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REVIEW

Nutritional characterization and measurement of dietary carbohydrates

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Dietary carbohydrate characterization should reflect relevant nutritional and functional attributes, and be measured as chemically identified components. A nutritional classification based on these principles is presented, with a main grouping into 'available carbohydrates', which are digested and absorbed in the small intestine providing carbohydrates for metabolism, and 'resistant carbohydrates', which resist digestion in the small intestine or are poorly absorbed/metabolized. For the available carbohydrates, the chemical division into the starch and total sugars categories does not adequately reflect the physiological or nutritional attributes of foods. Characterizing carbohydrate release from starchy foods provides insight into some of the inherent mechanisms responsible for the varied metabolic effects. Also, a pragmatic approach to product signposting consistent with guidelines to limit free (or added) sugars is proposed. The most prominent of the resistant carbohydrates are the non-starch polysaccharides (NSP) from plant cell walls, which are characteristic of the largely unrefined plant foods that provide the evidence base for the definition and measurement of dietary fibre as 'intrinsic plant cell-wall polysaccharides' as proposed in conjunction with this paper and endorsed by the scientific update. Indigestibility in the small intestine was not considered to be an adequate basis for the definition of dietary fibre, as there is insufficient evidence to establish public health policy by this approach and concerns have been raised about potential detrimental effects of high intakes of rapidly fermentable resistant carbohydrates. Functional ingredients such as resistant starch and resistant oligosaccharides should therefore be researched and managed separately from dietary fibre, using specific health or function claims where appropriate. This structured approach to the characterization of nutritionally relevant features of dietary carbohydrates provides the basis for establishing population reference intakes, nutrition claims and food labelling that will assist the consumer with properly informed dietary choices.
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Introduction

The classification and measurement of dietary carbohydrates requires a systematic approach that describes both the chemical and functional properties of carbohydrates in foods. The objective of this paper is therefore to provide an in-depth examination of the issues essential to the achievement of a suitable approach to the nutritional characterization of dietary carbohydrates. As summarized in Figure 1, there are a number of reasons why information on the type and amounts of carbohydrates present in foods is required including, nutrition labeling, food composition tables, product development and nutrition research. These applications all have an impact on public health, which must be considered the ultimate purpose of providing appropriate carbohydrate characterizations.

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Nutritional considerations for the classification and measurement of carbohydrates

Foods can contain a range of chemically distinct carbohydrate substances, which have varied gastrointestinal and metabolic properties. In addition, biological origin and food processing have an important role in determining the overall attributes of the food matrix and the physico-chemical properties of carbohydrates in foods, which can have a major impact on their physiological handling (Figure 2). This

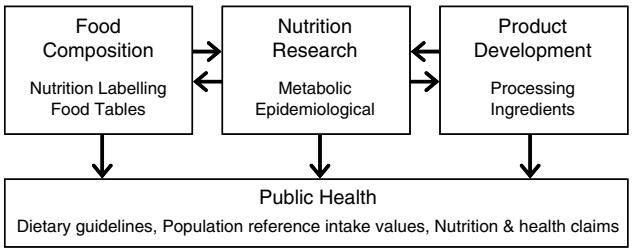

Figure 1 Requirements for carbohydrate measurements. A range of carbohydrate measurements are used in diverse but interrelated fields within nutrition and food technology. Carbohydrate characterizations should evolve to describe specific functional attributes of carbohydrate containing foods as these are identified by nutrition research. This then informs the development of appropriate measurements that can be applied in food composition and product development, which in turn stimulates further research. These activities contribute to the scientific evidence base on which dietary guidelines are formulated. Appropriate carbohydrate characterizations can assist the consumer with informed diet selections and thereby can make a significant contribution to public health.

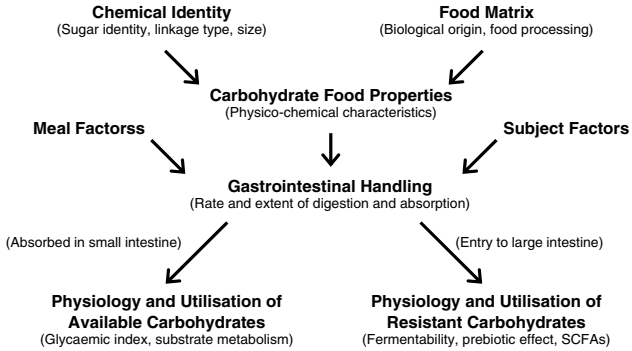

Figure 2 Determinants of gastrointestinal fate of dietary carbohydrates. Largely depending on their physiochemical properties, different carbohydrate foods can exert a range of physiological effects, including varied rates of digestion and absorption in the small intestine and varied fermentability and profile of fermentation products in the large intestine. Even though there will be a degree of variation in gastrointestinal handling due to meal and subject factors, the emphasis from a nutritional perspective must to be on describing the inherent physico-chemical characteristics of the foods, reflecting both the chemical identity of carbohydrates and the influence of the food matrix on functional attributes.

variability in functionality needs to be considered in the nutritional characterization of dietary carbohydrates, taking into account issues of both food matrix and chemical identity (Englyst and Englyst, 2005). Based on current knowledge of the mechanisms by which dietary carbohydrates exert their influence on physiology and health, it is possible to describe these characteristics and incorporate them into an overall classification scheme (Table 1) that can evolve as new evidence becomes available. Other papers in this report have dealt with physiology and health aspects of dietary carbohydrates in detail. The key nutritional issues connected with the characterization of carbohydrates are summarized here.

Gastrointestinal fate and metabolizable energy

In terms of providing the body with metabolizable substrates, the digestion of carbohydrates should be considered as an event of both the upper (absorption of metabolizable carbohydrate) and lower (fermentation providing short-chain fatty acids (SCFA)) gastrointestinal tract. Therefore, for calculation of the contribution to energy, there is a need to know the gastrointestinal and metabolic fate of carbohydrates (which is discussed in more depth in the accompanying physiology paper by Elia and Cummings (2007). Although a number of essentially synonymous terms have been used to describe this division, the terms available carbohydrate and resistant carbohydrate should probably be considered the most informative and these can be defined as follows.

Available carbohydrates are those that are absorbed in the small intestine and provide carbohydrate for metabolism. This definition is equivalent to the glycaemic carbohydrate term used in the 1998 Food and Agriculture Organization/World Health Organization report on carbohydrates, but which has not been widely used. The 'available carbohydrate' term is long established and has been widely adopted (McCance and Lawrence, 1929; FAO, 2003).

Resistant carbohydrates are those that resist digestion in the small intestine, or are poorly absorbed and/or metabolized. This definition is based on a similar one proposed to distinguish gastrointestinal fate (Rumessen, 1992), but includes the aspect of poorly metabolized to more effectively encompass the polyols. In essence, the resistant carbohydrate definition is equivalent to the indigestible, unavailable and non-glycaemic terms, but provides a relatively more accurate description of this grouping of carbohydrates.

Metabolism of different types of sugars

The available carbohydrates are absorbed and metabolized as the monosaccharides glucose, fructose and galactose, although lactase deficiency can result in malabsorption of lactose (Gudmand-Hoyer, 1994). While glucose is utilized by all tissues, the majority of fructose and galactose metabolism occurs in the liver, with an estimated 50–70% hepatic extraction of fructose from the portal vein. Although it is difficult to draw firm conclusions regarding the nutritional implications of the metabolic effects of ingesting different types of sugar, enough concern has been raised, particularly regarding fructose, to warrant monitoring the intake of individual sugars (Daly, 2003; Fried and Rao, 2003; Gross *et al*., 2004).

Rate of digestion and absorption

The influx of exogenous carbohydrate for metabolism is determined by the rate that carbohydrates become available for absorption at the epithelium of the small intestine. This is influenced by numerous gastrointestinal factors, including the rate that carbohydrates leave the stomach and the

Table 1 Nutritional characterization of dietary carbohydrates. Modified from Englyst and Englyst 2005

Main categories	*Chemical components*	*Nutritional grouping*	*Physiology and health*
Available carbohydrates	Sugars	Lactose	Malabsorbed by those with lactase deficiency
		Fructose (including from sucrose)	Largely metabolized by liver. Possible detrimental effect on lipid metabolism
	Starch	Available glucose from sugars, maltodextrins and starch. Rate of release measured as RAG and SAG	RAG and SAG reflect the rate of glucose release from food, which is a main determinant of the GI. Evidence to suggest that metabolic response associated with slow-release carbohydrates are most conducive to optimal health
Resistant carbohydrates		RS	Varied rate and extent of fermentation. Insufficient knowledge of effect on health
	NSP	Dietary fibre (intrinsic plant cell wall polysaccharides)	Marker for minimally refined plant foods that are rich in micronutrients and shown to be beneficial to health
		Added NSP	Varied rate and extent of fermentation. Some have specific functional properties
	RSCC	Present naturally and added	Varied rate and extent of fermentation. Some have specific functional properties
	Sugar alcohols	Present naturally and added	Partly absorbed and metabolized, and partly fermented

Abbreviations: GI, glycaemic index; NSP, non-starch polysaccharides; RAG, rapidly available glucose; RS, resistant starch; RSCC, resistant short-chain carbohydrates; SAG, slowly available glucose.

diffusion of released sugars from the alimentary food bolus. The rate that carbohydrates are released from food, through the disruption of the food matrix and the action of endogenous amylases on starch, is therefore an important determinant of carbohydrate entry to the portal vein. Carbohydrate type, biological origin and food processing all contribute to the food properties that can influence the rate of carbohydrate release (Figure 2). The nutritional significance of the rate of carbohydrate digestion and absorption is the impact it has on postprandial blood glucose homeostasis and the associated metabolic and endocrine responses. Although the glycaemic index (GI) has been the subject of some controversy, the majority of the metabolic and epidemiological evidence lends support to an increase in the consumption of slow-release carbohydrates in place of their rapidly absorbed high GI counterparts (Jenkins *et al.*, 2002; Willett *et al.*, 2002; Brand-Miller *et al.*, 2003).

Functional properties of resistant carbohydrates

Nutritionally, the most prominent resistant carbohydrates are the intrinsic plant cell-wall polysaccharides, which, as described in later sections, provide the only definition of dietary fibre consistent with the plant-rich diet. There are numerous other sources of resistant carbohydrates that occur naturally in small amounts or that have been developed as functional ingredients. These include extracted polysaccharides such as gums, oligosaccharides such as fructans, polydextrose, resistant maltodextrins and high resistant starch (RS) ingredients. Depending on their physico-chemical properties, these heterogeneous resistant carbohydrates have a range of properties that include viscosity in the upper gastrointestinal tract (Ellis *et al.*, 1996), fermentation and fermentation products (Wong *et al.*, 2006), prebiotic effects (Roberfroid, 2005; Macfarlane *et al.*, 2006) and mineral absorption (Abrams *et al.*, 2005). However, as functional ingredients can be incorporated into foods in high amounts, there has also been concern about potentially detrimental effects of large amounts of easily fermentable carbohydrate reaching the large intestine (Goodlad, 2007). There is therefore a requirement to research and evaluate how these substances should be managed from a health promotion perspective.

Food properties

The nutritional role of dietary carbohydrates cannot be adequately addressed without consideration of the overall characteristics of the foods themselves. Therefore, although fruit, vegetables and whole grains are considered carbohydrate-rich foods, their health benefits can often be attributed to their low-energy density and high content of micronutrients and phytochemicals (Southgate and Englyst, 1985; Liu *et al.*, 2000a, b, 2001; Liu, 2002; Englyst and Englyst, 2005). These overall nutritional attributes of foods need to be recognized within dietary guidelines and perhaps, just as importantly, they ought to be supported by consistent public health messages relating to dietary carbohydrate consumption.

One approach is to use a prefix identifying the food source of the carbohydrate, with the most recognized example being the division between intrinsic and extrinsic (free) sugars, which describes whether or not they are contained within cellular structures. At face value, it seems rather contradictory that consumption of the intrinsic sugars from

fruit and vegetables should be promoted, while the chemically identical extrinsic sugars are restricted. Of course, it is not the intrinsic sugars themselves that are being promoted, but rather the overall health benefits associated with the fruit and vegetable food group.

Measurement of dietary carbohydrates

Carbohydrate determinations should describe chemical composition accurately, and provide information of nutritional relevance, thereby complementing dietary guidelines. The traditional calculation of carbohydrate 'by difference' does not conform to either of these criteria as (i) it combines the analytical uncertainties of the other macronutrient measures as well as any unidentified material present and (ii) a single value for carbohydrate cannot reflect the range of carbohydrate components or their diverse nutritional properties. For this reason, carbohydrates should instead be categorized based on relevant nutritional properties, and measured as the sum of chemically identified components (Table 1). The analytical challenge is to apply chemical, physical and enzymatic approaches to exploit these characteristic differences to achieve determinations of each carbohydrate fraction. The measurement principles for the main carbohydrates are described in Table 2. It is always preferable to apply rational methods (which specifically measure the component of interest), rather than empirical methods (which are defined by the methodology) or proximate analysis, which are either prone to errors or limited in their interpretation.

The basic requirements of analytical methodologies for the determination of dietary carbohydrates as the sum of their constituent sugars can be described as follows.

Sample preparation. Preparation techniques should ensure sample homogeneity and facilitate the extraction of the nutrients of interest. For compositional analysis, this is typically achieved by freeze-drying and milling the food. The exception is for measurements of carbohydrates that reflects their digestibility (for example, RS). Such samples need to be prepared and analysed 'as eaten'.

Isolation of specific fractions. The first stage of the analysis is to ensure that the fraction of interest is completely extracted from the food matrix in its native form (for example, sugars), or dispersed to such an extent that it can be hydrolysed and measured as its component parts (for example, total starch). Interfering compounds can be accounted for by a sample blank measurement, although it is preferable to remove these by enzymatic or physical approaches, especially when the sample blank is high or the compound of interest is present in only small amounts.

Hydrolysis to constituent sugars. Once appropriately isolated, oligosaccharides and polysaccharides may be subjected to enzymatic (adds specificity) or acidic (when appropriate enzyme unavailable) hydrolysis to release their constituent sugars.

Detection. Separation by gas chromatography or high-performance liquid chromatography (HPLC) can be used to measure specific monosaccharaides, disaccharides and small oligosaccharides. Colorimetric assays can be used to measure sugars with reducing groups. Enzyme-linked colour reactions can be used for the determination of individual sugar species (for example, glucose oxidase-linked assays).

The following sections describe the specific methodological issues in the measurement of individual fractions of available carbohydrates and resistant carbohydrates.

Determination of available carbohydrates

Sugars. The common available sugars are glucose, fructose, galactose, maltose, sucrose and lactose. Aqueous extraction of these sugars is readily achieved from disrupted food matrices by a short period of heating and mixing. The solubility of sugars and the insolubility of proteins and polysaccharides in 80% ethanol can be used to further isolate them. When several sugars are present in a sample, quantification of the individual mono- and di-saccharides is best achieved by chromatography.

Maltodextrins. The short-chain α-glucan maltodextrins occur naturally in plants in only small amounts, but can be manufactured from starch by hydrolysis with acid, heat or enzymes. Analytically, maltodextrins are usually included within the total starch value, but a separate value can be obtained by measuring the glucose released by amyloglucosidase (EC 3.2.1.3) in the supernatant fraction of an 80% aqueous ethanol extraction, with a correction for glucose and maltose content.

Starch and starch digestibility. Starch, the storage polysaccharide of many plants, consists of α 1–4 linked glucose monomers and occurs as linear polymers (amylose) or as macromolecules of shorter chains with α-1–6 branch linkages (amylopectin). Historically, starch has been determined by approaches including polarimetry and the formation of starch–iodine complexes, but their use is generally considered inappropriate for complex food systems. Therefore, for quantitative determination, starch should be hydrolysed and measured as the component glucose monosaccharide units released, applying a 0.9 hydration factor to convert them to the polysaccharide on a weight basis. This is achieved most conveniently with a combination of amylolytic enzymes (typically amylase (EC 3.2.1.2) and amyloglucosidase (EC 3.2.1.3)) that hydrolyse the starch polymer, including the α-1–6 branch linkages, and the final cleavage of maltose and isomaltose to glucose. Procedures for the accurate measurement of total starch need to include a

Table 2 Principles of carbohydrate measurement

Carbohydrate	*Types and dietary occurrence*	*Extraction and isolation*	*Hydrolysis and quantification*
Sugars	The main dietary sources are *Fruit and vegetables*: sucrose, glucose, fructose *Milk and dairy*: lactose, galactose *Added sugars*: mainly sucrose, glucose, fructose	Recovered by aqueous extraction and can be further isolated from other macronutrients in an 80% ethanol fraction.	Monosaccharides and disaccharides can be determined specifically by chromatography. Enzyme linked colorimetric assays can measure individual monosaccharides.
Starch	*Starch*: consists of α-1–4-, α-1–6-linked glucose, as amylose (short linear chains) and amylopectin (larger, more branched molecules). Principle sources are cereal grain, legumes and tubers. *Maltodextrins*: Mainly from hydrolysed starch. *RS*: defined as 'the starch and starch degradation products that on average resist digestion in the small intestine'. Starchy foods have a range of physico-chemical characteristics, influencing their rate and extent of digestion.	*Total starch including maltodextrins*: Majority dispersed in aqueous conditions. Some high amylose and retrograded starch requires chemical dispersion. *Maltodextrins*: Conveniently isolated as the α-glucans (DP > 3) soluble in 80% ethanol. *RS*: isolated from samples prepared 'as eaten' with digestible starch removed by conditions that correlate with *in vivo* data. For all starch fractions glucose from sugars must be accounted for.	Starch and its components are determined as glucose released by enzymatic hydrolysis applying a hydration factor of 0.9. *Total starch including maltodextrins*: Measured as the sum of glucose released following complete dispersion of starch *Maltodextrins*: Measured as glucose released from the α-glucans (DP > 3) soluble in an 80% ethanol. *RS*: measured as glucose released from RS fraction following physical and alkali dispersion.
NSP	NSP are a grouping of several types of polysaccharide that do not have the α 1–4 glucosidic linkage characteristic of starch. *Intrinsic plant cell-wall NSP*: this component is defined as Dietary Fibre as it consistent reflect the health benefits of plant-rich diets. *Other NSP*: usually added extracts.	NSP is isolated by the dispersion and enzymatic hydrolysis of starch, which is then removed along with sugars by precipitating the NSP in 80% ethanol.	Following acid hydrolysis NSP constituent sugars are determined individually by chromatography or as a total by a colorimetric assay. *Dietary fibre*: For the majority of products total NSP provides a measure of dietary fibre *Other NSP*: Should be accounted for separately from dietary fibre. Identified by NSP sugar profiles.
RSCC	*Fructans*: Natural inulin (e.g., onions) and fructooligosaccarides from hydrolysed inulin or synthesized. *α-Galactosides*: Sucrose with galactose units, raffinose (+1), stachyose (+2), verbascose (+3). *Other RSCC*: Mainly manufactured by synthesis or by polysaccharide hydrolysis. Examples are galacto- and xylo-oligosaccharides, resistant maltodextrins, polydextrose.	*Fructans*: Aqueous extraction. Need to account for glucose and fructose, including that from sucrose. *α-Galactosides*: Aqueous extraction. If determined as monosaccharide components need to account for glucose, fructose and galactose. *Other RSCC*: Aqueous extraction and isolation from polysaccharides in an 80% ethanol fraction. Sugars, sugar alcohols, maltodextrins, fructans and raffinose family must be accounted for.	*Fructans*: Hydrolysed by fructanase and measured as fructose (and glucose) components. *α-galactosides*: Determined specifically by chromatography or alternatively hydrolysed enzymatically and measured as their components. *Other RSCC*: Determined as the monosaccharide components released by enzymatic or acid hydrolysis. Intact species can be determined by chromatography, but is less specific.
Polyols (sugar alcohols)	Occur naturally in small amounts. Manufactured and added to foods.	Polyols are easily extracted in aqueous conditions.	Polyols can be determined directly by chromatography.

Abbreviations: DP, degree of polymerization; NSP, non-starch polysaccharides; RS, resistant starch; RSCC, resistant short-chain carbohydrates.

heating step at 100°C for the gelatinization of starch granules and treatment with either dimethyl sulphoxide or sodium/potassium hydroxide for the dispersion of RS (Englyst *et al*., 1982, 1992).

It is the physico-chemical characteristics that make the enzymatic digestion of starch nutritionally interesting, and more challenging to deal with appropriately from an analytical perspective. The rate and extent that starch is hydrolysed is determined by the accessibility of the amylolytic enzymes, which explains why food processing, sample preparation and analytical methodology can all influence various aspects of starch determination.

The influence of the physico-chemical characteristics of foods on starch digestibility can be described by measuring the rate and extent of glucose released by amylolytic enzymes under *in vitro* conditions controlled for pH, temperature, viscosity and mixing (Englyst *et al*., 1992). The terms rapidly available glucose and slowly available glucose are used to describe rate of release characteristics and their physiological relevance has been confirmed by the

Table 3 Carbohydrate digestibility fractions for a selection of foods

Food	*g/100 g as eaten*				*SAG in %*
	Fru	*RAG*	SAG	*RS*	*available CHO*
Long grain rice (brown)	0.0	17.2	12.5	1.7	42.2
Rice pudding (canned)	2.5	10.9	0.2	0.2	1.7
Spaghetti	0.2	17.4	13.0	1.8	42.4
Wholemeal spaghetti	0.3	18.1	9.4	0.9	33.8
Brown bread	0.3	41.9	1.3	2.3	3.0
Wholemeal bread	0.4	35.5	1.5	1.9	3.9
Rye bread	1.7	27.5	5.1	2.8	15.0
Swiss roll	16.4	41.5	1.0	1.5	1.7
Crispbread-rye	1.6	59.5	5.4	2.5	8.1
Oatcakes	0.7	47.8	8.4	2.6	14.7
Water biscuit crackers	0.9	72.7	5.1	0.2	6.5
Digestive biscuits	4.8	44.2	11.4	1.1	18.9
Shortbread	8.2	31.1	21.9	1.8	35.7
Corn flakes	3.8	81.7	2.5	3.6	2.8
Muesli	17.1	38.6	4.7	1.3	7.8
Shredded wheat	1.2	68.7	2.8	2.4	3.9
Potato salad	1.7	8.0	2.1	1.0	17.8
Boiled potatoes	0.6	13.9	0.4	0.5	2.6
Potato crisps	0.2	50.7	1.8	1.0	3.4
Chick peas	0.5	4.2	11.7	3.5	71.0
Green split peas	0.5	6.9	8.7	3.4	54.2
Red kidney beans	0.8	6.4	8.3	3.4	53.3
Haricot beans	0.4	4.6	7.9	3.6	61.1
Baked beans	5.7	11.9	1.8	1.8	9.5
Sweetcorn	1.5	13.2	4.0	0.6	21.4

demonstration of strong correlations between their content in foods with glycaemic response and GI values (Englyst *et al.*, 1999, 2003). Table 3 shows the carbohydrate-release characteristics for a range of products.

Determination of resistant carbohydrates

Polyols (sugar alcohols). The polyols are hydrogenated carbohydrates, which include sorbitol, mannitol, xylitol and maltitol. Typically, there is only a small intake of naturally occurring polyols, principally in the form of sorbitol from apples and pears. However, the various types of polyols can be manufactured and are used as sugar replacers, which can be present in large amounts in some products, particularly 'sugar-free' confectionery. Different polyol types can be absorbed and metabolized to varying extents, although a proportion of most polyols enters the large intestine as fermentation substrates (Livesey, 2003). As it is not practical to encompass the varied fate of polyols within a classification scheme, polyols are usually considered within the resistant carbohydrate grouping.

The sugar alcohols are easily extracted in aqueous or ethanol fractions and can be measured directly by HPLC. They are more stable than sugars in alkali conditions, a feature that can be utilized to isolate and measure sugar alcohols specifically by gas chromatography without interference from sugars (Quigley *et al.*, 1999).

Resistant short-chain carbohydrates. This fraction encompasses all fructans, and the resistant carbohydrates that are soluble in 80% ethanol, other than the sugar alcohols. The resistant short-chain carbohydrate term was developed in order not to be constricted by the chemical definition of oligosaccharides as degree of polymerization (DP) 3–9, as molecules of considerably higher DP may be included, depending on branching (Englyst and Hudson, 1996; Quigley *et al.*, 1999). The terms non-digestible oligosaccharides and resistant oligosaccharides are synonymous with the RSCC term and in practice all describe the same substances.

This diverse group of substances are typically consumed in only small amounts from naturally occurring sources, principally as fructans present in onions, Jerusalem artichoke, wheat and chicory, and as the small α-galactosides (raffinose family) from legumes, with more complex galactooligosacchides found in breast milk. A range of resistant oligosaccharides have also been developed as functional ingredients. There is a need to have a consistent approach to the determination of all RSCCs, so that their dietary content and relevance can be identified, and to prevent the potential for gaps in carbohydrate classification and measurement. There are several approaches to the determination of RSCC depending on the chemical characteristics of the individual species.

It is convenient to measure fructans as their fructose and glucose constituents after specific hydrolysis with fructanase (EC 3.2.1.7) (McCleary *et al.*, 2000). The α-galactosides can either be determined individually by chromatography or by their monosaccharide constituents after enzymatic hydrolysis with α-galactosidase (EC 3.2.1.22) and α-glucosidase (EC 3.2.1.20) (Vinjamoori *et al.*, 2004).

Other RSCC can be measured by a single method based on the acid hydrolysis of RSCC isolated in an 80% ethanol extract and determination of constituent sugars by gas chromatography (Quigley *et al.*, 1999). This approach is suitable for the determination of a wide range of substances, including, but not restricted to isomalto-oligosaccharides, xylo-oligosaccharides, galacto-oligosaccharides and resistant maltodextrins.

There are also methods for the determination of RSCC by the chromatographic separation of intact species. However, as discussed later, this type of method can lack specificity and there is the possibility of overlap between different methods, which would lead to double counting and an overestimate of the total RSCC content.

Non-starch polysaccharides. This group of carbohydrates is defined as the polysaccharides that do not contain the α-1–4-linked glucose that is characteristic of starch. There are various types of non-starch polysaccharides (NSP) that differ in their sugar composition and glycosidic linkages, which are important features in determining their physico-chemical properties. The NSP present in plant cell walls have a structural function in defining the integrity of plant cells and tissues. NSP can also occur as gums and mucilages, some

Table 4 NSP in a selection of foods

Food	*NSP g/100* g *as eaten*	*NSP constituent sugars*							
		Rha	*Fuc*	*Ara*	*Xyl*	*Man*	*Gal*	*Glu*	*Uronic acid*
Bran, wheat	37.2	0.0	0.0	8.8	16.8	0.1	0.6	9.8	1.1
Bread, wholemeal	5.2	0.0	0.0	1.5	2.0	0.1	0.2	1.3	0.2
Bread, white	1.6	0.0	0.0	0.5	0.7	0.1	0.1	0.3	0.0
Bread, Rye	7.3	0.0	0.0	1.9	3.2	0.1	0.2	1.9	0.1
Cornflakes	0.9	0.0	0.0	0.1	0.3	0.0	0.0	0.4	0.1
Meal, oats	7.0	0.0	0.0	0.8	1.1	0.0	0.1	4.9	0.2
Spaghetti, White	1.2	0.0	0.0	0.4	0.5	0.0	0.1	0.2	0.0
Spaghetti, Wholewheat	3.5	0.0	0.0	1.0	1.3	0.1	0.1	1.0	0.1
Beans, French	3.1	0.0	0.0	0.2	0.2	0.1	0.4	1.2	0.9
Peas, Cooked	5.2	0.1	0.0	1.1	0.2	0.0	0.1	3.0	0.7
Potato	1.2	0.0	0.0	0.1	0.0	0.0	0.4	0.5	0.2
Cabbage, Winter	3.7	0.2	0.0	0.8	0.2	0.1	0.4	1.2	0.9
Cauliflower, Cooked	1.6	0.1	0.0	0.3	0.1	0.0	0.2	0.6	0.4
Broccoli	3.0	0.1	0.0	0.4	0.2	0.1	0.3	1.1	0.9
Carrots, Cooked	2.5	0.1	0.0	0.3	0.0	0.1	0.4	0.9	0.8
Tomato	1.1	0.0	0.0	0.1	0.1	0.1	0.1	0.5	0.3
Cucumber	0.5	0.0	0.0	0.0	0.0	0.0	0.1	0.2	0.1
Apple, Cox	2.0	0.1	0.0	0.3	0.1	0.1	0.1	0.7	0.6
Peach	1.5	0.0	0.0	0.3	0.1	0.0	0.1	0.5	0.5
Banana	3.3	0.1	0.0	0.4	1.0	0.1	0.2	0.4	1.2
Orange	2.1	0.0	0.0	0.3	0.1	0.1	0.3	0.5	0.9
Melon, Honeydew	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.2
Kiwi fruit	1.7	0.0	0.0	0.0	0.2	0.1	0.1	0.7	0.6
Hazelnuts	6.5	0.2	0.0	1.4	0.5	0.0	0.5	2.3	1.6

of which are extracted and used as food additives for their technological properties. Biological variation between plant species means that different food groups have characteristic profiles of NSP, as identified by their spectrum of constituent sugars (Table 4). The NSP glucose is present in all food types, occurring mainly in the form of cellulose, but some cereal products, such as oats and barley, can contain considerable amounts of the more soluble β-glucan. Galacturonic acid is the main component of pectin found in fruit and vegetables. Xylose is found predominantly as arabinoxylans in cereals. Arabinose, mannose and galactose are present in all food types, with rhamnose and fucose being present in only small amounts in some fruits and vegetables.

As NSP is a chemically defined substance, its occurrence in foods can be measured directly and specifically. This is achieved by the extraction and isolation of these polysaccharides from other carbohydrate components, with their subsequent hydrolysis to constituent sugars for colorimetric or chromatographic determination. Some individual NSP such as β-glucans, can be determined as their constituent sugars released by hydrolysis with the relevant substrate-specific enzymes.

Currently, the most convenient determination of total NSP is achieved by hydrolysing the extracted NSP by an acid treatment with measurement of the released monosaccharides (Englyst *et al*., 1982, 1994). Extraction involves the complete dispersal of starch with dimethylsulphoxide and aqueous gelatinization, making it susceptible to hydrolysis by a combination of amylolytic enzymes. The NSP is isolated by precipitation in acidified 80% ethanol, which removes hydrolysed starch and any sugars present in the sample. If information on NSP solubility is required, then the insoluble fraction can be extracted by precipitation in pH 7 phosphate buffer, which together with a total NSP value can be used to calculate the soluble fraction. While 2 M sulphuric acid is sufficient for the hydrolysis of most types of NSP, an initial 12 M sulphuric acid step is needed for the hydrolysis of cellulose, and indeed omitting this step can be used to calculate the cellulose content of a sample. Pectin is difficult to hydrolyse, requiring a subsequent incubation with pectinase. The sugars released may be measured by gas liquid chromatography or HPLC to obtain values for individual monosaccharides. A single value for total sugars may be obtained by colorimetric determination of reducing groups (Englyst *et al*., 1994). Further issues relating to NSP are considered in the sections on the definition and measurement of dietary fibre.

Resistant starch. Starch digestion by endogenous enzymes is a continuous process during passage through the small intestine, which, for a number of different reasons may not go to completion. Starch can escape digestion in the small intestine because it is physically inaccessible, within the food matrix (RS1), or within starch granules (RS2), or because it is present as retrograded starch (RS3) produced during food manufacture and preparation. For many foods, a small

proportion of the starch present will be RS (typically 0–5% of starch in most cereal products) although for some foods such as legumes this is higher (typically 10–20% of starch for some beans), and food processing influences the amount present. In addition, starch can be chemically modified to a form that is resistant to enzymatic digestion (sometimes termed RS4), which includes starch that has been etherized, esterified or cross-bonded. Various high RS preparations have become available, through either plant breeding to increase amylose contents, or by physical and chemical manufacturing processes that increase the RS fraction of starches. As each RS preparation has its own specific physico-chemical characteristics that can influence the rate and site of fermentation in the colon, it is appropriate that each preparation should be considered on its own merits, and the properties associated with one preparation cannot necessarily be extrapolated to others.

RS is physiologically defined on the basis of *in vivo* studies. Quantifying the amount of starch entering the human colon presents considerable technical problems (Champ *et al.*, 2003). Hydrogen excreted in breath has been used as an indicator of fermentation in the colon, but it is considered to lack the sensitivity required for quantification. Intubation has been used to sample digesta from the ileum, but this technique is restricted to liquidized meals. Several studies have used human ileostomy subjects as a model to investigate the digestive physiology of the small intestine. Carbohydrate analysis of the ileostomy effluent allows determination of the amount of starch escaping digestion in the small intestine, which was found to vary between individuals by $\pm$20% around the mean (Englyst and Cummings, 1985, 1986, 1987; Silvester *et al.*, 1995). It is from these studies that the currently accepted definition of RS is derived as 'the sum of starch and starch degradation products that, on average, reach the human large intestine' (Englyst *et al.*, 1992). The challenge for *in vitro* methods that attempt to quantify the RS content of foods is to reflect the mean values obtained by the *in vivo* studies.

Except for the modified RS4, resistant starch is not chemically different from starch digested and absorbed from the small intestine although products high in amylose have a higher propensity to form RS. The amounts of RS in foods is largely dependent on the degree of food processing, which can result in an increase or a decrease in the RS values from those found in the raw product. Therefore, RS needs to be measured in foods as they would normally be eaten, and values cannot be derived by summing the RS contents of raw ingredients, or indeed be measured in samples that have undergone laboratory preparation (freeze-drying/milling) before analysis, as this can influence the RS content.

Numerous RS methods have been proposed which have been reviewed (Champ *et al.*, 2003). The basis of methods for the determination of RS is the measurement of starch remaining unhydrolysed after a defined period of incubation with amylolytic enzymes. Measurements of total RS must include RS1, RS2 and RS3. However, due to inappropriate sample preparation and gelatinization of starch during the procedure, many methods measure only RS3, or only RS2 and RS3 fractions. In addition, few of the methods have been developed and validated in conjunction with *in vivo* studies. Consequently, highly variable RS values have been reported for similar foods.

In summary, methods designed to investigate the rate and/or the extent of starch digestion in the human gut should incorporate the following principles; analysis of foods prepared 'as eaten', reproducible disruption of the physical structure of food, standardized conditions of amylolytic hydrolysis, development and validation of methods in conjunction with *in vivo* studies. The analytical procedure for rapidly available glucose, slowly available glucose and RS determination conforms to these principles, and has been tuned to yield values for RS that match the mean proportion of starch recovered in ileostomy studies for both single foods and mixed meals (Englyst *et al.*, 1992; Silvester *et al.*, 1995).

Dietary fibre

The term dietary fibre has been applied by different researchers and in varied disciplines within the field of nutrition to describe a diverse range of substances. This has resulted in often disparate interpretations of what is meant by the term, a situation that has not been helped by the fact that the phrase 'dietary fibre' does not in itself provide an unambiguous description of what it consists of. At best, 'dietary' infers that this is a food component, and 'fibre' implies a fibrous, coarse or structural nature. It is agreed that it was originally used as shorthand for plant cell walls and that it was intended as a nutritional term describing a potentially beneficial characteristic of the diet.

In the intervening period, a large number of definitions have been proposed. Suggestions have included one or more of the following characteristics: chemical identity; intrinsic material occurring naturally in foods; resistance to digestion in the small intestine; demonstration of a specific physiological effect; and material recovered by a particular methodology. Where inconsistencies between definitions have emerged, these can be related directly to which inclusion criteria have been applied. In striving for a usable approach, it is important that any limitations associated with proposed definitions are identified so that any potential conflict with dietary guidelines and health issues can be assessed.

In essence, the current situation can be summarized by two contrasting approaches. One firmly retains the link with plant foods with the definition 'dietary fibre consists of intrinsic plant cell-wall polysaccharides', which remains true to the original concept. The other approach has 'indigestibility in the small intestine' as its central feature and encompasses a wider range of substances from diverse sources. In November 2006, the Codex committee on nutrition and foods for special dietary uses (CCNFSDU) was

asked to consider both the 'plant-rich diet' and the 'indigestibility' approaches to dietary fibre definition.

As the substances included within these two approaches to definition is not the same, the potential public health implications associated with each of them will also differ. Its prominent position in guidelines and the associated health messages has led to good consumer recognition of the dietary fibre term. Therefore, dietary fibre is very much a public health term, and a foremost consideration must be to ensure that consumers can interpret dietary fibre values and any associated nutrition claims in a manner that assists them with informed choice in diet selection, and that does not present the opportunity for the misrepresentation of products.

An important aspect of nutrition is the requirement to describe clearly defined nutritional components. To this end, methods of analysis are a secondary issue, where suitability should be assessed on how well they measure the intended component. This principle should apply to the definition and measurement of every nutrient, but for dietary fibre there has been an inappropriate emphasis on methodology, to the extent that some proposed definitions have been based on the material recovered by a particular analytical procedure. This is an unacceptable situation with respect to describing food composition. Analytical methods should be 'fit for purpose', which for dietary fibre can be assessed by the following criteria: (1) whether the material described in the respective stated aims of methods are suitable as a measure of dietary fibre; (2) the degree to which the methods actually measure the material described within their respective stated aims.

The sections that follow evaluate the rationales behind the 'plant-rich diet' and 'indigestibility' approaches to the definition of dietary fibre, including discussion of their perceived limitations (summarized in Table 5). This includes an assessment of the methodological approaches available for the determination of the substances included in each definition, with a comparison of the main NSP and enzymatic–gravimetric methods provided in Table 6.

The plant-rich diet approach to dietary fibre definition

Associated definition. By this approach, dietary fibre is a characterized component of plant foods, providing a consistent indicator of the minimally refined plant-rich diet promoted by the food-based guidelines for dietary fibre consumption. As part of the Food and Agriculture Organization/World Health Organization scientific update on carbohydrates in human nutrition, held in Geneva July 2006, it was agreed that the definition of dietary fibre should maintain this clear link to fruits, vegetables and whole grain cereals and the following definition was subsequently endorsed on behalf of the carbohydrate scientific update for consideration by CCNFSDU.

'*Dietary fibre consists of intrinsic plant cell-wall polysaccharides*'

This definition, together with its rationale and associated measurement, was proposed at the Geneva meeting in the presentation relating to this paper, and is described in the following sections. The definition cannot easily be misinterpreted, as demonstrated when evaluating its component parts:

- *'Intrinsic'*—This emphasizes that the health benefits of plant-rich diets is not restricted just to the plant cell wall (or its polysaccharide component), but may relate as well to the overall profile of associated micronutrients and phytochemicals.
- *'Plant cell wall'*—This identifies the food component of interest and therefore specifies that it is the structural polysaccharides of the plant cell wall that should be determined.
- *'Polysaccharides'*—This establishes that dietary fibre is a carbohydrate term, providing the required chemical element that should form an essential part of any definition.

Rationale and implications of the plant-rich diet approach. The modern concept of dietary fibre as a protective nutritional component has stemmed largely from observations that diets rich in unrefined plant foods were associated with a lower incidence of certain diseases including diverticular disease, colon cancer and diabetes (Burkitt, 1969; Trowell, 1972; Trowell *et al.*, 1985). When compared with their refined counterparts, the most prominent identifying characteristic of these foods and diets was the presence of largely unprocessed plant cell-wall material, which is composed predominantly of structural polysaccharides. The need to distinguish this carbohydrate fraction on the basis that it did not provide the same energy as starch and sugars was already recognized (McCance and Lawrence, 1929). However, rather than the issue of energy alone, it has been the prospect of an association with more direct health benefits that has driven the demand for a dietary fibre term. The prominent public health status of dietary fibre and the positive nutritional message it conveys is largely the result of the consistent advice within dietary guidelines to increase consumption of dietary fibre in the form of fruits, vegetables and whole grains (Department of Health, 1991; WHO, 2003; USDA/DHHS, 2005). Furthermore, the reference intake values and nutrition claims relating to dietary fibre have been established from the health benefits that have been associated with the intake of these naturally high fibre foods. For example, the current American reference intake values are based mainly on three prospective studies on the association with cardiovascular disease (Pietinen *et al.*, 1996; Rimm *et al.*, 1996; Wolk *et al.*, 1999; IOM, 2002).

There are important distinctions between advice that specifies increased intake of dietary fibre from specific food groups, as opposed to a solely nutrient-based approach. For instance, their high water content means that fruits and vegetables may not at first appear to be particularly good sources of dietary fibre when they are considered in terms of g/100 *g* as consumed. In fact, it is precisely this quantitatively small amount of plant cell wall material that confers the

Table 5 Comparison of the plant-rich diet and indigestibility approaches to the definition of dietary fibre

Plant-rich diet approach	*Indigestibility approach*
Definition: 'Intrinsic plant cell-wall polysaccharides'.	*Definition*: 'Indigestible carbohydrate (DP > 3) and lignin.'
Rationale: This definition is targeted specifically at the fruits, vegetables and whole grain products that are consistently linked with health benefits. These foods have the characteristic feature of containing plant cell walls, which mainly consist of structural polysaccharides, which can be quantified in chemical terms. Other non-carbohydrate components are not included as they can neither be determined specifically nor would their inclusion enhance the definition as an indicator of these foods. The definition recognizes that the benefits of a natural fibre-rich diet are not due to any single component, but rather the effect of synergistic elements including micronutrients, phytochemicals and low energy density.	*Rationale*: There are numerous versions of this definition, which have the common feature of placing the emphasis on escaping digestion in the small intestine. The definition is not restricted to carbohydrates as it encompasses lignin and other substances associated with the plant cell wall. In addition to the plant cell-wall polysaccharides, the indigestibility criterion has the implication of including RS and other extracted or synthesized carbohydrates, including resistant oligosaccharides. However, as this grouping can include a wide range of substances it has been suggested that there should also be a demonstrated physiological effect for a specific material to be included.
Scientific evidence for rationale: This is a food-based rationale, which is strongly supported by the epidemiological evidence for the health benefits of fruits, vegetables and whole grain products. Retaining a distinct dietary fibre term identifying plant-rich diets with their unique health benefits reinforces the food-based dietary guidelines. This distinction allows the properties of other resistant carbohydrates to be researched and if appropriate promoted in their own right.	*Scientific evidence for rationale*: For the existing epidemiological evidence relating to the last few decades this definition provides a reasonable indicator of plant-rich diets, as supplementation with resistant carbohydrate preparations was uncommon. However, this is not always the case for manufactured products developed recently. Specific physiological properties have been associated with individual supplements, but these vary depending on type, making it difficult to consider them within a single definition. The long term health effects/ safety remains to be established.
Nutrition labelling: A dietary fibre value describing intrinsic plant cell-wall polysaccharides would guide consumers to the selection of plant-rich foods. If other sources of resistant carbohydrates are present, then there would be scope for these to be labelled specifically.	*Nutrition labelling*: By the indigestibility approach the fibre label would not provide a consistent indicator of plant-rich foods that may mislead consumers who have this expectation. By grouping all indigestible carbohydrates within a single undifferentiated nutrition label, there is less opportunity to identify any added functional ingredients.
Nutrition and health claims: The claims for dietary fibre are largely based on the epidemiological evidence, which relates to fibre from plant-rich diets. When appropriate, specific health claims should be established for individual resistant carbohydrate functional ingredients, thereby acknowledging their specific properties and taking account of variations in their effective and safe dosages.	*Nutrition and health claims*: The epidemiological evidence for dietary fibre cannot be extrapolated to a definition that includes enzymatic-gravimetric values of unknown composition, as well as a range of supplemented materials with varied functional properties. There is the potential for inappropriate nutrition claims for materials with either no effect or detrimental properties, which would undermine the position of dietary fibre as a beneficial food component.
Population reference intakes: The population reference intake values for dietary fibre are largely based on the epidemiological evidence that minimally refined plant-rich diets are associated with a lower incidence of several diseases. The intrinsic plant cell-wall polysaccharide definition ensures that dietary fibre intakes contributing towards the reference value would consistently reflect both the epidemiological evidence and the intended message of the dietary guidelines.	*Population reference intakes*: The use of this definition could result in a situation where the consumer selects supplemented products on the basis that they will contribute towards the reference intake value, although in reality this would not be a true reflection of the intention of the dietary guidelines. This raises two concerns (1) that the supplemented product is unjustly promoted on the back of the epidemiological evidence; and (2) that if direct substitution of products occurs, then the consumption of the intended target of plant-rich food groups may be diminished.
Impact on food industry: Although values for 'intrinsic plant cell-wall polysaccharides' are generally lower than those for the indigestibility approach, this should not make a difference to the marketing of the majority of products, as population reference intakes and claims would be based on the plant-based approach. The emphasis would be on manufacturers to incorporate minimally refined plant ingredients into products to achieve nutrition claims for dietary fibre. There would be a positive opportunity to market other types of resistant carbohydrates with respect to their specific functional properties. For food labelling purposes, there would be cost savings with the analysis of NSP compared to the enzymatic-gravimetric and supplementary analysis.	*Impact on food industry*: With this definition, there would be less impetus for the manufacturer to incorporate unrefined plant ingredients, as it would be possible to elevate the dietary fibre content through processing or supplementation instead. However, it would be difficult for the consumer to distinguish between these different types of product if they carried identical nutrition claims. This may be perceived as conflicting with the intended aim of reference intake values and dietary guidelines which are targeted at plant-rich diets. Grouping varied functional ingredients together limits the opportunities for manufacturers to promote the specific properties of individual products. As gravimetric values are influenced by food processing, food labelling cannot be based on food table values of component ingredients.
Impact on nutrition research: Maintaining 'intrinsic plant cell-wall polysaccharides' as a distinct definition of dietary fibre facilitates research	*Impact on nutrition research*: The indigestibility approach groups diverse substances including plant cell-wall material, retrograded starch,

Table 5 *Continued*

Plant-rich diet approach	*Indigestibility approach*
into the benefits of plant-rich diets, and encourages specific research into types of resistant carbohydrate preparations. Only with detailed information on distinct substances will it be possible for future epidemiological studies to establish the intakes and effects of different types of resistant carbohydrates.	supplements and non-carbohydrate artifacts in unknown proportions. This single undifferentiated grouping will not provide the detailed information required by future epidemiology studies to establish the intakes and health effects of different types of resistant carbohydrates. Nutrition research is better served by detailed information on specific food components.

DP, degree of polymerisation.

Table 6 Comparison of the principles and analytical issues relating to the principal methods associated with the plant-rich diet approach (NSP method) and the indigestibility approach (enzymatic–gravimetric methods) to the definition of dietary fibre

NSP Method	*Enzymatic–gravimetric methods (AOAC 985.29 & 991.43)*
General principles	*General principles*
Stated aim: To measure polysaccharides that do not contain the α 1–4 glucosidic linkages characteristic of starch (i.e., NSP) *Analytical principle*: Complete dispersion and enzymatic hydrolysis of starch. Precipitate residue in 80% ethanol and isolate by centrifugation. Hydrolyse and measure NSP as constituent sugars by colorimetry or chromatography. *Information provided*: Values for total, soluble and insoluble NSP, with the option of detailed information on constituent sugars by the GC version. *Effect of food processing*: As a chemically distinct food component, NSP is minimally affected by normal food processing. *Is stated aim achieved*: Yes. The procedure completely removes starch and sugars and provides a specific determination of NSP.	*Stated aim*: To measure the sum of indigestible polysaccharides and lignin. *Analytical principle*: Partial enzymatic hydrolysis of starch and protein. Precipitate residue in 80% ethanol and isolate by filtration. Record total residue weight and then determine and subtract ash and protein contents. *Information provided*: Weight of total, soluble and insoluble residue containing carbohydrate and non-carbohydrate material in unknown proportions. *Effect of food processing*: A range of materials are recovered in the residue, which is highly dependent on food processing (e.g., retrograded starch, Malliard reaction products). *Is stated aim achieved*: No, not consistently. In addition to NSP, this procedure measures a variable amount of RS, which may not relate to the true extent of physiological starch digestion. In addition to lignin, the non-carbohydrate part can include food processing artifacts.
Analytical issues	*Analytical issues*
Specific reagents and equipment: *Enzymes*: Heat stable amylase, (EC 3.2.1.1), pullulanase (EC 3.2.1.41), pancreatin (these enzymes should be devoid of NSP hydrolytic activities), pectinase (EC 3.2.1.15). *Analysis vessels*: screw cap test tubes. *Equipment*: Centrifuge and either spectrophotometer or GC system. *Practical issues*: All the steps of this procedure are conducted in test tubes, which makes it well suited to the analysis of large batch sizes. It is important to ensure complete starch dispersion and hydrolysis, which is achieved by a combination of physical, chemical and enzymatic steps. The chemical end-point determination techniques are those used in the measurement of other carbohydrates (e.g., sugars, starch). The procedure takes 1 day with colorimetric measure or 1.5 days for GC measure. *Environmental impact*: Only small amounts of solvent waste generated. *Suitability for use in developing countries*: The NSP procedure only requires standard laboratory equipment including a spectrophotometer for the colorimetric version. *Traceability*: The primary standard is a representative mixture of the individual monosaccharides of NSP. *Method specificity*: Only NSP is measured, with no interference from other substances. *Method reproducibility*: A range of certified reference materials are available (e.g., BCR). Method CV $<5\%$.	*Specific reagents and equipment*: *Enzymes*: Heat stable amylase, (EC 3.2.1.1), protease, amyloglucosidase (EC 3.2.1.3). These enzymes should be devoid of NSP hydrolytic activities. *Analysis vessels*: 400 ml beakers and fritted glass crucibles. *Equipment*: vacuum manifold, muffle furnace and Kjeldahl equipment. *Practical issues*: Batch sizes are limited by the difficulties of handling large numbers of 400 ml beakers. The selective removal of starch other than RS is difficult or impossible to achieve within this procedure. The method is labour intensive due to: preparation and repeated weighing of the crucibles; numerous pH checks; manual transfer and filtration of residues; subsidiary ash and Kjeldahl methods. The procedure takes 1.5–2 days or more with longer filtration times. *Environmental impact*: Large amounts of solvent waste are generated. *Suitability for use in developing countries*: The gravimetric procedure requires specialist glassware, muffle furnace and Kjeldahl equipment for the measurement of nitrogen. *Traceability*: No primary standard is available as the procedure does not measure a chemically distinct component. *Method specificity*: Any added material or food processing artefacts recovered in the residue are a potential source of interference. *Method reproducibility*: A range of certified reference materials are available (e.g., BCR). Method CV $<5\%$.

Table 6 *Continued*

NSP Method	*Enzymatic–gravimetric methods (AOAC 985.29 & 991.43)*
Suitability as a measure of dietary fibre	*Suitability as a measure of dietary fibre*
Potential discrepancies with definitions: For plant foods, the NSP content is a measure of 'intrinsic plant cell-wall polysaccharides'. In a few plants NSP can occur as gums and alginates, but these are not typical foods and are more likely to occur as ingredient extracts. When extracted or synthesized NSP are present in products then these will be known by the manufacturer and can be deducted from the NSP measurement to obtain a value for the intrinsic plant cell-wall polysaccharides. The presence of specific extracts can often be identified by their NSP constituent sugar profile. With the plant cell-wall polysaccharide definition, resistant oligosaccharides and RS are separate groupings. Their content in foods is measured specifically and they do not conflict with the NSP measurement. *Evaluation of method*: The intrinsic plant cell-wall polysaccharide definition provides a clear link to the plant-rich diet shown to be beneficial to health. The NSP procedure provides measurements that are suitable for this definition.	*Potential discrepancies with definitions*: As the AOAC gravimetric procedure measures a range of indigestible materials of varied composition and origin it does not provide a consistent measure of plant cell-wall material. It can include non-carbohydrate food processing artifacts (e.g., Maillard reaction products) that are not part of any dietary fibre definition. The residual starch recovered can be misleading, as it does not relate to physiologically RS, for which separate measurement is required. It does not recover resistant oligosaccharides, resistant maltodextrins or all RS, and therefore by itself does not provide a measure of the indigestible carbohydrates proposed for inclusion. These substances require separate analysis if they are to be included. *Evaluation of method*: The indigestible carbohydrate and lignin definition does not consistently identify plant-rich diets. Neither does the AOAC gravimetric procedure provide a consistent measurement of the material included in this definition.

Abbreviation: GC, gas chromatography.

high water-holding ability of fruit and vegetables, which in turn is responsible for their low energy density. In addition, the plant cell walls have a central role in defining the high nutrient density with respect to vitamins, minerals and phytochemicals, which are considered as closely associated companion nutrients. These are unique nutritional properties associated with the dietary fibre in these food groups, and therefore food-based guidelines for dietary fibre are always applicable even if, as is the case with fruits and vegetables, their contribution to dietary fibre intake is often modest compared with that derived from whole grain products.

The benefits of plant cell-wall-rich foods is supported by prospective observational studies that identified significant inverse relationships between intake of fruits, vegetables and whole grains and incidence of cardiovascular disease, diabetes and some cancers (Jacobs *et al*., 1998; Liu *et al*., 1999, 2003; van Dam *et al*., 2002; Bazzano *et al*., 2003, 2005; Rissanen *et al*., 2003; Slavin, 2003; Steffen *et al*., 2003; WHO, 2003). To ensure consistency for the public health message being conveyed, it is essential that any measure of dietary fibre is a true representation of the unrefined plant food-based diet endorsed by the epidemiological evidence and dietary guidelines.

The nutritional relevance of 'intrinsic plant cell wall polysaccharides' can be considered at various levels (1) as a distinct carbohydrate component of the food, (2) as a provider of cell wall structures and (3) as a marker of a diet rich in micronutrients. When present as an intrinsic part of plant foods, these elements connected with cell-wall polysaccharides cannot be disassociated from one another, with the implication that it is not possible to assign the benefits of dietary fibre-rich diets to just one of these attributes. In other words, the proposal to define dietary fibre as the 'intrinsic plant cell-wall polysaccharides' is based on the fact that this is the only component that is consistently associated with the plant-rich diet linked with reduced disease incidence.

Determination of intrinsic plant cell-wall polysaccharides. This approach describes a chemically defined food component that can be determined by an enzymatic–chemical method. The stated aim of this method is to measure the polysaccharides that do not have the α-(1–4) glucosidic linkages characteristic of starch. Therefore, the method is designed to disperse and remove all starch, with NSP measured as the sum of chemically identified NSP constituent sugars (Englyst *et al*., 1994).

The enzymatic–chemical method for the analysis of NSP is an extension of the pioneering work of McCance and Lawrence (1929) and later of Southgate (1969), which recognized the importance of the direct measurement of the various types of carbohydrates for nutrition composition purposes. NSP forms part of the unified scheme to classify and measure all food carbohydrates (Table 1). To evaluate it as a measure of dietary fibre, the NSP method is assessed here in terms of its suitability to measure 'intrinsic plant cell wall polysaccharides'.

In typically consumed unsupplemented foods, the entire NSP component will be derived from the intrinsic plant cell wall. The advantage of NSP as a chemically distinct substance is that it is not in itself created or destroyed by normal food preparation or storage techniques, which means that NSP can be used as a fairly consistent indicator of plant cell-wall material. When added preparations of NSP are present in foods, these too are measured as their carbohydrate components, and will contribute to the total NSP value. Manufacturers' data on the amount and type of carbohydrate preparation used will normally be sufficient to

account for any supplemented material present. However, for the purpose of traceability and authenticity checks, it would in most cases be possible to identify the presence of specific preparations by their profile of constituent NSP sugars. For example, the presence of guar gum in a product can be identified by higher galactose and mannose compared with the NSP sugar profile of the unsupplemented food.

Unfortunately, a lack of understanding of the practical issues involved has led to inaccurate statements about the complexity of the NSP method. The actual situation is that the enzymatic–gravimetric method, promoted as part of the 'indigestibility approach', is more time consuming, resource demanding and subsequently more expensive to perform than the NSP procedure. The NSP method has been subjected to successful collaborative trials (Wood *et al*., 1993; Pendlington *et al*., 1996) and for routine purposes, including food labelling, NSP can be determined by colorimetry with a simple spectrophotometer. Furthermore, the NSP method is well suited to the analysis of large batch sizes as it uses test tubes as the reaction vessel, compared with cumbersome 400 ml beakers and filtration crucibles used in the enzymatic-gravimetric methods.

The indigestibility approach to dietary fibre definition

Associated definition. By this approach, the primary defining characteristic is indigestibility in the small intestine, thereby grouping together diverse substances. In addition to cell-wall polysaccharides, such a grouping would include non-structural carbohydrates that are normally absent, or present only in small amounts in most foods (for example, inulin), and in the case of RS is largely dependent on food processing. Also included would be extracted, synthesized, or otherwise manufactured polysaccharides and oligosaccharides that could be added to individual foods in considerable amounts. A proposed definition based on this approach has been considered by CCNFSDU in the context of providing guidelines for the use of nutrition claims. The proposed definition states as follows:

Dietary fibre means carbohydrate polymers with a DP not lower than 3, which are neither digested nor absorbed in the small intestine. A DP not lower than 3 is intended to exclude mono- and disaccharides. It is not intended to reflect the average DP of a mixture.

Dietary fibre consists of one or more of edible carbohydrate polymers naturally occurring in the food as consumed, carbohydrate polymers, which have been obtained from food raw material by physical, enzymatic or chemical means, synthetic carbohydrate polymers.

In addition, this definition is associated with a lengthy footnote included to justify the use of specific enzymatic–gravimetric methods, which are acknowledged to recover a wide range of non-carbohydrate materials that would otherwise fall outside the stated definition.

Also linked with the definition is a statement relating to the physiological properties generally considered to be associated with dietary fibre and the recommendation that 'where a declaration or claim is made with respect to dietary fibre, a physiological effect should be scientifically demonstrated', with the exception of naturally occurring polymers for which no such justifying criteria were deemed necessary. With respect to the application of this definition, it goes on to raise issues as to the food safety requirements, diverse efficacies of different substances purporting to be dietary fibre, and the consumer perception of fibre as being of plant origin.

Due to the diverse nature of the substances included, there is no single analytical method currently available that will provide an accurate and comprehensive determination of the material encompassed within the indigestibility approach. Instead, 10 methods of analysis are stated in connection with this definition, with one of two versions of an enzymatic–gravimetric technique considered to be the principal method.

Rationale and implications of the indigestibility approach. As evidenced by the length of the above definition and associated conditions, the rationale for this indigestibility approach is necessarily more complex as it tries to amalgamate issues of food composition, analytical methodologies and physiological attributes. The primary basis for the indigestibility approach is the fundamental difference in the physiological handling of carbohydrates depending on their gastrointestinal fate.

However, although it may be possible to group diverse substances by a shared attribute such as indigestibility, this does not mean that such groupings should necessarily form the basis for dietary advice. The relation between the amount and type of fermentable substrate reaching the colon and related physiological parameters is incompletely understood, with both beneficial and potentially adverse effects having been reported. There is insufficient evidence to suggest that all sources of resistant carbohydrates should be actively promoted, or that it would be desirable to set a single population reference value for total resistant carbohydrate intake. Nevertheless, this would in essence be the prospect with the indigestibility based dietary fibre definition.

As the characteristic of 'being neither digested nor absorbed in the small intestine' does not in itself equate to a health benefit, it is implied that a further level of justifying criteria are needed for functional ingredients to be considered as dietary fibre by the indigestibility approach. This therefore relies on an evidence base of specific physiological properties being associated with individual substances. Although a range of physiological parameters have been investigated, it is not always clear to what extent these translate into actual health benefits. What is apparent is the diversity of the substances and their efficacies with respect to physiological outcomes varies widely. For example, the varied impact of different resistant carbohydrates on stool weight and prebiotic effects are reviewed in the physiology paper (Elia and Cummings, 2007).

The common attribute of the substances included in the indigestibility approach is that they provide potential substrates for colonic fermentation, stimulating bacterial growth and the production of SCFA, which have a range of physiological effects (Macfarlane *et al.*, 2006; Wong *et al.*, 2006). Different amounts and types of substrate vary in the rate, site and extent of fermentation and the profile of SCFA produced. Although butyrate has been proposed as protective against colon cancer, the effects it has are complex and somewhat contradictory (Sengupta *et al.*, 2006). Some studies have found butyrate providing substrates have had adverse effects (Burn *et al.*, 1996; Wacker *et al.*, 2002), and it seems that the amount, site, whether promixal or distal, and underlying conditions all influence the effect of butyrate. The desirability of providing large amounts of easily fermented resistant carbohydrate substrates has been questioned (Wasan and Goodlad, 1996; Goodlad, 2007), with concern about the impact of a feast or famine scenario on gut health. The epidemiological evidence base for a protective effect of resistant carbohydrates against colon cancer has been inconclusive (Bingham *et al.*, 2003; Park *et al.*, 2005), and so far, intervention studies have tended to show either no effect or a worsening in outcomes (Alberts *et al.*, 2000; Bonithon-Kopp *et al.*, 2000). On reviewing the evidence Food Drug Administration concluded that dietary fibre did not protect against colon cancer (FDA, 2000).

It should be noted that the exclusion of DP <3 by the indigestibility linked definition demonstrates a lack of consistency, as resistant sugars such as lactulose and some polyols share similar physiological attributes to those suggested as the basis for characterizing carbohydrates with DP >3 as dietary fibre, for instance prebiotic effects (Gostner *et al.*, 2006), and promoting calcium absorption (van den Heuvel *et al.*, 1999). In addition, by this approach it is not clear how to handle carbohydrate ingredients that are indigestible, but for which no beneficial physiological attributes have been demonstrated.

While the addition of resistant carbohydrates to products has no direct impact on the food-based guidelines to consume dietary fibre as fruits, vegetables and whole grains, there is a greater potential for confusion with respect to the population reference intake values. For dietary fibre, these have predominantly been derived from epidemiological evidence linking plant-rich diets with reduced disease incidence. However, unless supplemented foods are clearly identified as such, then a potential conflict arises if the consumer perceives that such preparations are directly equivalent to the dietary fibre present in unsupplemented foods. This could result in a situation where the consumer selects supplemented products on the basis that they will contribute towards the reference intake value, although in reality this would not be a true reflection of the intention of the dietary guidelines.

This argument does not preclude that some resistant carbohydrate preparations cannot have a position within diets, but it should be emphasized that such formulations are researched and if shown beneficial promoted on the basis of the usually very specific functional properties that they may have. If resistant carbohydrate preparations were included as dietary fibre, the population reference intake values established by the epidemiology evidence base would become redundant, as there would be no clear link between the food label and the guideline.

It should also be considered that the bulking effect that acts to self-limit the intake of foods with a naturally high dietary fibre content, represents much less of a constraint for some resistant starches and resistant oligosaccharides that can be formulated within products in relatively high amounts, and could make a considerable contribution to some diets. This highlights the need to consider potential detrimental effects and whether safe upper intake limits for resistant carbohydrates are required.

Determination of substances included within the indigestibility approach. The principle of determining carbohydrates grouped on the basis of the physiological attribute of indigestibility in the small intestine has already been addressed within the earlier section on the measurement of resistant carbohydrates. Although NSP constitutes the major resistant carbohydrate fraction in most foods, this is typically not determined specifically by this approach, but instead forms part of an enzymatic–gravimetric measurement that includes other materials in unknown amounts. The values obtained by these enzymatic–gravimetric techniques have previously been presented as 'total dietary fibre' measures, but a number of supplementary methods have since been proposed to determine other substances included within the indigestibility approach. The principal enzymatic–gravimetric techniques and the other complementing methods are discussed in turn.

Enzymatic–gravimetric procedures. The stated aim of these methods is to measure the sum of indigestible polysaccharides and lignin as the weight of a residue, corrected for ash and crude protein content, which remains after treatment with protease and amylolytic enzymes and washing with 80% ethanol. These methods therefore do not focus on plant cell-wall material, but seek to include RS, which may be present in large amounts as the result of food processing or the addition of RS preparations. Non-carbohydrate materials are also recovered, with the given justification of measuring lignin and other non-carbohydrate cell-wall components, although in practice it also recovers food processing artefacts such as Maillard reaction products, which may have adverse physiological effects (Tuohy *et al.*, 2006).

This approach has evolved from the early methods used for measurement of 'crude fibre'. The different versions of the enzymatic–gravimetric technique can be considered as modifications of the method proposed by Prosky and coworkers (AOAC method 985.29; AOAC, 2005). The most common variation is AOAC method 991.43, which uses a different buffer system. Briefly, they utilize 400 ml beakers as

reaction vessels, with successive treatments with amylase, protease and amyloglucosidase, each step requiring individual pH adjustments. Four volumes of ethanol are added and the precipitated material is transferred to a filtration crucible where it is dried and weighed. For each sample separate residues are collected for determination of ash and protein.

Contrary to normal conventions of nutrient definition, the material recovered by these methods has been presented by some as a '*de facto*' definition of dietary fibre (AACC, 2001). Of the various materials that may be determined, only the intrinsic cell-wall polysaccharides are a consistent feature of natural unrefined plant foods, and for many plant-based products this will be the main component of the gravimetric fibre value. However, as this methodological approach can include some RS and other substances formed as the result of food processing or during sample preparation for analysis, it is not possible to identify how much, if any, of the 'fibre' value is plant cell wall material (Ranhotra *et al.*, 1991; Theander and Westerlund, 1993; Rabe, 1999). A detailed assessment of the influence of food processing on materials recovered by enzymatic–gravimetric approaches has been provided elsewhere (Englyst *et al.*, 1996).

This method cannot be considered as 'fit for purpose' in meeting the requirement to consistently reflect natural unrefined plant foods. Neither does it meet its own stated aim of measuring indigestible polysaccharides, as the RS recovered in the residue may have little bearing on what is present in the food. Lignin should be excluded from further consideration as part of a dietary fibre measure on the grounds that (1) it is not a carbohydrate, (2) it is not present in the human diet in significant amounts, (3) there is no specific routine method for is analysis, (4) its inclusion has often inappropriately been used to justify the presence of unidentified material in the gravimetric fibre residue.

In terms of practicality for the analytical chemist, the enzymatic–gravimetric approach is excessively cumbersome. It requires considerable time and reagent resources and is not well suited to large batch sizes, increasing the cost of this analysis.

Complementary procedures for the indigestibility approach. The intended purpose of the other stated methods associated with this approach are to give additional information about individual resistant carbohydrate fractions and provide determinations of those substances that are incompletely recovered by precipitation in 78% ethanol with the enzymatic–gravimetric techniques. Several of these methods determine carbohydrates as their constituent sugar components released by hydrolysis, along the principles described in the carbohydrate determination section.

Similar to the NSP procedure outlined earlier, the AOAC 994.13. method is primarily based on the determination of sugars released by the acid hydrolysis of polysaccharides isolated by precipitation in ethanol. It differs in that by this technique starch is only partially dispersed and hydrolysed, so unlike the NSP method it does not describe a chemically distinct grouping of carbohydrates. It also includes a determination of Klason lignin as the material recovered in an acid hydrolysis resistant residue. The reality is that Klason lignin can include a considerable amount of artifact material including Maillard reaction products formed during food processing.

AOAC 2002.02 is a resistant starch method based on treatment with amylolytic enzymes and precipitation of unhydrolysed starch in 80% ethanol, which is then chemically dispersed and determined as glucose released by hydrolysis. As discussed in the resistant carbohydrate determination section, the degree of starch hydrolysis is influenced by the analytical conditions, and for this method these have principally been designed only to determine retrograded starch (RS3) and some RS2 in starch granules. Therefore, this method does not consistently provide a total RS determination, and neither does it measure the same starch fraction recovered by the enzymatic–gravimetric methods, making it difficult to integrate the values obtained by these methods. Furthermore, small starch degradation products resulting from the enzyme hydrolysis could potentially be lost in the 80% ethanolic supernatant, and therefore would not be included as RS.

The AOAC 995.16 method determines β-glucans and is an example of the measurement of an individual NSP species by selective enzymatic hydrolysis. The AOAC 999.03 and 997.08 methods determine fructans (inulin and fructo-oligosaccharides) as the fructose (and small amount of glucose) released after fructanase treatment. AOAC 997.08 uses HPLC to measure the increase in released sugars on top of the sugars already released by hydrolysis of sucrose and starch, leading to a high uncertainty when dealing with small quantities of fructans. Although AOAC 999.03 attempts to address this issue by the removal of sugars by chemical reduction prior to the fructan hydrolysis, this approach results in an incomplete recovery of lower DP fructooligosaccharides as their reducing end groups are also affected by the reduction step. Along similar principles, the AOAC 2001.02 method determines *trans*-galactooligosaccharides in an aqueous extract as the galactose released after treatment with β-galactosidase (EC 3.2.1.23), with a separate measurement and correction for galactose from lactose, which is also hydrolysed by this enzyme.

The stated methods for the determination of polydextrose (AOAC 2000.11) and resistant maltodextrin (2001.03) do not measure their component glucose parts, but instead rely on quantification of intact oligosaccahrides by chromatography. AOAC 2000.11 is based on an aqueous extraction treated to hydrolyze those α 1–4 bonds of polydextrose that are accessible to an amylolytic enzyme, as well as removing any available starch and maltodextrins present. A fructanase treatment is also included to prevent fructans from co-eluting with polydextrose when it is separated by high performance anion-exchange chromatography. The AOAC 2001.03 method is actually an extension of the soluble/

insoluble version of the enzymatic–gravimetric method AOAC 985.29, and is intended to measure the resistant maltodextrins that remain in the solvent filtrate from the precipitation and washing of the water-soluble residue. This solvent filtrate, which can be up to 500 ml, is evaporated and then ion exchange resins are used to remove salts and proteins from the redisolved residue, which is then dried again and filtered before quantification by HPLC as units with DP>3. There will be crossover in the materials measured by the AOAC 2001.03 and 2000.11 methods, and although fructans can be removed when present, the reality is that any resistant oligosaccharides and possibly other substances, may co-elute and therefore inflate the values obtained.

Taken as a whole, the enzymatic–gravimetric analysis and the complementing AOAC methods form a disjointed approach to the determination of resistant carbohydrates. There is specific concern about the double counting of the same substances by more than one of these procedures, which severely limits the integration of values. It has been suggested by some that the combined enzymatic-gravimetric and resistant maltodextrin method could provide an integrated approach to dietary fibre determination for the indigestibility approach. This must be viewed with some skepticism, as both these determinations would be prone to interference due to their empirical nature, and furthermore, there would be no primary standards available to reflect the diversity of materials recovered by the HPLC measurement. The other disadvantage of applying empirical methods recovering unidentified material is that it is not possible to indicate what material is present, or how much, if any, of it conforms to the qualifying criteria of exhibiting beneficial physiological properties.

Public health application of carbohydrate measurements

The nutritional characterization of dietary carbohydrates should acknowledge the heterogeneity in the functional properties of carbohydrate containing foods. This ranges from a consideration of the metabolizable energy provided, to their varied physico-chemical characteristics in the gastrointestinal tract and subsequent effects on physiology and metabolism, and to the more holistic consideration of the overall nutrient profile of the foods.

Describing these varied attributes in a consistent and nutritionally relevant manner has proved challenging, as chemical composition does not always adequately reflect functionality, especially in the context of the food matrix. The implication is that it has been difficult to assign population reference intake values and nutrition claims based on the commonly applied chemical divisions such as starch and sugars. There is therefore a requirement to incorporate additional nutritional descriptions into classification, measurement and public health messages relating to carbohydrate containing foods. Some of the challenges and potential solutions are commented on here.

Sugars

The nutritional considerations relating to sugar containing foods can be evaluated by their impact on dental caries, excess energy intake, nutrient:energy ratios, and physiology due to metabolic differences between sugars. The complexity in the nutritional description of sugars relates to the food groups from which they are consumed. This need to distinguish between sugar sources was recognized by the development of the term intrinsic sugars for those retained in intact cellular structures (Department of Health, 1991) and the terms free sugars, added sugars and non-milk extrinsic sugars which are essentially synonymous with each other. US Department of Agriculture have prepared a large database with values for added and total sugars for a wide range of products (Pehrsson *et al*., 2006).

There is no available evidence to suggest that there are any adverse effects on health outcomes in humans from the sugars consumed in the form of fruit, vegetables or milk. In any case, the bulky nature of fruits and vegetables tends to limit the absolute consumption of sugars from these sources. In contrast, free (or added) sugars have the potential to be consumed in large quantities and have a more direct impact on these related health issues (van Dam and Seidell, 2007). For this reason, guidelines have limited free sugar intake to <10% of energy (Department of Health, 1991; WHO, 2003). As there is no justification to have a specific limit for the consumption of intrinsic sugars from fruit, vegetables and milk, these are instead considered within the overall guidance to consume 45–60% of energy from carbohydrates.

However, apart from when dealing with primary food groups such as fruits, vegetables and milk, it can be difficult to identify the source of sugars in foods, particularly in products composed of multiple ingredients. Furthermore nutrition labels only state values for total sugars, and it is perceived as overly complex to include a division between intrinsic and free sugars. This poses a problem for the nutrient signposting and claims relating to sugar content. A practical solution has been proposed that establishes a high criteria for claim purposes based on the guidelines on free (or added) sugars (50 g for a 2000 kcal diet), but incorporating a small allowance (that is, 10 g) for the average consumption of intrinsic and milk sugars consumed from manufactured foods (FSA, 2006). This would allow the food labelling for total sugars to be used in the nutritional signposting of manufactured foods. This is a pragmatic approach that is consistent with the dietary guidelines for a selective restriction of free sugars without the reliance on an analytical distinction between intrinsic and free (or added) sugars, which has proved difficult for routine labelling purposes.

Starch and whole grains

Starch has been presented as a preferable source of carbohydrate to sugars. In reality this is an oversimplification, and

similar to the situation with sugars, the food source of starch needs to be considered when evaluating nutritional properties. A considerable amount of starch is consumed as refined cereal products where the germ and bran fractions have been lost along with the majority of the associated micronutrients and phytochemicals. This results in a lower nutrient/energy ratio for many refined cereal products when compared with their whole grain counterparts. Furthermore, the physicochemical characteristics of starch are very dependent on the biological origin and degree of processing, affecting both the rate and extent of digestion in the small intestine.

The consequence is that in isolation, a value for the total starch content in foods or diets is not necessarily very informative about the functional attributes of a carbohydrate food. Information on dietary fibre defined as 'intrinsic plant cell-wall polysaccharides' will help identify whether the starch is associated with refined or whole grain material, and the detailed profile of the carbohydrate-release characteristics will indicate the likely gastrointestinal and metabolic fate of the carbohydrate food.

Glycaemic index

The GI concept has provided insight into the physiological properties of carbohydrate containing foods, which are not apparent from chemical composition alone. This physiological ranking is often presented as a description of carbohydrate quality, and it is therefore appropriate to consider how the GI integrates with the overall strategy of characterizing the functionality of dietary carbohydrates.

In addition to the rate of carbohydrate digestion, other food-mediated effects on both gastrointestinal events and postabsorptive metabolism can influence the GI. Therefore, GI values do not represent a direct measure of carbohydrate absorption from the small intestine, but rather reflect the combined effect of all the properties of a food or meal that influence the rate of influx and removal of glucose from the circulation. However, the different mechanisms responsible for changes in the glycaemic response to a food or meal cannot be considered equally beneficial to health. For instance, it would be inappropriate to promote a food as low GI if the underlying mechanisms responsible were either high contents of fat or fructose. In such cases, any potentially detrimental nutritional attributes should take precedence over the physiological GI characteristic of the food or meal. Therefore, the GI measure should be applied only to foods with a high carbohydrate content, and there should be an overall consideration of the food and meal characteristics. The *in vitro* carbohydrate-release profiles, such as the measures of rapidly and slowly available glucose, can specifically identify the low GI products that are rich in slow-release carbohydrates, which have demonstrated health benefits.

As the GI measurement relates to the available carbohydrate component of foods, it is appropriate to calculate the portion sizes of test meals based on direct determinations of available carbohydrate, thereby reducing this aspect of GI measurement uncertainty and ensuring that resistant carbohydrates are not mistakenly included. The accompanying paper on GI addresses further issues relating to the determination and application of this physiological measure (Venn and Green, 2007).

Dietary fibre and other resistant carbohydrates

There has been considerable interest in the development and marketing of a number of resistant carbohydrates including polysaccharides (for example, retrograded and modified resistant starches, modified celluloses), oligosaccharides (for example, fructooligosaccharides, polydextrose and resistant maltodextrins) and sugars (for example, polyols and lactulose). As discussed earlier there has been debate about whether some or any of these materials should be encompassed within a dietary fibre term (that is, indigestibility approach), or whether they should be considered separately (that is, plant-rich diet approach). This debate is of considerable importance, as it impacts directly on the rationale for dietary fibre and on how different resistant carbohydrates can be managed from a public health perspective.

By the plant-rich diet approach, the definition very effectively supports the existing dietary guidelines to consume fruits, vegetables and whole grains. Likewise, this approach is consistent with the evidence base on which population reference intake values have been established. This need to differentiate between dietary fibre intrinsic to plant foods, and added preparations was also recognized by US academy of sciences (IOM, 2001).

By the indigestibility approach, the inclusion of materials other than 'intrinsic plant cell wall polysaccharides' as dietary fibre could adversely impact on the guidelines to consume fibre from plant foods, and it could be wrongly interpreted as inferring that the evidence for the benefit of the plant-rich diet can be extrapolated to these other substances. This issue is currently very pertinent, as although dietary guidelines are to consume at least five portions of fruit and vegetable and three or more portions of whole grains daily, average intakes are far lower in some populations. As a consequence, the average dietary fibre intakes are also less than the population reference values, and this has been represented by some as a 'fibre gap' that could be met through supplementation with other substances. However, this would be a fundamental misinterpretation of the evidence base for a fibre rich diet being beneficial to health and would potentially mislead the public in their selection of this diet.

As only intrinsic plant cell wall polysaccharides are included within the plant-rich diet definition, other sources of resistant carbohydrates would need to be described separately for labelling and nutrition claims purposes. Of course, it would only be necessary and appropriate to include additional categories such as resistant oligosaccharides or

resistant starch on nutrition labels if they were shown to be of sufficient relevance to public health.

The nutrition labelling of separate categories of resistant carbohydrates represents an opportunity for industry to stimulate product innovation through the development and promotion of functional ingredients based on their specific physiological properties. This would most appropriately take the form of health claims, for which there is established legislation to ensure that these substances are suitably evaluated and controlled (for example, European Commission Regulation EC No 1924/2006). The diversity in physiological attributes between different functional ingredients, and the variation in the quantities required to produce the nutritional or physiological effects, makes it unfeasible to group all these substances under a single set of conditions for nutrition or health claims.

Dietary recommendations and labelling

Food-based dietary guidelines have traditionally been the most effective approach to the public communication of nutrition. For carbohydrates, this is simply conveyed by the promotion of fruit, vegetable and whole grain consumption. However, the increasing predominance of manufactured products necessitates additional strategies that address the varied functional properties of these processed foods, thereby allowing either beneficial or potentially adverse attributes to be distinguished. Such functional parameters should reflect nutritional aspects of different types of ingredients and specific carbohydrate components, as well as any effects of processing on the overall food characteristics. These principles form the basis of the classification and measurement scheme for dietary carbohydrates presented in Table 1, which can be applied as a tool for further research into the link between dietary carbohydrates and health.

Recommendations

1. Food-based guidelines promoting the consumption of fruits, vegetables and whole grains are some of the most effective public health messages. Carbohydrate classification and measurements should support these dietary guidelines and provide the means with which to describe the component ingredients and functional properties of foods, including those attributed to the food matrix.
2. For energy calculation purposes, there is a need to describe carbohydrates in terms of their gastrointestinal and metabolic fate, as 'available', which provide carbohydrate for metabolism, and as 'resistant', which resist digestion in the small intestine or are poorly absorbed/metabolized. These categories should be further subdivided into types describing attributes of specific nutritional and functional relevance.
3. There should be a commitment to move away from empirical based methods that measure unspecified materials, towards rational methods providing direct and specific measurement of different types and categories of carbohydrates as their chemically identified components.
4. The selective restriction of free or added sugars is a useful food-based guideline. To support this, nutrition claims relating to the content of sugars in manufactured products should be provided in the context of the established maximum intake limit of free sugars (10% of energy), but with an additional small allowance for the intrinsic sugars provided by manufactured food groups. This allows the content of total sugars to be used for claims purposes and overcomes the need to distinguish between intrinsic and free sugars on nutrition labels. This effectively targets the restriction of free or added sugars, without adversely influencing intakes of fruits, vegetables or milk.
5. The GI and glycaemic load are useful nutritional terms providing insight into a physiological parameter that is not always apparent from chemical composition alone. Its application should be limited to foods with high carbohydrate contents and is most effectively interpreted in conjunction with information on other food/meal characteristics including detailed sugar composition and starch digestibility profiles.
6. Dietary fibre should be considered as a public health term supporting dietary guidelines to consume a plant-rich diet. The definition 'intrinsic plant cell wall polysaccharides' provides the only consistent link with the scientific evidence on which these guidelines are based. The physiological characteristic of indigestibility is therefore not considered to be an adequate basis for the definition of dietary fibre.
7. Resistant carbohydrates other than dietary fibre should be considered separately and by their own merits. Distinct categories are essential for functional ingredients to be managed effectively from a public health perspective. As there is the potential for large amounts of added resistant carbohydrates to be consumed, safe upper intake limits may need to be established.

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During the preparation and peer-review of this paper in 2006, the authors and peer-reviewers declared the following interests.

Authors

Dr Klaus Englyst: Director and share-holder in Englyst Carbohydrates Ltd which is a small research-oriented

company working on dietary carbohydrates and health. The UK Food Standards Agency is the main research partner and sponsor. In addition, Englyst Carbohydrates provide analytical assistance to universities and food industry worldwide, albeit on a small scale. The complete independence of Englyst Carbohydrates is maintained by not entering into any consultancy agreement.

Professor Simin Liu: Member of the Scientific advisory board for the EU Health Grain Project.

Dr Hans Englyst: Director and share-holder in Englyst Carbohydrates Ltd which is a small research-oriented company working on dietary carbohydrates and health. The UK Food Standards Agency is the main research partner and sponsor. In addition, Englyst Carbohydrates provide analytical assistance to universities and food industry worldwide, albeit on a small scale. The complete independence of Englyst Carbohydrates is maintained by not entering into any consultancy agreement.

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