

Department Drug Science and Technology

Plant Based Food Vegetals





Department Drug Science and Technology

Unit 1: Dietary fibre: definitions, chemical composition and nutritional characteristics. Definition of pre-biotic and pro-biotic (functional foods) and effects on human metabolism.

Unit 2: Anti-nutritional substances and toxic substances present in plants: protease inhibitors, lectins, phytates, glucosinolates, AA derivatives, alkaloids, cyanogenic glycosides, saponins).

Classifications



DI TORINO

Department Drug Science and Technology

Foods of plant origin, fresh and/or subjected to technological treatments can be classified as follows:

- ✓ "I gamma" Fresh products (e.g. fresh vegetables);
- ✓ "II gamma" Foods preserved through heat treatment (e.g. pasteurized milk);
- "III gamma" Frozen and deep-frozen foods (e.g. frozen fish fillets); fresh-cut foods - fresh, clean and ready-to-eat foods (e.g. washed and pre-cut vegetables);
- ✓ "V gamma" Food already cooked and vacuum preserved (e.g. vacuum cooked first courses).





In the EU, regulation 1169/2011 on the provision of food information to consumers, defines fibre as 'carbohydrate polymers with three or more monomeric units, which are neither digested nor absorbed in the human small intestine and belong to the following categories:

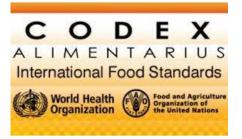
- \checkmark edible carbohydrate polymers naturally occurring in the food as consumed,
- edible carbohydrate polymers which have been obtained from food raw material by physical, enzymatic or chemical means and which have a beneficial physiological effect demonstrated by generally accepted scientific evidence,
- edible synthetic carbohydrate polymers which have a beneficial physiological \checkmark effect demonstrated by generally accepted scientific evidence'.





Similar to the EU, the United States (US) **Food and Drug Administration** (FDA) definition refers to 'non-digestible soluble and insoluble carbohydrates (with 3 or more monomeric units), and lignin that are intrinsic and intact in plants; isolated or synthetic non-digestible carbohydrates (with 3 or more monomeric units) determined by FDA to have physiological effects that are beneficial to human health'.

The EU and US definitions differ from the **Codex Alimentarius definition** on the number of monomers that constitute the carbohydrate polymer; while the EU and US includes three or more monomeric units, the Codex definition specifies ten or more, leaving national authorities to decide whether to include as fibre also carbohydrates with 3-9 monomers.



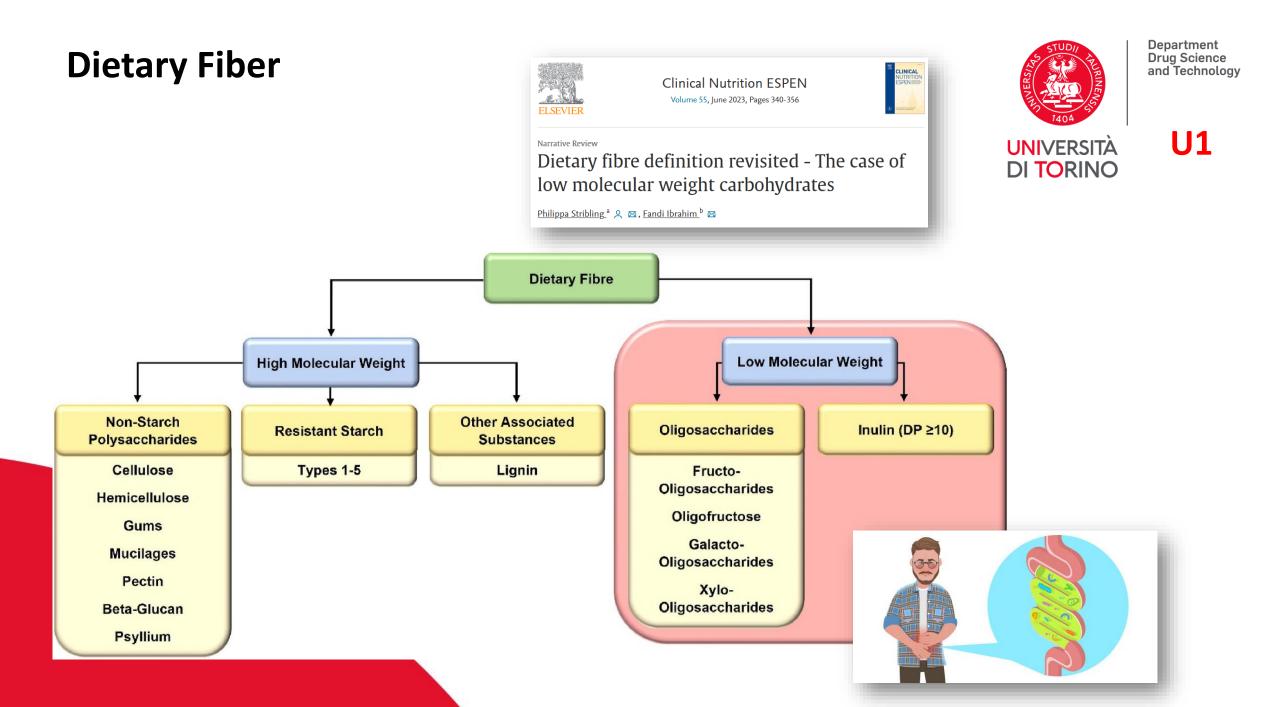
UNIVERSITÀ DI TORINO Department Drug Science and Technology

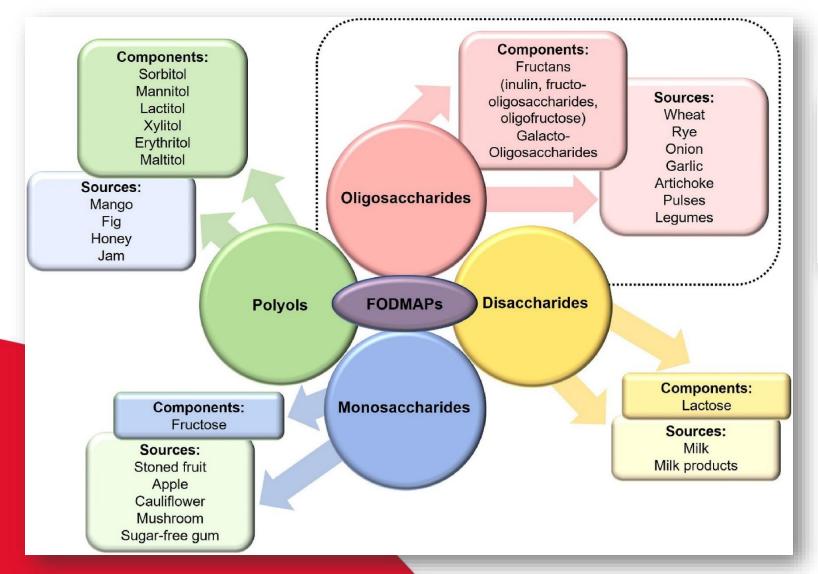
Department Drug Science and Technology **UNIVFRSITA** DI TORINO

111

Early definitions of dietary fibre included only non-digestible components of the plant cell wall (namely NSP), but more recent definitions now include any edible parts of the plant or analogous (extracted/synthetic) carbohydrates that are resistant to digestion in the small intestine and fermented in the large intestine. Such expansions have led to the inclusion of **non-digestible oligosaccharides** within many definitions.

<u>Oligosaccharides</u> are a category of <u>short-chain carbohydrates</u>, which are considered low molecular weight (LMW) dietary fibre and are plant energy reserves found naturally in wheat-based grains, pasta, bread, pulses, and legumes. Oligosaccharides are primarily associated with their microbial effects, rather than their effects on bowel function, and there has been great controversy regarding their classification as dietary fibre.





UNIVERSITÀ

Department Drug Science and Technology

U1



Clinical Nutrition ESPEN Volume 55, June 2023, Pages 340-356

DI TORINO



Narrative Review

Dietary fibre definition revisited - The case of low molecular weight carbohydrates

Philippa Stribling a 🝳 🖾 , Fandi Ibrahim ^b 🖂

The components of the low FODMAP (fermentable oligosaccharides, disaccharides, monosaccharides, and polyols) diet and respective sources. Oligosaccharides, including inulin, are the main components of the low FODMAP diet due to their presence in many commonly consumed foods.

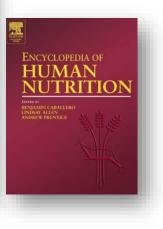


UNIVERSITÀ DI TORINO Department Drug Science and Technology

U1

Table 7 The physiological effects of resistant starch, or	ble 7 The physiological effects of resistant starch, oligosaccharides, and dietary fiber					
Physiological effect	Resistant starch	Oligosaccharides	Dietary fiber			
Energy supply	$8-13 \text{kJ} \text{g}^{-1}$	8–13 kJ g ⁻¹	$8-13 kJ g^{-1}$			
Increased glucose tolerance	Some foods	No	Some NSP ^a			
Decreased plasma cholesterol and triacylglyceride levels	No	Not known	Some NSP			
Fermentability	Complete	Complete	Variable			
Production of SCFA	Yes	Yes	Yes			
Increased butyrate production	High	High	Variable			
CO ₂ and H ₂ production	Yes	Yes	Variable			
Decreased fecal pH	Yes	Yes	Some NSP			
Decreased production of deoxycholate	Yes	Yes	Some NSP			
Increased colonocyte proliferation	Yes	Yes	Yes			
Increased fecal bulk	At high dose	No	Variable			
Faster whole gut transit time	At high dose	No	Yes			
Increased bacterial nitrogen and biomass	Yes	Yes	Yes			
Reduced mineral absorption in small intestine	No	No	Some NSP			
Increased mineral absorption in large intestine	Yes	Yes	Some NSP			
Possible prevention of colorectal cancer	Yes	Not known	Yes			

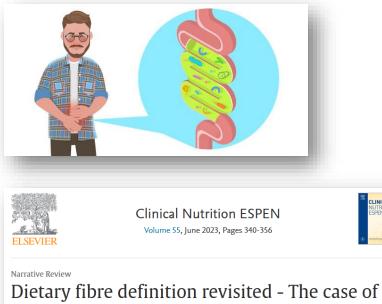
^aNSP, nonstarch polysaccharide.



LMW non-digestible carbohydrates, due to their <u>rapid</u> <u>fermentation in the proximal colon</u>, may cause deleterious effects in individuals with functional bowel disorders (FBDs) and, as such, <u>are excluded on the low FODMAP (fermentable</u> <u>oligosaccharides, disaccharides, and polyols) diet</u> and similar protocols.

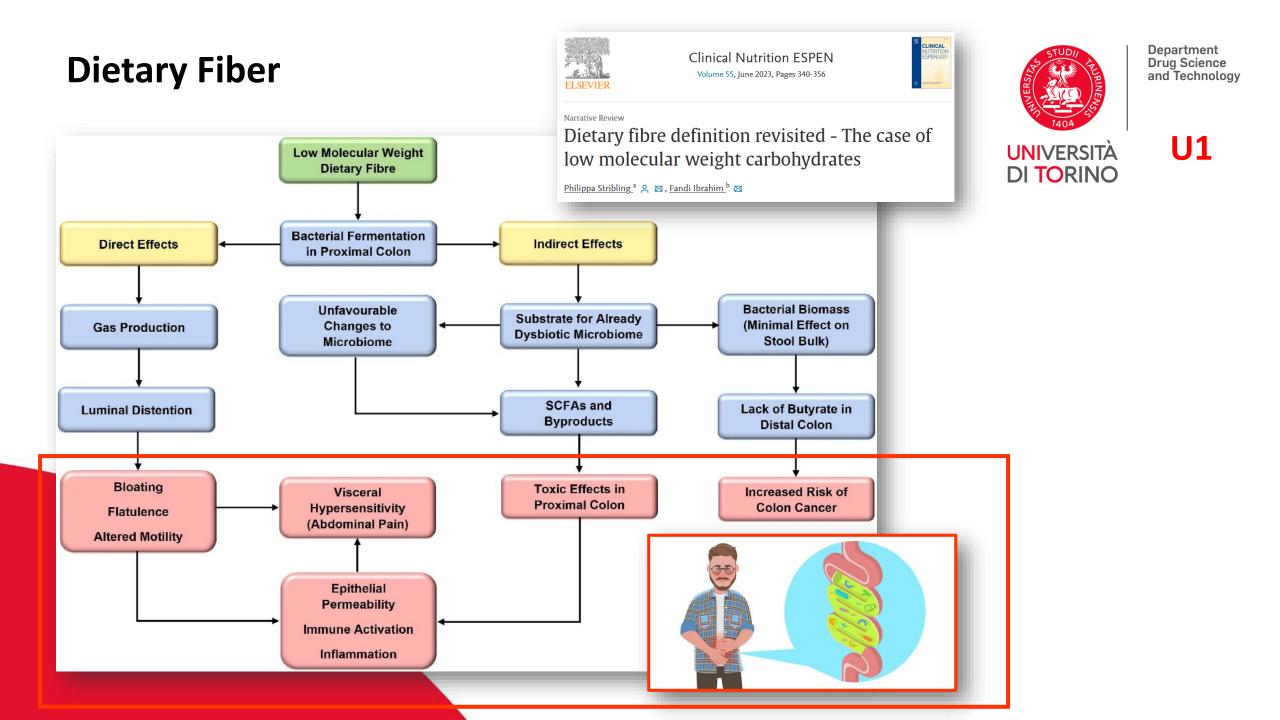
Their addition to food products as dietary fibre allows the use of associated nutrition/health claims, causing a paradox for those with FBDs, which is further complicated by lack of clarity on food labelling.





low molecular weight carbohydrates

Philippa Stribling.ª 🔉 🖾 , Fandi Ibrahim.^b 🖾



Source

[2]

EFSA 2010 [2]



Dietary recommendations for dietary fibre intake as described by food- and health- related organisations **Dietary recommendation** · 'Based on the available evidence on bowel function, the Panel considers dietary fibre intakes of 25 g per day to be adequate for normal laxation in adults'. • '...in adults there is evidence of benefit to health associated with consumption of diets rich in fibre-containing foods at dietary fibre intakes greater than 25 g per day...'. • 'There is limited evidence to set adequate intakes for children. [...] the Adequate Intake (AI) for dietary fibre for children should be based on that for adults with appropriate adjustment for energy intake. A fibre intake of 2 g per MJ is considered adequate for normal laxation in children from the age of one year'. 'Wholegrain cereals, fruits and vegetables are the preferred sources of non-starch FAO/WHO 2003 [7 (pdf), Mann J. 2007 polysaccharides (NSP)'.

> • 'The recommended intake of fruits and vegetables and consumption of wholegrain foods is likely to provide >20 g per day of NSP (>25 g per day of total dietary fibre)'.

https://knowledge4policy.ec.europa.eu/health-promotion-knowledge-gateway/dietary-fibre-recommendations-2 en

Department Drug Science and Technology

Pre-biotics vs. pro-biotics







Department Drug Science and Technology

U1

Classical **prebiotic effects** are mediated through co<u>nsumption of the</u> <u>substrate by specific groups within the microbiota, promoting their</u> <u>growth and metabolic activity</u>. Provision of substrate to select group/s of bacteria can also indirectly influence other bacterial groups within the microbiome – promoting growth through crossfeeding <u>interactions as well as inhibitory effects via pathogen displacement</u>. Resulting <u>changes in microbial composition</u> and metabolite concentrations from prebiotic administration <u>impact host epithelial</u>, <u>immune, nervous, and endocrine signalling and mediate health</u> <u>benefits such as improvements in bowel function, immune response,</u> <u>glucose and lipid metabolism, bone health, and regulation of appetite</u> <u>and satiety</u>.

Chief by-products of bacterial prebiotic metabolism are the <u>SCFAs</u> <u>acetate</u>, <u>butyrate</u>, <u>and propionate</u>, which are well recognised to interact with these host systems and facilitate many prebiotic effects.

In addition to nutritive effects on microbes, <u>prebiotic molecules are</u> <u>also recognised to interact directly with host receptors, modulating</u> <u>immune and gut epithelial cell signalling</u> with local effects on inflammation and barrier function. **Probiotics** interact with both the host and the microbiome via **molecular effectors** present on the cell structure or secreted as metabolic products. Probio<u>tic metabolites can act on the microbiota by crossfeeding interactions,</u> <u>changes in the gastrointestinal microenvironment (e.g., pH lowering),</u> <u>competition for nutrients and binding sites, and inhibition of growth via the</u> <u>production of strain-specific antibacterial compounds including bacteriocins</u>. Such microbiota-directed effects contribute to the ability of probiotics to mediate health benefits in pathogen overgrowth states.

With regard to host cells, <u>probiotic effector molecules can interact directly</u> <u>with receptors in intestinal epithelial, enteroendocrine, and immune cells as</u> <u>well as vagal afferent fibres</u>. These interactions produce local gut effects, such as enhancement of intestinal barrier integrity and inflammation (e.g., via Tolllike receptors), as well as systemic effects via host immune, endocrine, and nervous system.

Probiotics can also <u>perform enzymatic metabolism of host compounds such</u> <u>as bile salts and ingested xenobiotics</u>. Specific probiotic surface-associated effector molecules include pili, lipoteichoic acids, exopolysaccharides, and various surface-layer proteins, many of which are strain-specific and therefore mediate the delivery of strain-specific effects.

Pre-biotics *vs.* **pro-biotics**

Microbiology

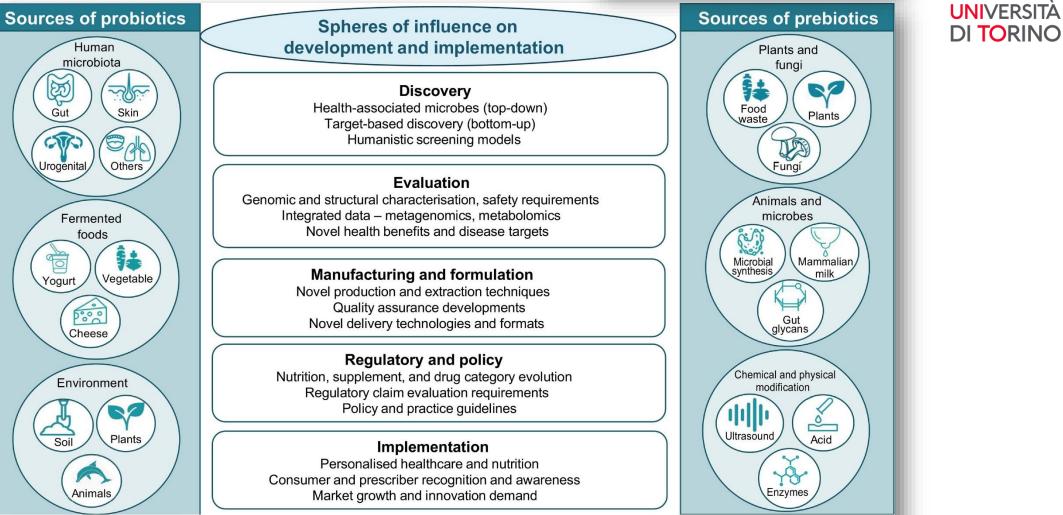
Special Issue: Regulation of Human Health by the Microbiota Feature Review

Trends in

Shaping the Future of Probiotics and Prebiotics

CelPress

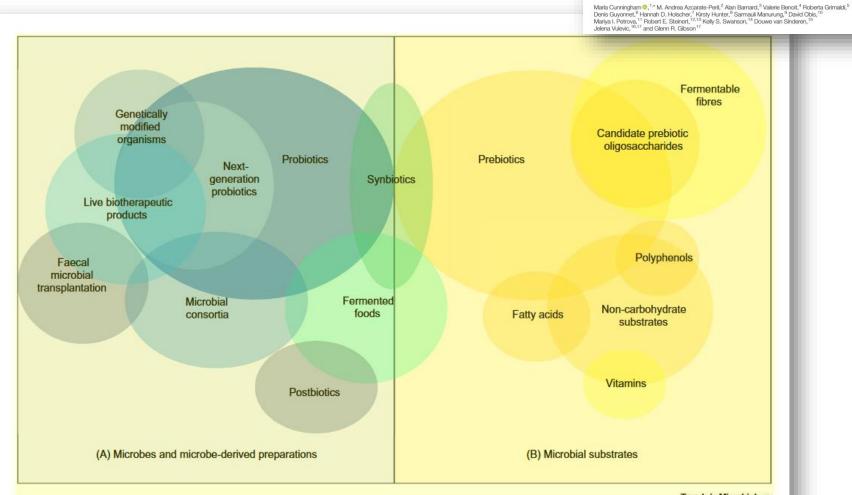
Marla Cunningham 9,1.* M. Andrea Azcarate-Peril,2 Alan Barnard,3 Valerie Benoit,4 Roberta Grimaldi,5 Hatas Guringret, Hanan D. Holscher,² Kirsty Hunter, "Scarnavi, Yuaeio C. Sham, "Iobert Denis Guyoner," Hanan D. Holscher,² Kirsty Hunter, "Samauli Manung," David Obis," Martya I. Petrova, ¹¹ Robert E. Steiner, ^{12,12} Kelly S. Swanson, ¹⁴ Douwe van Sinderen, ¹⁵ Jelena Vulevi, ^{6,15} and Glann R. Gibson¹⁷



Trends in Microbiology



Pre-biotics *vs.* **pro-biotics**





CelPress

Department Drug Science and Technology

U1

Trends in Microbiology

Trends in

Microbiology

Feature Review

Special Issue: Regulation of Human Health by the Microbiota

Shaping the Future of Probiotics and Prebiotics

Figure 2. Probiotics, Prebiotics, and Adjacent Fields. Many areas of new discovery intersect and are adjacent to the currently defined fields of probiotics and prebiotics. The figure depicts a conceptual map of these established and emerging concepts, with (A) microbes and microbe-derived preparations represented in shades of blue; (B) microbial substrates represented in shades of yellow; and preparations containing both represented in green. Current and potential relationships between fields are depicted by the overlap of shapes (note: the size and shape representing each field is for visual purposes only). Refer to the Glossary for definitions of each field.

Anti-nutritional substances and toxic substances

Vegetables may contain substances capable of interfering with the absorption, distribution and/or metabolism of nutrients. These substances are defined as "anti-nutritional" and have different targets depending on their structure and mechanism of action.

Some examples of anti-nutritional substances are:

Inhibitors of proteases, amylases, lectins (in legumes they cause malabsorption), phytates (they reduce the absorption of some micronutrients - metals) etc...

When a **real toxicity mechanism** is established (which can have different connotations depending on the exposure - acute/chronic) then we are **referring to toxic compounds**. In this category it is possible to include the **glucosinolates** contained in the *Brassicaceae* which have goitrogenic activity, some **glycosidic alkaloids** of legumes which cause hemolytic anemia in individuals with hereditary enzyme deficiencies, the **saponins** present in legumes with haemagglutinogenic activity, the pyrrolizidine alkaloids present in derivatives such as honey with marked hepatotoxicity, the **cyanogenic glycosides** which through hydrolysis release HCN, a powerful inhibitor of cellular respiration, amino acid derivatives such as β -N-oxalyl-L-a,b-diaminoproprionic acid contained in the wild pea (Lathyrus sativus) and responsible for lathyrism (limb paralysis lower and damage to the CNS), **erucic acid** (present in rapeseed oil) which leads to metabolic disorders. Not to be forgotten are the proteins capable of triggering allergic responses.

Follow some examples of toxic and/or anti-nutritional substances, also discussing possible technological treatments and/or measures that can reduce the impact on human health.



DI TORINO

Enzyme inhibitors

The main enzyme inhibitors to be considered real anti-nutritional factors are **protease inhibitors**; they are proteins whose sequence is known, as well as the active sites, which reduce the activity of pancreatic enzymes, leading to pancreatic hypertrophy and decreasing protein absorption.

They are particularly present in legumes (beans, peas and soya) but also in eggs and cereals; to reduce its activity, denaturation by heat treatment (cooking) is necessary.

Some protease inhibitors found in legumes, also called Bowman-Birk inhibitors, have two different active sites capable of blocking enzymes such as trypsin and chymotrypsin. These compounds are particularly resistant to normal treatments aimed at inactivating them.

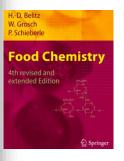
		Inhibition of ^a						
Source/Inhibitor	Molecular weight	T CT		Р	Bs	AP	SG	PP
Animal tissues								
Bovine pancreas								
Kazal inhibitor	6153	+	_	_				
Kunitz inhibitor	6512	+	+	_	_	_	+	
Chicken egg								
Ovomucoid	27-31,000	+	_		_			
Ovoinhibitor	44-52,000	+	+	_	+	+		
Ficin-papain-inhibitor	12,700	—	_	+	_			
Plant tissues								
Cruciferae								
Raphanus sativus ^b	8-11,200	+	\pm	_	+	+		
Brassica juncea ^b	10-20,000	+	\pm					
Leguminosae								
Arachis hypogaea ^b	7500-17,000	+	+					
Cicer arietinum ^b	12,000	+	+					
Glycine max								
Kunitz inhibitor	21,500	+	$^+$	_	_			
Bowman–Birk inhibitor	8000	+	+	_	+			
P. coccineus ^c	8800-10,700	+	+					
P. lunatus ^c	8300-16,200	+	+	_	_	\pm		
P. vulgaris ^c	8-10,000	+	+	_	_			
Pisum sativum ^b	8-12,800	+	+					
Vicia faba ^b	23,000	+	+					
Convolvulaceae								
Ipomoea batatas ^b	23-24,000	+	_	_	_	_		
Solanaceae								
Solanum tuberosum ^b	22-42,000 ^c	\pm	\pm	_	\pm	\pm	\pm	\pm
Bromeliaceae								
Ananas comosus ^b	5500	+	+					
Gramineae								
Hordeum vulgare ^b	14-25,000	\pm	_	_	\pm	\pm	\pm	
Oryza sativa ^b		\pm	_	+				
Secale cereale ^b	10-18,700	+	+	_				
Triticum aestivum ^b	12-18,500	±	_	_				
Zea mays ^c	7-18,500	+	+	_				

^a T: trypsin, CT: α -chymotrypsin, P: papain, Bs: Bacillus subtilis proteases, AP: Aspergillus spp. proteases, SG: Streptomyces griseus proteases, PP: Penicillium spp. proteases, +: inhibited, -: not inhibited, \pm : inhibited by some inhibitors of the particular source.

^b The properties of different inhibitors are combined.

c Subunits 6-10,000.

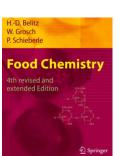
Department Drug Science and Technology



Enzyme inhibitors

The resistance of protease inhibitors to stomach pH is quite significant (especially for Bowman-Birk inhibitors). Thermal treatments are generally the most effective way to reduce their effectiveness and therefore limit their effects on health.

Table 16.16. Destruction of trypsin inhibitors by heating					
Sample	Process	Destruction (%)			
Soy flour	Live steam, 100 °C, 9 min	87			
Soy bean	10% Ca(OH)2, 80 °C, 1 h	100			
Navy bean	Autoclaving, 121 °C, 5 min	80			
	Autoclaving, 121 °C, 30 min	100			
	Dry roasting, 196–204 °C, 20–25 s	75			
Navy bean	Pressure cooking, 15 min	89			
Winged bean	Autoclaving	92			
-	Soaking + autoclaving	95			
Chickpea	Autoclaving	54			
Broad bean	Autoclaving, 120 °C, 20 min	90			
Horse bean	Autoclaving	100			
Black gram	Cooking, 100 °C, 10 min	15			
·	Autoclaving, 108 °C, 10 min	27			
	Autoclaving, 116 °C, 10 min	38			
Cow pea	Cooking, 90-95 °C, 45 min	52			
•	Autoclaving, 121 °C, 15 min	11			
	Toasting, 210 °C, 30 min	44			
	Toasting, 240 °C, 30 min	22			
	Extrusion cooking	19			
Peanut	Moist heat, 100 °C, 15 min	100			



UNIVERSITÀ

DI TORINO

Department Drug Science and Technology

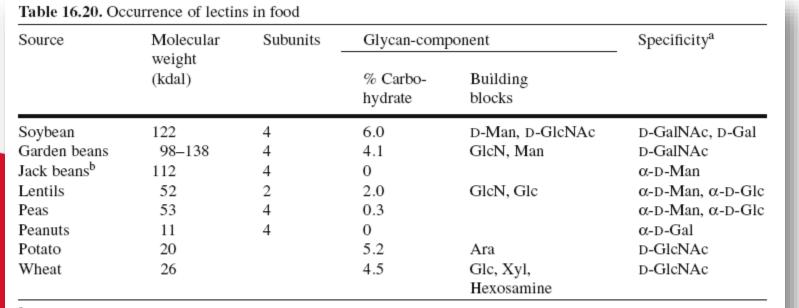
U2

Source/Inhibitor	Remaining activity ^b (%
Soybean, Kunitz inhibitor	0
Bowman-Birk inhibitor (BBI)	100
extract	30-40
Lima bean, BBI-type inhibitor	70-93
Kidney bean, BBI-type inhibitor	100
Kintoki bean, BBI-type inhibitor	100
Lentil, BBI-type inhibitor	83-100
Chick pea, inhibitors	100
Broad bean, trypsin-chymotrypsin	
inhibitor	100
Moth bean, trypsin inhibitor	91
Broad bean, trypsin inhibitor	100

^b Against bovine and human trypsin and chymotrypsin.

Lectins

Legumes contain a group of proteins, many of which have links to carbohydrate residues (glycoproteins), called **lectins** which are able to bind to erythrocytes causing hemoagglutination. There are other toxic effects on humans including: morpho-functional damage to the intestinal epithelium, pancreatic and hepatic hypertrophy, thymic atrophy and in general an increase in the catabolism of proteins and fats.



Soy lectins are inactivated by heat with treatment at 100°C for 10'.

^a Precipitates biopolymers that contain the given building blocks (polysaccharides, glycoproteins, lipopolysaccharides).

^b Canavalia ensiformis.



H.-D. Belitz

W. Grosch P. Schieberle

> 4th revised and extended Edition

Food Chemistry

Department Drug Science and Technology

Phytates

Cereals and legumes contain significant quantities (up to 1% in cereals) of phytic acid. It is capable of binding up to 70% of the phosphorus contained in the vegetal matrix, in the caryopsis it is compartmentalized in the aleurone layer and its concentration in the flours will depend on the degree of buttering. During the formation of gluten, due to hydration of the flours, the phytases are activated and hydrolyze the substrate to myoinositol and phosphoric acid. The anti-nutritional activity of this compound is linked to the ability to sequester metals such as Zn, Fe, Ca and Mg, thus removing them from absorption.





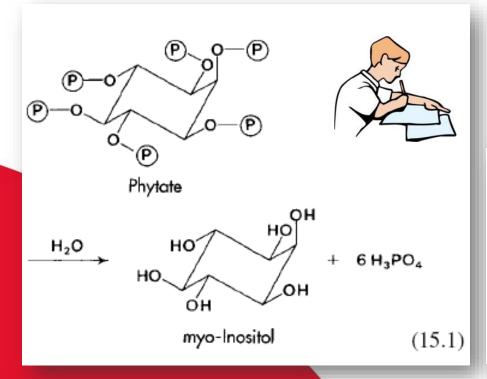
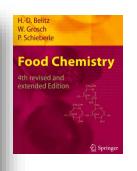


Table 15.20.1	Phytate content in wheat	flour
Degree of grinding	Phytate (mg/kg) ^a	
70%	53	
85%	451	
92%	759	

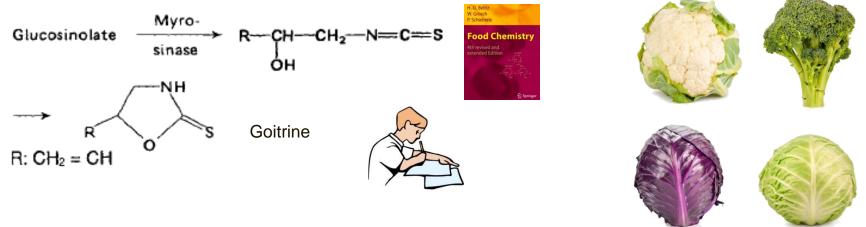


Glucosinolates

The *Brassicaceae* contain a class of compounds called glucosinolates which, following enzymatic hydrolysis by specific enzymes - myrosinase -, form oxazolidine derivatives defined as rhodanides which, by interfering with the uptake of iodine by the thyroid tissue, act as goitrogens, especially if administered in iodine-poor diets.

Amino acid derivatives

Some varieties of pea, of the genus *Lathyrus sativus*, particularly widespread in India where they are grown during the dry period, contain a compound, structurally similar to glutamic acid, β -N-oxalyl-L-a,b-diaminopropionic acid. This compound, which can be eliminated by soaking the legumes in water for 12 hours, targets the CNS, causing chronic toxic effects such as paralysis of the lower limbs (neurolathyrism).





UNIVERSITÀ

DI TORINO

Alkaloids from Vicia faba

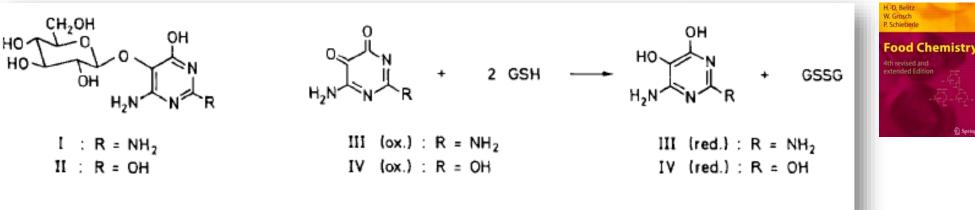
Among the legumes, the fruits of *Vicia faba*, the broad beans, compete for some glucoside alkaloids (vicin and convicin – I and II) which, attacked by the β -glycosidases of the gastro-intestinal tract, release the aglycones (divicin and isouramyl – III and IV respectively) which by interacting with glutathione at the erythrocyte level (GSH) cause its oxidation. In individuals with hereditary deficiency of the enzyme G6PDH (glucose-6-phosphate dehydrogenase) this causes hemolytic anemia, the so-called favism.



UNIVERSITÀ

DI TORINO





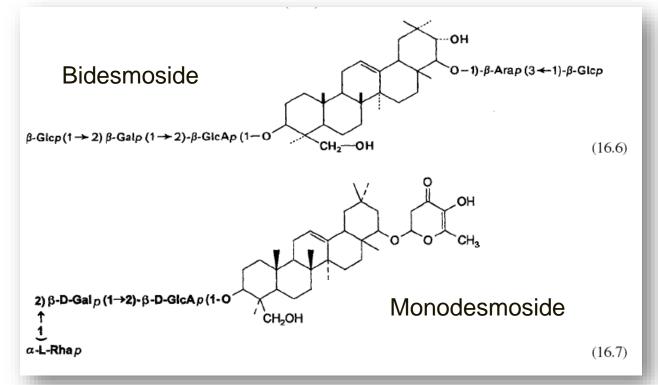
Saponins

Saponins are specialized plant metabolites with a pentacyclic structure, belonging to the triterpene class. They have an aglyconic portion and a glycosidic portion in which from 1 to 8 carbohydrate and/or uronic acid residues are present. They have surfactant properties and in legumes they are responsible for the characteristic flavour. They are divided into two classes, monodesmosides and bidesmosides, depending on the number of glycoside chains they present.

The toxic activity translates into a **hemolytic effect**, present only for some compounds, given the reduced absorption in the GI tract the toxicity of saponins is not relevant.

nistry

Table 10.27. 5	aponin content in foods	W. Grosch P. Schieberle
Food	Saponin (g/kg solids)	Food Cher
Chick peas	56	4th revised and extended Edition
Soybeans	43	- o- (m
Garden beans	4.5-21	
Peanuts	6.3	
Lentils	3.7-4.6	
Broad beans	3.5	
Peas	11	
Spinach	47	
Asparagus	15	
Oat bran	1.0	





DI TORINO

Department Drug Science and Technology

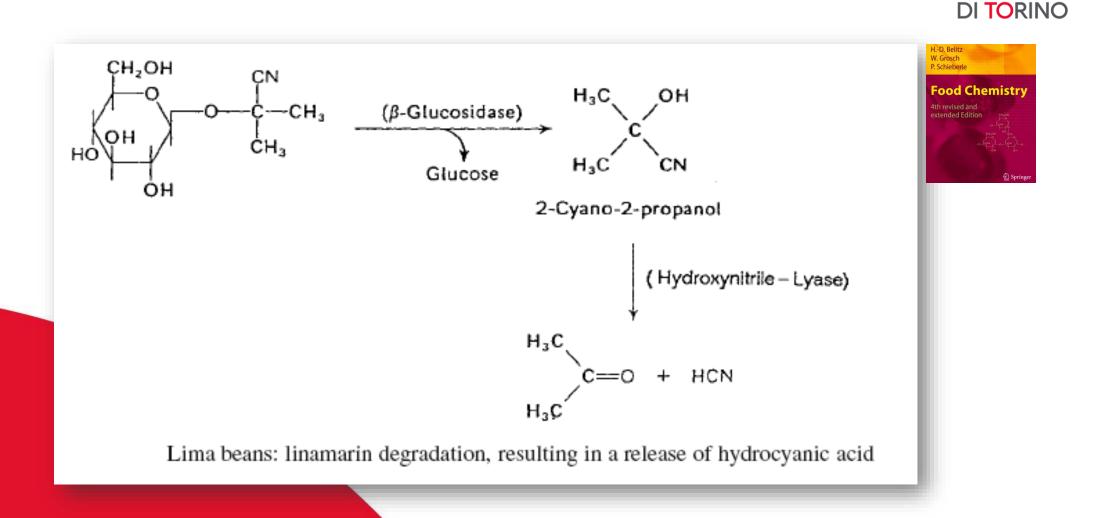
The cyanogenic glycosides present in the seeds of green beans (Lima bean – *Phaseolus lunatus*) and other vegetables including Cassava, sweet almonds, peaches, apples etc. are compounds which, hydrolysed by b-glucosidases, release HCN.

Technological treatments such as crushing and hydrating the seeds lead to the hydrolysis of the glycosides and the subsequent heating promotes the removal (through evaporation) of the hydrocyanic acid formed.





	$\begin{array}{c} R_2 \\ O - \beta - sugar \\ C \\ R_1 \\ C \\ N \end{array}$					
Glycoside Name	Structure		Sugar	Amino acid precursor	Occurrence (seeds)	
	R ₁	R_2			. ,	
Linamarin	CH3	CH3	Glucose	Val	Lima bean Linseed (flax) Cassava	
(R)-Lotaustralin	C_2H_5	CH_3	Glucose	Ile	like Linamarin	
(R)-Prunasin	Phenyl	Н	Glucose	Phe	Prunes	
(R)-Amygdalin	Phenyl	Н	Gentiobiose	Phe	Bitter almond Apricots Peaches Apples	
(S)-Dhurrin	HO-Phenyl	Н	Glucose	Tyr	Sorghum sp.	



Department Drug Science and Technology

U2

UNIVERSITÀ

Avvelenamenti dei bovini da sorgo foraggero, evento legato alla siccità

SANITÀ ANIMALE

August 2022





Department Drug Science and Technology

U2

The causes are attributable to the drought, which on the one hand would have pushed farmers to use fodder more resistant to water shortages, such as **sorghum**, and on the other hand this year's weather and climate factors would have favored greater production of this toxin by fodder sorghum. This year's levels are probably such that they frequently reach the lethal dose, causing the poisoning of cows. The analyzes conducted on a sample of sorghum highlighted the presence of over 10,000 mg/kg of **dhurrin**. These quantities, taking into account the transformation factor from durrin to hydrocyanic acid, correspond to the presence of over 900 mg/kg of hydrocyanic acid. The limit for feed is set by the National Official Control Plan on Animal Nutrition at 50 mg/kg. The quantity found in this case is almost 20 times higher than the limit.

.OH

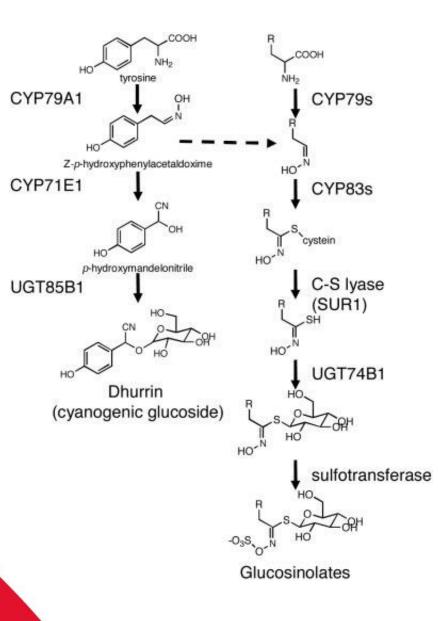
<text><text><text><image>

ЮH

C III N

HO

HC





Department Drug Science and Technology