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Application News

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Gas Chromatography

Quantification of higher alcohols and ethyl acetate in wine using liquid extraction and GC-2030 with FID

▪ Introduction

The taste and aroma of wines can cohere to the amount of different alcohols and solvents like methanol, butanol and ethyl acetate [1]. Therefore winemakers are interested in a quantification of these substances. Controlling fermentations for better taste, and even more importantly for decreasing health damages by other alcohol than drinking alcohol, is a main concern.

Determining these compounds in the newest Shimadzu wine „Science Selection – Grauburgunder Spätlese“ with the gas chromatograph GC-2030 and a flame ionisation detector (FID) is a fast and efficient way to measure the amount of ethyl acetate, isoamyl alcohol, n-propanol, isobutanol and even methanol without need for headspace usage. A second brand of wine, dry red wine, was also tested and compared with the Shimadzu white wine. The sample preparation was done by liquid extraction with organic solvent in a milliliter scale. 2-Butanol as an internal standard (ISTD) and response factors are used for calibration.

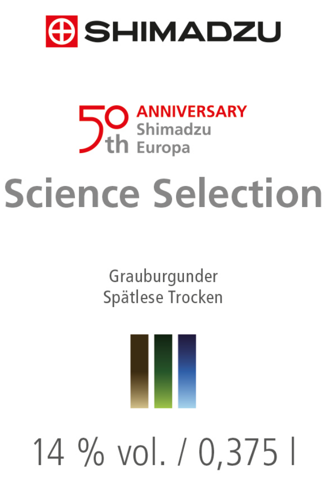

Figure 1: Shimadzu wine „Science Selection – Grauburgunder Spätlese“

▪ Experimental

A standard solution and an internal standard solution are mixed for identification and quantification of the alcohols. To create both stock solutions, the alcohols are weight into a volumetric flask and filled up with ethanol to the concentrations given in table 1.

ISTD solution	Standard stock solution [g/L]		Calibration solution [mg/L]
2-butanol:	isoamyl alcohol:	10	100
10 g/L (stock)	ethyl acetate:	10	50
in sample	methanol:	20	200
and calibration:	n-propanol:	5	50
100 mg/L	isobutanol:	5	50

Table 1: Concentrations of the internal standard and the standard stock solutions as well as the calibration solution.

To improve reliability of the results, the calibration is performed in matrix. As wine matrix a solution with 12 vol. % ethanol in water with pH = 3.5, adjusted with tartaric acid, is used. 50 µL of each standard solution (ISTD, Standard) are added to 4.9 mL of this matrix, then mixed with 0.5 g NaCl and 2 mL dichloromethane for 30 seconds with a vortex mixer. This results e.g. in a concentration of ca. 100 mg/L for ethyl acetate in this calibration solution. The dried organic phase (lower phase) is injected into the GC.

The preparation of a real sample is similar. This procedure needs 5 mL cold wine sample and 50 µL internal standard solution. After vortexing with NaCl and dichloromethane, the phase separation is done at room temperature for 10 min followed by 5 min centrifugation at 2900 rpm.

Parameter	Value
injector volume	1 µL, Split 1:10
injector temperature	220 °C
carrier gas	He
carrier gas speed	30 cm/s, linear velocity mode
column oven program	40 °C, 3min, 5 °C/min, 50 °C, 4 min, 5 °C/min, 70 °C, 3 min, 45 °C/min, 240 °C, 5 min
FID temperature	260 °C

Table 2: Method parameters for alcohols and ethyl acetate in wine

The dried organic phase is ready for injection. Table 2 shows the GC method parameters. As column a SH-Rxi-624Sil MS 30 m length, 0.25 mm ID and 1.4 µm df was used. The analysis time was optimized to be 25 min.

Calculation

The calculations of the concentrations are done via response factors RF of each analyte with following equation (1)

$$RF = \frac{c_{alcCAL} \times A_{ISTD-CAL}}{c_{ISTD-CAL} \times A_{alcCAL}} \quad (1)$$

c_{alcCAL} and A_{alcCAL} are the concentration and the area of each alcohol in the calibration solution. ISTD-CAL stands for the internal standard values (2-butanol) of the calibration.

With these response factors RF of the calibrations, the concentrations c_i of the alcohols in the sample are calculated by equation (2) using the area and concentration values of the internal standard ($A_{ISTD-sample}$, $c_{ISTD-sample}$), which was added to the sample, and the measured area of the alcohol in the sample ($A_{alc-sample}$)

$$c_i = RF \times \frac{c_{ISTD-sample} \times A_{alc-sample}}{A_{ISTD-sample}} \quad (2)$$

With the values given in table 3, for ethyl acetate in Shimadzu wine e.g. the RF is 0.929. With a concentration of 97 mg/L in the calibration solution the concentration in the sample is determined to be 58 mg/L.

Results

Table 4 summarizes the results for both wines measured and calculated with the method above. Figure 2 shows a chromatogram of the analyzed Shimadzu wine.

Both wines tested show similar values for n-propanol and ethyl acetate. The main differences were found for methanol, isobutanol and isoamylalcohol of which the red wine contains much higher amounts. These results correlate to average concentrations of methanol in red and white wines stated in the literature.

	2-butanol (ISTD)		Ethyl acetate	
	Area (A)	concentration (c) [mg/L]	Area (A)	concentration (c) [mg/L]
calibration (CAL)	242158	100	250688	97
sample	246513	100	152138	58

Table 3: Areas and concentrations of ethyl acetate and ISTD for example calculation (based on realistic weighing)

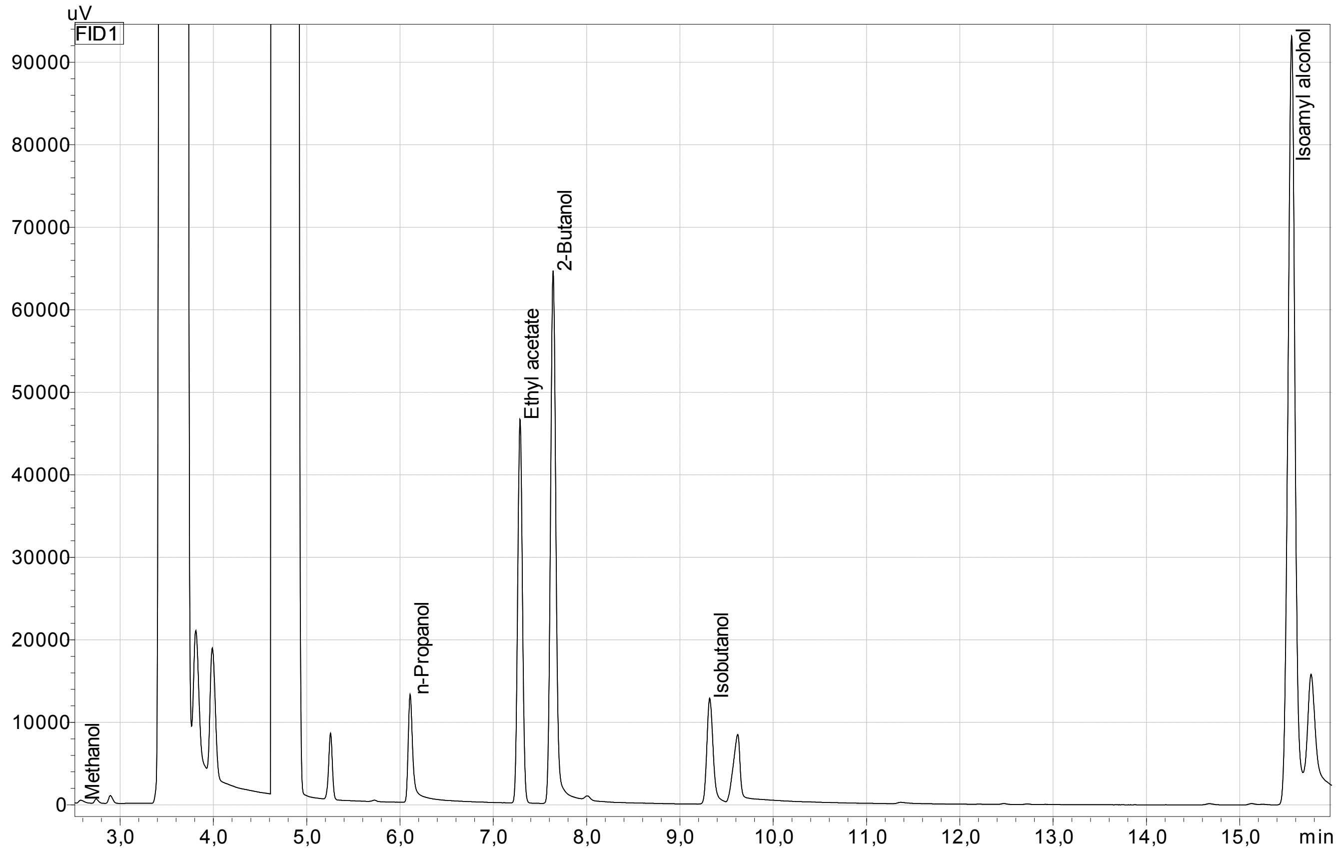

Figure 2: Chromatogram of the analyzed Shimadzu wine

Compound	Red wine [mg/L]	Shimadzu wine [mg/L]
methanol	170	28
n-propanol	34	39
ethyl acetate	68	58
2-butanol (ISTD)	100	100
isobutanol	41	24
isoamylalcohol	241	120

Table 4: Concentration in mg/L of alcohols and ethyl acetate in both wines analyzed

According to [2] there is about 60 to 230 mg/L methanol in red wines and 17 to 100 mg/L in white wines. Furthermore, a common method from the OIV (International Organisation of Vine and Wine) for methanol provided similar results in their validation tests of the methanol analytic method for GC.

▪ Conclusion

With the identification of alcohols and other organic compounds it is possible to support the aroma profiling for wines and monitor their quality. GC-2030 with FID detection offers an efficient way to analyze these without the need for advanced equipment and complicated sample preparation.

▪ Literature

[1] Oenology research, Winetech Technical, How higher alcohols and volatile phenols impact on key aromas, Russell Moss, 2015

[2] https://glossar.wein-plus.eu/methanol 2018/6/8

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