

## A STUDY ON THE CONTENT OF TERPENIC COMPOUNDS IN THE CULTIVAR 'MORAVIAN MUSCAT' (*VITIS VINIFERA* L.)

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### ABSTRACT

Terpenic represent a very interesting group of aromatic substances. They occur in aromatic grapevine cultivars and create muscat and flower aromas. In 2013, their contents as well as contents of other aromatic substances were determined in juices made of the Moravian Muscat cultivar after 5, 12 and 24 of maceration. Pedigree: Muscat Ottonel x Prachtraube. As Control, fresh juice analysed immediately after the pressing was used. The aim of this experiment was to find out which terpenic compounds are present in grapes of this cultivar and which length of maceration would be the most suitable for making wine. Attention was paid also to levels of monoterpenes, phenols, ethyls, alcohols and acids as well as to basic analytical parameters. Results of a chemical analysis indicated that an optimum is the maceration of grapes for 24 hours because after this time interval the highest amount of terpenic substances was released.

*Keywords:* terpenes, maceration, antioxidant activity, volatile compounds, analytical parameters

## 1. INTRODUCTION

Terpenic compounds are members of an important group of aromatic compounds and are characterized by floral, muscatel or fruity aromas that are synthesized in berries and stored in their skin. This aroma of a wine depends on its content of volatile compounds, over 680 of which have been identified in wines from some white grape varieties (PEINADO *et al.*, 2004). Although terpenes mainly give off a pleasant aroma some of them may show a negative effect on quality of wine. So, for example, yeast of the genus *Streptomyces* may synthesize sesquiterpenes either on cork or in the barrel. Their presence may jeopardize the sensory quality of wine (JACKSON, 2008). The aroma of young wines is the product of a biochemical and technological sequence. Its formation derives from the grapes and juice production (grape de-stemming, crushing, and pressing technology), and is decisively influenced by the fermentation procedure. All of these parameters will determine the complexity of the wine aroma. (TAO *et al.*, 2008) Its quality and quantity influenced by the cultivar, soil, climate and viticultural practices. (RIBÉREAU-GAYON *et al.*, 2006, SANTIAGO *et al.*, 2011) The experiment monitored the occurrence of terpenic compounds in juice samples after different periods of maceration. Terpenes contribute to some white wines aroma, especially these produced from Muscat grapes and others aromatic ones of high terpene contents (Gewürtztraminer, Traminer, Huxel, Sylvaner). (DZIADAS and JELEŇ, 2010) The variety which was chosen is 'Muscat Moravsky'. Pedigree: Muscat Ottonel x Prachtraube. It was chosen for its good reputation in the South Moravia (Czech Republic). There are many preconcentration techniques to obtain volatile compounds, such a terpenes, subsequently analyzed by gas chromatography. The most widely used are Headspace sorptive extraction techniques (HSSE), such a SPE or SPME, purge and trap technique – sample is stripped by inert gas, which is concentrated in sorption column, steam distillation, continuous distillation and extraction. All this preconcentration techniques needs expensive equipment, or their performance needs high temperature, which can lead to terpene cyclization. Technique liquid-liquid extraction (LLE) used in this experiment is easy to perform, even it use high temperature. For purpose of this experiment, where we compare the length of maceration of similar samples is this sample preparation sufficient. The product of terpene cyclization is  $\alpha$ -terpinol, so in each sample we can compare its increasing. The aim of this experiment was to found out if (and to which extent) the length of maceration shows an effect on the increase in the content of terpenic and other compounds in produced wine. Terpenes, because of their high concentrations and low aroma thresholds, are the principal components responsible for the characteristic aroma of a wine (CARBALLEIRA LOIS *et al.*, 2001).

## 2. MATERIALS AND METHODS

### 2.1. Sampling site and procedure

Grapes used in the experimental part of this study originated from vineyards situated in the locality Velké Bílovice (wine-growing subregion of Velké Pavlovice). In this region, the average annual precipitations and temperature are 550 mm and 9.5 °C, respectively. Phenological data of the grapes and vine in year 2013:

Shooting: (BBCH05) – 25.4.2013,

Full anthesis: (BBCH 65) – 12.6.2013,

Verasion: (BBCH 81) – 9.8.2013,

Ripening: (BBCH 88) – 11.9.2013.

Rootstock CR2 typical for Czech Republic was used. The Czech grape training is modified German training specially Rhone\_Hessen, nevertheless fruitful wood is horizontal tying. Clasp planting: 1x1.2 mB. High of trunk is 0.75 m. Berries were collected random from the top, middle, and bottom of selected clusters. In order to obtain representative sample, colored berries were not favoured over greens. Berries were stored in a sealed plastic bags in the refrigerator until processing.

## **2.2. Experimental variants of maceration and processing of samples**

In this experiment, grapes of the variety Moravian Muscat' were used. These grapes were harvested on the 11 September 2013. Their sugar content was 19 °NM (i.e. 19 kg of natural sugars in 100 liters of juice). Harvested grapes were crushed and destalked in a stainless destalking-crushing machine, macerated for 0 (Variant 1); 5 (Variant 2); 12 (Variant 3) and 24 hours (Variant 4) at the temperature of 14 °C, and finally pressed.

Wine was made from each juice sample and used for the estimation of the following parameters: pH and contents of alcohol, titratable acidity and sugars, respectively. In individual wine samples, the content of aromatic compounds was estimated as well.

## **2.3. Estimation of total titratable acidity (EEC No 2676/90)**

The content of total titratable acidity was estimated by titration in an automatic titrator TITROLINE EASY (manufacturer SI Analytics GmbH, Germany). Titrations were performed with NaOH (0.1 mol.L<sup>-1</sup>) as the titration reagent. For analyses, a 10 ml sample was used; this sample was diluted with 10 ml of distilled water. Because of a subsequent formol titration, the sample was not titrated up to the usual pH value of 7.0 but up to the value of 8.1 (the resulting deviation was thereafter considered to be a systematic error). The detection of pH was assured by means of a pH-electrode SenTix 21. After the end of titration, the consumption of NaOH solution in milliliters was read, with the accuracy of two decimal positions, on the titrator's display. The content of total titratable acidity (in g.L<sup>-1</sup>) was calculated as follows: the amount of consumed NaOH solution was multiplied by the factor of the NaOH solution used for the titration; the product of this multiplication was thereafter multiplied by and by the coefficient 0.75.

## **2.4. Estimation of pH**

The pH value was estimated in an undiluted sample using a pH-meter WTW pH 526 and a pH electrode SenTix 21 (both manufactured by the company WTW, Germany).

Estimation of residual sugar and alcohol

Contents of residual sugar and alcohol were estimated in the apparatus ALPHA. The ALPHA apparatus is a compact FTIR analyzer that uses the ATR sampling technique. This technique helped to process samples before the analysis. Before the first measurement, the spectrometer was thoroughly rinsed with deionized water and the background was determined using a blank sample (i.e. of deionized water). For analyses, 1 ml samples were taken with a syringe; of this sample, 0.5 ml was used for rinsing of the system while the remaining volume of 0.5 ml was analyzed three times. Depending on the calibration used, the measured values were evaluated automatically using a special software.

## 2.5. Estimation of contents of aromatic compounds in berries by means of gas chromatography

### 2.5.1 Preparation of samples

Contents of aromatic compounds present in berries were estimated after their extraction by an organic solvent. In each sample, altogether 100 g of berries were mixed with 100  $\mu\text{L}$  of 1M  $\text{K}_2\text{S}_2\text{O}_8$  solution (to prevent oxidation) and 10  $\mu\text{L}$  of the GC internal standard. Thereafter, the mixture was homogenised in a manual mixer and the juice was separated from the mush using a filter paper. The pH value of the juice was adjusted to 3.0 with 10 M  $\text{H}_3\text{PO}_4$ . The adjusted juice was thereafter poured into a volumetric flask of the volume of 25 ml. The extraction was performed after the sample incubation in the boiling water bath for 1 hour. It is necessary to mention in this context that each heating results in a disintegration of glycosides and a release of aromatic compounds. The sample heating promote also the terpenic cyclization, but this preparation of sample is easy to perform and for purpose of this experiment is convenient. The product of terpene cyclization is  $\alpha$ -terpinol, so in each sample we can compare its increasing. The extraction was performed using 1 ml of methyl terc-butyl ether that contained 1 % of cyclohexane. After the separation, the phase of organic matter was dried up with anhydrous magnesium sulphate and used for the GC-MS analysis.

### 2.5.2 Analysis of aromatic compounds by means of gas chromatography

Concentrations of individual volatile compounds in wine were determined according to until now unpublished method of extraction with methyl-t-butylether. Into a 25-ml volumetric flask, 20 ml of wine was pipetted together with 50  $\mu\text{l}$  of 2-nonanol solution in ethanol; this compound was used as an internal standard (in concentration of 400  $\text{mg}\cdot\text{L}^{-1}$ ) and 5 ml of a saturated  $(\text{NH}_4)_2\text{SO}_4$  solution. The flask content was thoroughly stirred and thereafter 0.75 ml of the extraction solvent (MTBE with an addition of 1% cyclohexane) was added. After another thorough stirring and separation of individual phases, the upper organic layer was placed into a micro test tube together with the produced emulsion centrifuged and the clear organic phase was dried up with anhydrous magnesium sulphate. Extract samples, adjusted in this way, were thereafter used for the GC-MS analysis.

Instruments: Shimadzu GC-17A, Autosampler: AOC – 5000, Detector: QP-5050A, Software: GCsolution. Program: LabSolutions, GC MS solution. Version 1.20, Conditions of separation: column: DB-WAX 30m  $\times$  0.25mm; 0.25 $\mu\text{m}$  stationary phase (polyethylene glycol). Voltage of the detector 1.5 kV. Individual compounds were identified on the base of MS spectrum and retention time using NIST 107 library, which contains 107,886 spectra.

## 2.6. Estimation of antioxidant activity by the DPPH method

150  $\mu\text{L}$  volume of the reagent (0.095 mM 2,2-diphenyl-1-picrylhydrazyl - DPPH $\cdot$ ) was incubated with 15  $\mu\text{L}$  of wine sample. The absorbance was measured at 505 nm for 10 minutes and the output ratio was calculated as a difference between absorbance values measured at the 10<sup>th</sup> minute and the 2<sup>nd</sup> minute of the assay procedure.

## 3. RESULTS AND DISCUSSIONS

For experiments, aromatic cultivar Moravian Muscat was used. The weight of 50 berries was 67.8 g at harvest time. The juice was quartered, each of these four parts was macerated

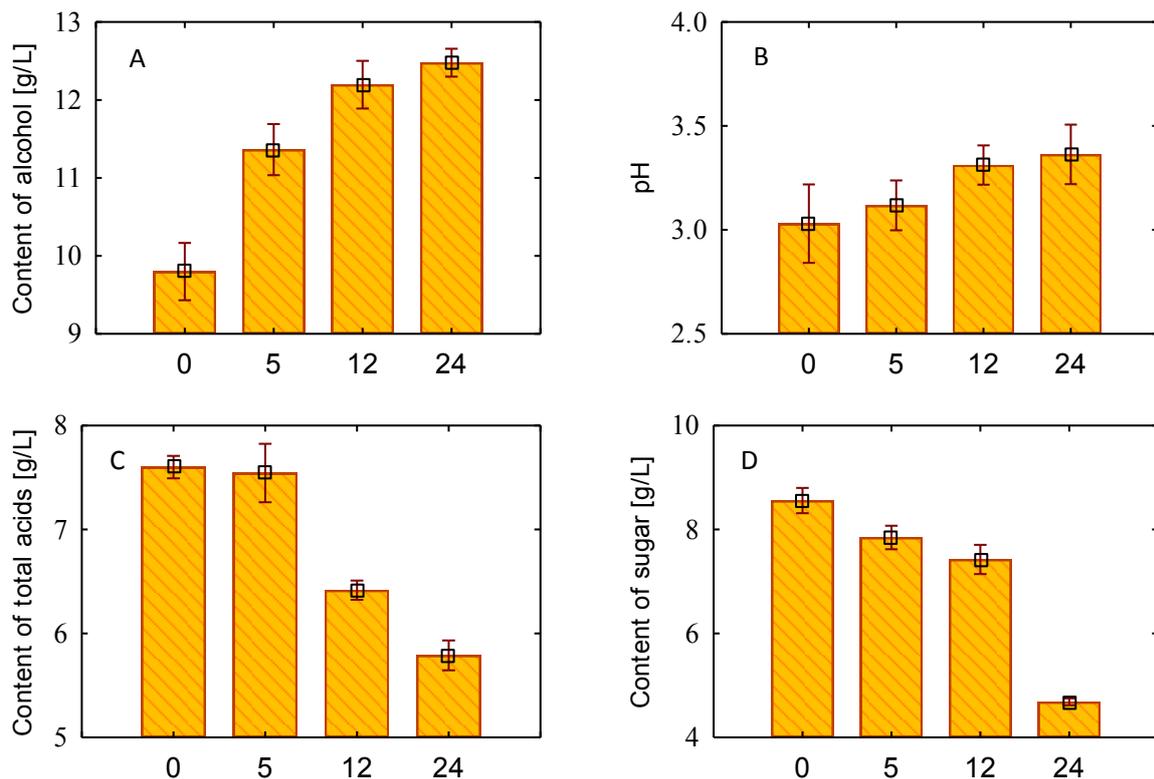
for a different time interval and thereafter used for wine making. The following qualitative parameters were estimated in each part of experimental juice: weight of berries, sugar content, content of titratable acids, pH, content of yeast assimilable nitrogen and aromatic compounds. Contents of aromatic compounds were estimated also in wine made from individual parts of berries. The aim of this experiment was to find out if the length of the maceration period influenced the content of terpenic substances in final wine product.

### 3.1. Estimation of basic analytical parameters

Values of basic analytical parameters are presented in Fig. 1.

The lowest content of alcohol was determined in Variant 1 (9.79 g.L<sup>-1</sup>); this value was also correlated with the highest level of residual sugars (8.55 g.L<sup>-1</sup>). The lowest content of residual sugars and the highest content of alcohol was found out in Variant 4 (4.68 and 12.47 g.L<sup>-1</sup>, respectively).

The highest and the lowest contents of total titratable acids (i.e. 12.47 g.L<sup>-1</sup> and 5.79 g.L<sup>-1</sup>) were found out in Variants 1 and 4, respectively. In this context it can be concluded that the longer the period of maceration, the lower the amount of total titratable acids in produced wine. This means that their contents decreased with the period of maceration. On the other hand, however, the pH value increased with the period of maceration. The highest pH (3.36) was found out in Variant 4.



**Figure 1.** A - Content of alcohol, B - pH, C - content of total acids, D - content of sugar; in period of maceration: 0, 5, 12 and 24 hours.

### 3.2. Monoterpenes

Linalool is a naturally occurring terpene alcohol that has a characteristic floral scent with spicy and lemon tones. It can be found in the pulp of berries of muscat cultivars. Its content does not change too much during the alcoholic fermentation. Linalool is changed because its oxidation to  $\alpha$ -terpineol that occurs in grapes only in smaller amounts. It can be identified only with difficulties and it does not influence the aroma of wine. The odour of nerol resembles roses and thyme but it is considered to be a little fresher. In the course of fermentation, the content of this monoterpene decreases and it is gradually changed to  $\alpha$ -terpineol. It has a scent of roses and citrus fruit. A rose-like scent of this compound participates significantly in the aroma of muscat wines; however, it was not identified in other varieties. During the alcoholic fermentation, its content is slowly decreasing. In the course of wine ageing, geraniol is transformed to  $\alpha$ -terpineol (JACKSON, 2008).

In general, the content of monoterpenes increased in dependence on the duration of macerations. The content of linalool doubled within 24 hours of maceration. The content of ho-trienol increased from 78.6 to 155.5  $\mu\text{g}\cdot\text{L}^{-1}$  while that of  $\alpha$ -terpineol rose only from 81.7 to 142.4  $\mu\text{g}\cdot\text{L}^{-1}$  and those of nerol and geraniol rose from 5 to 17.1  $\mu\text{g}\cdot\text{L}^{-1}$  and from 20 to 49.8  $\mu\text{g}\cdot\text{L}^{-1}$ , respectively.

Results are presented in Table 1.

**Table 1.** Content of monoterpenes.

Substance		Time of maceration							
		0 hours		5 hours		12 hours		24 hours	
		Avg.	SD	Avg.	SD	Avg.	SD	Avg.	SD
Linalool	$[\mu\text{g}\cdot\text{L}^{-1}]$	362.40	7.49	381.25	5.73	491.63	7.48	724.21	8.48
Ho-trienol	$[\mu\text{g}\cdot\text{L}^{-1}]$	78.67	1.67	92.52	1.39	117.61	1.80	155.55	3.18
$\alpha$ -terpineol	$[\mu\text{g}\cdot\text{L}^{-1}]$	81.73	0.47	89.17	2.03	85.00	2.55	142.41	3.73
$\beta$ -citronellol	$[\mu\text{g}\cdot\text{L}^{-1}]$	2.96	0.07	4.98	0.03	5.02	0.03	8.05	0.09
Nerol	$[\mu\text{g}\cdot\text{L}^{-1}]$	5.08	0.10	5.92	0.07	8.03	0.12	17.17	0.29
Geraniol	$[\mu\text{g}\cdot\text{L}^{-1}]$	20.07	0.70	26.35	0.60	29.60	0.69	49.81	1.06
Epoxyllinalol 1	$[\mu\text{g}\cdot\text{L}^{-1}]$	129.25	3.03	144.96	4.40	187.47	4.27	346.00	3.46
Epoxyllinalol 2	$[\mu\text{g}\cdot\text{L}^{-1}]$	78.67	1.67	106.08	2.08	131.77	2.05	206.61	2.40
2,6-dimethyl-3,7-Octadiene-2,6-diol	$[\mu\text{g}\cdot\text{L}^{-1}]$	607.01	6.01	701.95	6.95	954.45	9.45	1181.55	17.81

### 3.3. Phenols

Volatile phenolic substance are produced step by step in the course of wine making by means of enzymatic decomposition of phenolic acids present in berries. The principal compounds are the following: 4-vinylphenol, 4-vinylguaiacol, 4-ethylphenol and 4-ethylguaiacol. In white wines, vinylphenols (4-vinylphenol and 4-vinylguaiacol in concentrations ranging from 10 to 490 and from 70 to 1,150  $\mu\text{g}\cdot\text{L}^{-1}$ , respectively) are predominating while in reds the principal compounds are ethylphenols (4-ethylphenol up to 6 000  $\mu\text{g}\cdot\text{L}^{-1}$ ). In case that the content of ethylphenol is higher than 400  $\mu\text{g}\cdot\text{L}^{-1}$  (sigma 4-ethylphenol + 4-ethylguaiacol), the quality of wine may be negatively influenced; this is usually manifested as an unpleasant wine aftertaste resembling horse sweat (RAPP and VERSINI, 1996).

The content of phenols is presented in Table 2.

The content of 4-vinylguaiacol increased in the course of the whole maceration period. After 24 hours, the initial value of 118.5  $\mu\text{g}\cdot\text{L}^{-1}$  increased to 24 as much as 292.9  $\mu\text{g}\cdot\text{L}^{-1}$ . The content of 4-vinylphenol was fluctuating; however, the difference between the initial and final values, i.e. at the beginning and to the end of maceration (0 h and 24 h) was 992  $\mu\text{g}\cdot\text{L}^{-1}$ .

**Table 2.** Content of phenols.

Substance		Time of maceration							
		0 hours		5 hours		12 hours		24 hours	
		Avg.	SD	Avg.	SD	Avg.	SD	Avg.	SD
Methionol	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	0.39	0.01	2.58	0.03	1.48	0.03	0.51	0.01
2-Methyltetrahydro- phen-3-one	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	0.68	0.02	0.90	0.04	0.59	0.01	0.51	0.01
4-Vinylguaiacol	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	118.58	2.42	208.03	4.43	267.96	6.03	292.85	4.41
4-Vinylphenol	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	155.43	4.15	1029.88	15.47	653.60	14.90	1147.81	28.79
4-Ethylguaiacol	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	10.93	0.23	10.07	0.21	9.87	0.15	8.11	0.18
4-Ethylphenol	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	16.94	0.26	16.11	0.49	13.13	0.13	12.16	0.18
1,1-Diethoxyethane	[ $\mu\text{g}\cdot\text{L}^{-1}$ ]	565.00	22.60	211.15	4.94	630.00	12.60	677.49	7.77

### 3.4. Alcohol and ethyls

According to Dennis (DENNIS *et al.*, 2012) the content of precursors of acetate esters is dependent on their concentration in the juice and on the technology of processing of grapes. These precursors involve above all alcohols. Their concentration increases with the duration of pre-fermentation maceration. These precursors are thereafter transformed to aforementioned ethyls and these influence the aromatic profile of wine.

The content of ethylacetate increased within the whole period of maceration, from the initial value of 32.6  $\text{mg}\cdot\text{L}^{-1}$  to that of 59.7  $\text{mg}\cdot\text{L}^{-1}$ . Contents of ethyl butyrate and ethyl octanoate increased by 150 and 120  $\mu\text{g}\cdot\text{L}^{-1}$ , respectively. The content of isoamyl acetate increased by 92 % to the value of 7.6  $\mu\text{g}\cdot\text{L}^{-1}$ . As far as 2-phenylethyl acetate was concerned, its content increased 6 times within the first 5 hours and thereafter decreased to the level that corresponded with 2.5 multiple of its initial concentration. The initial content of 1-hexyl acetate was 261.7  $\mu\text{g}\cdot\text{L}^{-1}$  and decreased to 43.7  $\mu\text{g}\cdot\text{L}^{-1}$  within the first 5 hours; thereafter, its level increased to final 199  $\mu\text{g}\cdot\text{L}^{-1}$  at the end of the 24 hour period of maceration. The content of alcohols is presented in Table 3.

As far as individual alcohols are concerned, the content of methanol is also important. Its level increased within the whole period of maceration. Within 24 hours, this value increased from 12 to 42  $\text{mg}\cdot\text{L}^{-1}$ . Within the first 5 hours, the content of (E)-3-hexen-1-ol increased. However, after 12 hours it began to decrease again and reached approximately the initial level. The content of (Z)-3-hexen-1-ol increased within the whole 24-hour period of maceration: the initial value of 71  $\mu\text{g}\cdot\text{L}^{-1}$  doubled and was as much as 146  $\mu\text{g}\cdot\text{L}^{-1}$ .

The content of alcohols is presented in Table 4.

**Table 3.** Content of ethyls.

Substance		Time of maceration							
		0 hours		5 hours		12 hours		24 hours	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD
Ethyl acetate	[mg.L <sup>-1</sup> ]	32.63	0.74	36.59	0.87	47.32	0.54	59.70	1.20
Ethyl propionate	[µg.L <sup>-1</sup> ]	109.27	1.27	109.08	1.87	107.71	1.24	85.55	1.81
Ethyl isobutyrate	[µg.L <sup>-1</sup> ]	28.90	0.17	42.14	1.06	46.92	0.92	46.69	0.54
Ethyl butyrate	[µg.L <sup>-1</sup> ]	402.18	8.51	470.00	18.80	505.00	8.66	550.80	9.35
Ethyl hexanoate	[µg.L <sup>-1</sup> ]	518.22	8.05	228.47	4.79	536.90	8.34	573.68	9.84
Ethyl octanoate	[µg.L <sup>-1</sup> ]	489.36	2.83	512.00	10.24	624.24	12.24	608.91	12.76
Ethyl decanoate	[µg.L <sup>-1</sup> ]	54.45	0.95	62.16	0.96	1.00	0.02	125.87	2.56
Ethyl lactate	[mg.L <sup>-1</sup> ]	5.56	0.12	2.71	0.01	2.72	0.03	2.61	0.01
Diethyl uccinate	[mg.L <sup>-1</sup> ]	0.60	0.00	1.01	0.02	1.71	0.01	1.99	0.06
Diethylmalate	[mg.L <sup>-1</sup> ]	3.14	0.07	3.50	0.04	3.09	0.08	2.47	0.05
Monoethyl succinate	[mg.L <sup>-1</sup> ]	2.62	0.04	5.41	0.19	4.63	0.05	3.32	0.10
Gama-butyrolactone	[mg.L <sup>-1</sup> ]	7.70	0.09	21.36	0.25	20.35	0.43	19.70	0.53
Isoamyl acetate	[mg.L <sup>-1</sup> ]	3.94	0.10	5.49	0.10	5.82	0.07	7.60	0.08
2-Phenylethyl acetate	[µg.L <sup>-1</sup> ]	151.50	2.31	932.45	22.12	552.35	12.42	384.28	13.45
1-Propyl acetate	[µg.L <sup>-1</sup> ]	66.33	0.67	39.78	0.78	43.14	1.51	90.70	3.20
Isobutyl acetate	[µg.L <sup>-1</sup> ]	116.61	2.94	198.53	4.04	249.00	9.96	283.05	4.34
1-Hexyl acetate	[µg.L <sup>-1</sup> ]	261.73	7.94	43.71	0.25	93.69	2.37	199.00	0.00
(Z)- 3-Hexen-1]-yl acetate	[µg.L <sup>-1</sup> ]	6.04	0.12	14.19	0.21	14.28	0.24	14.19	0.16

**Table 4.** Content of alcohol.

Substance		Time of maceration							
		0 hours		5 hours		12 hours		24 hours	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD
Methanol	[mg.L <sup>-1</sup> ]	12.34	0.31	13.91	0.29	32.57	0.57	42.11	1.29
Isoamylalcohol	[mg.L <sup>-1</sup> ]	56.19	0.32	136.65	4.78	134.64	4.71	109.92	3.33
Isobutylalcohol	[mg.L <sup>-1</sup> ]	6.33	0.07	28.08	0.48	25.00	0.25	22.17	0.78
2-Phenylethanol	[mg.L <sup>-1</sup> ]	9.73	0.20	68.21	1.21	52.29	0.81	19.80	0.00
1-Propanol	[mg.L <sup>-1</sup> ]	19.69	0.41	9.96	0.24	12.63	0.15	35.14	0.36
1-Butanol	[µg.L <sup>-1</sup> ]	1240.67	14.42	1247.15	29.19	1967.58	38.58	7130.90	146.01
1-Hexanol	[µg.L <sup>-1</sup> ]	1508.00	45.24	1292.80	12.80	915.85	19.19	888.00	20.78
(E)-3-Hexen-1-ol	[µg.L <sup>-1</sup> ]	84.15	1.47	114.00	3.42	101.00	1.00	88.16	2.01
(Z)-3-Hexen-1-ol	[µg.L <sup>-1</sup> ]	71.76	1.10	126.00	5.04	147.46	2.53	146.03	1.71
3-Methyl-1]-pentanol	[µg.L <sup>-1</sup> ]	6.02	0.03	18.68	0.40	13.95	0.49	11.11	0.19
Benzylalcohol	[µg.L <sup>-1</sup> ]	111.72	2.28	123.22	3.23	305.00	0.00	410.63	2.38
2,3-Butandiol	[mg.L <sup>-1</sup> ]	346.61	5.37	552.57	6.29	1309.57	26.81	1497.02	14.82
Propandiol	[mg.L <sup>-1</sup> ]	6.60	0.07	8.94	0.19	23.64	0.50	19.05	0.29

### 3.5. Antioxidation activity

As compared with red wines, a lower antioxidant capacity of white ones is caused by a lower content of phenolic compounds (VINSON and HONTZ, 1995). A higher content of phenolic in red wine results is caused by the period of maceration during which phenolic compounds are released from skins, seeds, stalks and pulp of berries (FUHRMAN *et al.*, 2001). Because in white wines the maceration usually does not take place, their content of phenolic substances is limited and also their antioxidant activity is reduced (LAMUELA-RAVENTOS and DE LA TORRE-BORONAT). For that reason the maceration represents an interesting (and natural) step when producing white wines because it enables extraction of phenolic compounds and, thus, production of wine with strong antioxidant properties.

In the course of the first 12 hours of maceration, the antioxidant activity (as measured by the DPPH assay, increased and thereafter remained without changes (i.e. constant) during 24 hours of measuring. After 12 hours, it was even more than three times higher than at the beginning. Dependence of antioxidant activity on length of maceration is depicted in Fig. 2.

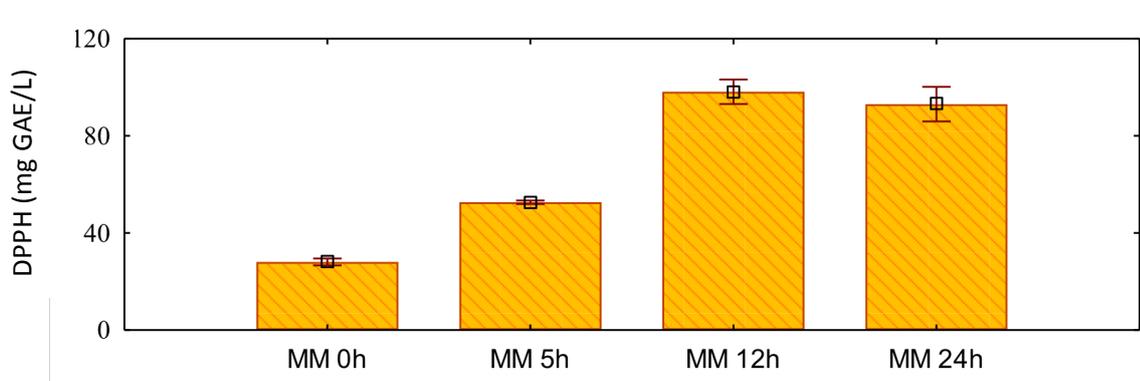


Figure 2. Antioxidation activity.

### 3.6. Time of maceration

Correlations existing between contents of individual aromatic substances and the time of maceration are presented in Table 5. They are expressed as the Pearson's correlation coefficient and characterize the tightness of individual relationships. Values between 0.1 and 0.3 indicate a weak correlation while those between 0.4 and 0.6 and between 0.7 – 0.8 indicate medium and strong correlations, respectively. Values above 0.9 mean that the correlation is very strong.

**Table 5.** Correlations existing between contents of individual substances and the period of maceration. Significant correlations ( $p < .05000$ ) are in red.

Linalool	0.982	Ethyl butyrate	0.939	1-Propanol	0.712
Ho-trienol	0.998	Ethyl hexanoate	0.467	1-Butanol	0.926
$\alpha$ -terpineol	0.891	Ethyl octanoate	0.823	1-Hexanol	-0.894
$\beta$ -citronellol	0.961	Ethyl decanoate	0.531	(E)-3-Hexen-1-ol	-0.169
Nerol	0.963	Ethyl lactate	-0.679	(Z)-3-Hexen-1-ol	0.784
Geraniol	0.977	Diethyl succinate	0.951	3-Methyl-1-pentanol	0.105
Epoxylinolol 1	0.969	Diethylmalate	-0.834	Benzylalcohol	0.967
Epoxylinolol 2	0.994	Monoethyl succinate	-0.039	2,3-Butandiol	0.935
2,6-dimetyl-3,7-octadiene-2,6-diol	0.990	Gama-butyrolactone	0.575	Propandiol	0.745
Methionol	-0.250	Isoamyl acetate	0.969	Acetic acid	0.884
2-Methyltrathiophen-3-on	-0.698	2-Phenylethyl acetate	-0.019	Propionic acid	0.569
4-Vinylguaiacol	0.903	1-Propyl acetate	0.587	Butyric acid	0.953
4-Vinylfenol	0.711	Isobutyl acetate	0.923	Isobutyric acid	0.967
4-Ethylguaiacol	-0.966	1-Hexyl acetate	-0.004	Isovaleric acid	-0.315
4-Ethylphenol	-0.941	(Z)- 3-Hexen-1-yl acetate	0.655	2-methylbutanoic acid	-0.003
1,1-Diethoxyetane	0.532	Methanol	0.962	Hexanic acid	0.347
Ethyl acetate	0.994	Isoamylalcohol	0.386	Octanoic acid	0.144
Ethyl propionate	-0.899	Isobutylalcohol	0.451	Decanoic acid	-0.111
Ethyl isobutyrate	0.779	2-Phenyletcanol	-0.110	DPPH GA	0.880

#### 4. CONCLUSIONS

In these experiments, interesting volatile compounds characterizing the cultivar Moravian Muscat. Wine production from aromatic cultivars such a Moravian Muscat is a complex process depended on the health condition of grapes, temperature and length of the maceration period. Results of this study indicate that the content of individual compounds is changing in the course of maceration. For that reason it is important to pay attention to its length and create favourable conditions enabling the development of wine character. The content of terpenic compounds (especially of monoterpenes) is increasing above all with the increasing time of maceration. This increases not only the antioxidant activity of wine but also the content of ethyls that can show a negative effect on the aromatic profile of produced wine. However, the content of total acids decreases. The only exception represents the content of acetic acid that markedly increases with the length of maceration so that the quality of produced wine is deteriorated.

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