

Are you made of sugar?

Carbohydrates are derived from photosynthesis and are the main constituent of biomass.

Carbohydrates constitute the biggest part of the biomass on Earth. They are produced by photosynthesis and are present in all plants and plant-based materials. The amount and composition of carbohydrates in a sample can reveal a wide range of different information, depending on the context. As a result, they are subject to analysis in various industries.

Carbohydrates are everywhere

In the food industry, carbohydrate and sugar content are notable for being key factors in determining the nutritional value of food and drink. In environmental analysis – to mention but one example – the anhydrosugar levoglucosan which is produced by the pyrolysis of cellulose and acts as a tracer for biomass combustion is determined in aerosols. These are just two of the many applications of carbohydrate analysis.

Carbohydrates are composed of one or more monosaccharide units, each of which has a carbonyl group (aldehyde or ketone group) and several hydroxyl groups¹. Because mono-, di-, and oligosaccharides are water-soluble, ion chromatography, which is performed in the aqueous phase, is particularly suitable for analyzing them. It does not require extraction to the organic phase; thus, determination can be performed directly. However, a high-capacity column is necessary because sugars are relatively large molecules which are in many cases similar in structure (e.g., glucose and galactose; see Figure 1). For this purpose, Metrohm now offers the Metrosep Carb 2 column.

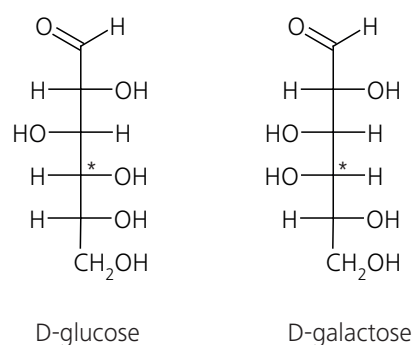


Figure 1. Structural formulae of glucose and galactose. The molecules differ only in the position of the OH group at the C4 atom (highlighted with an asterisk).

Sugars in foods

From December 2016, the European Union (EU) requires that nutritional values are indicated on all foodstuffs, with the exception of unprocessed products and products sold loose (regulation no. 1924/2006). What is already established practice, i.e., indicating the calorific value and certain nutrients including sugar and carbohydrates, is set to become mandatory.

Along with starch, which is a polymer of glucose, the usable carbohydrates found in foodstuffs are largely in the form of sugars. According to the EU definition, this includes all mono- and disaccharides with the exception of polyvalent alcohols. The majority of sugars in foodstuffs is made up of the monosaccharides glucose, fructose, galactose, and the disaccharides sucrose, lactose, and maltose.

Apple juice analysis

The chromatogram in Figure 2 was taken after the injection of apple juice, which was diluted with ultra-pure water. Apart from that, no sample preparation is necessary. The alkaline eluent (100 mmol/L sodium hydroxide/10 mmol/L sodium acetate) ensures that the sugars are present in dissociated form (as anions) and can therefore be separated in the column using the ion exchanger.

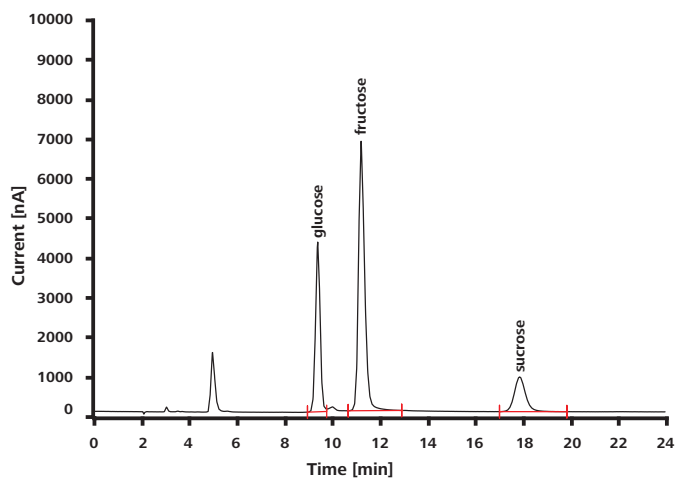


Figure 2. Determination of glucose, fructose, and sucrose in apple juice. Except for simple dilution, no sample preparation is required.

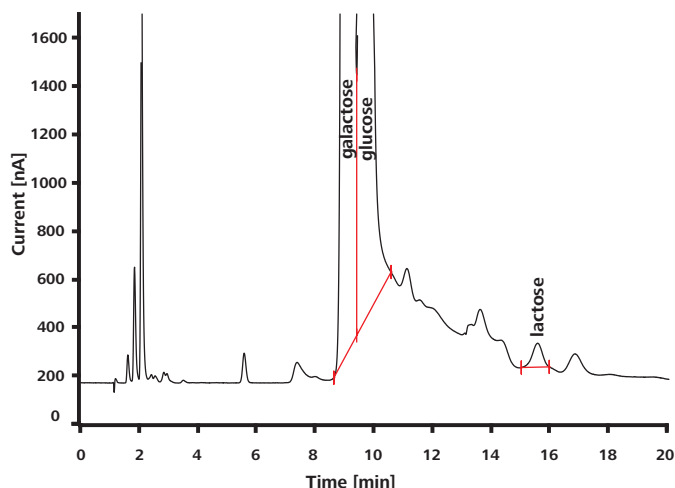
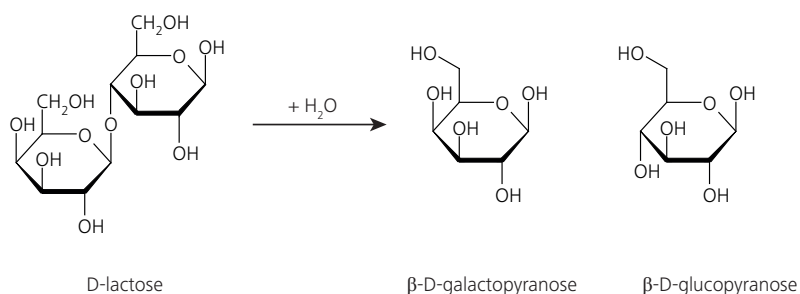


Figure 3. Determination of lactose traces in milk declared lactose-free, spiked with 100 mg/L lactose.

Because carbohydrates are electrochemically active, they can be detected amperometrically. During amperometric detection, the analytes are oxidized to a working electrode by applying a potential to the latter. This results in an electrical current that reveals the concentration. Over time, however, carbohydrates form deposits on the working electrode when a continuous potential is applied. The amperometric detector is therefore operated in PAD mode (pulsed amperometric detection). Here, a three-stage cyclic potential ensures that after measuring the current, i.e., after the determination stage, the electrode is cleaned from the adsorbed molecules and eventually conditioned.

Residual lactose in «lactose-free» products

A key part of the quality control of products declared lactose-free is the determination of residual lactose. The ion chromatogram in Figure 3 illustrates the determination of lactose in «lactose-free» milk to which 100 mg/L lactose was added. Again, the separation takes place under strongly alkaline conditions (eluent of 5 mmol sodium hydroxide/2 mmol sodium acetate) and the analyte is detected by pulsed amperometry. The high concentrations of galactose and glucose illustrated in the chromatogram are a result of the enzymatic breakdown of lactose into these very monosaccharide constituents (Figure 4). Because of its protein-rich matrix, milk must undergo dialysis before being analyzed; with the Metrohm Inline Sample Preparation, this is a fully automated process, and therefore does not involve any additional effort.

Figure 4. Lactose is composed of the monosaccharides galactose and glucose. The hydrolysis of lactose illustrated here is catalyzed by the enzyme lactase.

Carbohydrates as tracers in environmental analysis

Fine dust limit values, which are used as health protection measures, are regularly being violated in many places. When looking for the culprit, the usual suspects are traffic and industry, but residential wood burning used for heating, too, has been linked to high fine dust values². The tracer levoglucosan (Figure 5) is often determined in order to detect wood combustion. Figure 6 shows the determination of a standard solution in which, in addition to levoglucosan, mannosan, and galactosan – all products of wood combustion – were analyzed, as well as several biological sugars, alcohols, etc., which are typically found on aerosol particles. The high-capacity column achieves good separation of all substances which thus can be determined in a single analysis.

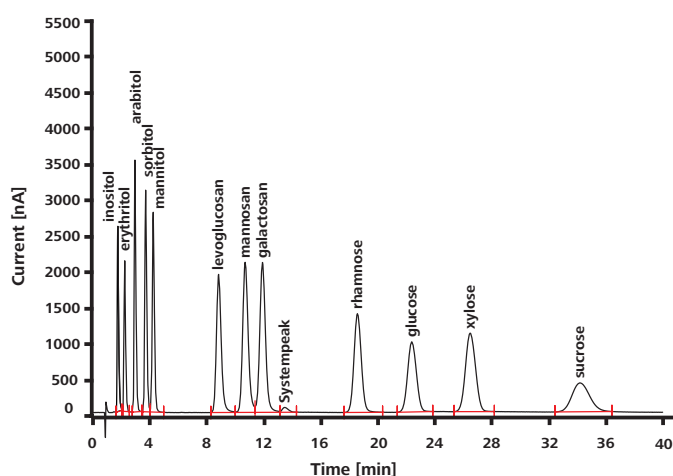


Figure 6. Determination of indicators for wood combustion (levoglucosan, mannosan, and galactosan) and biological sugars and alcohols, which are found in aerosols such as pollen.

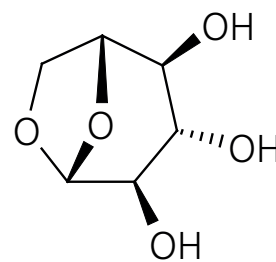


Figure 5. Levoglucosan (1,6-anhydro- β -D-glucopyranose) is produced in the pyrolysis of cellulose and is therefore commonly used as an indicator for biomass combustion.

The new «carbohydrate column»

The Metrosep Carb 2 chromatography column excels with its high ion exchange capacity, i.e., with the high number of ion exchange groups contained in its carrier material. This allows clean separation of the various sugars. Applications are found in a wide range of industries: water and environmental analysis, the pharmaceutical and food industry, forensics, the cosmetic industry, and the quality control of biofuels. In addition to carbohydrate analysis, the Metrosep Carb 2 is also suitable for determinations in samples with high salt content where lower-capacity columns fail, e.g., seawater.

References

- [1] Ebermann, R., Elmadfa, I. (2008): Lehrbuch Lebensmittelchemie und Ernährung. Vienna: Springer Verlag.
- [2] German Federal Institute for Risk Assessment (2015): «Analyse des quantitativen Einflusses der Holzverbrennung auf die Feinstaubkonzentration in Berlin und Brandenburg anhand des Tracers Levoglucosan (Levoglucosan)». Retrieved on May 18, 2015 from http://www.bfr.bund.de/de/analyse_des_quantitativen_einflusses_der_holzverbrennung_auf_die_feinstaubkonzentration_in_berlin_und_brandenburg_anhand_des_tracers_levoglucosan__levoglucosan_-193056.html.



The «carbohydrate column» Metrosep Carb 2 is available in three models: 250 mm, 150 mm, and 100 mm.