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Chemical characterization and differentiation of Montenegrin plum spirits obtained by two techniques of traditional batch distillation

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Abstract: Volatile compounds of plum spirit have a decisive influence on its quality and are closely related to the method of production. In this paper, minor and major volatile of plum spirits produced in small artisanal distilleries of Montenegro, obtained by two batch distillation techniques traditionally used (single and double distillation in alembic of the same construction) were analyzed by GC-FID and GC-FID-MS methods. All plum spirits contained all 8 major volatile compounds analyzed by GC-FID method, but out of a total of 138 minor aromatic compounds detected, only 32 are common to all samples. Wide ranges of concentrations of most volatile compounds indicated the great heterogeneity of plum spirit production methods in Montenegro, included, among others, two different distillation techniques. The principal component analysis have shown that differentiation of plum spirits obtained by single or double batch distillation is not possible using the all compounds analysed by GC-FID or GC-FID-MS, as well as compounds belonging the same chemical class, but rather based on the content of the volatile compounds typical for batch distillation tail fraction.

Keywords: GC-FID-MS; volatile compounds; alembic; single distillation; double distillation, PCA.

INTRODUCTION

In the last five decades, studies aimed to characterize and determinate quality and authenticity of plum spirits collected from the market and from small artisanal producers is carried out periodically in plum-producing European countries. Besides other chemical methods of analysis (including those from classical volumetric and ultra-violet/visible spectrophotometric methods to synchronous

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fluorescence spectroscopy and stable isotope ratio analyses), GC-FID and GC-MS methods were widely used for more detailed characterization of plum spirit.¹⁻⁷ Volatile fingerprinting based on GC-MS analysis is an excellent method for plum spirits authentication, which can be very useful for distinguishing among the plum spirits that were obtained from different plum cultivars⁸ and by different production methods⁹ or originated from different regions.¹⁰

Plum is the most grown fruit species in Montenegro, primarily around the Lim River (Lim Valley - Upper Polimlje).¹¹ Almost the entire annual plum crop is processed into plum spirit, exclusively in small household artisan distilleries. Distilleries use traditional method of plum spirit production, with its small variations depending on the knowledge of the distiller, the equipment of the distillery, and the preferences of final consumers. Traditional Montenegrin plum spirits obtained by classic single distillation are characterized by pronounced acidic taste and by low ethanol concentrations (< 30 % v/v). However, contemporary consumers are usually interested in consumption of plum spirits that are free from any sensory quality defects, including sharp, acidic, often unpleasant odour and taste. As consumer preferences change, manufacturers must periodically adjust characteristics of their products. Some authors have suggested that producers must adapt distillation operating recipes to meet consumer preferences.¹² This led to the modification of the traditional, classic method of single distillation in order to produce plum spirits with an alcohol content of over 40 % (v/v) and a reduced acid content, but only to the degree necessary to maintain the pleasing freshness and acidity that distinguish Montenegrin plum spirits. Anyway, the aim of plum spirit producer is to preserve traditional character of product, by using traditional raw materials and traditional production methods with minimal modification in order to preserve the plum fruits aroma and enable fulfilled requirements of law regulation governing the content of certain toxic compounds (*e.g.*, methanol) and, at the same time, avoidance the occurrence of sensory defects of product.

Batch distillation in alembic is considered as a most important step in the production of high quality spirit drinks (*e.g.* cognac).^{13,14} Double distillation in the alembic is necessary to obtain a high-quality product because it allows for stronger purification and obtaining cleaner, less impressive, but sufficiently aromatic distillates. However, a unified plum spirit distillation technique does not exist in the north of Montenegro. Just like in some other producing areas of other Balkan countries (Serbia, Bosnia and Herzegovina and Croatia), Montenegrin plum spirits can be obtained by single or double distillation techniques in copper pot still, called alembic.

Single distillation (S) technique in the simple alembic of Charentais type (with a capacity of about 100 L), directly yields consumable plum spirits with 42-47 % (v/v) of ethanol. This distillation technique is carried out as follows. The boiler of

traditional alembic (useful volume of about 100 L) is filled with about 70 to 80 kg of fermented plum mash and about 20 L of water or tail fraction stored from the previous year. During the distillation, about 0.5-0.7 L of the head fraction is discarded, and the heart fraction (between 5 and 10 L, with an ethanol content between 42 and 47 % v/v) and the tail fraction (about 15-20 L with ethanol content between 10 and 15 % v/v) are collected separately. The next batch in the alembic is also filled with 70-80 kg of fermented mash and with tail fraction obtained from the previous distillation batch. Distillate fractions are separated as in the previous distillation. Thus, the head fraction is always discarded, the middle fraction with ethanol content about 42-47 % (v/v) is used for direct consuming, and the tail fraction is recycled (returned) to the next batch.

Double distillation (D) technique is carried out in the same type of Charentais type alembic. In the first distillation (without separating fractions), the first distillate with an ethanol content between 23 and 29 % (v/v) is obtained from the fermented plum mash. For the second distillation (redistillation), the first distillate is poured into the boiler of the same pot still. During the second distillation, the first fraction (head) is separated in the amount of 0.5-0.7% calculated on the volume of the first distillate poured into the boiler of alembic. The cut-off point between the middle fraction (heart) and the tail fraction varies by manufacturer. The middle fraction is collected with ethanol content between 45 and 68 % (v/v). The tail fraction is not recirculated to the next distillation batch. Middle fractions with less than 50 % (v/v) are consumed directly, and those with more than 50 % (v/v) must be diluted with deionized water to an ethanol concentration below 50 % (v/v).

According to Spaho,¹⁵ plum spirit obtained by single stage distillation in alembic is very aromatic and contains higher concentration of congeners (acetic acid, esters, aldehydes, higher alcohols) than plum spirit produced by double distillation in same type of alembic. Some of these congeners are not desirable, and in high concentrations can give the unpleasant sensory character of plum spirits. However, some consumers in Montenegro emphasize that plum spirits obtained by single distillation have a more fruity smell and a softer taste than those obtained by double distillation. In other words, although the science and art of distillation favour plum spirits obtained by double distillation in alembic, producers must sometimes take into account the preference of certain consumers who like the taste and smell of distillates obtained by single distillation in alembic.

In this study, we aimed to characterize the aromatic profile of Montenegrin plum spirits based on GC-FID and GC-FID-MS analysis, with a special emphasis on finding chemical markers that would allow distinguishing plum spirit samples obtained by single and double distillation techniques in the traditional alembic, considering that both techniques are equally represented in distilleries in Montenegro.

EXPERIMENTAL

Plum spirits samples

Twelve plum spirits (6 produced by single distillation and 6 produced by double distillation) were collected in the spring of 2021 from small, artisanal producers of the Upper Polimlje region in the north of Montenegro. Characteristics of collected samples are shown in Table I. All plum spirits were produced from spontaneously fermented mashes of locally plum varieties in fermenters of 500-2000 L in volume. The fermented mashes were usually stored for more than a month after the fermentation is completed, even up to 6 months. Single and double distillation techniques were carried out in alembics, with the separation of fractions, in the manner specified in the Introduction. Three-member expert panel (using the 20-point Buxbaum method⁷) founded that all analyzed plum spirits were of good sensory quality (sensory ratings range 17.55-18.30), with a dominant, more or less pronounced, fruity note, without defects in odour and taste (Table I). The plum spirits had a fresh, pleasant full taste, which was somewhat rounder and more pronouncedly acidic in the case of single-distilled plum spirits. Plum spirits obtained by single distillation usually had a higher content of total acids than those obtained by double distillation. The plum spirits obtained by two distillation techniques are consumed with ethanol content ranging between 42 and 50 % v/v.

TABLE I. Plum spirit samples obtained by single and double distillation techniques

Plum spirit code ^a	Ethanol content in consumable plum spirit (% v/v)	Ethanol content in heart fraction of distillate (% v/v)	Additional information on samples (cultivar ^b /stones ^c /mash storage ^d)	Total acids (mg L ⁻¹)	Sensory quality (points)
Single distillation technique (S)					
S1	47.0	47.0	P/W/60 days	1104.0	17.90
S2	45.0	45.0	P/W/180 days	1732.8	18.25
S3	43.0	43.0	C/W/30 days	1216.8	17.70
S4	46.0	46.0	C/W/45 days	1944.0	18.30
S5	44.0	44.0	S/WO/60 days	820.8	18.20
S6	42.0	42.0	SC/W/60 days	1920.0	17.55
Double distillation technique (D)					
D1	49.0	63.0	T/WO/70 days	529.0	17.85
D2	50.0	55.0	P/W/55 days	676.8	17.80
D3	48.0	60.0	P/W/10 days	912.0	17.65
D4	45.0	45.0	C/W/180 days	1533.6	18.05
D5	45.0	53.0	C/W/30 days	496.8	17.85
D6	45.0	68.0	S/W/70 days	223.2	18.05

^aS1-S6 – samples obtained by single distillation, D1-D6 – samples obtained by double distillation; ^bcultivars (P – Požegača, C – Čačanska Rodna, S – Stanley, SC – Čačanska Rodna+Stanley, T – Turgulja); ^cW – mash with stones, WO – mash without stones; ^dduration of fermented mash storage;

GC-FID and GC-FID-MS analysis of volatile compounds in plum spirits

Sample preparation and GC-FID analysis of major volatile compounds (acetaldehyde, ethyl acetate, methanol, 1-propanol, 2-methyl-1-propanol, 1-butanol, 2/3-methyl-1-butanol, and 1-hexanol) and GC-FID-MS analysis of minor volatile compounds in plum spirits were carried out by the methods briefly described in the Supplementary material.⁹

Statistical analysis

Descriptive statistics, correlation analysis, and principal component analysis (PCA) were performed using Statistica 7 Software (StatSoft Inc., Tulsa, OK, USA).

RESULTS AND DISCUSSION

According to the high sensory score, all analyzed plum spirits, regardless of distillation technique applied, are considered to be spirits of good sensory quality, without defects, with a more or less expressed fruity aroma, medium to full body and pleasant taste. In general, plum spirits obtained by single distillation had a slightly higher content of total acids and a more pronounced freshness and fruity character of odour and taste than plum spirits obtained by double distillation.

Volatile compounds in plum spirits

Eight major volatile compounds were quantified in plum spirits by GC-FID method (Table II). In Tables S-I to S-XII (shown in Supplementary material), concentration ranges of 138 minor volatile compounds (10 alcohols, 9 aldehydes, 2 ketones, 3 lactones, 10 acetals, 12 acids, 59 esters, 2 benzenoids, 15 terpenoids, 5 sesquiterpenoids, 4 isoprenoids, 7 hydrocarbons) analyzed by GC-FID-MS method are shown. In all samples, regardless of distillation techniques, following volatile compounds were found: all 8 major volatile compounds (acetaldehyde, ethyl acetate, methanol, 1-propanol, 2-methyl-1-propanol, 1-butanol, 2/3-methyl-1-butanol, and 1-hexanol) and only 32 minor volatile compounds – 4 alcohols (1-hexanol, benzyl alcohol, 1-octanol, and 2-phenylethanol), 3 aldehydes (nonanal, furfural, and benzaldehyde), 5 acids (3-methylbutanoic acid, hexanoic acid, 2-hydroxy-4-methylpentanoic acid, octanoic acid, and decanoic acid), 3 acetals (1,1-diethoxy-3-methylbutane, 1,1,3-triethoxypropane, and 1,1-diethoxyhexane), 15 esters (ethyl butanoate, ethyl hexanoate, ethyl octanoate, ethyl nonanoate, ethyl decanoate, ethyl dodecanoate, ethyl hexadecanoate, ethyl 2-methylbutanoate, 3-methylbutyl acetate, 2-phenylethyl acetate, ethyl lactate, ethyl benzoate, ethyl (*E*)-cinnamate, ethyl (*Z*)-2-butenate, and diethyl succinate), and 2 terpenoids ((*E*)-linalool oxide (furanoid) and linalool). These compounds form the basic aromatic pattern of plum spirits from Upper Polimlje. Other minor volatile compounds analysed were not found in all plum spirit samples. Vyviurska et al. found a very similar aromatic profile in plum spirits obtained from 25 different plum cultivars.⁸ Monovarietal plum spirits in the study of mentioned authors contained between 95 and 195 identified volatile compounds, depending on the cultivar; the common compounds for all samples were 4 major volatile compounds (1-propanol, 2-

methyl-1-propanol, 3-methyl-1-butanol, 1-hexanol) and 27 minor volatile compounds (9 alcohols, 2 aldehydes, 2 acids, 13 esters and 1 terpenoid). In French Mirabelle brandies, 175 volatile compounds were quantified, but only 105 were found in all samples analysed, although the Mirabelle brandy production has been standardized in terms of cultivar and production methods.¹

TABLE II. Major volatile compounds contents in plum spirits

Compound ^a	RT (min)	Single distillation (n=6)			Double distillation (n=6)		
		Min	Max	Mean±SD	Min	Max	Mean±SD
acetaldehyde	1.124	125.44	226.37	188.76±40.79	91.11	263.88	153.32±65.47
ethyl acetate	1.738	1101.12	2907.50	1623.75±654.31	658.71	3136.54	1558.03±890.52
methanol	1.825	4.27	8.95	6.82±1.62	4.13	8.49	6.69±1.55
1-propanol	3.686	1137.93	1693.50	1350.89±196.48	208.86	1735.89	890.90±532.20
2-methyl-1-propanol	5.483	225.38	340.09	294.04±38.05	186.49	414.89	325.38±88.52
1-butanol	7.725	30.28	528.39	170.03±190.67	49.40	201.50	88.97±61.32
2/3-methyl-1-butanol	11.329	828.18	1090.25	975.84±120.26	777.72	1709.25	1145.79±415.73
1-hexanol	17.795	13.47	115.60	43.37±39.70	12.38	55.98	23.65±16.06

^acontents are given in mg L⁻¹a.a., except the methanol content (in g/L⁻¹a.a.);

Major volatile compounds

According to the content, major volatile compounds (Table II) form the basic body of all plum spirits analyzed, besides ethanol and water. The contents of the analyzed compounds were in the ranges that are characteristic for plum spirits originated from Southeast Europe.¹⁶ Some of these compounds (acetaldehyde, ethyl acetate, 1-propanol, 2-methyl-1-propanol, 1-butanol, 2/3-methyl-1-butanol) are mostly formed by microbiological activity during alcoholic fermentation and storage of fermented mash before distillation. The others are formed by enzymatic degradation of fruit ingredients during fruit processing and fermentation – methanol is generated by pectin methylesterase activity on pectic substances of fruit, whereas 1-hexanol is formed from linoleic acid through lipoxygenase pathway.¹⁷ Since the concentration of most major volatile compounds of the first distillate obtained by the distillation of fermented plum mash, except total acids, cannot be significantly reduced by using even traditional double distillation,^{14,18,19} manufacturers should pay great attention to the selection of plum variety, method of primary processing and alcoholic fermentation of plum mash and its storage until distillation, as well as the cut-off points between the heart fraction and the tail fraction during double distillation.²⁰⁻²² Heterogeneity in these pre-distillation steps in the production of Montenegrin plum spirits caused the contents of certain major compounds were higher in plum spirits obtained by double distillation than in those obtained by single distillation.

Analyzed plum spirits had no sensory defects, *e.g.* ester-like tone, tail-like tone, head-like tone, impure odor, which, according to Scholten and Kacprowski,²³ most commonly occur in fruit spirits. The contents of above-mentioned major compounds were lower than contents that these authors considered to be thresholds for the appearance of sensory defects. Due to its potential toxicity, maximal concentration (12 g L⁻¹a.a.) of methanol is limited by the Regulation EC.²⁴ All samples contained significantly lower methanol than prescribed.

Minor volatile compounds

As mentioned previously, 138 minor volatile compounds were quantified in Montenegrin plum spirits (Tables S-I to S-XII). Previous research showed that number of quantified minor volatile compounds in plum spirits varied between 89 and 195, depending on the country of origin, the plum cultivar, the plum spirit production method, and the GC-MS techniques used.^{1,3,4,7,8,9} Wide ranges of concentrations of minor compounds in analysed plum spirits obtained by the same distillation technique indicate the great heterogeneity of the production method of plum spirit from Upper Polimlje in Montenegro. In other words, in addition to the distillation technique, the composition of plum spirit can be strongly influenced by other factors, as the locality,¹⁰ variety^{8,9,20}, and pre-distillation steps.²¹

It is interesting that ethyl pentanoate, ethyl heptanoate, ethyl salicylate, methyl decanoate, ethyl phenyllactate, ethyl linoleate, (*Z*)-linalool oxide (furanoid), α -terpineol, linalool acetate were found only in all plum spirits obtained exclusively by single distillation technique. On the other hand, γ -decalactone, γ -dodecalactone, 1,1-diethoxy-nonane, 2-methyl-propanoic acid, 2-methylbutyl acetate, ethyl phenylacetate, ethyl tetradecanoate, 4-vinylanisole were found in all 6 plum spirits produced by double distillation technique. These results confirm the findings of other authors,^{13,14} that is some constituents of the distillate are formed during distillation, while others are broken down, which in the case of Montenegrin plum spirits could probably be connected with the applied batch distillation technique. In other words, during distillation, especially in alembic, numerous reactions occur due to high temperatures (hydrolysis, esterification, acetalization, Maillard reactions and thermal degradation of pentoses), resulting in the formation of numerous compounds (furfural, esters, aldehydes, acetals, terpenoids and norisoprenoids). These reactions are especially intense during the first distillation - the distillation of the fermented raw material.

The odor attributes of minor volatile compounds can be pleasant or unpleasant, depending not only on the type of compound, but also on its concentration and odor threshold. The majority of the minor compounds, which belong to the classes of alcohols, aldehydes, lactones, acetals, esters, terpenoids and isoprenoids, are characterized by different nuances of a pleasant fruity and flowery smell.²⁵ Benzaldehyde is a characteristic ingredient of stone fruit spirits and has a typical bitter almonds odour (the fruit stone odour).¹⁷ Esters of long-

chain fatty acids (saturated and unsaturated) have a so-called stearin-like smell.¹⁷ According Ledauphin *et al.*,²⁰ 4-vinylanisole gives to calvados a tone that that can be described as perspiration like, delicatessen tone. Small amounts of some phenols or anisoles can contribute to pleasant fruit characteristic aroma, but the exceeded sensorial threshold can lead to unpleasant taste and aroma (cork defect or leather odor), and some of them can give phenolic or “Brett” character of alcoholic drinks.²⁶ Some volatile compounds can negatively affect the sensory characteristics of plum spirit. For instance, the majority of volatile acids have extremely unpleasant smells: of stable, sweat, marsh, *etc.*²⁰

Pearson's correlation analysis (Correlations between common volatile compounds)

For further characterization of Montenegrin plum spirits, it would be interesting to determine the correlation between the contents of the individual volatile compounds. A significant correlation was found between the content of some of 38 volatile compounds (6 major and 32 minor volatile compounds) found in all 12 samples (Table S-XIII in the Supplementary material). Based on correlations, the contents of some volatile compounds can be used to predict the contents of others. However, on the basis of these correlation coefficients, it was not possible to determine the difference between plum spirits obtained by single or double distillation.

Principal component analysis (PCA)

Principal component analysis based on the results of the GC-FID analysis of 8 major (Fig. 1) and GC-MS analysis of 32 minor (Fig. 2) compounds common to all samples, indicated that clustering of plum spirits obtained by various distillation techniques in alembic is not possible.

Minor volatile compounds belonging to the same chemical class (shown in Tables S-I to S-XII) have different origins and/or different dynamics of distillation, which has already been mentioned above. Therefore, PCAs based on contents of all 138 minor compounds or content of compounds that belong to the individual classes of chemical compounds (alcohols, carbonyl compounds, lactones, acetals, esters, terpenoids, sesquiterpenoids, isoprenoids, benzenoids and hydrocarbons, respectively) did not indicate a good discrimination between samples obtained by two different distillation technique in alembic (results not shown). The results of PCAs showed that the composition of plum spirit is more decisively influenced by pre-distillation steps (such as the choice of plum variety, the method of preparing fruits for fermentation or the fermented mash storage time) than the applied distillation technique itself. This also indicates that the same plum variety and the same pre-distillation steps should be used in future research, in order to more clearly observe the differences in the composition of plum spirits obtained by single or double distillation techniques. But,

since the production methods of Montenegrin plum spirits are very heterogeneous, especially in terms of pre-distillation steps, chemical markers should be found that, regardless of these differences, could indicate whether the plum spirits were produced by single or double distillation.

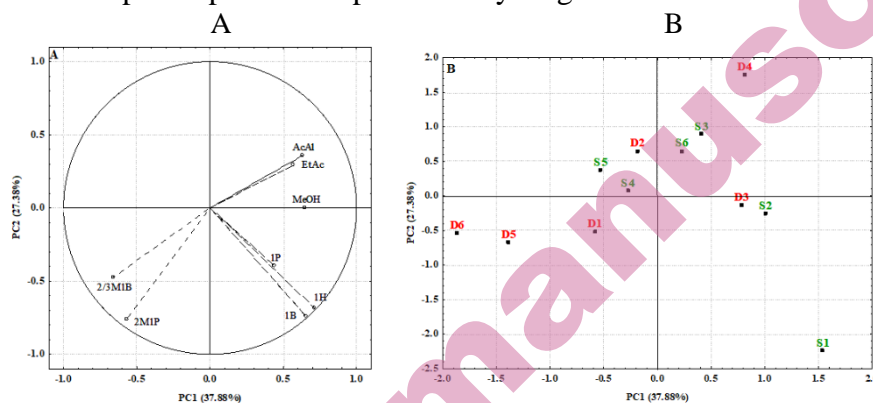


Fig. 1. Principal component analysis (PCA) calculated on 8 major volatile compounds of plum spirits; A) variable loadings, acetaldehyde (AcAl), ethyl acetate (EtAc), methanol (MeOH), 1-propanol (1P), 1-hexanol (1H), 1-butanol (1B), 2-methyl-1-propanol (2MIP), 2/3-methyl-1-butanol (2/3MIB); B) sample scores, plum spirits obtained by single distillation (S1-S6), plum spirits obtained by double distillation (D1-D6).

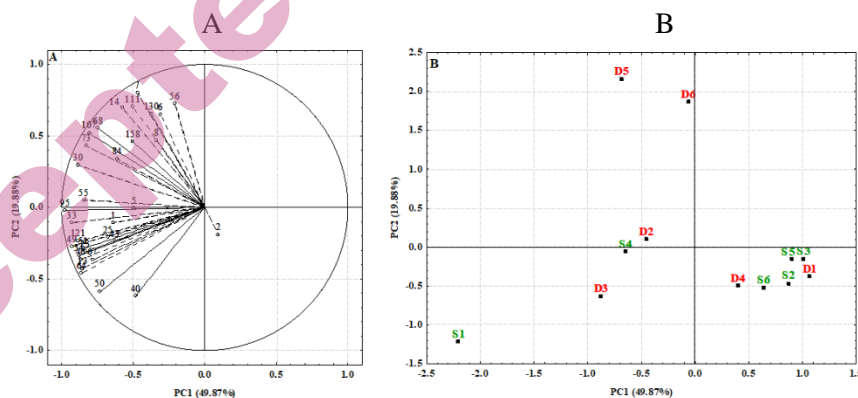


Fig. 2. Principal component analysis (PCA) calculated on 32 minor volatile compounds common for all plum spirit samples; A) variable loadings, alcohols (13, 40, 46, 56), aldehydes (5, 25, 55), acetals (23, 49, 51), acids (6, 30, 43, 68, 107), esters (1, 2, 7, 8, 14, 33, 64, 67, 73, 84, 95, 111, 121, 130, 158), terpenoids (50, 54); B) sample scores, plum spirits obtained by single distillation (S1-S6), plum spirits obtained by double distillation (D1-D6).

During the distillation in alembic, the dynamics of distillation of most volatile compounds are similar in both the first and second distillations.¹⁴ Some volatile compounds pass into the distillate at the beginning of the distillation and some

later, so the first group is considered as characteristic compounds for the head fraction and others for the heart or tail fractions. By the single distillation technique still used in Montenegro, the tail fraction separated during the first distillation is returned to the next batch of distillation together with the mash, so it could be expected that the plum spirit obtained in this way should contain an increased amount of compounds typical of the tail fraction compared to plum spirit obtained by the double distillation technique in alembic. Previous studies have shown that the compounds typical of the tail fraction during distillation in alembics are octanoic acid, decanoic acid, dodecanoic acid, 2-methylpropanoic acid, 2-methylbutanoic acid, 3-methylbutanoic acid, 2-phenylethanol, furfural, 2-phenylethyl acetate, ethyl lactate, and diethyl succinate.^{14,27} These 11 compounds may be interesting for differentiation of the plum spirits produced by a single distillation (absence of adequate separation of heart and tail fractions) and of the plum spirits produced by double distillation (the tail fraction is separated, but the distillation cut points between heart fraction and tail fraction in the second distillation are different). In this case, the PCA showed that the first two principal components (Fig. 3) explain 62.11% of the total variance; PC1 (40.83%) and PC2 (21.28%).

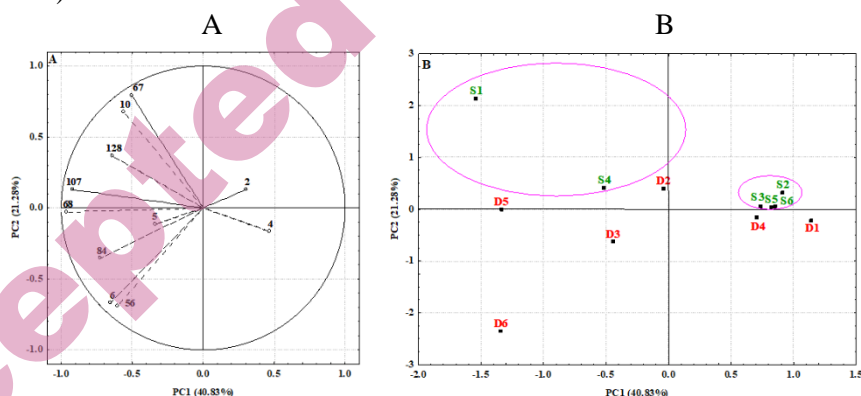


Fig. 3. Principal component analysis (PCA) calculated on 11 minor volatile compounds that are characteristic for tail fraction; A) variable loadings, ethyl lactate (2), 2-methyl-propanoic acid (4), furfural (5), 3-methyl-butanoic acid (6), 2-methyl-butanoic acid (10), 2-phenylethanol (56), diethyl succinate (67), octanoic acid (68), 2-phenylethyl acetate (84), decanoic acid (107), dodecanoic acid (128); B) sample scores, plum spirits obtained by single distillation (S1-S6), plum spirits obtained by double distillation (D1-D6).

Plum spirits obtained by a single distillation (S2, S3, S5, S6, as well as S1 and S4) were scattered in the positive region PC2. In these plum spirits, the contents of ethyl lactate, as well as those of diethyl succinate, 2-methylbutanoic acid, dodecanoic acid, and decanoic acid, were higher than in some plum spirits obtained by double distillation. Samples D4, D2, and D5, obtained by double

distillation, also contained high concentrations of certain tail compounds, which can be explained by the late cut off point between heart and tail fractions in the second distillation. Namely, the ethanol content in the heart fraction in production of these three double distilled plum spirits was less than 60 % (v/v), *i.e.* 45 %, 55 %, and 53 % (v/v), respectively, indicating that the compounds typical of the tail fraction were transferred, to a certain extent, also to the heart fraction. On the other hand, Spaho et al.²² have shown that the contents of a typical tail component, ethyl lactate, are not statistically different in heart fractions if, during the second distillation, the distillation cuts are done so that the ethanol content in the heart is greater than 60 % (v/v). Although the recycling of the tail fraction used in the single distillation technique increases the contents of the so-called tail compounds, their contents in the final plum spirit will depend not only on their distillation dynamics but also on their origin. Thus, the most interesting are the compounds of plum spirit that are mainly formed by bacteria during the fermentation and storage of the fermented mash (*e.g.*, ethyl lactate). This component, depending on the used pre-distillation steps, can be much more present in some mashes, so that even the double distillation technique cannot remove them to a significant extent. In other words, plum spirits obtained from bacterially infected mashes distilled by double distillation technique may contain more of these compounds than plum spirits from mashes that were not bacterially infected and were distilled by single distillation technique.

CONCLUSION

There is a great heterogeneity in the plum spirits production methods in the north of Montenegro, included the two techniques of alembic batch distillation (single and double distillation). GC-FID and GC-FID-MS analysis defined for the first time the profile of volatile compounds in good-quality plum spirits from the north of Montenegro, which can serve as a basis for their authentication. The principal component analysis, based on the content of specific chemical markers, such as compounds typical of distillation tail fraction, can be used to define more closely the manner of production of plum spirits and more efficient control of the Montenegrin plum spirits quality in the future.

SUPPLEMENTARY MATERIAL

Additional data are available electronically at the pages of journal website: <https://www.shd-pub.org.rs/index.php/JSCS/article/view/13040>, or from the corresponding author on request.

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ИЗВОД

ХЕМИЈСКА КАРАКТЕРИЗАЦИЈА И ДИФЕРЕНЦИЈАЦИЈА ЦРНОГОРСКИХ ШЉИВОВИЦА ДОБИЈЕНИХ ДВЕМА ТЕХНИКАМА ТРАДИЦИОНАЛНЕ ШАРЖНЕ ДЕСТИЛАЦИЈЕ

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Испарљиве компоненте шљивовице одлучујуће утичу на њен квалитет и блиско су повезане са начином производње. У овом раду, GC-FID и GC-FID-MS методама су анализирани шљивовице доброг сензорног квалитета произведене у малим занатским дестилеријама Црне Горе, добијене два традиционално коришћеним техникама шаржне дестилације (једноструком и двоструком дестилацијом у аламбику исте конструкције). Све шљивовице су садржале свих 8 најзаступљенијих испарљивих компоненти (анализираних GC-FID методом), али само 32 од 138 мање заступљених испарљивих компоненти (анализираних GC-FID-MS методом). Широки распони концентрација већине испарљивих једињења указивали су на велику хетерогеност метода производње шљивовице у Црној Гори, укључујући, између осталог, две различите технике дестилације. Анализа главних компоненти (PCA) је показала да није могућа диференцијација шљивовица, добијених једноструком или двоструком шаржном дестилацијом, базирана на свим компонентама анализираних методама GC-FID или GC-FID-MS, као и компонентама које припадају појединим хемијским класама, већ само на основу садржаја испарљивих компоненти типичних за паточну фракцију при шаржној дестилацији.

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REFERENCES

1. J. Ledauphin, C. Le Milbeau, D. Barrilier, D. Hennequin, *J. Agr. Food Chem.* **58** (2010) 7782 (<https://dx.doi.org/10.1021/jf9045667>)
2. L. Adam, J. Meinel, N. Christoph, G. Versini, *Kleimbrennerei* **9** (1995) 188
3. W. Brandes, M. Karner, R. Eder, *Mitt. Klosterneuburg* **57** (2007) 63
4. J. Velišek, F. Pudil, J. Davidek, V. Kubelka, *Z. Lebensm. Unters. For.* **174** (1982) 463
5. R. Winterova, R. Mikulikova, J. Mazač, P. Havelec, *Czech J. Food Sci.* **26** (2008) 368 (doi: 10.17221/1610-cjfs)
6. T. E. Coldea, C. Socaciu, Z. Moldovan, E. Mudura, *Not. Bot. Horti. Agrobi.* **42** (2014) 530 (<https://doi.org/10.15835/nbha4229607>)
7. V. Tešević, N. Nikićević, A. Jovanović, D. Djoković, L. Vujisić, I. Vučković, M. Bonić, *Food Technol. Biotech.* **43** (2005) 367 (<http://www.ftb.com.hr/images/pdfarticles/2005/October-December/43-367.pdf>)

8. O. Vyviurska, F. Matura, K. Furdikova, I. Španik, *J. Food Sci. Tech.* **54** (2017) 4284 (<https://doi.org/10.1007/s13197-017-2900-5>)
9. S. Ivanović, K. Simić, V. Tešević, L.; L. Vujisić, M. Ljekočević, D. Godevac, *Molecules* **26** (2021) 1391 (<https://doi.org/10.3390/molecules26051391>)
10. M. Filatova, K. Bechynska, J. Haslova, M. Stupak, *Lebensm-Wiss. Technol.* **167** (2022) 113864 (<https://doi.org/10.1016/j.lwt.2022.113864>)
11. D. Božović, V. Jačimović, *J. Pomol.* **45** (2011) 117 (in Serbian)
12. D. Osorio, J.R. Perez-Correa, L.T. Biegler, E. Agosin, *J. Agric. Food Chem.* **55** (2005) 6326 (<https://doi.org/10.1021/jf047788f>)
13. P. Awad, V. Athes, M.E. Decloux, G. Ferrari, G. Snakkers, P. Raguenaud, P. Giampaoli, *J. Agric. Food Chem.* **65** (2017) 7736 (<https://doi.org/10.1021/acs.jafc.7b02406>)
14. G. Zanghelini, P. Giampaoli, V. Athès, S. Vitu, V. Wilhelm, M. Esteban-Decloux, *Food Research International*, **178** (2024) 113977 (<https://doi.org/10.1016/j.foodres.2024.113977>)
15. N.Spaho, *Distillation Techniques in the Fruit Spirits Production*, in *Distillation – Innovative Applications and Modeling*, M. Mendes, Ed., IntechOpen, London, UK, 2017, p. 129 (<https://doi.org/10.5772/66774>)
16. J. Mrvčić, A. Trontel, K. Hanousek-Čiča, N. Vahčić, N. Nikićević, N. Spaho, M. Mihaljević-Žulj, A. Brodski, V. Jurak, M. Krajnović, R. Sahor, V. Rubeša Vili, R. Petrović, D. Stanzer, *Glasnik zaštite bilja* **6** (2021) 80 (<https://doi.org/10.31727/gzb.44.6.9>)
17. L. Nykänen, I. Nykänen, *Distilled beverages*, in *Volatile Compounds in Food and Beverages*, H. Marse, Ed., Marcel Dekker, Inc., New York, NY, 1991, p. 547 (<https://doi.org/10.1002/food.19910351027>)
18. N. Nikićević, B. Popović, V. Tešević, O. Mitrović, M. Kandić, N. Miletić, I. Urošević, *J. Pomol.* **48** (2014) 21 (in Serbian)
19. I. Lukić, S. Tomas, B. Milićević, S. Radeka, Đ. Peršurić, *J. Inst. Brew.* **117** (2011) 440 (<https://doi.org/10.1002/j.2050-0416.2011.tb00491>)
20. B. Popović, O. Mitrović, A. Leposavić, S. Paunović, D. Jevremović, N. Nikićević, V. Tešević, *J. Serb. Chem. Soc.* **84** (2019) 1381 (<https://doi.org/10.2298/JSC190307061P>)
21. B. Popović, O. Mitrović, N. Nikićević, V. Tešević, I. Urošević, N. Miletić, S. Milojević, *Processes* **11** (2023) 863 (<https://doi.org/10.3390/pr11030863>)
22. N. Spaho, P. Dürr, S. Graba, E. Velagić-Habul, M. Blesić, *J. Inst. Brew.* **119** (2013) 48 (<https://doi.org/10.1002/jib.62>)
23. G. Scholten, M. Kacprowski, *Kleinbrennerei* **6** (1995) 130
24. Regulation (EC) no. 110 of the European Parliament and of the Council of 15 January 2008 on the definition, description, presentation, labelling and the protection of geographical indications of spirit drinks and repealing Council Regulation (EEC) no. 1576/89, *Off. J. Eur. Commun. Law* **39** (2008) 16
25. N. Christoph, C. Bauer-Christoph, *Flavour of Spirit Drinks: Raw Materials, Fermentation, Distillation, and Ageing*, in *Flavours and Fragrances. Chemistry, Bioprocessing and Sustainability*, R. G. Berger, Ed., Springer-Verlag, Berlin, Germany, 2007, p. 219 (https://doi.org/10.1007/978-3-540-49339-6_10)
26. M. Jakubikova, J. Sadecka, K. Hrobonova, *Eur. Food Res. Technol.* **245** (2019) 1709 (<https://doi.org/10.1007/s00217-019-03291-3>)
27. S. Guan, H. J. Pieper, *Dtsch. Lebensm. Rundsch.* **95** (1999) 1.