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Correlation of an automated discrete analysis sulfur dioxide method to standardized para-rosaniline methods in the analysis of beer

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Keywords

Discrete Analyzer, Photometric Analyzer, Gallery Analyzer

Goal

To compare the total sulfur dioxide test to standardized *p*-rosaniline methods for beer analysis

Introduction

In beer, sulfur dioxide (SO_2) originates from yeast metabolism and reacts with carbonyl compounds to form hydroxysulfonates. Hydroxysulfonates react with the carbonyl compounds in beer and produce a stale, unwanted flavor. Sulfur dioxide also plays an important role as an antioxidant and is known to exert antimicrobial properties at high concentrations. Its concentration is controlled at the end of beer production to ensure beer quality.

Sulfur dioxide is typically measured by the European Brewery Commission (EBC) Method 9.25.3 or by the similar American Society of Brewing Chemists (ASBC) Beer-21 para-rosaniline (*p*-rosaniline) method. This study shows correlation between the *p*-rosaniline method and the total SO_2 method that is based on 5, 5′-dinitrobenzoic acid (DTNB) measurement at 405 nm. Sulfur dioxide can be found in beer in its free forms, SO_2 (gas) and bisulfate ion (HSO_3^-), or bound to compounds that incorporate a carbonyl group, such as acetaldehyde. Free forms of SO_2 are pH and temperature dependent and because of the acidic nature of beer, SO_2 is usually present and measured as a bisulfate ion (HSO_3^-) with results reported as SO_2.

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Commercial beer and cider samples were analyzed by the Thermo Scientific™ Gallery™ discrete analyzer using the total sulfur dioxide test, a rapid two-reagent method that does not require sample pretreatment. A fully automated table top photometric analyzer enables simultaneous analysis of multiple parameters from the same sample.

According to this study, all samples analyzed by the total SO_2 method with the Gallery discrete analyzer show results similar to EBC (9.25.3) and ASBC Beer-21 *p*-rosaniline methods. The method is linear from 2 to 10 mg/L without dilution and with automated dilution total.

Experimental

Materials and methods

Instruments

A Gallery discrete analyzer (shown in Figure 1) from Thermo Fisher Scientific, Vantaa, Finland was used.

Reference values were obtained using a spectrophotometer in a third party beer analysis laboratory.

Figure 1. Thermo Scientific Gallery discrete analyzer

Total SO_2 method principle

The Thermo Scientific method is based on a reaction between SO_2 and DTNB in basic conditions and is performed at 37 °C, using a 405 nm filter for detection. A 700 to 750 nm filter is used for a side wavelength. The purpose of measuring a side wavelength is to eliminate variations, e.g., the effect of bubbles which may appear in the cuvettes. The side wavelength is measured from the area of the spectrum where no reaction occurs.

Application for beer samples

The automated Gallery SO_2 Total application consists of two reagents, an end-point measurement with a sample blank, and a linear calibration curve used for the calculation of results. The measuring range is from 2 to 50 mg/L. In this method 120 µL of SO_2 Total Blank reagent (a buffer pH ≥ 7.0) and 15 µL of sample are incubated for 180 seconds and then a blank measurement is taken. After a 50 µL addition of SO_2 Total Reagent and 360 seconds incubation, the reaction is measured at 405 nm with a side wavelength of 700 nm.

Application for cider samples

The automated Gallery SO_2 Total high application consists of one reagent, an end-point measurement, and a linear calibration. The same SO_2 Total kit is used to analyze cider samples but is adapted to samples with high concentrations. The measuring range is from 2 to 300 mg/L. The application uses a true sample blank that blanks the reaction by replacing the reagent with blank reagent. A true sample blank uses two cuvette positions to eliminate the effect of turbidity and color. Initially, 120 µL of SO_2 Total reagent is incubated for 120 seconds. After a 3 µL addition of sample and 300 seconds incubation, the reaction is measured at 405 nm with a side wavelength of 700 nm.

Reagents and calibrator

A Thermo Scientific™ Gallery™ system reagent SO_2 Total kit (P/N 984345) was used. Reagent 1 (R1) is buffer (pH ≥ 7.0) and reagent 2 (R2) includes buffer (pH ≥ 7.0) and DTNB (< 0.2 mM).

Preparing the calibrator

Dissolve 2.5 g of citric acid and add 400 µL of acetaldehyde in 500 mL of distilled water to form diluent A. Weigh precisely 0.0303 g of sodium metabisulfite ($Na_2S_2O_5$ MW = 190.11 g/mol, purity ≥ 98%) into a 100 mL volumetric flask and fill to the mark with diluent A. The resulting solution has a SO_2 concentration of 200 mg/L and must be made fresh daily.

Samples

Twelve commercial beer samples and two cider samples were analyzed. Reference values were determined using the *p*-rosaniline method by a third party. No sample pretreatment was used. The samples were homogenous liquids requiring no centrifugation or degassing and results could be repeated with precision. The buffer included in the reagents adjusts the pH in the samples to levels optimized for the reaction.

Results and discussion

Correlation between the Gallery system reagent SO_2 Total and reference values of randomly selected beer and cider samples measured by the *p*-rosaniline method are shown in Table 1.

The total SO_2 method shows good precision for all samples and for most samples CV% is less than 1.5%. However, beer sample #5 showed relatively high variation at 10.2%. The combination of a low SO_2 concentration and a very dark sample color (> 220 EBC units) was determined to decrease the method sensitivity. The most common beer color is a pale amber (EBC units ≤ 12). Because of the low concentrations of SO_2 in beer, samples cannot be diluted. In addition, most color removing agents will also remove SO_2 or react with the reagents further invalidating the test.

Linearity is shown in Figure 2. Water-based standards were used to determine method linearity. The method is linear from 2 to 10 mg/L without dilution and from 2 to 50 mg/L with an automated sample dilution of 1:4. As a result, the method is suitable for all beer samples tested with no additional predilutions.

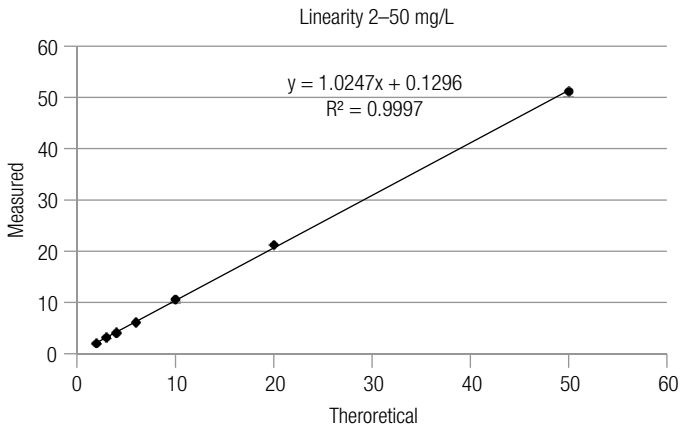

Figure 2. Method linearity

Cider samples were analyzed with a separate high application which is also suitable for determining SO_2 in white wines.

Table 1. Value correlation between the Gallery system reagent SO_2 Total and reference values measured by the *p*-rosaniline method

Sample	SO_2 Total (mg/L)	Ref. Value (mg/L)	Bias	CV% (n=10)
Beer 1	9.1	10	–0.9	1.4
Beer 2	4.8	5	–0.2	0.8
Beer 3	7.9	8	–0.1	0.9
Beer 4	3.6	2	1.6	2.5
Beer 5	2.4	2	0.4	10.2
Beer 6	2.8	2.3	0.5	3.4
Beer 7	2.9	1.9	1.0	3.5
Beer 8	5.3	4.4	0.9	1.4
Beer 9	3.5	2.7	0.8	1.1
Beer 10	5.1	3.8	1.3	1.3
Beer 11	1.2	0.8	0.4	1.4
Beer 12	5.4	5.7	–0.3	0.9
Cider 1	54.7	57	–2.3	3.3
Cider 2	76.3	80	–3.7	2.5

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Conclusions

According to this study, all samples tested by the SO_2 Total method showed results similar to the EBC (9.25.3) *p*-rosaniline method. The Gallery method was easy to use and provided a robust method for SO_2 Total measurement for both cider and beer samples. Ready-to-use Thermo Scientific system reagents eliminated reagent preparation, which saved time, and the volume-optimized methods minimized reagent waste. Thermo Scientific system reagents along with the fully automated Thermo Scientific discrete analyzers provide a total system solution for food and beverage analysis.

References

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