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Carbohydrate fractions of legumes: uses in human nutrition and potential for health

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Starch and fibre can be extracted, using wet or dry processes, from a variety of grain legumes and used as ingredients for food. α-Galactosides can be isolated during wet processes from the soluble extract. Starch isolates or concentrates are mostly produced from peas, whereas dietary fibre fractions from peas and soyabean are commercially available. The physico-chemical characteristics of fibre fractions very much depend on their origin, outer fibres being very cellulosic whereas inner fibres contain a majority of pectic substances. Inner fibres are often used as texturing agents whereas outer fibres find their main uses in bakery and extruded products, where they can be introduced to increase the fibre content of the food. Most investigations on impacts on health have been performed on soyabean fibres. When positive observations were made on lipaemia, glucose tolerance or faecal excretion, they were unfortunately often obtained after non-realistic daily doses of fibres. Legume starches contain a higher amount of amylose than most cereal or tuber starches. This confers these starches a lower bioavailability than that of most starches, when raw or retrograded. Their low glycaemic index can be considered as beneficial for health and especially for the prevention of diseases related to insulin resistance. When partly retrograded, these starches can provide significant amount of butyrate to the colonic epithelium and may help in colon cancer prevention. α-Galactosides are usually considered as responsible for flatus but their apparent prebiotic effects may be an opportunity to valorize these oligosaccharides.

Legumes: Dietary fibre: Starch: α-Galactosides: Human nutrition: Fractionation

Introduction

Grain legumes are considered to be good for health due to their mutual compatibility with cereals and for their properties in disease prevention, including cardiovascular diseases, type 2 diabetes, obesity and, possibly, colon cancer. The nutritional potential of the seeds from this group of plants is based on their high level of protein and, depending on species, a high proportion of either starch or oil (Table 1). Along with macronutrients, leguminous seeds contain appreciable amounts of some vitamins and minerals as well as dietary fibre. The most common legumes for human consumption are bean, lentil, pea, chickpea and faba bean. Most grain legumes are consumed after simple processing, as vegetables, salads, soups, mashed and cooked seeds.

However, grain legume seeds can be fractionated to obtain protein and starch concentrates and isolates, and as a by-product of the process, dietary fibre. Starch, protein and dietary fibre are indeed the main fractions of most European grain legumes, the main exceptions being lupin and soya, which are both rich in fat (Table 1).

Fractions isolated from grain legumes can be used in the food-processing industry as simple ingredients, techno-functional ingredients or additives. Apart from these technological interests, we wonder whether such products could have real nutritional properties for humans.

Dietary fibre content varies according to the species, the variety and processing of legume seeds. In most grain legumes consumed as pulses by humans, the content ranges from 8 to 27·5 %, with soluble fibre in the range 3·3–13·8 %. Dietary fibre, or cell wall material, content in the cotyledon of legume seed is generally low compared to that of the testa. Indeed, the cell walls accounted for about 90 % of the testa dry weight.

Brillouet & Carré (1983) reported values of dietary fibre contents for pea, broad pea and soyabean cotyledons in the range 6·9–9·3 % (on a dry weight basis). Lupin species

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Abbreviations: ACF, aberrant crypt foci; GI, glycaemic index; RS, resistant starch; SCFA, short-chain fatty acid.

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Table 1. Chemical composition (g/100 g dry matter) of some legume seeds (from Bagger *et al.* 1998)

Legume seed	Protein ($N \times 6{\cdot}25$)	Crude fat	Dietary fibre	Starch	Sucrose
Vicia faba	26–34	2–4	15–24	40–50	2·1–2·3
Pisum sativum	23–31	2–3	15–21	20–50	0·7–5·7
Lupinus luteus, *L. angustifolius*, *L. albus*	33–42	4–12	25–40	1–2	1·5–3·5
Glycine max	38–42	18–22	7–15	1–2	4·7–7·6

have a special position within the *Leguminosae* family by containing a high amount of cell wall material in the cotyledons (in the range 7·5–32·1 %) in the form of rather thick cell walls (Brillouet & Riochet, 1983). This could ascribed to the high amount of galactans stored in the cell walls.

Starch content varies between genera, from negligible amounts in *Glycine max* to half the dry seed weight in a wild-type, round-seeded, pea (*Pisum sativum*) (Table 1). Mutations that affect the activities of enzymes of the starch biosynthetic pathway can profoundly affect not only starch content but also its composition. For instance, in the pea, which is one of the species that has been extensively genetically manipulated, mutations at the *r* locus, which encodes starch-branching enzyme I, reduce starch content to 30 % of dry weight and reduce the amylopectin content, whereas those at *rug3*, which encodes plastidial phosphoglucomutase, can completely eliminate starch (Casey, 1998).

α-Galactosides are oligosaccharides which are not digested in the upper part of the gastrointestinal tract, due to the absence of α-galactosidase among human endogenous enzymes, and are therefore available for bacterial fermentation in the colon. Overall α-galactoside content is within the range of 2–10 g/100 g dry matter, and stachyose is the prevalent oligosaccharide in most pulses (*Phaseolus vulgaris*, *Pisum sativum*, *Lens esculenta*, etc.) (Table 2). Lupin seeds seem to contain the highest concentration of α-galactosides among grain legume species. In faba beans, mung beans, pigeon pea, and some varieties of chickpeas, verbascose is the main oligosaccharide in the seed. Raffinose content is lower than 1·5 g/100 g in all pulses (Rackis, 1975; Fleming, 1981; Oboh *et al.* 1998). Ajugose is present in significant amounts in lupin seeds (0·3–2·0 g/100 g) (Reddy *et al.* 1984). The α-galactoside content of soyabean is quite similar to that of most pulses; for example, raffinose, stachyose and verbascose comprise 0·7–1·3, 2·2–5·4 and 0·0–0·3 %, respectively, of seed dry matter (Reddy *et al.* 1984).

Fractionation of grain legumes

Dry and wet separation processes have been used to fractionate grain legumes for experimental purposes but also for industrial applications (Kozlowska *et al.* 1998; Czukor *et al.* 2001). Wet separation processes are used to produce high-purity protein isolates while dry separation results in enriched fractions.

In conventional wet process, for food applications, the hulls are removed because they can contain antinutritional compounds that can be released during the extraction process. The dehulled seeds are pen milled and the legume flour is pulped with an aqueous decomposing agent (generally alkaline solution) for extracting protein (Colonna *et al.* 1981; Gueguen, 1983; Sosulski & McCurdy, 1987). The proteins are isolated from this extract by acidic precipitation or by ultrafiltration. The wet protein isolates are then dried. The liquid phase contains the α-galactosides as well as many other soluble contaminants. These oligosaccharides can be further isolated in 80 % ethanol. The solid phase left after protein separation is suspended in water and is screened through a series of sieves (Schoch & Maywald, 1968; Colonna *et al.* 1981). The starch is recovered from the under flow fraction, with the fraction rich in cell wall material remaining on the screens. The starch isolate contains 0·04–0·40 % protein, less than 4 % of cell wall material and about 0·1–1·0 % of lipid as impurities. The fibre fraction contains small amounts of proteins (4–8 %) and lipids (0·5–1·5 %).

The ‘dry’ process consists in disintegration of the dehulled seeds on pill mill and air classification into starch and protein fractions (Sosulski, 1979; Colonna *et al.* 1980; Tayler *et al.* 1981; Sosulski *et al.* 1985; Sosulski & McCurdy, 1987). The starches and dietary fibre are concentrated mostly in the light, fine fraction, and the proteins and lipids, in the heavy, coarse one. Dry processes have been carried out more successfully with grain legumes, where starch is the main storage compound rather than oil. The main advantages of air classification

Table 2. Amount of α-galactosides in the main pulse species compared to a single main source (%, dry matter)*

	Phaseolus vulgaris	*Lens esculenta*	*Cicer arietinum*	*Pisum sativam*	*Vicia faba*	*Lupinus albus*	*Glycine max*
Raffinose	<0·05–0·93	0·3–1·0	0·4–1·2	0·3–1·6	0·1–0·3	0·5–1·1	0·5–1·3
Stachyose	0·5–4·1	1·7–3·1	2·0–3·6	1·3–5·5	0·7–1·5	0·9–7·4	2·2–4·3
Verbascose	0·06–4·0	0·6–3·1	0·6–4·2	1·6–4·2	1·7–3·1	0·6–3·4	0·0–0·3
Total	2·6–6·6	3·0–7·1	7·4–7·5	5·1–8·7	3·1–4·2	7·4–9·5	2–6

* Source: Harding *et al.* (1965), Cristofaro *et al.* (1974), Rackis (1975), Matheson & Saini (1977), Eskin *et al.* (1980), Gueguen *et al.* (1980), Iyer *et al.* (1980), Olson *et al.* (1982), Quemener & Brillouet (1983), Reddy *et al.* (1984), Sathe *et al.* (1984), Savage & Deo (1989), Leakey *et al.* (1998), Frias *et al.* (2001), Vidal-Valverde *et al.* (2001).

are reduced energy and water consumption. However, all fractions, and especially protein fractions, are contaminated by α-galactosides (Sahasrabudhe *et al.* 1981).

Because legumes have a variety of characteristics and differ, for example, in oil content, some modifications of the standard procedures have been adopted for optimizing the process for a particular legume (Nickel, 1988; Czuchajowska & Pomeranz, 1994; Kovacs, 1996; Krikken, 1999; Dijkink & Langelaan, 2001).

Physical and chemical characteristics of fibre and starch fractions and derivatives

The chemical structure and physico-chemical properties of starch and fibre are important for functional behaviours in food use and for diet-related health effects. Depending on the application, the use of enriched fractions may be a good alternative to high-purity isolates.

Fibre fractions

The properties of dietary fibre that influence their techno-functionality are the fibre dimensions, porosity, hydration, rheological and fat-binding properties. The colours and flavour are also of importance.

Dietary fibre preparations commercially available arise mainly from pea and soya from either the cotyledons (inner) or from the hulls (outer) (Table 3). Preparations are generally richer in dietary fibre when obtained from hulls. Indeed, preparations from cotyledons contain variable amounts of starch and protein. Inner-fibre fractions exhibit higher water retention capacity than outer fibres (Table 4). Oil-binding capacity is in the same range for both inner and outer fibres (Table 4). Inner fibres are smooth, while outer fibres have a sandy character. The inner fibre products are, in general, in the form of powder low in odour and flavour (Table 4). The outer fibres are generally available at different particle sizes. They are generally light in colour and flavour (Table 4).

Processing can be applied to improve the functional characteristics of fibre. For example, a mixture of cellulase and carbohydrase has been applied to improve the sensory properties, including the mouth-feel characteristics and smoothness of the soya fibre material (Lin Santa *et al.* 1996).

The composition of the dietary fibre fraction depends very much on its localization in the seed coat (outer fibre) or the cotyledons (inner fibre) (Table 5). A major difference between the inner and outer dietary fibre is the relative content of cellulosic and non-cellulosic polysaccharides. The cell walls of the cotyledons contain a range of polysaccharides, including pectic substances (about 55 %), cellulose (about 9 %) and non-starchy non-cellulosic glucans (in the range 6–12 %; Brillouet & Carré, 1983; Brillouet & Riochet, 1983; Al-Kaisey & Wilkie, 1992; Petterson, 1998; van Laar *et al.* 1999, 2000), while the seed coat contains large quantities of cellulose (ranging from 35 to 57 %) and lower amounts of hemicelluloses and pectins (Brillouet & Riochet, 1983; Weightman *et al.* 1994; van Laar *et al.* 1999). The cell walls from the cotyledons are non-lignified.

Testa from pea and lupin species have been shown to contain a low amount of lignin. The concentrations reported were 6·6 mg/g for alcohol-insoluble residues from pea hull, and between 0·4 and 1·7 % for hulls from various lupin species (Brillouet & Riochet, 1983; Weightman *et al.* 1994). The lignin values were unrelated to hull colour.

Starch fractions

Most starches from grain legumes have a relatively high amylose content compared to most starches (Table 6). As a consequence their X-ray diffraction pattern is type C, which is considered to be intermediate between types A and B (Gallant *et al.* 1992) (Table 6). However, wrinkled peas are known to exhibit a B-type X-ray diffraction pattern which is due to its high level of amylose (Table 6).

Legume starches are kidney-like or ovoid with well-defined shells centred along an elongated hilum. Some exceptions are also known, such as the compound starch granules of the wrinkled pea, in which sphero-pyramidal units are associated (Gallant *et al.* 1992). Enzymatically treated pea starches (C-type starch) exhibit a characteristic similar to B-type starches, which is the presence of highly resistant and large blocklets (4–500 nm diameter at the peripheral level of the granules). These blocklets would explain the resistance of B and C starch granules to hydrolysis (Gallant *et al.* 1992).

There is a much variability in amylose content among

Table 3. Chemical composition of some commercial dietary fibres (dry basis, %)

	Pea		Soyabean		Lupin	
	Cotyledon*	Hull*	Cotyledon†	Hull*	Cotyledon*	Hull*
TDF	55	89	80	75	80	80
SDF	14	7	21	10	8	8
Protein	17	5	13	8	15	14
Lipid	0·5	1·5	0·2	2	0·5	2
Available C	23	2	nd	6	3	1
Mineral	4	4	5	5	1	4

Available C, available carbohydrates; nd, not determined; TDF, total dietary fibre; SDF, soluble dietary fibre.

* Pfoertner & Fischer (2001).

† Dubois *et al.* (1993).

Table 4. Properties of some commercial dietary fibres (from Pfoertner & Fisher, 2001)

Dietary fibre	Colour	Flavour	WRC	Oil retention
Cotyledon				
Pea	White	Neutral beany	9–11	1·5–2·0
Soya	Light cream	Bland	7–8	4·0
Lupin	White	Nearly neutral	8–11	1·5–2·0
Hull				
Pea	Creamy white	Neutral	4–5	1·8–2·0
Soya	Light tan	Neutral nutty	3–5	1·4–1·7
Lupin	Creamy light	Neutral nutty	7–8	1·6–1·7

WRC, water retention capacity.

genotypes of peas (Skrabanja *et al.* 1999). Indeed, the wild type contains 30 % amylose whereas *rb* and *r* mutants, for instance, contain, respectively, 20 and 65 % amylose (Bergthaller *et al.* 2001). According to Bergthaller *et al.* (2001), these same mutants differ widely in starch extractability from cotyledons in a wet milling process but also in the purity of the extracted starch, which varied from 85·3 to 97·5 % (dry matter basis) for *r* and *rug4* mutants. Temperatures of gelatinization do not seem to be significantly different from 'normal' cereal or potato starches, as they range between 55 and 97°C (Table 6).

The procedure of preparation of the starch can modify the characteristics of the initial starch present in the grain. Intense damaging of the starch granules can be observed in some industrial starches (Soral-Śmietana *et al.* 2001*b*). As a consequence, commercial native starches may have different properties. Apparently, Gel-Flow (native pea starch from Parrheim Foods, Canada) crystallinity, estimated by X-ray diffraction, is lower from that of Nastar (native pea starch from Cosucra S.A., Belgium) (Soral-Śmietana *et al.* 2001*a*,*b*). As a consequence, resistant starch (RS) contents of the native pea starches are different: 43 and 19 %, respectively, for Nastar and Gel-Flo. The latter starch exhibited slightly higher water and oil adsorption capacities than the former (Soral-Śmietana *et al.* 2001*a*,*b*).

Starch has also been isolated from chickpeas. One of the starches that has been isolated seems to have similarities with native maize starch, with a relatively high temperature of gelatinization (67°C), whereas another had a low temperature of gelatinization (60°C) (Meares *et al.* 2001).

Ionic (cationic, anionic and amphoteric) pea starch derivatives have been developed to correspond to the demand of industries in various non-food applications. The introduction of ionic substituents into the starch molecules significantly affects their physico-chemical properties, such as gelatinization temperature, swelling characteristics, solubilization and iodine complexation (Lewandowicz *et al.* 2001).

α-Galactosides

α-Galactosides are derived from sucrose and contain 1–3 units of galactose linked by α-1,6 linkages. They are highly soluble in aqueous media and very rapidly fermented by colonic microflora. The role of α-galactosides seems to be multiple, as they are an energy source for the plant and disappear during germination. They are stored by seeds during the final stage of ripening when they dry. They seem to help protect against abiotic stresses such as cold and desiccation (Jones *et al.* 1998). Some agricultural factors seem to affect α-galactoside content in faba bean seeds. For example, the concentration of stachyose and verbascose can decrease when optimal irrigation is applied (Szukala *et al.* 2001).

Soaking, the most common treatment for partial elimination of α-galactosides from grains, becomes more efficient when bicarbonate is added, due to the greater

Table 5. Sugar composition of the cell walls (% of total cell wall sugars)

	Pea		Soyabean		Lupin	
	Cotyledon*	Hull†	Cotyledon*	Hull†	Cotyledon‡	Hull‡
Rha	2·0	1·1	1·8	1·2	1·0	0·5
Fuc	0·9	0·2	3·1	0·5	nd	nd
Ara	42·7	5·2	17·9	7·6	13·3	8·4
Xyl	4·5	10·8	5·1	12·6	5·5	21·6
Man	nd	0·2	1·6	7·2	0	1·3
Gal	5·4	1·5	34·4	3·7	62·4	1·8
Glc	25·9 (57)§	64·0	13·8 (11)	53·5	9·5 (9)	56·5
Uronic acid	18·7	17·1	22·3	13·6	8·2	10·0

Rha, rhamnose; Fuc, fucose; Ara, arabinose; Xyl, xylose; Man, mannose; Gal, galactose; Glc, glucose; nd, not determined.

* Brillouet & Carre (1983).

† Lo (1989).

‡ Brillouet & Riochet (1983).

§ Percentage of glucose released by mild acid hydrolysis corresponding to non-cellulosic glucose.

Table 6. Some characteristics of legume starches compared to other common starches

Species	X-ray diffraction pattern	Amylose content (% of starch)	Temperature of gelatinization (°C)	References
Pisum sativum				
Smooth	C	31–35	55–65	Colonna & Mercier (1985), Colonna & Champ (1990)
Wrinkled	B	66–72	55–125	Colonna & Mercier (1985)
Phaseolus vulgaris	C	33–35		Colonna & Champ (1990)
Lens esculenta	C		95	Colonna & Mercier (1985)
Cicer arietinum	C	23·9–27·8	60·1–67·3	Meares *et al.* (2001), Soral-Śmietana *et al.* (2001*a*)
Vicia faba	C	24	97	Duprat *et al.* (1980), Colonna & Champ (1990)
Wheat	A	22·7	82–94	Colonna & Champ (1990), Soral-Śmietana *et al.* (2001*a*)
Maize (normal)	A	22·1	55–70	Colonna & Mercier (1985), Soral-Śmietana *et al.* (2001*a*)
Potato	B	25·0	66–90	Colonna & Champ (1990), Soral-Śmietana *et al.* (2001*a*)

permeability obtained by partial solubilization of the cell wall (Ibrahim, 2002).

The addition of α-galactosidase (of commercial origin) lowered α-galactoside content in lentil and pea flours (Frias *et al.* 2001). Germination is one of the most efficient biological treatments for removing α-galactosides. After germination for 48 h at 20°C 40–60 % of pea oligosaccharides disappeared (Dostalova *et al.* 2001). The combined effect of germination and microwave treatment and/or conventional drying further decreased the α-galactoside content of germinated peas (Kadlec *et al.* 2001).

De Lumen (1992) has suggested the use of biotechnological and genetic engineering approaches for α-galactoside removal. Price *et al.* (1988) and Leakey (1994) considered that breeding against flatulence could be useful.

Instead of eliminating α-galactosides during food preparation, it is possible to prevent flatus by different means. Hall *et al.* (1981) found that orally administrated activated charcoal was effective in preventing a large increase in the number of flatus events and raised normal breath hydrogen concentrations following a gas-producing meal. However, according to Potter *et al.* (1985), activated charcoal does not seem to influence gas formation after ingestion of a baked bean meal. An oral α-galactosidase solution (Beano) has also been proposed to prevent flatus (Ganiats *et al.* 1994) after consumption of pulses.

Main uses of fibre and starch fractions

Fibre fractions

Inner fibres are generally used as texturing or bulking agents. The high water-binding capacity, fat-binding and texturing effect allow the control of migration in food preparation by providing stability towards industrial manufacturing and storage processes as well as desirable texture. They may, in many cases, replace food additives, offering the benefit of a ‘clean labelling’. The dosage rate is 1–5 % of the final product weight. They are used in bread and baked goods, particularly biscuits. They can also be used to enrich mousses, jellies and drinks to provide tasty desserts. These could be part of the diet of dysphagic groups and other groups who might otherwise have low fibre intake. Outer fibre is used primarily to enrich the fibre content of food without modifying the technical properties of the end products. It finds applications in bakery and extruded products, snacks, cereals or diet specialities.

Starch fractions

Starch from peas is used in deep-frozen dishes, dressings, extruded bakery products, instant soups and puddings. It can also been used for non-food applications (the paper and board industry, detergent manufacture, water-treatment industry, textiles, plastics and pharmaceutical production), as it is the case for maize and potato starches (Kozlowska *et al.* 1998). The production of legume starches is still small compared the overall production of starch, which is over 6 millions tonnes each year (Kozlowska *et al.* 1998). However, the characteristics of legume starches, and particularly their amylose content, offer a large potential for new applications, both in non-food uses and in human nutrition.

Wrinkled-pea starch seems to be favourable for the functional properties of bioplastics (Funke & Lindhauer 1994; Colonna *et al.* 1995). Legume starches also have potential in agrochemical and pharmaceutical industries as an encapsulation agent, binding material to make up tablets, or a disintegrating agent (Kozlowska *et al.* 1998). The nutritional potential of legume starches will be discussed below.

α-Galactosides

Due to their characteristics, it is highly probable that prebiotic properties of α-galactosides will be confirmed in the future, as has been demonstrated for fructooligosaccharides, for instance. These properties will be discussed below.

Is there a potential use of fibre and starch fractions for health?

There is a large literature on the nutritional aspects of grain legumes, including the digestibility of main nutrients (mainly proteins, starch and dietary fibre), colonic fermentation, post-prandial glycaemia and insulinaemia and some data on lipid metabolism. These observations are, for the most part, very positive and would tend to demonstrate that grain legumes should be promoted as part of a healthy diet. We wonder if such beneficial effects could be, at least

Table 7. Physiological effects of fibre preparation obtained from grain legumes

Source of dietary fibre	Patients or animals	Dose	Adaptation period	Results	Remarks	References
Glucose response and tolerance						
Soya cotyledon fibre	Obese NIDDM patients	10 g/d	Post-prandial study	Improved glucose tolerance		Tsai *et al.* (1987)
Pea cotyledon fibre	Healthy subjects	33 g/d	2 weeks	No effect on glycaemic response Lower PP insulin	DF added to a normal diet	Sandström *et al.* (1994)
Soya hulls	Healthy subjects	26 g/d	30 d	Improved glucose tolerance		Munoz *et al.* (1979)
	NIDDM patients	26 g and 52 g/d	4 weeks	Improved glucose tolerance		Mahalko *et al.* (1984)
Soya fibre (?)	NIDDM patients	40 g/d	4 and 12 weeks	Improved glucose tolerance Insulin levels unchanged Decreased fasting glucose (12 weeks) (when 130 mg/100 ml at time 0)	SF in bread more efficient than powder	Madar *et al.* (1988)
	ob/ob obese mice	11–12 % of the diet	90 and 180 d	Decreased fasting glucose Improved glucose tolerance Decreased fasting insulin		Madar *et al.* (1985)
Pea hulls	Healthy subjects	10 g DF	Post-prandial study	No effect on glucose and insulin response	DF added to a complex meal	Dubois *et al.* (1993)
		30 g (15 g DF)	Post-prandial study	Lower glucose and insulin response	DF added to a test meal	Hamberg *et al.* (1989)
Lipaemia						
Soya cotyledon fibre	Hypercholesterolaemic patients	25 g/d	9 weeks	Lowering effect on cholesterol (total, LDL)		Lo *et al.* (1986)
	Obese NIDDM patients	10 g/d	Post-prandial study	Decreased rise of pp plasma TG		Tsai *et al.* (1987)
	Normocholesterolaemic, mildly or moderately hypercholesterolaemic patients			No effect on cholesterolaemia		Tsai *et al.* (1983), Schweizer *et al.* (1983)
				Decreased cholesterolaemia		Shorey *et al.* (1985), Lo & Cole (1990)
Pea cotyledon fibre	Healthy subjects	33 g/d	2 weeks	Decreased fasting and PP plasma TG No effect on fasting cholesterolaemia (total, LDL and HDL)	DF added to a normal diet	Sandström *et al.* (1994)
Soya hulls	Healthy subjects	26 g/d	28–30 d	Decreased cholesterolaemia	Typical American diet	Munoz *et al.* (1979)
	NIDDM patients	52 g/d	4 weeks	No effect on plasma TG and TC Increased HDL–cholesterol		Mahalko *et al.* (1984)
	Rats	5 g/100 g	4 weeks	Lower plasma and tissue TC (LDL and VLDL) and TG No effect on HDL-cholesterol	Hypercholesterolaemic diet	Uberoi *et al.* (1992)
Pea hulls	Normolipaemic subjects	10 g	Post-prandial study	No effect on PP plasma TG Decreased PP cholesteroaemia (mostly esterified cholesterol)	High-fat, high-cholesterol diet	Dubois *et al.* (1993)

Mineral absorption						
Soya fibre	Healthy subjects	25–30 g/d		No effect on mineral absorption and excretion		Fischer *et al.* (1985), Heymsfield *et al.* (1988), Taper *et al.* (1988), Tsai *et al.* (1983)
	Rats	30 % of the diet	3 weeks	Enhanced absorption of Ca and Mg in the large intestine		Levrat *et al.* (1991)
Large intestine physiology						
Soya cotyledons fibre	Healthy subjects	25 g/d	17 d	Increased stool weight No effect on TT, faecal DM and energy	Low DF diet	Tsai *et al.* (1983)
	Healthy subjects	21 g/d	3 weeks	Increased stool weight No effect on TT and stool frequency	Normal diet	Schweizer *et al.* (1983)
	Healthy subjects	30–60 g/d	10 d	Increased stool weight and frequency Decreased TT	Liquid diet	Slavin *et al.* (1985)
	Constipated, tube-fed non-ambulant patients	20–22·3 g/d	2 weeks	Increased stool weight No effect on TT	Enteral preparation	Fischer *et al.* (1985)
	Diarrhoeic, tube-fed non-ambulant patients	21 g/l per d	15 d	No reduction of diarrhoea	Enteral preparation	Dobb & Towler, (1990)
	Infants with acute diarrhoea (>6 months old	0·7 g DF. D per kg infant's weight	10 d	Reduction of the duration of the diarrhoea	Liquid diet	Vanderhoof *et al.* (1997)
Soya hulls	Healthy subjects	26 g/d	28–30 d	Increased number of stools per week Increased stool wet weight		Munoz *et al.* (1979)
Pea hulls	Healthy subjects	30 g/d	15 d	Increased stool weight No effect of fine particles (<100 μm) on TT No effect in subjects with short TT	Normal diet	Cherbut *et al.* (1991)
	Healthy subjects	15 g/d	3 weeks	No effect of fine particles (<100 μm) on colonic motor profile		Guédon *et al.* (1996)
	Rats	5 % of the diet	nm	Increased faecal bulk Coarse particles decrease mean TT		Focant *et al.* (1990)

DF, dietary fibre; DM, dry matter; NIDDM, non-insulin-dependent diabetes mellitus; PP, post-prandial; SF, soluble fibre; TC, total cholesterol; TG, triacylglycerols; TT, transit time; nm, not mentioned in the paper.

partly, attributed to the starch and/or the dietary fibre (total or one specific fraction) and thus if purified or semi-purified starches and fibres could be of interest for health.

Fibre fractions

The literature on isolated dietary fibre from legume seeds is very scarce, and mainly concerns soya and, to a lesser extent, pea fibres. Most of these studies are summarized in Table 7. It is very difficult to draw definitive conclusions from these data as doses of fibre, duration and design of studies are not comparable. However, there are the following tendencies:

1. according to several studies, consumption of fibre from pea or soya would improve glucose tolerance;
2. some of the studies indicated a positive effect of soya and pea fibre on cholesterolaemia and/or postprandial triglyceridaemia but others did not show any effect;
3. most studies, performed with healthy subjects, showed an increased of the stool weight when they ate soya or pea fibres (usually more than 20 g/d).

Part of the physiological effects of these fibres could be explained by their fermentation pattern. The fermentability of fibres has been studied *in vitro* in batch systems (Cherbut *et al.* 1991; Titgemeyer *et al.* 1991; Bourquin *et al.* 1993; Barry *et al.* 1995; Cloutour, 1995; Guillon *et al.* 1995; Casterline *et al.* 1997; Lebet *et al.* 1998; Van Laar *et al.* 2000). The results showed that fibres from cotyledons were highly degraded (percentage of fermentability: 57–91 %) while fibres from hulls were fermented only to a limited extent (percentage of fermentability: 22–41 %). Acetate was always the major short-chain fatty acid (SCFA) of the medium (about 47–89 %), followed by propionic (7–45 %) and then butyric acid (6–22 %), the other acids being present in very low concentration. Compared to other sources of highly fermented dietary fibre such as pectins, sugarbeet or apple fibres, soyabean fibre (inner fibre) fermentation was generally characterized by relatively high proportions of propionic acid and butyric acid (Table 8). *In vivo* in rats, Levrat *et al.* (1991) reported that fermentation of soya fibre yielded a high level of propionate and low level of butyrate in the caecum. *In vivo,* the increase in SCFA in the caecum of rats fed soyabean fibre was accompanied by an increase in SCFA absorption (Levrat *et al.* 1991; Key & Mathers, 1993). The caecum was enlarged and the wall hypertrophied. There was a moderate and transitory induction of enzymes (ornithine decarboxylase and thymidine kinase) involved in proliferative processes of the colonic epithelium (Levrat *et al.* 1991). Nevertheless, the degree of induction of the enzymes was less than that reported for other fermentable substrates (Calvert *et al.* 1989).

Starch fractions

Bioavailability of native and cooked starches from grain legumes is known to be relatively poor compared to most cereal starches (Hildebrandt & Marlett, 1991; Granfeldt *et al.* 1992; Björck *et al.* 1994; Tovar, 1996; Brighenti *et al.* 1998; Seewi *et al.* 1999). When inside an intact grain, starch granules are entrapped inside the cell wall, making them unavailable in the upper part of the digestive tract unless these cell walls are disrupted during the preparation of the food or by chewing. The low bioavailability of these starches is also explained by the starch itself, which is relatively rich in amylose. Indeed, (1) when raw, it is more resistant than most cereal starches due to its higher crystallinity; (2) it needs a higher temperature to be fully gelatinized and has a higher risk of being insufficiently cooked; (3) after appropriate cooking, it has a higher capacity to retrograde than most starches with a lower content in amylose. This retrogradation is a recrystallization of the linear chains of amylose and, later on, of the branched chains of amylopectin. As a consequence, part of these starches can be ‘resistant’ to digestion in the small intestine (Noah *et al.* 1998) and ferment in the colon, producing SCFA and gases (CO_2, H_2 and, in some, CH_4). The fraction that is digested in the small intestine is slowly available and contributes to the low glycaemic index of the wholegrain legumes. The resistant starch fraction seems to be interesting due to the production of a large amount of butyrate (one of the three main short-chain fatty acids) during colonic fermentation. This nutrient is the

Table 8. Short-chain fatty acid production during *in vitro* fermentation of substrates (mmol/g original substrate dry matter)

		Molar ratio			
Substrate	Total SCFA (mmole/g)	Acetate	Propionate	Butyrate	Reference
Better basic 872 oat fibre	0·42	61·9	16·7	21·4	Bourquin *et al.* (1993)
Bleached oat hull	0·37	87·3	11·6	1·1	Titgemeyer *et al.* (1991)
Oat bran	2·66	49·6	36·1	14·3	Casterline *et al.* (1997)
Fibrim® soya fibre	5·75	64·5	19·5	16·0	Bourquin *et al.* (1993)
Fibrim® soya fibre	1·94	71·4	20·6	8·1	Titgemeyer *et al.* (1991)
Fibrim® soya fibre	2·55	46·7	44·7	8·6	Casterline *et al.* (1997)
Duo sugarbeet fibre	1·16	92·6	6·6	0·8	Titgemeyer *et al.* (1991)
Apple fibre	1·56	67·9	26·3	5·8	Casterline *et al.* (1997)
Arabic gum	8·19	65·7	22·8	11·3	Bourquin *et al.* (1993)
Arabic gum	6·94	66·2	25·3	8·4	Casterline *et al.* (1997)
Centara II pea fibre	0·76	74·1	16·7	9·2	Titgemeyer *et al.* (1991)
Centara III pea fibre	1·99	73·4	21·1	5·5	Casterline *et al.* (1997)

SCFA, short-chain fatty acid.

main fuel of the colonocyte and seems to play a role in the prevention in a number of colonic diseases, including colon cancer.

Recently Soral-Śmietana *et al.* (2001*a*) proposed using native pea starches to produce resistant starches. They obtained an RS preparation from two commercial peas starches (Nastar and Gel-Flo) with 38 % RS (% dry matter), the initial native pea starches containing respectively 43 and 19 % RS (% dry matter).

Pure pea starch (NASTAR, Cosucra BV, Rosendaal, The Netherlands) elicited less hyperglycaemia (− 47 %), hyperinsulinaemia (− 54 %), and C-peptide secretion (− 37 %) as compared to corn starches (modified and unmodified) ($P < 0{\cdot}05$) in healthy subjects (Seewi *et al.* 1999). No differences in flatulence nor breath hydrogen were observed between both starches, suggesting no significant differences in RS content between the two starches.

In a study on pigs, van der Meulen *et al.* (1997) concluded that ileal digestibilities of native maize and pea starches were equal, but that the rate of appearance of glucose in the portal vein was higher for maize starch. Net portal glucose flux was lower for pea starch, but after 8 h post-prandially, portal glucose flux was significantly higher than with maize starch. Net portal SCFA flux, being higher with pea starch than with maize starch, did not apparently confirm the results of ileal digestibility, but that could be explained by different uptakes of SCFA by the colonic mucosa as already suspected with other RS (Martin *et al.* 2000).

Mung bean (*Phaseolus aureus*) and waxy maize starches were incorporated in the diet of rats to provide, respectively, a low- and a high-glycaemic-index (GI) diet (Kabir *et al.* 2000). After 12 weeks, the group fed the low-GI food had higher plasma leptin and ob mRNA than the other group, but no effect was observed on food intake, basal plasma glucose, insulin or triacylglycerols. It was suggested that leptin sensitivity was increased in the ‘high-GI group’ and that this step might precede weight gain and increase in fat mass. Earlier, the same group (Kabir *et al.* 1998) showed that the high-GI diet stimulated fatty acid synthase activity and lipogenesis, and might have undesirable long-term metabolic effects.

The same mung bean starch has been compared with corn starch and glucose in humans to quantify the rate of net post-hepatic appearance of glucose after ingestion of both starches (Lang *et al.* 1999). Glycaemic indices of maize and mung bean starches were respectively 95 ± 18 and 51 ± 13. Post-hepatic appearance of glucose from glucose, corn and mung bean starches represented respectively 79·4 ± 5·0, 72·6 ± 4·0 and 35·6 ± 4·6 % of the glucose load after 4.5 h post-prandially. This big difference has been attributed to the fact that mung bean starch contains about 11 % of RS, and to a long absorption period of mung bean starch which was probably not finished 4.5 h after the meal.

Recently, Fukushima *et al.* (2001) observed the lowering of serum total cholesterol in rats by starches of two different varieties of beans (*Phaseolus vulgaris*) (15 g/kg). The total cholesterol:HDL–cholesterol ratio in these bean starch groups was also significantly lower than in the control group (maize starch) at the end of the 4-week feeding period. This effect was attributed to the enhancing effect of RS on the hepatic LDL receptor mRNA level.

The pattern of large intestine SCFA after consumption of starchy grain legumes has been confirmed to be rich in butyrate. Indeed, Key & Mathers (1993) observed a considerable increase in butyrate between 1 and 3 d and up to 14 d of adaptation of rats to a diet containing cooked *P. vulgaris*. No RS was recovered in the faeces of the animal, showing the complete colonic fermentation of this fraction.

α-Galactosides

Due to their high fermentability, α-galactosides induce the production of gases (mainly CO_2, H_2 and, in some populations, CH_4) responsible for the digestive discomfort related to pulse consumption. These oligosaccharides are quite characteristic of grain legumes and are present in all species, with large variabilities among different varieties. Although α-galactosides are claimed to be solely responsible for flatulence in soyabeans or pulses, the flatulence activity of some grain legumes (e.g. smooth-seeded field peas) may also be due to indigestible oligosaccharides and cell wall fibre constituents (Fleming, 1981).

Besides causing digestive discomfort in most populations, flatus production may be a more acute problem in individuals with colonic pathologies such as irritable bowel syndrome. For instance, the local bean diet has been identified as one of the most common aggravating factors of irritable bowel syndrome in Nigerians (Atoba, 1988). Flatus production needs to be lowered in these patients by reducing consumption of fermentable carbohydrates such as beans or lentils (Friedman, 1991).

For most of the population, it might seem desirable to remove α-galactosides from pulses by technological or genetic means. However, these non-digestible oligosaccharides have been identified as prebiotic agents (Van Loo *et al.* 1999), i.e. food ingredients potentially beneficial to the health of consumers. At the present time in Europe, the main prebiotics are inulin-type fructans, characterized by the presence of fructosyl units bound to the β-2,1 position of sucrose. Prebiotics escape enzymatic digestion in the upper gastrointestinal tract and enter the caecum without change to their structure. None are excreted in stools, which indicates that they are fermented by colonic flora to produce a mixture of SCFA (acetate, propionate and butyrate), L-lactate, CO_2 and H_2. Their stimulation of bifidobacteria may have several beneficial implications for health:

1. Potential protective effects against colorectal cancer and infectious bowel diseases through inhibition of putrefactive (*Clostridium perfringens*) and pathogenic bacteria (*Escherichia coli*, salmonella, *Listeria* sp. and shigella), respectively. The effects of prebiotics on colon carcinogenesis and tumour growth have been evaluated in animals (mostly on azoxymethane- or dimethylhydrazine-treated rats), and development of colonic aberrant crypt foci (ACF) is the marker most often described. ACF are putative preneoplastic lesions

from which adenomas and carcinomas may develop in the colon (Reddy, 1999).

2. Improvement of carbohydrate and lipid metabolism. This possibility has gained support from the observation that dietary oligofructosaccharides cause suppression of hepatic triacylglycerol and VLDL synthesis in animals, resulting in marked reductions in triacylglycerol and, to a lesser extent, cholesterol levels (Taylor & Williams, 1998).
3. Providing fibre-like properties by decreasing renal nitrogen excretion.
4. Increasing the availability of essential minerals.
5. Acting as a low cariogenic factor.

It has been shown clearly in human volunteers that non-digestible oligosaccharides, particularly fructooligosaccharides, stimulate the growth of bifidobacteria selectively, modifying the composition of the colonic microbiota significantly (Roberfroid, 1997).

These potentially beneficial effects have been largely studied in animals, but have not really been tested in humans (Roberfroid, 1998; Grizard & Barthomeuf, 1999; van Loo *et al.* 1999). Human clinical trials are likely to broaden our insight concerning the importance of prebiotics in health and disease.

Although prebiotic effects have been demonstrated extensively with oligofructose, fructooligosaccharides and inulin, there is no evidence (a lack of studies) of such effects with α-galactosides. However, like low molecular weight fructans, they are quickly fermented (Bradburn *et al.* 1993) in the colon and could be expected to have beneficial properties similar to those of fructooligosaccharides.

Conclusion

Apart from being eaten as a vegetable, grain legumes can be considered as a source for raw material for the processing industry. Legume starch and fibre both have useful functional properties and can be used readily in food products. Procedures have been developed for isolating these fractions. Legume starch has unique properties, having a good stability to high temperature and high point viscosity compared with equivalent cereal or tuber starch. The technofunctional properties can be further improved by starch processing, including chemical and biotechnological methods. Moreover, mutant genes affecting starch synthesis may enlarge the spectrum of starch that could suit a wide range of food and non-food applications. The fibre fraction from the seed embryo has excellent water hydration properties that could be utilized to structure food and replace fat in areas such as confectionery, dressings or meat.

Starch and fibre can also be proposed as ingredients for their beneficial health effects. Legume seed starch is a source of RS, which seems to be interesting for the production of a large amount of butyrate on its fermentation by colonic bacteria. Fibre provides a broad range of positive effects, both physiological and metabolic, at least in subjects suffering from disorders. These effects are related to the source of fibre (from cotyledon or hull), are dose related and depend on the form in which it is ingested.

Scientific research must be undertaken to substantiate the potential long-term positive effects of legume starch and fibre ingestion on health, in both normal individuals and subjects suffering from disorders. Fundamental knowledge of the behaviour of starch and fibre fractions in complex food systems is still required in order to be able to propose ingredients and adaptations to formulations for appetizing foods with good nutritional properties.

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