Cereal Chemistry

SHORT RESEARCH ARTICLE OPEN ACCESS

Evaluation of AOAC-Method 2017.16: Detection of Oligosaccharides as Low Molecular Weight Soluble Dietary Fiber

Rebekka Elke Schmidt | Mirko Bunzel

Institute of Applied Biosciences, Department of Food Chemistry and Phytochemistry, Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany

Correspondence: Mirko Bunzel (mirko.bunzel@kit.edu)

Received: 20 December 2024 | **Revised:** 25 March 2025 | **Accepted:** 28 March 2025

Funding: Federal Ministry for Economic Affairs and Climate Action (BMWK)

Keywords: AOAC-method 2017.16 | dietary fiber analysis | low molecular weight soluble dietary fiber | oligosaccharides | size exclusion chromatography

ABSTRACT

Background and Objectives: In many countries, the Codex Alimentarius definition of dietary fiber has been adopted with the optional inclusion of oligosaccharides with a degree of polymerization (dp) of ≥ 3. These oligosaccharides are usually captured as low molecular weight soluble dietary fiber (LMWSDF). The suitability of the AOAC-method 2017.16 for the determination of LMWSDF was evaluated, focusing on the correct differentiation between LMWSDF and mono- and disaccharides using different groups of oligosaccharides.

Findings: Desalting of LMWSDF with ion exchange resins resulted in an expected, almost complete loss of uronic acid-based oligosaccharides. Due to their elution behavior in size exclusion chromatography, pentotrioses (arabinotriose, xylotriose) were excluded from LMWSDF. Differently, 1,6-linked hexobioses such as melibiose eluted earlier than 1,4-linked hexobioses (such as the standard maltose) and were incorrectly captured as LMWSDF.

Conclusions: A precise determination of LMWSDF content using AOAC-method 2017.16 depends on the sample to be analyzed. As LMWSDF cannot always be analyzed correctly, these data raise additional questions about the suitability of the dietary fiber definition.

Significance and Novelty: This study indicates that data of the AOAC-method 2017.16 applied to samples containing rather unusual oligosaccharides have to be carefully interpreted.

1 | Introduction

Many countries adopted the Codex Alimentarius (2015) definition of dietary fiber (DF). In a footnote of this definition, national authorities are given the option whether to include oligosaccharides with a degree of polymerization (dp) of 3–9 within their DF definition. Accordingly, within the European Union, for example, DF consists of carbohydrate polymers with at least three monomer units, which can neither be digested nor absorbed in the small intestine of humans (Regulation (EU) No 1169/2011). DF can be divided into insoluble (IDF) and soluble DF (SDF) depending on their solubility in water. SDF can be further grouped into ethanol-insoluble (high molecular weight) SDF and ethanol-soluble low molecular weight (LMW) SDF. LMWSDF mainly consist of oligosaccharides, which are inconsistently defined as saccharides containing at least 3 and up to 10 or 19 monomers (Moss et al. 1995; Mussatto and Mancilha 2007).

Oligosaccharide-enriched fiber preparations can be produced from side-streams such as cereal brans or fruit pomaces by degrading cell wall polysaccharides (e.g., arabinoxylans, cellulose,

or pectic polymers such as arabinans and homogalacturonans) to oligosaccharides (e.g., arabino-xylooligosaccharides, cellooligosaccharides, arabinooligosaccharides, or galacturonic acid oligosaccharides). Prebiotic effects were postulated for oligosaccharides, including aforementioned arabino-, cello-, and xylooligosaccharides (Liu et al. 2024; Nieto-Domínguez et al. 2017; Prandi et al. 2018), which makes them potentially valuable ingredients for the fortification of foods with DF. The correct determination of these and other oligosaccharides as LMWSDF is particularly important for the labeling of these foods.

The first official method to analyze the total content of DF as sum of IDF, SDF, and LMWSDF was AOAC-method 2009.01 (McCleary et al. 2013). Analysis of LMWSDF was optimized in AOAC-method 2017.16 by adding glycerol as an internal standard, adjusting the desalting step, and by improving the HPLC-RI based analysis (McCleary et al. 2019). In the latest AOAC-method 2022.01 determination was extended to separately determine IDF and SDF (McCleary and McLoughlin 2023). In all methods, LMWSDF is determined from the ethanolic supernatant after the previous separation of IDF and SDF. Buffer salts are removed by ion exchange resins, and LMWSDF is analyzed by HPLC-RI based on separation by size exclusion. Retention times of maltose and maltotriose are used to differentiate between saccharides with a dp of 2 and 3. Thereby the method assumes that all di- and trisaccharides have the same elution behavior. However, different saccharides differ in their structure, for example, the position of the glycosidic linkage. Therefore, the hydrodynamic volume of these saccharides is probably also different, so that it is problematic to evaluate all potential DF saccharides on the basis of maltooligosaccharides.

To the best of our knowledge, there are no systematic studies that demonstrate a correct differentiation between LMWSDF and di- and monosaccharides as well as a complete detection of structurally different oligosaccharide groups. Here, we focus on these potential issues by evaluating the AOAC method 2017.16 with respect to the coverage of oligosaccharides as LMWSDF.

2 | Materials and Methods

2.1 | Materials

Standard compounds (arabinobiose, arabinotriose, cellobiose, cellotriose, 3^1-β-cellobiosylglucose, mono-, di- and trigalacturonic acid, galactobiose, 1-kestose, 4′-galactosyllactose, 6′-galactosyllactose lactose, laminaribiose, laminaritriose, maltose, maltotriose, maltulose, isomaltose, isomaltulose, isomaltotriose, mannobiose, mannotriose, raffinose, sucrose, stachyose, 3^2-α-L-arabinofuranosyl-xylobiose, xylobiose, and xylotriose were purchased from Alfa Aesar, Acros Organics (Geel, Belgium), Biosynth (Bratislava, Slovakia), Carbosynth (UK), Carl Roth GmbH & Co. KG (Karlsruhe, Germany), Genaxxon Bioscience (Ulm, Germany), Megazyme (Bray, Ireland), Riedel de Häen, Sigma Aldrich (Schnelldorf, Germany), or Supelco analytical (Bellefonte, USA). Invertase (EC 3.2.1.26 from *Saccharomyces cerevisiae*, ≥ 300 U/mg) was purchased from Sigma Aldrich, and ion exchange resins Ambersep 200 (H+) and Amberlite FPA53 (OH−) from Megazyme. All other chemicals were of analytical purity and were purchased from Carl Roth GmbH & Co. KG, Honeywell Chemicals, Merck KGaA (Darmstadt, Germany) or VWR-Chemicals (Radnor, PA, USA). A mixture of melibiose and manninotriose was produced from a solution with raffinose and stachyose by complete enzymatic hydrolysis with invertase.

2.2 | Studies on the Need of Desalting Before HPLC-RI Analysis

Phosphate buffer (0.2 M; pH 6.0) and maleate buffer (0.35 M; pH 6.0) as well as aqueous solutions of maltose, maltotriose (both 0.5 mg/mL), and glycerol (10 mg/mL) were prepared. Solutions and buffers (injection volume: 50 µL) were analyzed with HPLC (Hitachi, Merck, Darmstadt, Germany) equipped with RI detector (Knauer, Berlin, Germany) using a guard column (TosohTSK-gel PW_{XL} 40 mm × 6.0 mm, particle size 12 µm) and two size exclusion columns in series (2× TosohTSK-gel $G2500PW_{XL}$ 300 × 7.8 mm, particle size 17 µm). Separation was performed isocratically with ultrapure water at a flow rate of 0.4 mL/min at 80°C as described by McCleary et al. (2019).

2.3 | Recoveries of Neutral and Acidic Sugars After Desalting

Phosphate buffer (pH 6.0) was adjusted to pH 4.0 with hydrochloric acid to reflect pH conditions of LMWSDF solutions after removing IDF and SDF. This buffer was used to prepare the following standard solutions: (1) arabinobiose, arabinotriose, and isomaltotriose (200 µM each), (2) cellobiose, cellotriose, and raffinose (200 µm each), and (3) mono-, di-, and trigalacturonic acid (200 µM each) and isomaltotriose (100 µM). Raffinose or isomaltotriose, respectively, were added as internal standard to monitor the reproducibility of the desalting process. According to AOAC-methods 2017.16 and 2009.01, desalting is done by (dispersive) SPE with ion exchange resins (McCleary et al. 2010, 2019). Even if both methods differ in methodological details (e.g., 5 mL of fraction with LMWSDF desalted with 1.5 g of each ion exchange resin vs. 20 mL of fraction with LMWSDF desalted with 25 g of each ion exchange resin), interactions between the sample solutions and ion exchange resins should be comparable. Here, desalting of the three solutions was performed on small scale as described by McCleary et al. (2013). Briefly, anion and cation exchange resins (1 g of each) were mixed and washed with ultrapure water (5 times, 10 mL each) followed by addition of 1 mL of standard solution. The suspension was allowed to stand for 15 min. Standard compounds and internal standards were eluted with 40 mL (eluate 1), 20 mL (eluate 2), and 20 mL (eluate 3) of ultrapure water. Eluates were individually dried by rotary evaporation (max. temperature of 50°C), the residue was redissolved in a defined volume of ultrapure water, and raffinose (standard solutions (1) and (3)), or isomaltotriose (standard solution (2)), respectively, was added as a second internal standard for chromatography. Eluates were analyzed by high-performance anion exchange chromatography with pulsed amperometric detection (HPAEC-PAD, Dionex ICS-5000 system, Thermo Fisher Scientific, Dreieich, Germany) using an analytical CarboPac PA200 column (2500 × 3 mm, 6 µm particle size, Thermo Fisher Scientific) with AminoTrap (50 × 3 mm, 6.5 µm particle size, Thermo Fisher

Scientific) as pre-column. Separation was performed at 25°C with a flow rate of 0.4 mL/min after an injection of 25 µL sample as described previously (Wefers and Bunzel 2016). Ultrapure water (A), 100 mM NaOH (B), and 100 mM NaOH with 500 mM NaOAc (C) were used as eluents. Gradients were adapted to the analytes (see Supporting Information S1: Tables S1–S3). To calculate recoveries of the standard compounds, the aqueous standard solutions (1), (2), and (3) were also injected without prior desalting.

2.4 | Elution Patterns of Different Di- and Trisaccharides

Aqueous standard solutions (0.5 mg/mL) of di- and trisaccharides were prepared and individually analyzed with HPLC-RI as described in Section 2.2.

3 | Results and Discussion

3.1 | Studies on the Need of Desalting Before HPLC-RI Analysis

The first step of the analytical procedure to determine DF comprises the enzymatic degradation of protein and starch. Depending on the method used, different buffer salts are applied. While phosphate buffer is used in earlier methods based on the initial AOAC-method 985.29, maleate buffer is used in AOAC-methods 2009.01 and 2017.16 (McCleary et al. 2013). After filtration of IDF and precipitation of SDF in 78% ethanol (as separate steps to determine SDF and IDF individually or in one step [filtration only after precipitation] for a combined SDF/IDF determination), the content of ethanol-soluble LMWSDF is determined by size exclusion chromatography following desalting with anion and cation exchangers, using glycerol as an internal standard (McCleary et al. 2019). The retention time-based differentiation of saccharides with dp 2 and 3 is related to the elution behavior of maltose (dp 2) and maltotriose (dp 3). Compounds that elute faster than or at the same time as maltotriose are captured as LMWSDF.

Because desalting adds an additional step to the procedure and also discriminates (uronic) acid containing oligosaccharides, as described below, the need of a desalting step was evaluated. Since size exclusion chromatography is monitored by RI detection, eluting buffer salts may affect the chromatograms. As shown in Figure 1, phosphate coeluted with maltotriose and maleate with glycerol. Because defined pH conditions are required for enzymatic starch and protein degradation, the use of water without the addition of buffer salts is not an option. To prevent over- or underestimation of LMWSDF contents, phosphate and maleate buffer must be removed before analysis with HPLC-RI by using a desalting process. Alternatively, maleate buffer can also be used in combination with another suitable internal standard. Here, further studies are required to identify such an internal standard and to adapt and validate a corresponding method. In AOAC-method 2022.01, for example, diethyleneglycol is used instead of glycerol (McCleary and McLoughlin 2023).

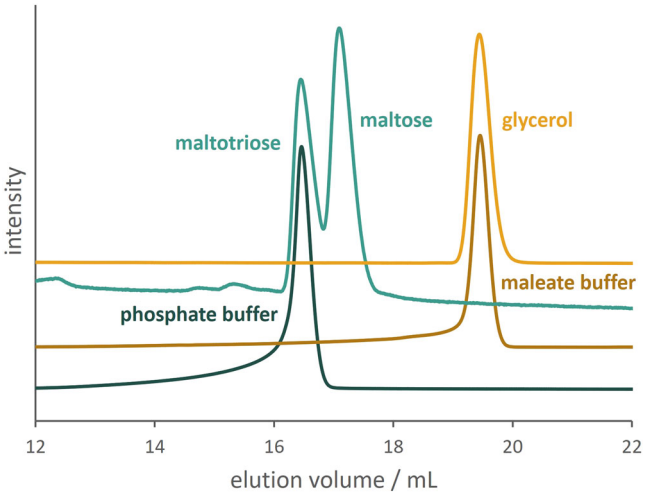

FIGURE 1 | Normalized HPLC-RI-chromatograms of phosphate and maleate buffer compared to maltotriose, maltose, and glycerol. [Color figure can be viewed at wileyonlinelibrary.com]

3.2 | Recoveries of Neutral and Acidic Sugars After Desalting

As desalting is required, the influence of the desalting process on the recovery of various di- and trisaccharides (cellobiose and -triose as being hexose-based, arabinobiose and -triose as being pentose-based, mono-, di- and trigalacturonic acids as being uronic acid-based) was evaluated (Table 1). Cellobiose, -triose and arabinobiose were recovered to a high percentage (90%–93%) in eluate 1. Only the recovery of arabinotriose (86%) was slightly lower. Eluates 2 and 3 contained less than 1% of both di- and trisaccharides. Thus, the elution volume recommended by McCleary et al. (2010) was sufficient for complete elution.

As expected, mono-, di-, and trigalacturonic acids showed very low recoveries (8%–33%) after desalting using ion exchange resins. Losses increased with dp. Galacturonic acids that bind to the anion exchange resin cannot be completely eluted by the conditions used. Increasing the elution volume (eluates 2 and 3) did not result in increased elution of uronic acid containing oligosaccharides. Therefore, the application of AOAC method 2017.16 to samples containing acidic oligosaccharides leads to underestimation of the LMWSDF content. However, as described in Section 3.1, omitting the desalting process would require additional modifications of the current AOAC method.

3.3 | Elution Patterns of Different D-i- and Trisaccharides

Depending on the monosaccharides involved (e.g., pentoses vs. hexoses), their ring form and anomeric configuration as well as the position of the glycosidic linkages, the hydrodynamic volume of different di- and trisaccharides may vary. Therefore, the elution behavior of selected di- and trisaccharides (for structures, see Supporting Information S1: Figure S1) in size exclusion chromatography was analyzed (Figure 2). These di- and trisaccharides either occur natively or are products of the (enzymatic) degradation of the corresponding poly- or oligosaccharides present in

TABLE 1 | Recoveries of the three eluates (1, 2, 3) in percent (%) of cellobiose, cellotriose, arabinobiose, arabinotriose, mono-, di-, and trigalacturonic acid after desalting using ion exchange resins ± standard deviation ($n = 3$).

Eluate	Cellobiose	Cellotriose	Arabinobiose	Arabinotriose	Galacturonic acid	Digalacturonic acid	Trigalacturonic acid
1	92.2 ± 2.9	93.3 ± 3.3	90.3 ± 4.0	86.2 ± 5.1	33.4 ± 10.3	9.3 ± 5.1	8.2 ± 5.8
2	0.5 ± 0.1	0.4 ± 0.1	0.9 ± 0.2	0.6 ± 0.1	0.2 ± 0.0	—	—
3	0.1 ± 0.0	0.1 ± 0.0	0.3 ± 0.2	0.2 ± 0.2	0.2 ± 0.1	—	—

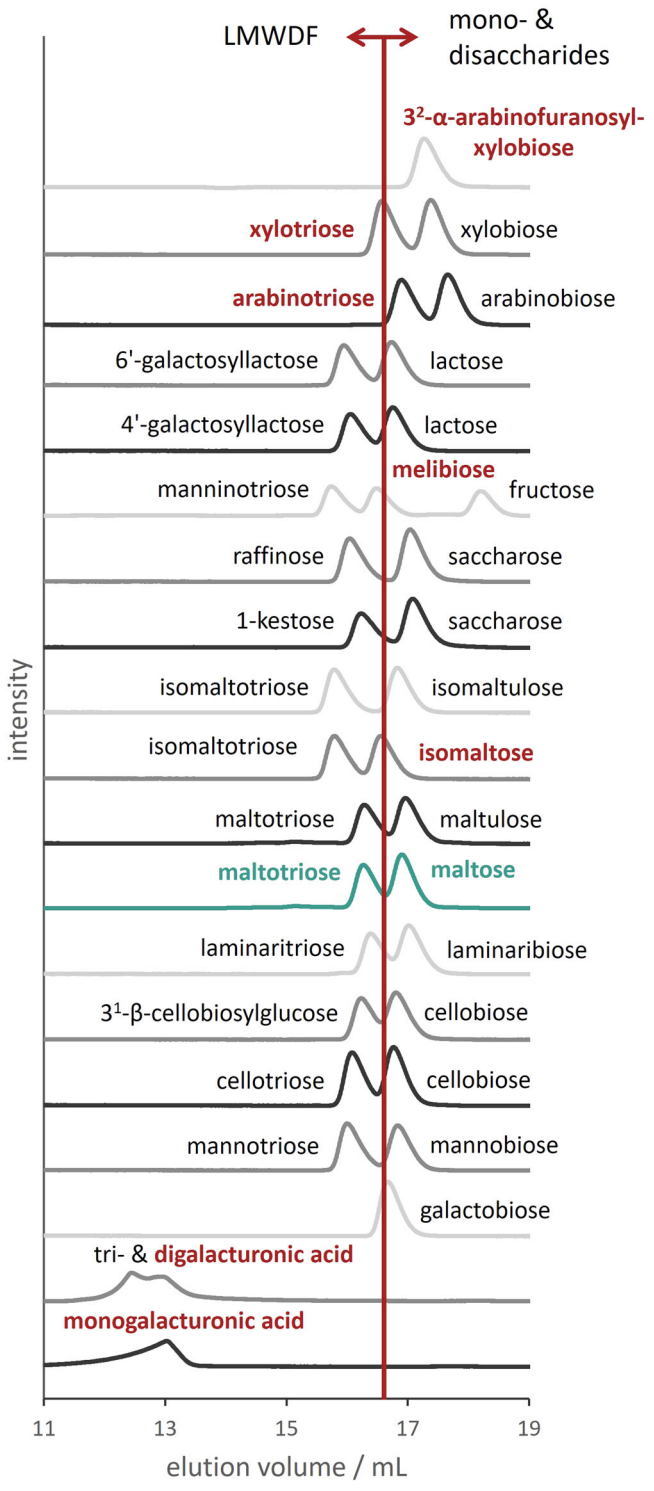

FIGURE 2 | Elution behavior of selected mono-, di-, and trisaccharides as analyzed by size exclusion chromatography in comparison to maltotriose and maltose (green) as cut-off (red line) markers to differentiate between low molecular weight soluble dietary fiber (LMWSDF) and mono- and disaccharides. Incorrectly categorized di- and trisaccharides are highlighted (red). [Color figure can be viewed at wileyonlinelibrary.com]

(plant/algae-based) food ingredients/foods (Table 2), thus being potentially relevant LMWSDF of food products. Sucrose, lactose, isomaltose, and isomaltotriose are not DF saccharides. They were nevertheless analyzed as sucrose is the natural precursor of

TABLE 2 | Occurrence and linkage types of different food-relevant di- and trisaccharides.

Dimer	Linkage	Trimer	Linkage	Occurrence
Maltose[a]	α-1,4	Maltotriose[a]	α-1,4	Starch degradation products
Maltulose[(a)]	α-1,4			Formation from maltose when heated, for example, in beer and honey (Li et al. 2020; Petersen et al. 2014; Swallow and Low 1990)
Isomaltose[a]	α-1,6	Isomaltotriose[a]	α-1,6	Starch degradation products
Isomaltulose[a]	α-1,6			Synthetic, sweetener, for example, found in honey, formation in sugar cane due enzymatic degradation (Sawale et al. 2017; Swallow and Low 1990)
Arabinobiose	α-1,5	Arabinotriose	α-1,5	Hydrolysis products of polysaccharides (arabinan, (arabino)galactan, cellulose, (glucuronoarabino)xylan, homogalacturonan, mannan, mixed-linked β-glucans) of the plant cell wall
Cellobiose	β-1,4	Cellotriose	β-1,4	
		3^1-β-Cellobiosylglucose	β-1,4 + β-1,3	
Digalacturonic acid	α-1,4	Trigalacturonic acid	α-1,4	
Galactobiose	β-1,4			
Mannobiose	β-1,4	Mannotriose	β-1,4	
Xylobiose	β-1,4	Xylotriose	β-1,4	
		3^2-α-Arabinofuranosyl-xylobiose	β-1,4 + α-1,3	
Sucrose[a]	α,β-1,2	1-Kestose	α,β-1,2	In plants, from fructooligosaccharides
		Raffinose	α,β-1,2 + α-1,6	In legumes
Melibiose	α-1,6	Manninotriose	α-1,6	In legumes, invertase hydrolysis products of raffinose and stachyose
Lactose[a]	β-1,4	4′-Galactosyllactose	β-1,4	In milk products, from galactooligosaccharides
		6′-Galactosyllactose	β-1,4 + β-1,6	
Laminaribiose	β-1,3	Laminaritriose	β-1,3	Hydrolysis products of the algae cell wall polysaccharide laminarin, formation in honey and beer through enzymatic degradation (Petersen et al. 2014; Swallow and Low 1990)

[a]Carbohydrates are usually fermentable in the human small intestine.

raffinose and 1-kestose, lactose is the dimeric reference for 4′-galactosyllactose and 6′-galactosyllactose, and isomaltose/isomaltotriose were used to detect structural similarities in the elution behavior of 1,6-linked oligosaccharides.

All α- and β-1,4-linked hexobioses and -trioses (cellobiose, cellotriose, galactobiose, lactose, 4′-galactosyllactose, maltulose, mannobiose, mannotriose, and the retention time markers maltose and maltotriose) showed the same elution behavior. Also, α- and β-1,3-linked (laminaribiose, laminaritriose, 3^1-β-cellobiosylglucose) and α,β-1,2-linked (sucrose, raffinose, 1-kestose) hexobioses and -trioses had a similar elution behavior as maltose and maltotriose. However, α- and β-1,6-linked hexobioses and -trioses (isomaltose, isomaltotriose, 6′-galactosyllactose, melibiose, manninotriose) tended to elute earlier than maltose and maltotriose, respectively, except isomaltulose. Hence, it can be assumed that 1,6-linked di- and trisaccharides have a higher hydrodynamic volume than 1,4-, 1,3- and 1,2-linked di- and trisaccharides. As a consequence, melibiose is incorrectly (partially) captured as LMWSDF.

Both isomaltose and isomaltulose are 1,6-linked disaccharides with the difference that the reducing end in isomaltose is glucose and in isomaltulose fructose, with the reducing ring form being preferentially glucopyranose and fructofuranose, respectively (Dreissig and Luger 1973). Based on the earlier elution of isomaltose compared to isomaltulose, it is expected that the hydrodynamic volume of pyranoses is greater than that of furanoses. A similar elution behavior was observed for arabinobiose and -triose (furanoses) as compared to xylobiose and -triose (pyranoses).

In addition, it was observed that pentobioses and -trioses eluted generally later than maltose and maltotriose, respectively. An even later elution showed the "branched" trisaccharide 3^2-α-arabinofuranosyl-xylobiose. As a consequence, these pentose-based trisaccharides are not captured as LMWSDF. Differently, mono-, di-, and trigalacturonic acid eluted clearly before maltotriose. Thus, if the methodology would be adjusted to avoid desalting before size exclusion chromatography, an overestimation of LMWSDF will be obtained if uronic acids or uronic acid containing disaccharides are present in the analyzed samples.

Therefore, our results show that depending on the oligosaccharides present in the sample, the AOAC-method 2017.16 over- or under-estimates the content of LMWSDF. Pentose-based trisaccharides appear to be (partially) excluded from LMWSDF, whereas uronic acid-containing carbohydrates are always captured as LMWSDF (unless they are removed during the desalting step). In case of hexose/hexulose-based disaccharides, both the linkage position (especially 1,6-linkages) as well as the ring form affect the elution behavior in size exclusion chromatography, resulting in incorrect data. McCleary and McLoughlin (2023) also described an earlier elution of lactose and isomaltose. To solve this problem, they recommend an additional enzymatic hydrolysis of lactose and isomaltose to galactose and glucose, which was considered in AOAC 2022.01. Nevertheless, other hexobioses, pentotrioses, and galacturonic oligosaccharides were not taken into account.

The elution behavior of oligosaccharides with dp ≥ 4 was not analyzed here. However, based on the observed elution behavior order it is assumed that they elute before their corresponding trisaccharides and, thus, are captured as LMWSDF.

4 | Conclusion

Our studies reveal weaknesses of the AOAC-method 2017.16 in the determination of LMWSDF contents. The suitability of the method to analyze LMWSDF contents strongly depends on the structures of the analyzed oligosaccharides. Especially, oligosaccharides that are based on pentoses as well as uronic acid containing carbohydrates will be incorrectly classified. However, also hexose/hexulose-based carbohydrates may be falsely captured as LMWSDF. Thus, the results of this study recommend that the AOAC-method 2017.16 needs to be adapted depending on the oligosaccharides present in the sample to be analyzed. This will become especially important if food products will be enriched with fiber preparations produced from side-streams such as cereal brans (generation of (arabino-) xylooligosaccharides from arabinoxylans) or fruit pomaces (generation of pectin-based oligosaccharides such as uronic acid containing carbohydrates [from homogalacturonans] or arabinooligosaccharides [from arabinans]).

Author Contributions

Rebekka Elke Schmidt: data curation, investigation, methodology, visualization, writing – original draft, review and editing original draft. **Mirko Bunzel:** conceptualization, resources, supervision, writing – review and editing.

Acknowledgments

This study was part of the IGF Project (21616N) of the FEI which was supported via AiF within the program for promoting the Industrial Collective Research (IGF) of the Federal Ministry of Economic Affairs and Climate Action (BMWK), based on a resolution of the German Parliament. Open Access funding enabled and organized by Projekt DEAL.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data presented in this study are available on request from the corresponding author.

References

Codex Alimentarius. 2015. *Guidelines on Nutrition Labelling CAC/GL 2-1985. Joint FAO/WHO Food Standards Programm*. Secretariat of the Codex Alimentarius Commission. FAO.

Dreissig, W., and P. Luger. 1973. "Die Strukturbestimmung der Isomaltulose, $C_{12}H_{22}O_{11}\cdot H_2O$." *Acta Crystallographica, Section B: Structural Crystallography and Crystal Chemistry* 29, no. 3: 514–521.

Li, M., J. Du, and K. Zhang. 2020. "Profiling of Carbohydrates in Commercial Beers and Their Influence on Beer Quality." *Journal of the Science of Food and Agriculture* 100, no. 7: 3062–3070.

Liu, X., K. K. Y. Hu, and V. S. Haritos. 2024. "Enzymatic Production of Cello-Oligosaccharides With Potential Human Prebiotic Activity and

Release of Polyphenols From Grape Marc." *Food Chemistry* 435: 137562.

McCleary, B. V., N. Ames, J. Cox, et al. 2019. "Total Dietary Fiber (CODEX Definition) in Foods and Food Ingredients by a Rapid Enzymatic-Gravimetric Method and Liquid Chromatography: Collaborative Study, First Action 2017.16." *Journal of AOAC International* 102, no. 1: 196–207.

McCleary, B. V., and C. McLoughlin. 2023. "Determination of Insoluble, Soluble, and Total Dietary Fiber in Foods Using a Rapid Integrated Procedure of Enzymatic-Gravimetric-Liquid Chromatography: First Action 2022.01." *Journal of AOAC International* 106, no. 1: 127–145.

McCleary, B. V., N. Sloane, A. Draga, and I. Lazewska. 2013. "Measurement of Total Dietary Fiber Using AOAC Method 2009.01 (AACC International Approved Method 32-45.01): Evaluation and Updates." *Cereal Chemistry* 90, no. 4: 396–414.

McCleary, B. V., J. W. De Vries, J. I. Rader, et al. 2010. "Determination of Total Dietary Fiber (CODEX Definition) by Enzymatic-Gravimetric Method and Liquid Chromatography: Collaborative Study." *Journal of AOAC International* 93, no. 1: 221–233.

Moss, G. P., P. Smith, and D. Tavernier, IUPAC. 1995. "Glossary of Class Names of Organic Compounds and Reactivity Intermediates Based on Structure (IUPAC Recommendations 1995)." *Pure and Applied Chemistry* 67: 1307–1375.

Mussatto, S. I., and I. M. Mancilha. 2007. "Non-Digestible Oligosaccharides: A Review." *Carbohydrate Polymers* 68, no. 3: 587–597.

Nieto-Domínguez, M., L. I. de Eugenio, M. J. York-Durán, et al. 2017. "Prebiotic Effect of Xylooligosaccharides Produced From Birchwood Xylan by a Novel Fungal GH11 Xylanase." *Food Chemistry* 232: 105–113.

Petersen, B. O., M. Nilsson, M. Bøjstrup, O. Hindsgaul, and S. Meier. 2014. "1H NMR Spectroscopy for Profiling Complex Carbohydrate Mixtures in Non-Fractionated Beer." *Food Chemistry* 150: 65–72.

Prandi, B., S. Baldassarre, N. Babbar, et al. 2018. "Pectin Oligosaccharides From Sugar Beet Pulp: Molecular Characterization and Potential Prebiotic Activity." *Food & Function* 9, no. 3: 1557–1569.

Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the Provision of Food Information to Consumers, Amending Regulations (EC) No 1924/2006 and (EC) No 1925/2006 of the European Parliament and of the Council, and repealing Commission Directive 87/250/EEC, Council Directive 90/496/EEC, Commission Directive 1999/10/EC, Directive 2000/13/EC of the European Parliament and of the Council, Commission Directives 2002/67/EC and 2008/5/EC and Commission Regulation (EC) No 608/2004 with EEA relevance, last amended on 01.01.2018 (OJ L 304, p. 18-63).

Sawale, P. D., A. M. Shendurse, M. S. Mohan, and G. R. Patil. 2017. "Isomaltulose (Palatinose)—An Emerging Carbohydrate." *Food Bioscience* 18: 46–52.

Swallow, K. W., and N. H. Low. 1990. "Analysis and Quantitation of the Carbohydrates in Honey Using High-Performance Liquid Chromatography." *Journal of Agricultural and Food Chemistry* 38, no. 9: 1828–1832.

Wefers, D., and M. Bunzel. 2016. "Arabinan and Galactan Oligosaccharide Profiling by High-Performance Anion-Exchange Chromatography With Pulsed Amperometric Detection (HPAEC-PAD)." *Journal of Agricultural and Food Chemistry* 64, no. 22: 4656–4664.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.