *Maraska* brandies had the highest concentration of ethyl octanoate which is considered one of the most important contributor to the aroma of alcoholic distillates.

Brandies produced from *Montmorencys* and *Kelleris* cherry varieties and fermented by classical process had better sensory characteristics, compared to the brandies fermented by immobilized yeast cells (KEL-CL had 97.5 total points, MON-CL 96.4, MON-IYC 93.05 and KEL-IYC 91.7 total points).

MAR-CL, MON-CL, KEL-CL and MAR-IYC were rated with a very high score by the examiners for a taste (44.1-45 of maximal 45 points). Those samples had high content of benzaldehyde (MAR-CL (2.27 mg/L) > MON-CL (1.69 mg/L) > KEL-CL = MAR-IYC (1.58 mg/L)). As mentioned earlier, benzaldehyde had significant effect on the aroma of cherry brandy.

Conclusions

All investigated cultivars and fermentation processes yield cherry brandies of very good to excellent quality. Cherry brandies produced by immobilized

yeast cells, had a higher content of aldehydes, but lower content of total acids, total extract, higher alcohols and esters compared to the samples produced by classical fermentation process. Cherry brandies produced by classical fermentation process had significantly higher content of benzaldehyde which has great influence on flavour of cherry brandies.

Ethyl octanoate which is considered one of the most important contributors to the aroma of alcoholic distillates was found in the highest concentrations in *Maraska* distillates.

The best evaluated sample was brandy produced from *Maraska* cherry variety fermented by immobilized yeast cells (99.1 total points of maximal 100 points) followed by brandy produced also from *Maraska* cherry variety, but by classical fermentation process (98.5 total points).

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