Dietary Fiber Analysis Solutions

For Food, Beverage & Ingredient Manufacturers

NEOGEN | Megazyme

Contents

Global Leaders in Dietary Fiber Analysis

Why Measure Dietary Fiber?

Dietary fiber forms an important part of our diet and is a common food additive due to its impact on digestion and health. Within the food industry, manufacturers invest research and resources into means of maximising the dietary fiber content of their products and providing accurate information for food labelling.

Dietary fiber content labelling in processed foods is strictly regulated and mandatory in some regions. It is therefore important for the industry to have reliable and accurate means of measuring dietary fiber in a product for labelling purposes. The correct analysis of fibers is key for accurate calorie values, appropriate fiber content claims, and to avoid litigations linked to improper labelling.

Our Solutions

Our Megazyme® range of assay kits and enzyme products support a broad range of customer needs for dietary fiber analysis. We have been at the forefront of developing new and optimized methods for dietary fiber analysis, allowing us to accurately measure fiber ingredients, such as β-glucan, starch and carbohydrates that would not be measured using more traditional methods. Our methods have been officially validated by AOAC through meticulous interlaboratory studies and have been recognised as reference methods by CODEX Alimentarius.

Why Choose our Megazyme Range?

Our knowledge in the dietary fiber area coupled with our exceptional technical support, in-house enzyme manufacturing, and added value service, makes us a valuable partner who can support your analysis needs, every step of the way.

- **Experienced** in dietary fiber analysis methods
- **International validated methods** recognised globally
- Excellent in-house **technical support**
- **Ultra-pure enzymes** for enhanced accuracy and **labelling compliance**
- Secure stock and **global supply chain**

Dietary Fiber Analysis Methods

We understand manufacturers' needs for high-quality diagnostic products that complement existing international standard methods for the analysis of dietary fiber and its components.

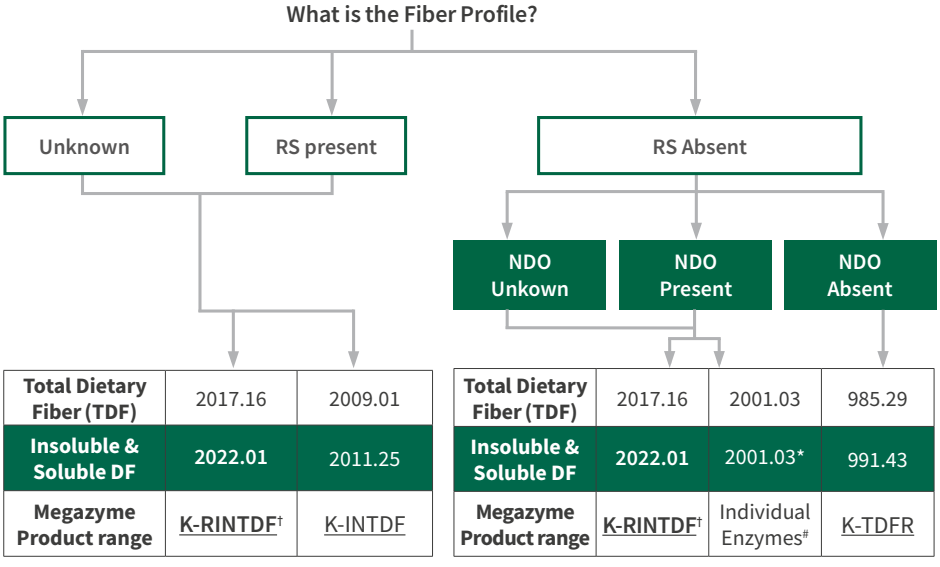
In addition to our product offering for fiber analysis according to the more traditional methods AOAC 985.29 and 991.43, we have **developed four additional methods for fiber analysis** to measure fiber according to the CODEX definition. These improved methods have been accepted as official AOAC standards and are also recognised by regulatory bodies worldwide as reference methods: **AOAC 2009.01**, **2011.25**, **2017.16**, and **2022.01**.

What components of Dietary Fiber does each method measure accurately?

AOAC Method	Megazyme Product range	Target Analytes				Issues	
		-	Soluble DF		Insoluble DF		
		Total Dietary Fiber		-			
		HMWDF	SDFS (NDO)	SDFP	IDF	Underestimated	Overestimated
985.29	K-TDFR	●				RS_2, RS_3 NDO not measured	RS_4
991.43				●	●		
2009.01	K-INTDF	●	●			RS_2, RS_4, FOS	Resistant maltodextrins artifacts
2011.25			●	●	●		
2017.16	K-RINTDF	●	●			NONE	NONE
2022.01			●	●	●		

DF: Dietary Fiber. **HMWDF**: High Molecular Weight Dietary Fiber. **RS**: Resistant Starch. **IDF**: Water insoluble Dietary Fiber. **SDFP**: Water soluble Dietary Fiber which precipitates in 78% ethanol. **SDFS**: Water soluble Dietary Fiber that remains soluble in 78% ethanol (= NDO).

Which AOAC method is most appropriate to measure Dietary Fiber?



DF: Dietary Fiber
RS: Resistant Starch
NDO: Non-digestible Oligosaccharides (=SDFS)

† K-RINTDF provides a more accurate measurement for resistant starch and removes the issue of fiber overestimation in starchy foods.
# E-AMGDFPD, E-BLAAM & E-BSPRPD
* Modification as per AOAC 991.43 to allow for soluble/insoluble DF determination

Dietary Fiber Range

Rapid Integrated Total Dietary Fiber Methods

Our latest dietary fiber method, the Rapid Integrated Total Dietary Fiber Method (RINTDF) has been accepted by AOAC as the official standard method: AOAC 2022.01, and is the most **recent**, **accurate**, and **encompassing method** for dietary fiber analysis allowing for the measurement of both **soluble** and **insoluble fiber separately**.

The K-RINTDF assay kit is the only commercially available product that allows for the correct analysis of Dietary Fiber content as defined by CODEX Alimentarius and is suitable for AOAC methods 2017.16 (Codex Type I method) and 2022.01.

These methods are different from all other dietary fiber methods for their enzymatic incubation conditions mimicking human digestion. Under these conditions, a more accurate measurement of resistant starch is obtained, including phosphate cross-linked starch (RS4) and no fiber overestimation occurs when analysing starchy foods.

Our kit contains the three enzymes required for the assay and also several standards to be used in the HPLC step for the analysis of the non-digestible oligosaccharide portion of the fiber. Additional products, such as Celite and desalting resins, necessary to run these methods are also available in the Megazyme range.

K-RINTDF Advantages

- Cost-effectiveness of fiber fortification by food manufacturers preventing under-estimation issues when using resistant starches and non-digestible oligosaccharides ingredients
- AOAC 2022.01 allows for the measurement of soluble and insoluble fiber separately and can improve the calorie profile of food products.
- Complies with the CODEX Alimentarius definition of dietary fiber which is supported by governing bodies worldwide
- Most encompassing method regardless of the fiber component profile of sample
- Improves labelling regulatory compliance for food manufacturers

Total Dietary Fiber Methods

The Total Dietary Fiber assay kit (K-TDFR) allows for the measurement of dietary fiber as specified in AOAC methods: 991.43, 991.42, and 985.29. These methods have been routinely used in the food industry and are recognised as the gold standard, however they fail to accurately measure certain fiber ingredients such as resistant starches and non-digestible oligosaccharides.

Our kit contains the three key enzymes required for this method, and we can also provide a bespoke pack-size suitable for use with the **ANKOM semi-automated fiber analysers**.

Some users choose to use AOAC 2001.03 in combination with the traditional AOAC methods (985.29 or 991.43) in order to measure the non-digestible oligosaccharides portion. While this is a known practice, this combination of methods may lead to issue with quantifying resistant starches (under and over-underestimation issues), is costly and more prone to human errors due to the use of two separate methods.

K-TDFR Advantages

- Ultra-pure enzymes allow for accurate results and minimise under-estimation issues.
- Can be used to measure dietary fiber following AOAC 985.29 and AOAC 991.43
- Enzyme products available to be used for AOAC method 2001.03
- Multiple pack sizes including the bespoke formulation for use with ANKOM instruments
- Secure stock & global supply chain

Integrated Dietary Fiber Methods

The Integrated Total Dietary Fiber assay kit (K-INTDF) allows for the measurement of dietary fiber following the official **AOAC methods: 2009.01** and **2011.25**. These methods follow the CODEX Alimentarius definition of dietary fiber and are recognised by selected government bodies as reference methods, however, they are not as universally applicable as AOAC 2017.16 and 2022.01 because they have under and over-estimation issues for some food types. These issues are linked to the long incubation conditions used in these methods which are not representative of human digestion.

Our kit contains the three enzymes required for the assay and several standards to be used in the HPLC step of these methods for the analysis of the non-digestible oligosaccharide portion of fiber. Our enzymes are also available for purchase as standalone products. Additional products such as Celite and desalting resins, necessary to run these methods are also available in the Megazyme range.

For those users that are interested in **measuring dietary fiber according to the CODEX Alimentarius definition, we would recommend the option of using the novel methods AOAC 2017.16 and 2022.01 with our K-RINTDF assay kit.**

Dietary Fiber Components
β-Glucan

Cereal β-glucan is a component of dietary fiber with recognized health benefits.

The β-glucan Assay Kit (K-BGLU) is suitable for the **measurement of cereal β-glucan** in cereal grains, milling fractions, malts, wort, beer, and food products following methods AOAC 995.16 and 992.28.

Regulatory bodies recognize β-glucan's cardiovascular health and cholesterol-lowering benefits, permitting associated health claims in different regions. These health benefits make this cereal-based food component of particular interest to the food industry.

Cereal β-glucan is also an **important analyte** in the **brewing industry** where elevated levels can lead to increased wort viscosity and cause processing challenges. It is also of relevance in the **animal feed industry** where **high levels of β-glucan** can lead to **reduced feed intake** and **decreased feed conversion**.

Our kit contains the enzymes required for the assay, and components to measure cereal β-glucan using a spectrophotometer, including a reference β-glucan standard.

K-BGLU Advantages

Accurately measures all cereal β-glucan content as per AOAC method 995.16 and serves as the reference method for health disclaimers on food packaging.

Improves labelling regulatory compliance for food manufacturers

Unique commercially available kit for β-glucan testing

Ultra-pure enzymes allow for accurate results and minimise under-estimation issues.

Fructan, Inulin & Fructo-oligosaccharides (FOS)

Fructan, Inulin and FOS are a family of polysaccharides that form a component of dietary fiber and are used in food manufacturing processes for fiber fortification

Fructans occur naturally in food, while FOS are approved fiber additives in infant formula in some countries. Selective analytical methods for fructan and FOS in food matrices and infant formula have been accepted as recognized standards, such as **AOAC 999.03**, **2016.14**, **and Chinese Standard GB 5009.255-2016**.

While the Fructan assay kit (K-FRUC) is only compatible with the AOAC methods, our Megazyme range does offer ultra-pure enzyme products that are suitable to be used when following the Chinese standard GB 5009.255-2016 method. When measured as part of dietary fiber and not as a single component, fructans including inulin and FOS are not fully measured by the traditional fiber methods AOAC 985.29 or 991.43 due to their low molecular weight which makes these ingredients non-digestible oligosaccharides. These fiber components can be measured in their entirety by the most recent methods for total dietary fiber AOAC 2017.16 and 2022.01.

Method	Megazyme Product Range
AOAC 2018.07	K-FRUC
AOAC 2016.14 (ISO/DIS 22579)	K-FRUC
AOAC 999.03 (AACCI 32-32.01 & Codex Alimentarius Type III)	K-FRUC
AOAC 2016.06	E-FRMXPD & E-SUCR
AOAC 997.08 (Codex Alimentarius Type II)	E-FRMXLQ & E-AMGFR
GB 5009.255-2016	E-FRMXLQ or E-FRMXPD & E-SUCR

Resistant Starch

Resistant starch is a type of dietary fiber component that resists the enzymatic breakdown occurring in the human small intestine during the digestion process.

The **Resistant starch assay kit (K-RSTAR)** is suitable for the measurement of resistant starch following method AOAC 2002.02 which has been widely adopted as the reference method. In addition to this kit, the Megazyme range has also released two additional kits for the measurement of resistant starch, **K-RAPRS (Rapid Resistant Starch Assay kit)** and **K-DSTRS (Digestible/Total/Resistant Starch Assay kit)**. While these two kits have not been officially validated, they use more physiologically relevant enzymatic conditions equivalent to those employed in K-RINTDF.

Four different types of resistant starch	
RS1	Physically inaccessible starch, such as that found in seeds or legumes and unprocessed whole grains.
RS2	Resistant starch that occurs in its natural granular forms, such as uncooked potato, green banana, flour and high amylose corn.
RS3	Resistant starch that is formed when starch-containing foods are cooked and cooled such as in legumes, bread, cornflakes, cooked-and-chilled potatoes, pasta salad or sushi rice. This treatment leads to retrogradation – the recrystallisation of amylose and amylopectin on cooling – which makes starch resistant to enzymatic hydrolysis.
RS4	Starches that have been synthetically modified to resist digestion. This type of resistant starch can exhibit a broad range of structural diversity.

K-RSTAR Advantages

Ultra-pure enzymes allow for accurate results and minimise under-estimation issues

Additional kits measure resistant starches and digestible starches using physiologically aligned enzymatic hydrolysis conditions

Enables accurate measurement of resistant starch and is compatible with the AOAC method 2002.02

Polydextrose

Polydextrose (PDX) is a glucose-based synthetic polymer commonly used as a low molecular weight dietary fiber (LMWDF) source in infant formulas and adult nutritional drinks.

To analyze polydextrose in food, specific methods have been accepted, such as **AOAC 2000.11 and the Chinese Standard GB 5009.245-2016**. These methods involve enzymatic digestion using ultra-pure enzymes and chromatography detection.

The Megazyme range provides **ultra-pure enzymes (E-AMGDF, E-EXOIAN, and E-ISAMY)** that can be used according to these official methods.

When assessing polydextrose as part of dietary fiber, it is not fully quantified by the traditional fiber methods AOAC 985.29 or 991.43, as it is considered a non-digestible oligosaccharide (NDO). However, the most recent methods for total dietary fiber, such as **AOAC 2017.16 and 2022.01, account for the measurement of polydextrose as part of the overall dietary fiber content**.

Available Carbohydrates

Available carbohydrates are the portion of carbohydrates that are digestible by humans in the small intestine and therefore are not dietary fiber.

The **Available Carbohydrates assay kit (K-AVCHO)** is suitable for the **determination of available carbohydrates** comprising total digestible starch (TDS) plus maltodextrins, maltose, sucrose, isomaltose, D-galactose, D-glucose, D-fructose and lactose. The method on which this kit is based, has been officially validated as **AOAC 2020.08**.

It is also a **perfect complement to AOAC methods 2017.16 and 2002.01 (K-RINTDF)**. Both methods use an equivalent enzyme incubation step that is physiologically relevant, making the kits ideal for simultaneous analysis; K-RINTDF measures dietary fiber, while K-AVCHO measures those carbohydrates that are digestible and therefore not fiber.

Laboratories that continue to work with **pre-CODEX definition of Dietary Fiber** are supported by our **Available Carbohydrates / Dietary Fiber Assay Kit (K-ACHDF)**, which measures TDF using AOAC 985.29/991.43 in addition to measuring available carbohydrates.

Key Megazyme Products

Available for the measurement of dietary fiber analysis

AOAC Methods	Codex Method Recognition	Target Analytes	Assay Kits
2022.01 2017.16	Type I (2017.16)	Insoluble, Soluble and Total Dietary Fiber in Foods	K-RINTDF
2011.25 2009.01	Type I (2011.25)	Insoluble, Soluble and Total Dietary Fiber in Foods	K-INTDF
991.43 985.29	Type I	Total Dietary Fiber in Foods	K-TDFR

Available for the measurement of dietary fiber components

AOAC Methods	Codex Method Recognition	Target Analytes	Assay Kits
2020.08	N/A	Available Carbohydrates	K-AVCHO
2018.17	N/A	Fructans in Animal Feed, Pet Food and Ingredients	K-FRUC
2016.14	N/A	Fructans in Infant Formula and Adult Nutrition	K-FRUC
2002.02	Type II	Resistant Starch in Starch and Plant Materials	K-RSTAR
2000.11	Type II	Polydextrose in Foods	E-AMGDF E-ISAMY E-EXOIAN
999.03 997.08	Type III (999.03) Type II (997.08)	Fructans in Foods	K-FRUC E-AMGFR & E-FRMXLQ
995.16 992.28	Type II (995.16)	β-D-Glucan in Oats and Barley	K-BGLU

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