

1. Introduction

After the fermentation process, beer contains a characteristic profile of residual sugars. Beer containing a high amount of such residual sugars is a full-bodied beer and often has a sweeter taste. Different sugars have specific characteristics and will influence the taste and mouthfeel of the beer. There are four major classes of carbohydrates in beer, namely monosaccharides, disaccharides, oligosaccharides and polysaccharides. Monitoring these sugars during the brewing process gives an indication of the fermentation status.

In this study, a HPLC separation of Glucose, Maltose, Maltotriose (DP-3), Maltotetraose (DP-4), Maltopentaose (DP-5), Maltohexaose (DP-6) and Maltoheptaose (DP-7) in beer samples was developed to enable monitoring of the brewing process.

2. Materials and Method

2.1 Sample pretreatment

10 mL commercially available beer samples were mixed with 10 mL acetonitrile/water (50:50 v/v). The mixture was degassed by sonication. After that, samples were filtered through a syringe filter (0.45 µm) and analyzed by HPLC (Fig.1).

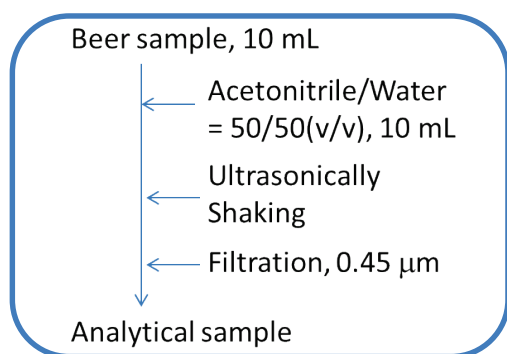


Figure 1: Beer sample pretreatment

The sugar standard (containing glucose, maltose, and DP3-DP7) was prepared in a concentration range of 200 - 2000 mg/L in acetonitrile/water (50:50 v/v).

2.2 Analytical conditions

System: Shimadzu Nexera X2 UHPLC system
 Column: Shim-pack GIST NH₂ (150 x 4.6 mm, 3 µm)
 Mobile Phase: 0.01 % H₃PO₄ in H₂O/ACN (30:70 v/v)
 Flowrate: 1.0 mL/min
 Column Temp.: 40 °C
 Injection Vol.: 5 µL
 Detection: RI detector

3. Results

3.1 Standard analysis

Separation of carbohydrate standards is shown in figure 2. where baseline separation of the sugars was achieved. The refractive index detector provided sufficient sensitivity. Retention stability was obtained by adding a small amount of phosphoric acid to the mobile phase.

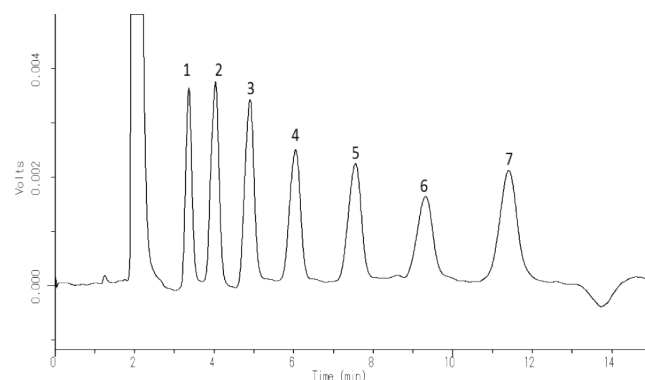


Figure 2: Chromatogram of 1g/L carbohydrate standard

(1. Glucose, 2. Maltose (DP-2), 3. Maltotriose (DP-3), 4. Maltotetraose (DP-4), 5. Maltopentaose (DP-5), 6. Maltohexaose (DP-6), 7. Maltoheptaose (DP-7)

Fig. 3 shows the calibration curves of each compound in the range of 0.1 – 2 g/L. All graphs demonstrate good linearity with an R² ≥ 0.997.

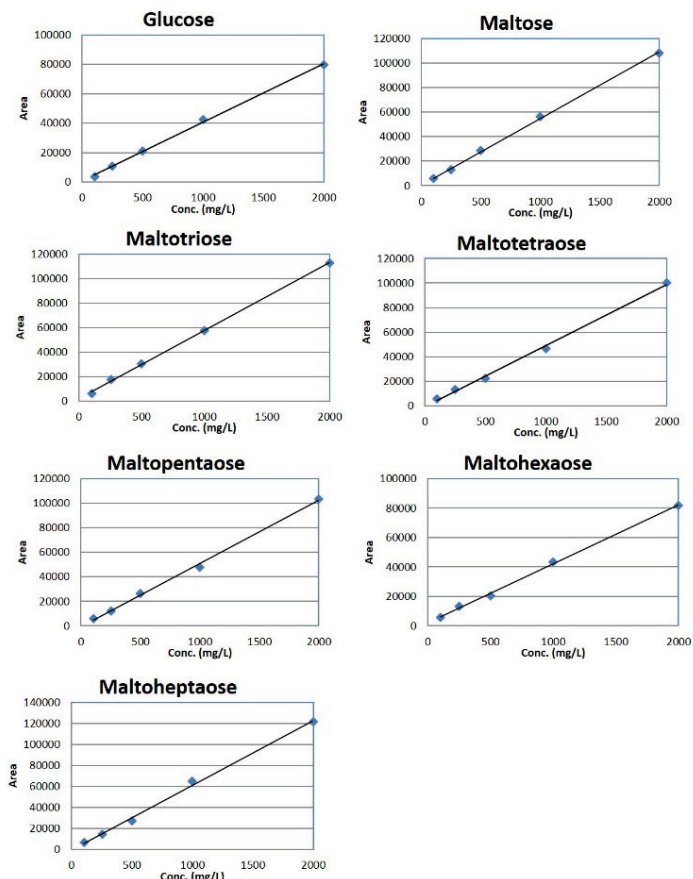


Figure 3: Calibration curves of carbohydrates in the range of 0.1 – 2 g/L with $R^2 \geq 0.997$

3.2 Beer samples analysis

Figure 4 shows the analysis of two different commercially available beer samples (A and B). Chromatograms of beer A and B show different sugar profiles.

Table 1 summarizes the quantitation of the seven carbohydrates analyzed (glucose and DP-2 - DP-7) in both beer samples. Maltoheptaose was not detected in either beer sample. Comparing the two, beer A (made in Switzerland) contains a higher concentration of all carbohydrates, indicating a sweeter taste.

Table 1: Quantification results for carbohydrates in beer

Carbohydrate	Conc. mg/L	
	Beer A	Beer B
Glucose	540	64
Maltose DP-2	1684	426
Maltotriose DP-3	875	702
Maltotetraose DP-4	3893	3171
Maltopentaose DP-5	1317	698
Maltohexaose DP-6	899	189
Maltoheptaose DP-7	ND	ND

(n = 1; variation not determined)

4. Conclusion

This study points out that the developed method is able to analyze different carbohydrates in beer by HPLC on a Shim-pack GIST NH₂ column. Using a small amount of phosphoric acid in the mobile phase helps to stabilize the retention and increase the sensitivity. Using this method the characteristic carbohydrate profile of different beer brands as well as the status of fermentation during the brewing process can be determined.

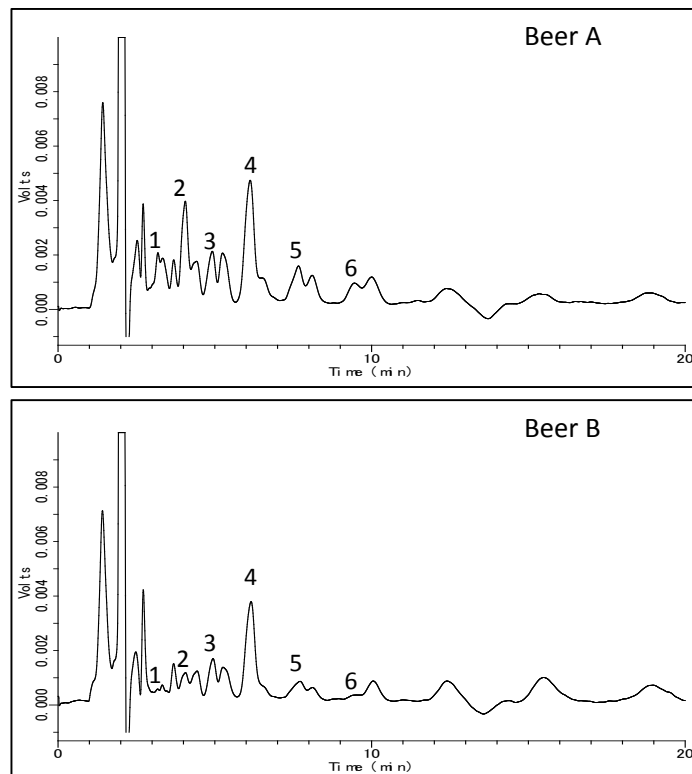


Figure 4: Chromatograms of analysis of carbohydrates in beer samples

1. Glucose, 2. Maltose (DP-2), 3. Maltotriose(DP-3), 4. Maltotetraose(DP-4), 5. Maltopentaose(DP-5), 6. Maltohexaose(DP-6), 7. Maltoheptaose(DP-7)

Beer A: Commercially available beer made in Switzerland

Beer B: Commercially available beer made in Germany