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Application News

No. AD-0168

Beer Analysis / HS GC-ECD

Quantitative Analysis of Vicinal Diketones Off-flavor in Beer by Static Headspace GC-ECD

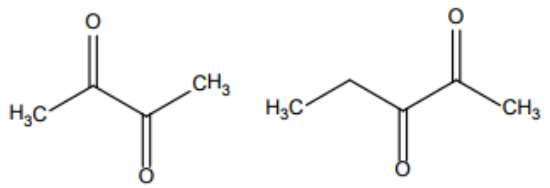
❑ Introduction

Flavor of beer is one of important quality attributes and analysis of flavor components by HS-GC-FID was established [1]. However, brewing process also generates off-flavor components in beer like vicinal diketones (VDKs), such as diacetyl and 2,3-pentanedione [2]. These off-flavor compounds cause undesirable buttery and honey/toffee tastes in beer [3,4]. Hence, brewers employ several strategies to minimize the amount of VDKs during fermentation [5]. Monitoring the concentrations of VDKs is therefore important to examine the beer quality. In this study, a GC-ECD coupled with static HS technique is employed. Electron capture detector (ECD) provides a high selectivity and sensitivity detection for low concentrations of diacetyl and 2,3-pentanedione in beer.

❑ Experimental

Materials and Analytical Conditions

The study was performed using GC -ECD coupled with headspace autosampler (HS). Vicinal diketones (VDKs) compounds, i.e. diacetyl and 2,3-pentanedione (Figure 1), along with internal standard (IS), 2,3-hexanedione, were separated using a SH-Rxi-624Sil MS column. Both data acquisition and processing were performed using Labsolutions workstation. Analytical conditions are presented in Table 1 (A & B).



Diacetyl 2,3-pentanedione

Figure 1: Diacetyl and 2,3-pentanedione are common off-flavor compounds present in beer

Headspace and GC Operation

Static headspace (HS) is an extraction method by heat, which would extract volatile compounds such as VDKs, without extracting the non-volatile materials from the beer matrix. The vapor containing VDKs was then transferred to gas chromatograph (GC) for separation and detection. Mixed standard or beer sample (10 mL) spiked with IS of 50 µg/L was added to a headspace vial and capped immediately before analysis.

Table 1(A): Static headspace (HS) condition

Incubation temp	60 °C
Equilibrating time	30 min
Sample line temp	150 °C
Transfer line temp	150 °C
Shaking level	Off
Pressurizing time	1 min (equilibrium time: 0.1 min)
Load time	0.5 min (equilibrium time: 0.1 min)
Injection time	1 min
Needle flush time	1 min
Pressurizing Pressure	60 kPa

Table 1(B): GC analysis condition

Injection mode	Split
Split Ratio	20
Carrier gas	He
Flow control mode	Linear velocity
Linear velocity	33.9 cm/s
Column	SH-Rxi-624Sil MS (60 m L, 0.32 mm ID, 1.8 µm df)
Column temp program	80^0C → rate: 10^0C/min → 120^0C (7 min)
Detector	ECD
Detector temp	150 °C
ECD current	1 nA
Make-up (N_2) flow	60ml/min

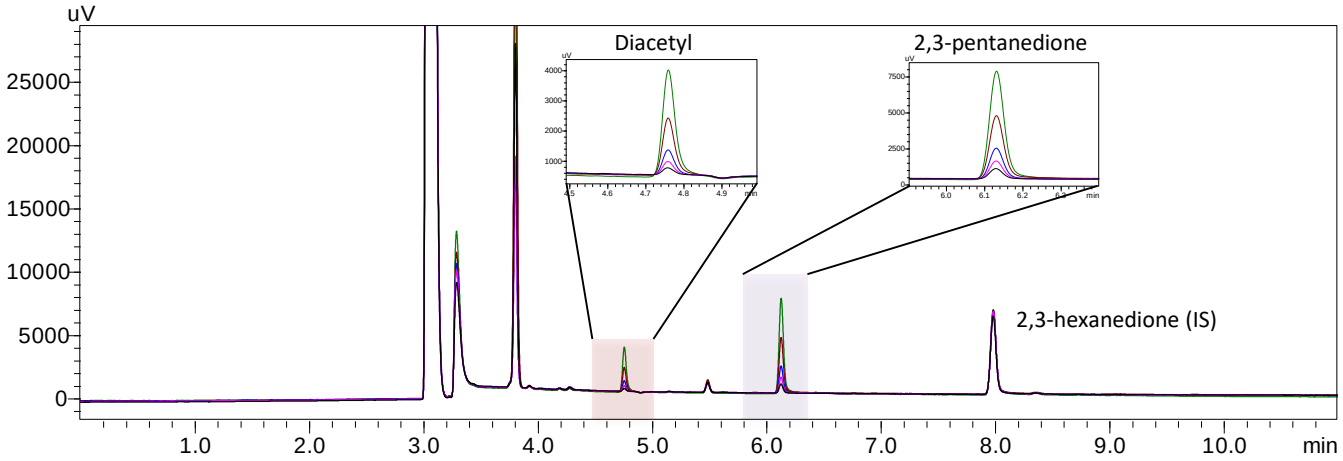


Figure 2: Overlay chromatograms of mixed standards (5, 10, 20, 50, 100 µg/L) with IS (50 µg/L)

❑ Results and Discussion

Calibration Curves

Figure 2 shows the chromatograms of mixed standards of VDKs and internal standard. Five-level internal standard (IS) calibration curves at 5, 10, 20, 50 and 100 µg/L were constructed for diacetyl and 2,3-pentanedione (Figure 3). Excellent linearity was obtained for both compounds.

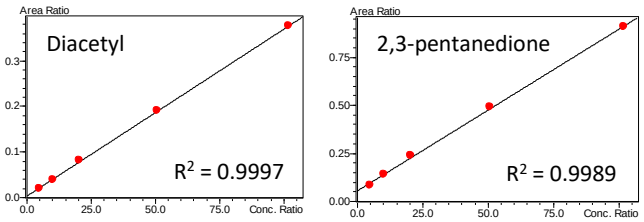


Figure 3: Five level calibration curves of Internal standard method (IS: 50 µg/L)

Quantitation Results

Quantitation of beer samples was carried out using the method. The quantitation results are displayed in Table 2. This shows that the concentrations of diacetyl and 2,3-pentanedione are different in various beer brands. These concentrations are much lower than the reported threshold levels of diacetyl (0.1-0.2 ppm in lager and 0.1-0.4 ppm in ales) and 2,3-pentanedione (0.9-1.0 ppm) [6].

Table 2: Quantitation results of three beer samples

Off-Flavor compound	Concentration (µg/L)		
	Beer 1	Beer 2	Beer 3
Diacetyl	31.32	6.964	17.465
2,3-pentanedione	16.348	Not Detected	24.359

❑ Conclusions

A static HS-GC-ECD method was developed for high sensitive determination of VDKs off-flavor compounds (diacetyl and 2,3-pentanedione) in beer. This method is appropriate to monitor the VDKs contents in various beer brands produced by different brewing conditions. The results can be used to review manufacturer brewing process and strategies to minimize VDKs in the finished beer products.

❑ References

1. Shimadzu Application News, AD-0166, *Quantitative Analysis of Flavours in Beer by Static Headspace-GC-FID* (2017)
2. He, Y., et al., *Wort composition and its impact on the flavour-active higher alcohol and ester formation of beer – a review*, Journal of the Institute of Brewing, Vol 120, Issue 3, 2014, 157-163.
3. Palmer, J., How to brew: everything you need to know to brew great beer everytime, Brewers Publications, 2017.
4. Krogerus, K., Gibson, B., *Influence of valine and other amino acids on total diacetyl and 2,3-pentanedione levels during fermentation of brewer's wort,* Appl Microbiol Biotechnol, Aug 2013, 97(15), pp. 6919-6930
5. Hui, Y., et al., Handbook for food products manufacturing, John Wiley & Sons, New Jersey, 2007.
6. Krogerus, K., Gibson B., 125th anniversary review: *diacetyl and its control during brewery fermentation*, J. Inst. Brew. 2013; 119: 86-97.

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Disclaimer: The Shimadzu GC-ECD and Headspace system and the data in this Application News are intended for Research Use Only (RUO). Not for use in diagnostic procedures.

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