Tannins

Tannins are a class of astringent, polyphenolic biomolecules that bind to and precipitate proteins and various other organic compounds including amino acids and alkaloids. The term tannin refers to the use of oak and other bark in tanning animal hides into leather. By extension, the term tannin is widely applied to any large polyphenolic compound containing enough hydroxyls and other suitable groups to form strong complexes with various macromolecules. The tannin compounds are widely distributed in many species of plants, where they play a role in protection from predation (including as pesticides) and might help in regulating plant growth. The astringency from the tannins is what causes the dry and puckery feeling in the mouth following the consumption of unripened fruit, red wine or tea. Likewise, the destruction or modification of tannins with time plays an important role when determining harvesting times. Tannins have molecular weights ranging from 500 to over 3,000 (gallic acid esters) and up to 20,000 (proanthocyanidins). Classification of tannins 1. Hydrolysable tannins 2. Condensed tannins 3. Pseudotannins 1. Hydrolysable tannins: These tannins are hydrolyzed by acids, or enzyme and produce gallic acid and ellagic acid. Chemically, these are esters of phenolic acid like gallic acid and ellagic acid. The tannins derived from gallic acid are known as gallitannins and from that of ellagic acid are known as ellagitannins. The gallic acid is found in rhubarb, clove and ellagic acid is found in eucalyptus leave and myrobalans and pomegranate bark. These tannins are treated with ferric chloride to produced blue or black colour. 2. Condensed tannins: These tannins are resistant to hydrolysis and they derived from the flavonols, catechins and flavan-3, 4-diols. On treatment with acids or enzymes they are decomposed into phlobaphenes. On dry distillation condensed tannin produce catechol. These tannins are called as catechol tannins. These tannins are found in cinchona bark, male fern, areca seeds, tea leaves and wild cherry bark, barks fruits, Amla, etc. they produce green colour with ferric chlorides. 3. Pseudotannins: These tannins are phenolic compounds of lower molecular weight and do not show the goldbeater's test. They are found in catechu and nux- vomica, etc. (i) Catechins: Catechu, cocoa, etc. (ii) Ipecacuanhic acid: Ipecacuanha (iii) Chlorogenic acid: Coffee and nux-vomica Properties of tannins: Tannins are freely soluble in water, alcohol, glycerol, and acetone and dilute alkalies. They are sparingly soluble in chloroform, ethyl acetate and other organic solvents. They have an astringent taste. They yield purple, violet or black precipitate with iron compounds. They are precipitated by number of metallic salts notably potassium dichromate, and lead acetate and sub acetate. They combine with skin and hide to form leather and with gelatin and isinglass to form an insoluble compound. They combine with alkaloids to form tannates, most of which are insoluble in water. Chemical tests of tannins 1. Gelatin test: To a solution of tannin, aqueous solution of gelatin and sodium chloride are added. A white buff coloured precipitate is formed 2. Goldbeater's skin test: A small piece of goldbeater skin is soaked in 20% hydrochloric acid, ringed with distilled water and placed in a solution of tannin for 5 minutes. The skin piece is washed with distilled water then transferred to a solution of ferrous sulphate. A brown or black colour is produced on the skin due presence of tannins. 3. Phenazone test: A mixture of aqueous extract of a drug and sodium acid phosphate is heated and cooled and filtered. A solution of phenazone is added to the filtrate. A bulky coloured precipitate is formed 4. Match stick test (Catechin test): A match stick is dipped in aqueous plant extract, dried near burner and moistened with concentrated hydrochloric acid. On warming near flame, the matchstick wood turns pink or red due to formation of phloroglucinol. 5. Chlorogenic acid test: An extract of chlorogenic acid containing drug is treated with aqueous ammonia. A green colour is formed on exposure to air. 6. Vanillin-hydrochloric acid test: Sample solution and added vanillin- hydrochloric acid reagent (Vanillin 1 gm, alcohol 10 ml, concentrated hydrochloric acid 10 ml). A pink or red colour is formed due to formation of phloroglucinol. Isolation of Tannins Both hydrolysable and condensed tannins are highly soluble in water and alcohol but insoluble in organic solvents such as solvent ether, chloroform, and benzene. Tannin compounds can be easily extracted by water or alcohol. The method for the extraction of tannic acid from various galls is either with water-saturated ether, or with mixture of water, alcohol, and ether. In such cases, free acids such as Gallic and ellagic acid go along with other tannin gets exracted in water. If the drug consists of chlorophyll or pigment, it may be removed by ether. After extraction, the aqueous and ethereal layers are separately concentrated, dried, and subjected to further isolation and purification using various separation techniques of chromatography Medicinal properties and uses Tannins occur in crude drugs either as major active constituent as in oak bark, or as a subsidiary component as in clove. In many cases, they synergistically increase the effectiveness of active principles. Tannins are medicinally significant due to their chelated properties. They promote rapid healing and the formation of new tissues on wounds and inflamed mucosa. Another major focus of the educational activities is to prepare graduate students and postdoctoral scholars for their future independent careers in science. Due to their astringent properties, Tannins are administered in cases of diarrhoea, intestinal catarrh and in cases of heavy metal poisoning as an antidote. Recently, these compounds have demonstrated their antiviral activities for treatment of viral diseases including AIDS. They are used as mordant in dyeing, manufacture of ink, sizing paper and silk, and for printing fabrics. They are used along with gelatin and albumin for manufacture of imitation horn and tortoise shell. They are widely used in the leather industry for conversion of hide into leather, the process being known as tanning. They are also used for clarifying beer or wine, in photography or as a coagulant in rubber manufacture. They are used for the manufacture of gallic acid and pyrogallol, and sometimes as a reagent in analytical chemistry.

Award Abstract # 1120323 Hydrolyzable Tannin Biosynthesis in Plants ABSTRACT Hydrolyzable tannins are a group of plant polyphenolic compounds that act as chemical defenses against microbes, insects and vertebrate herbivores. Hydrolyzable tannins play animpact soil nutrient dynamics and affect the germination, growth and development of other plant species. Besides imparting astringent taste sensations, hydrolyzable tannins also enhance the nutritional quality of plant foods due to their antioxidant, anticancer and cardioprotective activities. Despite these important functions that hydrolyzable tannins play in plants and humans, large and significant gaps remain in our understanding of the molecular and biochemical mechanisms underlying hydrolyzable tannin biosynthesis in plants. This project will investigate the key enzymatic steps leading to hydrolyzable tannin biosynthesis using strawberry and pomegranate as model systems. Cloning of hydrolyzable tannin biosynthetic genes will facilitate modification of plant composition for improved protein utilization in forage crops. Underpinning hydrolyzable tannin biogenesis will also broaden our understanding of plant defense mechanisms, as well as plant-soil and plant-plant (allelopathic) interactions. Broader Impacts In addition to the scientific impacts, various aspects of the research will be used to provide multidisciplinary training opportunities, encompassing molecular biology, biochemistry, and cell biology, to high school, undergraduate, and graduate students, and postdoctoral scholars. The educational activities at the undergraduate level will be focused on three areas: providing research opportunities to new UC Davis transfer students, promoting career access among women and underrepresented minority students, and retaining students from low-income families. Another major focus of the educational activities is to prepare graduate students and postdoctoral scholars for their future independent careers in science. An outreach program is currently being developed to expose high school students to research activities and biotechnological applications through internships in the PI's laboratory. PUBLICATIONS PRODUCED AS A RESULT OF THIS RESEARCH Note: When clicking on a Digital Object Identifier (DOI) number, you will be taken to an external site maintained by the publisher. Some full text articles may not yet be available without a charge during the embargo (administrative interval). Some links on this page may take you to non-federal websites. Their policies may differ from this site. Ono, N.N., Britton, M.T., Fass, J.N., Nicolet, C.M., Lin, D.W. and Tian, L. "Exploring the transcriptome landscape of pomegranate (Punica granatum) fruit peel for natural product biosynthetic gene and SSR marker discovery." Journal of Integrative Plant Biology , v.53 , 2011 , p.800 Ono, N.N. and Tian, L. "The multiplicity of hairy root cultures: prolific possibilities." Plant Science , v.180 , 2011 , p.439 Ono, N.N., Bandaranayake, P.G. and Tian, L. "Establishment of pomegranate (Punica granatum) hairy root cultures for genetic interrogation of the hydrolyzable tannin biosynthetic pathway." Planta , v.236 , 2012 , p.931-941 PROJECT OUTCOMES REPORT Disclaimer This Project Outcomes Report for the General Public is displayed verbatim as submitted by the Principal Investigator (PI) for this award. Any opinions, findings, and conclusions or recommendations expressed in this Report are those of the PI and do not necessarily reflect the views of the National Science Foundation; NSF has not approved or endorsed its content. Plants synthesize specialized chemicals that play important roles in ensuring optimal plant growth and effective defense against pathogens and predators; healthy plants can provide abundant food and medicine for human nutrition and health, as well as biomass for biofuel production. Our investigations centered on understanding how a group of specialized chemicals named hydrolyzable tannins are synthesized in plants. Although many beneficial activities have been reported for hydrolyzable tannins in animals and humans, there is a gap in our understanding of their production in plants, knowledge that is necessary for dissecting the mechanistic basis of their biological activities in mammals. Through our current research, we obtained genes involved in hydrolyzable tannin biosynthesis and demonstrated for the first time their functions in plants. Isolation of hydrolyzable tannin biosynthetic genes provides critical molecular tools for modification of hydrolyzable tannin composition and content in plants with the goal of breeding cancer suppression mechanisms of these specialized chemicals in humans or improving disease tolerance in forage crops for animal consumption. Elucidation of hydrolyzable tannin biosynthesis will also broaden our understanding of general plant defense mechanisms, as well as plant-soil and plant-plant interactions. Various aspects of this research were used to provide multidisciplinary training opportunities, encompassing molecular biology, biochemistry and cell biology, to high school, undergraduate, and graduate students and postdoctoral researchers. In addition, we focused our educational activities on promoting career access among women and underrepresented minority students, retaining students from low-income families and preparing graduate students and postdoctoral researchers for their future independent careers in science. The PI has also incorporated topics on phytonutrients and human health into her classroom curriculum and developed an outreach program that exposed high school students to research activities and biotechnological applications through internships in the PI's laboratory. Last Modified: 07/17/2015 Modified by: Li Tian Please report errors in award information to writing to: awardsearch@nsf.gov. The hydrolysable tannin family perhaps is the most fascinating group of tannins, since they show both structural simplicity and complexity within both monomeric and oligomeric forms Fig. 1. Examples of the structures of simple gallic acid derivatives (b–d) and gallotannin (e). Structurally, in most cases the most complex group of tannins are the hydrolysable tannins (HTs). HTs are first divided into three subclasses such as simple gallic acid derivatives, gallotannins (GTs) and ellagitannins (ETs). Simple gallic acid derivatives contain five or less galloyl groups (Fig. 1) that are most commonly esterified to either glucose (monogalloyl and pentagalloyl glucoses in Fig. 1b, d) or quinic acid (monogalloyl quinic acid in Fig. 1c). Gallic acid derivatives that contain six or more galloyl groups are defined as GTs and are further characterized by having one or more digalloyl groups (heptagalloyl glucose in Fig. 1e). Gallic acid derivatives and GTs are much rarer in plants than are ETs. The diverse ETs structures described to date (>500) may be divided into six subgroups: hexahydroxydiphenoyl (HHDP) esters (Fig. 2b), dehydro-HHDP esters (Fig. 2c) and their modifications (Fig. 2d), nonahydroxyterphenoyl (NHTP) esters (Fig. 2e), flavonoellagitannins (Fig. 2f), and oligomers with both varying degree of oligomerization and types of bonds between the monomers (Fig. 2g-h). Fig. 2. Examples of the structures of the ellagitannin subgroups. Condensed tannins and hydrolysable tannins are two types of tannins found in plants. Condensed tannins, also known as proanthocyanidins, are polymers of flavan-3-ols that are linked together. They are widely distributed in nature and can be found in different parts of plants, such as leaves, bark. These tannins are known for their ability to bind and precipitate proteins, which gives them astringent properties. They have been used for centuries in various industries, including leather tanning, wine production, and traditional medicine. Condensed Tannins Condensed tannins, also known as proanthocyanidins, are a type of tannin that are formed by the polymerization of flavan-3-ol monomers. They are commonly found in fruits, such as grapes, apples, and berries, as well as in the bark and wood of trees. Condensed tannins are characterized by their ability to form insoluble complexes with proteins, which makes them effective in preventing protein degradation and inhibiting microbial growth. One of the key attributes of condensed tannins is their antioxidant activity. They have been shown to scavenge free radicals and protect cells from oxidative damage. This property is believed to contribute to their health benefits, including their potential role in reducing the risk of cardiovascular diseases and certain types of cancer. Condensed tannins also have antimicrobial properties. They can inhibit the growth of various bacteria and fungi, making them useful in food preservation and as natural alternatives to synthetic antimicrobial agents. Additionally, condensed tannins have been found to have anti-inflammatory effects, which may be beneficial in the treatment of inflammatory diseases. Furthermore, condensed tannins have been shown to have positive effects on animal nutrition. They can improve the digestibility of proteins and carbohydrates, enhance the absorption of minerals, and reduce the risk of gastrointestinal disorders in livestock. These properties make condensed tannins valuable in animal feed formulations. However, condensed tannins can also have negative effects. In some cases, they can bind to proteins and reduce their bioavailability, leading to protein and nutrient deficiencies. Additionally, high levels of condensed tannins in certain plants can impart a bitter taste, which may limit their use in food and beverage applications. Hydrolysable Tannins Hydrolysable tannins, also known as ellagitannins, are another type of tannin that are formed by the esterification of gallic acid or ellagic acid with a sugar molecule. They are commonly found in plants such as oak, chestnut, and pomegranate. Hydrolysable tannins are characterized by their ability to undergo hydrolysis, which releases gallic acid or ellagic acid as a byproduct. One of the key attributes of hydrolysable tannins is their antioxidant activity. They have been shown to scavenge free radicals and inhibit oxidative stress, which is implicated in various diseases, including cancer, cardiovascular diseases, and neurodegenerative disorders. Hydrolysable tannins also have anti-inflammatory properties, which may contribute to their health benefits. Hydrolysable tannins have also been shown to inhibit the growth of cancer cells, induce apoptosis (programmed cell death), and inhibit angiogenesis (the formation of new blood vessels). These effects are believed to be mediated by their antioxidant and anti-inflammatory activities, as well as their ability to modulate signaling pathways involved in cell proliferation and survival. Furthermore, hydrolysable tannins have been found to have antimicrobial properties. They can inhibit the growth of various bacteria and fungi, making them useful in the treatment of infections. Hydrolysable tannins also have antiviral activity and have been shown to inhibit the replication of certain viruses. However, hydrolysable tannins can also have negative effects. In some cases, they can form complexes with dietary proteins and reduce their digestibility, leading to potential nutrient deficiencies. Additionally, high levels of hydrolysable tannins in certain plants can impart a bitter taste, which may limit their use in food and beverage applications. Conclusion Condensed tannins and hydrolysable tannins are two distinct types of tannins with different chemical structures and properties. While both types of tannins exhibit antioxidant, antimicrobial, and anti-inflammatory activities, they also have some unique attributes. Condensed tannins are known for their ability to form insoluble complexes with proteins, which makes them effective in preventing protein degradation and inhibiting microbial growth. Hydrolysable tannins, on the other hand, are characterized by their ability to undergo hydrolysis and release gallic acid or ellagic acid as a byproduct. Both condensed tannins and hydrolysable tannins have been extensively studied for their potential health benefits, including their role in reducing the risk of various diseases. However, they can also have negative effects, such as reducing the bioavailability of proteins and imparting a bitter taste. Further research is needed to fully understand the mechanisms of action and potential applications of these tannins in various fields, including medicine, food science, and agriculture. Comparisons may contain inaccurate information about people, places, or facts. Please report any issues. Tannins are non-nitrogenous organic compounds with astringent properties. They are a large group of compounds with a wider distribution in plants. Tannins are non-crystalline, oligomeric compounds with free phenolic groups in multiple units. They form a colloidal solution in water. They are soluble in water, alkali, alcohol, and glycerin. Tannins are partially soluble in ethyl acetate. They are soluble in acetate but insoluble in other organic solvents Tannins can combine with proteins that have inhibitory effects on enzymes. They function as protection to heartwood and bark. They are classified into two groups, based on the tanning test or Goldbeater's test. Tannins can combine with animal hides and are detected by their ability to absorb standard hide powder. This process is called tanning and this tanning test is known as Goldbeater's skin test. Tannins that show positive tanning are called true tannins and those who fall the test are called pseudotannins. High molecular weight compounds such as polyphenols are true tannins. These true tannins are classified based on its hydrolytic reaction and the phenolic nuclei in its structure. These are hydrolysable tannins and condensed tannins respectively. As the name suggests, hydrolysable tannins are hydrolyzes by enzymes such as tannase or various acids. They are also known as pyrogallol tannins, as the phenolic acids are converted into pyrogallol derivatives on dry distillation. They are water-soluble, and the solution turns blue with ferric chloride. Hydrolysable tannins mainly contain polyphenolic acids like gallic acid, ellagic acid, etc., which bond through ester linkages to a glucose molecule. They can be further classified based on the phenolic acids formed after hydrolysis. Gallotannins that contain gallic acid Ellagitannins contain hexahydrodphenic acid which later produces ellagic acid. These are condensed tannins that are non-hydrolysable by mineral acids or enzymes. They are also called proanthocyanidins. Compounds with condensed tannins containing phenolic nuclei are related to flavonoids. They can form more complex tannins with proteins and carbohydrates. On treatment with enzymes or mineral acids, these non-hydrolysable tannins produce insoluble red-colored phlobaphenes due to the polymerization reaction. These are characteristics of drugs like cinchona, cherry bark, etc. Condensed tannins are soluble in water and the solution turns green colored with ferric chloride. Catechin is flavan-3-ol and leucoanthocyanidin is flavan-3,4-diol. Non-hydrolysable tannins are commonly seen in plants of families Rosaceae, Leguminosae, Rubiaeae, Geraniaceae, Combretaceae, Polygonaceae, etc. Pseudo tannins are simple, phenolic tannins having low molecular weight. They do not respond to Goldbeater's skin test. They are found in dead tissues and dying cells. Eg. Chlorogenic acid, gallic acids, catechin, etc. Tannins are extracted using water or alcohol since they are insoluble in all other organic solvents such as benzene, ether, or chloroform. Water-saturated ether is used to extract tannic acid from all galls. A mixture of water, ether, and alcohol may also be used. When the mixture of all three solvents is used, free acids like ellagic and gallic acids go along with other while the true tannins go with water. Ether removes chlorophyll and other pigments. After extraction, the ethereal and aqueous layers are separated, concentrated, dried, and subjected to further purification by various methods. Isolation of tannins from plants can be done using different methods. Solvent extraction uses an appropriate solvent and the dried plant powder is mixed in. The solution is allowed to sit for a while and the tannins are extracted through osmosis. Decoction method uses a heat source to extract tannins from the solvent. The heat is usually 100°C and then it is allowed to cool down before filtering it. Pressurized water extraction uses static or dynamic water pressure to extract tannins from the solvent. Ultrasound method uses sound waves to destroy plant structures and yield tannins. Microwave-assisted extraction as the name suggests uses a microwave as the heat source. Ionic liquid-based microwave-assisted extraction uses ionic solution to soak the material and is heated in a microwave. Infrared-assisted extraction is a macerate process where the heat source is the infrared light and there is no solvent used here. Gamma-assisted extraction subjects the solvent-dissolved tannins to Co-60 gamma radiation Enzyme-assisted extraction uses cellulase enzymes in sea plants that have higher stability. Reference [6] Further solid extraction is an eco-friendly extraction method used for hydrolysable tannins Soxhlet extraction method is not widely used due to higher cost. Shah, Biren N, Avinash Seth. (2010). Textbook of Pharmacognosy and Phytochemistry. Elsevier . Tannins: Extraction from Plants. DOI: 10.5772/intechopen.86040 As a library, NLM provides access to scientific literature. Inclusion in an NLM database does not imply endorsement of, or agreement with, the contents by NLM or the National Institutes of Health. Learn more: PMC Disclaimer | PMC Copyright Notice . 2016 Oct 21;174(11):1244–1262. doi: 10.1111/bph.13630 Tannins are a heterogeneous group of high MW, water-soluble, polyphenolic compounds, naturally present in cereals, leguminous seeds and, predominantly, in many fruits and vegetables, where they provide protection against a wide range of biotic and abiotic stressors. Tannins have been extensively studied for their antioxidant, radical scavenging activity as well as antimicrobial, anti-cancer, anti-nutritional and cardio-protective properties. They also seem to exert beneficial effects on metabolic disorders and prevent the onset of several oxidative stress-related diseases. Although the bioavailability and pharmacokinetic data for these bioactive compounds are, at present, far from clear, absorption of these compounds across the intestinal barrier is inversely correlated with the degree of polymerization. Further studies are mandatory to better clarify how these molecules and their metabolites are able to cross the intestinal barrier and exert their biological properties. This review summarizes the current literature on tannins, focusing on the main, recently proposed mechanisms of action that underlie their pharmacological and disease-prevention properties, as well as their bioavailability, safety and toxicology. This article is part of a themed section on Principles of Pharmacological Research of Nutraceuticals. To view the other articles in this section visit GSE grape seed extract HTs hydrolysable tannins NOAEL no-observed-adverse effect level OPCs oligomeric proanthocyanidins Tannins are a heterogeneous group of water-soluble polyphenolic compounds of high molecular weight (500–3000 Daltons) – with as many as 20 hydroxyl groups – and more than 1 or 2 carboxyl groups per 100 relative molecular mass units (Khanbabaee and van Ree, 2001) naturally present in plants, foods and beverages (de Jesus et al., 2012). Being phenolic compounds, tannins are chemically reactive and form inter- and intra-molecular hydrogen bonds that are able to interact with, and precipitate macromolecules, such as proteins and carbohydrates. They are also responsible for the astringent taste of many fruits and vegetables (de Jesus et al., 2012; Lamy et al., 2016). With one to five hydroxyl groups, astringency is increased, while from seven groups upwards, it decreases, given that steric hindrance starts to counterbalance the strength of the hydrogen bonds (Zou et al., 2015). Tannins can be classified into two groups: hydrolysable tannins and condensed tannins (also named catechin tannins or proanthocyanidins). Hydrolysable tannins can be further divided into gallotannins, which provide sugar and gallic acid on hydrolysis, and ellagitannins, which on hydrolysis do not yield just sugar and gallic acid but also ellagic acid (see Lamy et al., 2016). These compounds, as the name suggests, are hydrolyzed by weak acids and decomposed by high temperatures to yield pyrogallol, a hepatotoxic and highly irritant compound (Jiménez et al., 2014). The second class, condensed tannins, also referred to as proanthocyanidins, is the most abundant plant-derived polyphenols. They are oligomers of flavan-3-ol (catechin monomers) and/or flavan-3,4-diol, usually linked by C-C (4-8 or 6-8) and occasionally by C-O-C bonds with a wide structural diversity (de Jesus et al., 2012; Lamy et al., 2016) and are also called oligomeric proanthocyanidins (OPCs). These compounds are not readily hydrolyzed; they decompose in acidic alcoholic conditions giving red pigments called phlobaphenes. To date, though, the chemistry of proanthocyanidins is only partly known. Despite their abundance in our diet (estimated daily intake 0.1–0.5 g), tannins have received little structural complexity (Serrano et al., 2009). In recent years, considerable attention has been paid to proanthocyanidins and their monomers due to the potential beneficial effects on human health, including immunomodulatory, anti-inflammatory, anticancer, antioxidant, cardio-protective and antithrombotic properties (Nile and Park, 2014; Smeriglio et al., 2016a; Sieniawska, 2015). However, to assess their precise role in human health, it is essential to gather further knowledge on their pharmacological and toxicological behaviour. In fact, although many plant-derived proanthocyanidins are widely used now as nutritional supplements, evidence of their safety and potential for long-term toxicity is still lacking (Berry et al., 2016). Here we have reviewed the latest developments and knowledge on the occurrence, dietary intake and biological effects of tannins, with particular attention on the pharmacological and toxicological aspects. Tannins (proanthocyanidins and hydrolysable tannins) are one of the main secondary metabolites of these specialized chemicals (Vercruysse et al., 2009). In recent years, considerable attention has been paid to proanthocyanidins and their monomers due to the properties, solubility and presence of substituents (Okuda and Ito, 2011). Taking into account, for instance, the structural features of hydrolysable tannins, they can be identified as follows: gallotannins, ellagitannins and complex tannins. Main tannin families and related food sources Tannins (mg·100 g−1 or mg·100 mL−1) Species Proanthocyanidins Ellagitannins Gallotannins Fruits Cranberries Vaccinium oxycoccus L. 194–496 — — Chokeberries Aronia melanocarpa (Michx.) Elliott 553-2106 — — Plums Prunus domestica L. 32–334 — — Black diamond Prunus spp 210–267 — — Blueberries Vaccinium myrtillus L. 87–274 — — Vaccinium corymbosum L. 311–335 — — Black currants Ribes nigrum L. 105–255 3–6 — Red currants Ribes rubrum L. 30-61 — Blackberries Rubus fructicosus L. 5–46 150–270 — Crowberries Empetrum nigrum L. 153–173 — — Lingonberries Vaccinium vitis-idaea L. 175–545 — — Red Grapes Vitis Labrusca L. 8–75 — — Grape seeds Vitis Vinifera L. 2180-6050 — — Strawberries Fragaria × ananassa Duchesne 15–181 71–81 — Peaches Prunus persica L. 29–110 — — Apricot Prunus armeniaca L. 8–73 — — Raspberries Rubus occidentalis L. 3–74 160–326 — Pears Pyrus communis L. 5–81 — Apple Malus domestica Borkh. 46–278 — — Pomegranate Punica granatum L. — 58–177 — Guava Psidium spp. — 20–25 — Mango Mangifera indica L. — 30-160 Juices Cranberry – 20–21 — — Apple – 52–69 — — Grape – 45–47 — — Nuts Almonds Prunus dulcis (Mill.) D.A.Webb 67–257 — 20–34 Hazelnuts Corylus spp. 125-645 — — Pecans Carya illinoinensis (Wangenh.) K.Koch 238–695 11–33 — Pistachio nuts Pistacia vera L. 113-271 — — Walnuts Juglans regia L. 35–87 36–59 — Legumes Beans Phaseolus vulgaris L. 5–830 — — Carob fibre Ceratonia siliqua L. 1–19 — — Cowpeas Vigna unguiculata (L.) Walp. 17–319 — — Lentils Lens culinaris Medik. 1–2 — — Peanuts Arachis hypogaea L. 121–141 — — Cereal grains Barley Hordeum vulgare L. 59–153 — — Buckwheat Fagopyrum esculentum Moench 4-46 — — Sorghum Sorghum bicolor (L.) Moench 413-5333 — — Rice Oryza sativa L. 9–91 — — Beverages Wine – 1–53 2–5 — Tea – 1–5 — — Cacao beans Theobroma cacao L. 6100-8100 — — Chocolate – 828–1332 — — Proanthocyanidins are oligomers or polymers of flavan-3-ols, where the monomeric units are linked mainly by C-4 C-8 bonds, although less frequently also C-4 C-6 linkages can be found. These types of linkages lead to the formation of the so-called B-type proanthocyanidins. A-type proanthocyanidins, on the other hand, are characterized by an additional bond between C-2 C-7 of the basic flavan-3-ol subunits, known as proanthocyanidin monomers or catechins. The most common monomeric units are the diastereomers of (epi)catechin, (epi)afzelechin and (epi)gallocatechin. In this class of flavonoids, two aromatic homocyclic rings (A and B) and a heterocyclic ring (C) are present, and they are joined by a three-atom carbon bridge. Their C ring features a hydroxyl group at the C-3 position, and it lacks a double bond between C-2 and C-3. The most abundant proanthocyanidins in vegetables are those composed of catechin and epicatechin units (procyanidins), while much less common are propelargonidin or prodelphinidin containing (epi)afzelechin or (epi)gallocatechin basic units (Figure 1), present in foods such as barley, broad beans, red kidney beans, redcurrants, pinto beans, black tea and cinnamon (Landete, 2011; de Jesus et al., 2012; Mateos-Martín et al., 2012; Lamy et al., 2016). Procyanidins have been identified, in natural sources, as dimers (e.g. procyanidin B1-B8, procyanidin A1-A2), trimers (e.g. procyanidin C1 and C2, sellingerain A and B), tetramers and oligomeric structures with a degree of polymerization ranging from 5 to 11. The most common acyl substituent (as in the case of tea- or wine-derived species) is a galloyl group bound to the hydroxyl group in the C-3 position. The carbohydrate moieties are usually linked to the C-3 or the C-5 position in glycosylated proanthocyanidins. Basic chemical features of B- and A-type proanthocyanidins and their monomeric units. Gallotannins represent the simplest class of hydrolysable tannins (HTs), containing gallic acid substituents esterified with a polyol residue (mainly D-glucose). The β anomer of glucose is the dominant configuration, although rare examples of the α configuration can be found in natural derivatives. The glucose hydroxyl groups can be partly or totally substituted by phenolic products in order to produce initially β-glucogallin (1-O-galloyl-β-D-glucopyranose). The biosynthetic pathway, starting from this molecule and after the galloylation reaction, yields di-, tri-, tetra, penta-, hexa-, hepta- and octagalloylglucoses. Of these, 1,2,3,4,6-penta-O-galloyl-β-D-glucopyranose is usually employed as the prototypical molecule of gallotannins (Figure 2). The formation of meta- or para-depside bonds is characteristic of these molecules and involves esterification of the aromatic hydroxyl group rather than those present in aliphatic structures. However, many other derivatives can be found, for example gallotannins with 10 or more (up to 12) units of gallic acid esterified to a single glucose moiety, as in the case of tannins obtained from sumac (Rhus semialata) or oak galls (Quercus infectoria), utilized since ancient times to tan animal hides to produce leather. Although glucose is by far the most abundant polyol identified in tannins, glucitol, fructose, shikimic acid, xylose, hamamelose, saccharose, quercitol and quinic acid may constitute the core molecules for the subsequent galloylation process. However, these derivatives are not very common and have only been isolated in maple, chestnut and witch hazel. Chemical structure of pentagalloyl-glucose. Different from gallotannins (rarely found in nature), ellagitannins are present in many plant families, and almost 500 molecules have been isolated and identified to date (Okuda and Ito, 2011; Lamy et al., 2016). Although not all ellagitannins are hydrolyzable, they continue to be classified as hydrolyzable tannins for historical reasons. They can be found in numerous forms: monomeric (e.g. nupharin A, punicalagin, geraniin, eugeniin, davidiin, casuarictin and corilagin), dimeric (e.g. sanguiin), oligomeric (e.g. agrimonin, nupharin E, nupharin C and hirtellin A) and C-glycosidic (e.g. vescalagin, castalagin, casuarinin and stachyurin) molecules. Ellagitannins are formed from gallotannins, where at least two galloyl units, bound to adjacent solution spontaneously lactonizes into ellagic acid (Figure 3). Ellagitannins and ellagic acid have also been identified in both fruits as well as in nuts and seeds (Landete, 2011; see Table 1. The oxidative coupling commonly involves C-4 and C-6 (as in the case of eugeniin) and may extend to C-2 and C-3, leading to the formation of casuarictin (Figure 4). Other carbon atoms involved in this reaction are the C-3/C-6, C-2/C-4 or C-1/C-6 pairs. Hexahydroxydiphenoyl acid can undergo esterification not only with glucose but also with hamamelose and terpenoids. Ellagitannins can yield dimers following intermolecular oxidative coupling with other hydroxylable tannins (as in the case of euphorbins) or high MW oligomers. Formation of ellagic acid in aqueous solution. Chemical structure of eugeniin and casuarictin. The reaction of gallotannins or ellagitannins with catechin units yields complex tannins. Flavan-3-ols can link at C position of an open-chain glucose. Typical examples of complex tannins are camelliatannins A and B, malabatrin A and acutissimin A (Okuda and Ito, 2011). Tannins, in particular proanthocyanidins, are one of the most widespread compounds present in food ingested daily. They are commonly found in plants and have been identified in almost all their parts (fruits, leaves, roots, seeds, wood and bark). More than 1000 derivatives have been identified to date, and these compounds continue to be of interest to researchers due to the wide range of applications and the numerous biological processes influenced by their presence. These compounds are found in tea, wine, cereals, cacao and fruits in amounts of up to 184 mg·100 g−1, with epicatechin being the most abundant component, followed by catechin and B-type p-procyanidin (de Jesus et al., 2012; Lamy et al., 2016). They are also the principal compounds found in ripe fresh nectarines, peaches and plums (Table 1). Recently, the presence of epicatechin and B-type dimers and trimers of proanthocyanidins has been detected in the pulp of Annona cherimola Mill (Barreca et al., 2011). In red wine, the content of catechin, epicatechin, procyanidin B1, B2, B3 and B4 amounts to ~18 mg·100 g−1, more than double that found in white wine (Sanchez-Moreno et al., 2003). Catechin and procyanidin B2 were the two principal components found in cereals and fruits, as well as in tea and red wine. A complete list of proanthocyanidin content in foodstuffs can be found on the USDA database of Proanthocyanidin Content of Selected Foods (http:// www.nal.usda.gov/fnic/foodcomp/Data/PA/PA.html). Gallotannins are present only in selected woody and herbaceous dicotyledons. Cereals are sources of hydrolyzable phenols due to phenolic compounds being linked to cell wall components. β-Glucogallin has been reported in oats, while the main components are D-glucose derivatives esterified with gallic acid, such as penta-, hexa-, hepta-, octa-, nona-, deca-, undeca- and dodeca-galloyl-D-glucose (Lu et al., 2013). Ellagitannins are present in only a few fruits and nuts (especially pecans and walnuts) consumed in the Western diet, and dietary intake (mostly probably far below 5 mg·day−1) is mainly resulting from the consumption of red fruits (strawberries, raspberries and blackberries or beverages and jams made from these) (Okuda and Ito, 2011; see Table 1. The principal derivatives identified in foods so far are as follows: sanguiin H-6, punicalagin, sanguiin H-5, vescalagin, castalagin, roburin E, casuarictin, pedunculagin, potentillin and lambertianin C. Estimates of the amount of ellagitannins present in food are based on detection of ellagic acid (after hydrolysis of these compounds) followed by HPLC detection, although care is needed during quantification to avoid underestimating their solubility issues. Sanguiin H-6, casuarictin, potentillin and pedunculagin are the predominantly ellagitannins identified in strawberries, raspberries and blackberries (Zorenc et al., 2016). These compounds are mainly contained in the peel and seed, while very low quantities of traces have been detected in the pulp. Pomegranates (fruits, juices and jams) are also a rich source of ellagitannins, mainly punicalagin and ellagic acid (Masci et al., 2016). Ellagitannins have been found in significant amount (Table 1) also in walnuts and pecan nuts (Landete, 2011), with pedunculagin being the main derivative present in walnuts. Aside from the pharmacological effects exerted locally in the gut, tannins have to be bioavailable in order to exert their health effects, and therefore, absorption, bioavailability and metabolism of these compounds must be carefully evaluated. Although the pharmacokinetics of simple phenols have been extensively investigated in animal and human studies (Velderrain-Rodríguez et al., 2014), the same cannot be said for tannins, where results are still scarce and controversial. High MW tannins apparently are not absorbed intact, as demonstrated by Wiese et al. (2015), who reported that almost 90% of apple juice-derived procyanidins consumed were retrieved in ileostomy effluent and were able to enter the colon (Wiese et al., 2015). The polymerization rate appears to play a major role in the fate of these compounds and seems to be bioavailable only for the absorption of these molecules through the gut barrier and to their metabolism by gut microbiota to the monomers (i.e. catechin and epicatechin) (Zhang et al., 2016). Some in vivo and in vitro studies have investigated the hydrolysis mechanism of polymeric proanthocyanidins and whether it is possible for the derived oligomeric molecules to be absorbed by the small intestine; often the molecules that appear in biological fluids can be very different from those ingested (Zhang et al., 2016). Even though small quantities of proanthocyanidins monomers and dimers were found in the plasma compartment, methylated and glucuronidated derivatives seem to be the main metabolites. Furthermore, in the colon, the proanthocyanidins are catabolized by the gut microflora into a series of simpler metabolites such as phenyl valerolactone, phenylacetic and phenylpropionic acids (Zhang et al., 2016). However, these results are still controversial. Some in vitro experiments have suggested, for example that cocoa procyanidins are hydrolyzed into a mixture of epicatechin monomers and dimers under conditions similar to those present in the human stomach. Nevertheless, other studies have demonstrated that no significant depolymerization of cocoa procyanidins occurs in the stomach, thus suggesting a plausible further mechanism provided by the food bolus against the acid stomach environment (Ellam and Williamson, 2013; Kim et al., 2014). However, despite these conflicting observations, what is certain is that during digestion, high MW proanthocyanidins can form complexes with proteins, starches and digestive enzymes, resulting in the formation of less digestible complexes and this property is indirectly related to the degree of polymerization. Indeed, small molecules such as dimeric and trimeric proanthocyanidins can be more readily absorbed (Zhang et al., 2016).Absorption of OPCs has been reviewed recently by Ou and Gu, 2014. They reported that procyanidin A-type dimers, trimers and tetramers were slowly transported across Caco-2 cells monolayers (transport ratio of 0.6%, 0.4% and 0.2% respectively), suggesting their possible absorption after dietary intake. Furthermore, they reported similar permeability coefficients for (+)-catechin and a proanthocyanidin dimer and trimer, while the permeability of proanthocyanidins with high polymerization was found to be approximately 10 times lower. As described above, most ingested tannins reach the colon. At this level, the gut microbiota play a continue to be of interest to researchers due to the wide range of applications and the numerous biological processes influenced by their presence. These compounds are found in tea, wine, chocolate, legumes and cereal grains (Table 1). There are many ways to define tannins based on their properties, solubility and presence of substituents (Okuda and Ito, 2011). The low concentration of 14C-labelled metabolites detected indicates controversial results (Deprez et al., 2000). However, these underestimates may be due to the ability of some polyphenols to bind strongly to various molecules in gut cells and dietary fibres in the colonic lumen. There is currently no data regarding the intestinal bacteria responsible for the degradation of proanthocyanidins and their metabolites. Hydrolysable tannins are metabolized to gallic acid, pyrogallol, phloroglucinol and finally to acetate and butyrate via several bacterial enzymes (Marín et al., 2015). Despite some evidence to the contrary, the OPCs appear to be hydrolyzed into epicatechin in rat isolated small intestine (Zhang et al., 2016). Nevertheless, orally administered procyanidins in rat plasma and trace amounts of procyanidin dimers and trimers in urine (Li et al., 2013) were detected. These results suggest that proanthocyanidin trimers and dimers can be absorbed in vivo, also due to the similarity of the permeability coefficient to that of mannitol, and limited polymer bioavailability in the gut lumen (Ou and Gu, 2014). Moreover, 16 metabolites, including phenolic, phenylpropionic and phenylbutyric acids, have been detected in rat urine after administration of proanthocyanidin isolated from willow tree catechins. The total yields significantly decreased in relation to the polymerization degree of the precursors: catechin monomer > dimer > trimer > polymer (Brenes et al., 2016). Analysis of luminal content, after administration of grape seed extract (GSE) to Sprague-Dawley rats, showed distinct native and metabolite profiles for each region (caecum and proximal, mid and distal colon) allowing insight to be gained into the distribution and delivery of procyanidins and their microbial metabolites throughout the colon. In particular, procyanidins reached maximum concentrations approximately 3 h postgavage appearing earlier in the more proximal segments, while metabolites reached maximum concentrations from 3 to 18 h post-gavage appearing later in more distal regions (Goodrich et al., 2015). Recently, the systemic absorption and metabolism of dietary procyanidin B4, after oral administration, were evaluated in pigs, demonstrating that this compound is absorbed as an intact molecule and in part excreted in urine. In addition, it was degraded to the monomeric subunits catechin and epicatechin that were then further metabolized to methylated and glucuronidated conjugates (Bittner et al., 2014). Unlike condensed tannins, few studies on the bioavailability of hydrolysable tannins are currently available. However, the absorption of ellagic acid has been extensively documented. Gallic acid and its metabolites after oral administration of pomegranate juice detected low concentrations of ellagic acid in plasma after 0.5–3 h, while no trace of ellagitannins were detected (Nuñez-Sánchez et al., 2014). The low plasma concentration of ellagic acid is probably due to its lower solubility and to its ability to complex calcium and magnesium ions in the intestine and thus compromise gut absorption (Serrano et al., 2009). The main ellagic acid derivatives have been found in human plasma and urine (Nuñez-Sánchez et al., 2014). The intestinal absorption of gallic acid in humans is relatively slow (tmax, 87 min), as also found in rats (tmax, 60 min) (Serrano et al., 2009). Two different speeds of gastric absorption have been suggested for this simple phenol: rapid permeation for the unchanged molecule and slow permeation for the metabolites. The main metabolite of gallic acid observed in humans is 4-O-methylgallic acid (Serrano et al., 2009). A plausible mechanism for ellagitannin degradation by human microbiota, via hydrolysis into ellagic acid and its microbial transformation into urolithin B, has been proposed. A study performed on human volunteers fed with a single dose of ellagitannin-rich dietary sources demonstrated a large inter-individual variation in the metabolite production (Kamiloglu et al., 2016). These data correlate with some in vitro antioxidant properties in tannins (Brenes et al., 2016). It may be concluded that absorption through the gut barrier is probably limited to the absorption of tannins with a low degree of polymerization and of those metabolites formed in the colon. In view of this, it is critical to evaluate bioavailability studies in order to gain a better understanding of the effects of these phytochemicals on human health. In addition, the above findings highlight the necessity to choose appropriate biomarkers for the in depth study of the pharmacokinetics of these molecules. For some polyphenols, this has already been achieved (Espín et al., 2013; Stalmach et al., 2014). However, there is a great deal still to be done in this area. The biological properties and health effects of tannins have been extensively reviewed (Coppo and Marchese, 2014; Katiyar, 2015; Salvadó et al., 2015; Verstraeten et al., 2015). Many of the in vitro and in vivo (human and animal) studies performed suggest that tannin intake may prevent the onset of several chronic diseases, as reported by recent reviews, systematic reviews and meta-analysis (Holt et al., 2012; Blumberg et al., 2013; Wang et al., 2014; Salvadó et al., 2015; Turati et al., 2015). The recognized pharmacological activities have been ascribed to dietary monomeric flavan-3-ol derivatives and to various crude and purified tannins or proanthocyanidin fractions from the whole plant, fruits, leaves, rhizome and bark of various medicinal plants (Table 2). Tannins exert their biological effects not only as an un-absorbable (high MW) complex structure with binding properties that may produce local effects in the gastrointestinal tract but also as absorbable tannins (dimers and trimers) with their metabolites producing systemic effects (Jiménez et al., 2014; Nuñez-Sánchez et al., 2014; Ou and Gu, 2014; Velderrain-Rodríguez et al., 2014; Sieniawska, 2015). In addition to their well-documented free radical scavenging Scavengers of hydroxyl, superoxide, and peroxyl radicals (Proanthocyanidins); Stronger antioxidant activity than ascorbic acid or α-tocopherol (Procyanidins B1 and B2); Dose-dependent radical scavenging action of galloylated condensed tannins. Nile and Park, 2014 Skrovankova et al., 2015 Kancheva and Kasaikina, 2013 Sieniawska, 2015 Anti-cancer Apoptosis p53-dependent and Bax/Bcl-2 proteins and caspase-3 activation mediated in JB6 C141 cells (Grape seed proanthocyanidins); Reduction of UV-induced oxidative stress-mediated phosphorylation and activation of NF-κB in NHEK cells (Grape seed proanthocyanidins); Inhibition of cellular proliferation and expression of MMP-2 and −9 in DU145 and LNCaP cells (Grape seed proanthocyanidins); Dose-dependent inhibition of cell viability, proliferation and apoptosis in 4 T1, HT29 and LoVo cells (Grape seed proanthocyanidins); G0/G1 phase cell cycle arrest with increase in Cip1/p21 protein levels and decrease of cyclins and cyclin-dependent kinases in MCF-7 and A-427 cells (Grape seed proanthocyanidins); Inhibition of KB, CAL-27, MCF-7, HT-29, HCT116, DU145 and LNCaP cells growth and apoptosis induction (blackberry, black raspberry, blueberry, cranberry, red raspberry and strawberry proanthocyanidins); Decrease in UVB-induced immune suppression in mice with induction of IL-12 (Grape seed proanthocyanidins); Apoptosis induction and metastasis inhibition of 4 T1 murine mammary cancer cells (Grape seed proanthocyanidins); Inhibition of induced mammary tumors in Sprague-Dawley rats (Grape seed proanthocyanidins); Inhibition of prostate tumour growth and progression in TRAMP mice (Grape seed proanthocyanidins). Gollucke et al., 2013 Nile and Park, 2014 Skrovankova et al., 2015 Ouédraogo et al., 2011 Sieniawska, 2015 Turati et al., 2015 Antimicrobial Inhibition of E. coli adherence to uroepithelium (Cranberry proanthocyanidins); Activity against Staphylococcus aureus, Candida albicans and Campylobacter jejuni (Ellagitannins); Antiherpetic activity of cytophatic effects of HIV (hydrolyzable tannins); Inhibition of cytopathic effects of HIV (hydrolyzable tannins); Activity against Helicobacter pylori (hydrolyzable tannins). Marín et al., 2015 Nile and Park, 2014 Sieniawska, 2015 de Jesus, 2012 Anti-nutrient Inhibition of gastrointestinal enzymes (Proanthocyanidins); Reducing capacity on Cr, Fe and Cu with alteration of their absorption (Proanthocyanidins); Modulation of chronic inflammatory bowel diseases (Procyanidin B3). Sieniawska, 2015 Li et al., 2012 Dias et al., 2016 Cardioprotective Hypocholesterolemic effects (Proanthocyanidins); Preventing myocardial ischaemic injury in adult rats (Proanthocyanidins); Cardioprotective effect due to inhibition of differentiation of monocyte to macrophages (Oligomeric proanthocyanidins isolated from Crataegus oxyacantha L.); Protective effect on doxorubicin-induced cardiac toxicity in rats (Grape seed proanthocyanidins); Suppression of doxorubicin-induced electrocardiographic and biochemical changes (Grape-wine proanthocyanidins); Protection against acute ischaemic brain damage in rats (Proanthocyanidins); Vascular protective effects (Procyanidin B2); Improved serum antioxidant status and decreased serum C-reactive protein and plasma homocysteine concentrations (Proanthocyanidins); Decreased systolic and diastolic blood pressure (Grape-wine proanthocyanidins). Blumberg et al., 2013 Wang et al., 2014 Holt et al., 2012 Guler et al., 2011 Mohana et al., 2015 Ammar et al., 2013 Yunoki et al., 2014 Yang et al., 2013 Anti-diabetic and anti-obesity Inhibition of salivary and pancreatic α-amylase and α-glucosidase activity (Hydrolyzable tannins and proanthocyanidins); Stimulation of glucose uptake and glycogen and fatty acid synthesis in muscle and adipocytes (Proanthocyanidins); Reduced plasmatic insulin and improved homeostatic model assessment (HOMA) index (hydrolyzable tannins and proanthocyanidins); Modulation of the active glucagon-like peptide-1 (GLP-1) levels (hydrolyzable tannins and proanthocyanidins); Insulin anabolic-like and lipostabilizer properties (Grape seed proanthocyanidins); Insulin anabolic-like and lipostabilizer properties (Grape seed proanthocyanidins); Inhibition of diabetes-induced cataract formation in rats (Cacao proanthocyanidins); Increased antioxidant enzyme activity and decreased induction of metal chelation activity, reduced resistin formation, and inhibition or activation of transcriptional factors such as NF-kB and PPARγ (Proanthocyanidins). Gonzalez-Abuin et al., 2015 Banihani et al., 2013 Pinent et al., 2012 Gato et al., 2013 Mathew et al., 2012 Kooti et al., 2016 Cascaes et al., 2015 Cook, 2015 Stohs and Ray, 2015 Salvadó et al., 2015 Most activities of the proanthocyanidins and hydrolysable tannins, including antioxidant and free radical-scavenging capacity, largely depend on their chemistry; for example, an increase in anti-diabetic activity was observed with an increase in the degree of polymerization (Sieniawska, 2015). Proanthocyanidins are known to inhibit lipid peroxidation and lipoxygenases in vitro, and several studies have demonstrated their ability to scavenge hydroxyl, superoxide and peroxyl radicals, helping to restore the oxidative balance of the body (Georgiev et al., 2014). In some instances, some proanthocyanidins B1 and procyanidin B3 have been recognized to be stronger antioxidants than ascorbic acid or α-tocopherol (Iglesias et al., 2012). These results observed in vitro have also been confirmed by several in vivo studies on animal models (Kancheva and Kasaikina, 2013; Nile and Park, 2014; Sieniawska, 2015). Fushimi et al. (2015) have investigated the antioxidant effects of unripe and mature persimmon in rats. Results showed that plasma phosphatidylcholine hydroperoxide levels, a biomarker of membrane lipid peroxidation, were significantly lower in the unripe persimmon group than in the control group, while no significant difference was observed between the mature persimmon group and the control group. The authors suggested that soluble tannins, found more in unripe fruits, could contribute to the inhibition of the antioxidant effect observed (Fushimi et al., 2015). A few years earlier, another study had shown that several proanthocyanidins from peanut skin and persimmon peel, possess high dose-dependent antioxidant activity. B-type dimers showed the highest radical scavenging activity in aqueous systems while A and B-type dimers showed similar antioxidant potency in tissue or lipid systems highlighting the importance of the food matrix effect (Dong et al., 2013). Protection against lipid peroxidation has also been observed in several tannin-rich plants. For example, the ethanolic extract of Nigella sativa L. was found to counteract in vivo Fe(II)-induced lipid peroxidation (Hassan et al., 2016). Also, Centella asiatica L. and its fractions were found to reduce lipid peroxidation induced by quinolinic acid and sodium nitroprusside in the rat brain (Marques et al., 2015). Free radical-scavenging activity of OPCs from Rhodiola rosea L. has been evaluated in nitric oxide, liver, heart and kidney of rats, where the ethanolic extract subjected to in vitro studies on animal models reduced malondialdehyde (MDA) content (Zhou et al., 2014). Another interesting and recent study has evaluated the protective effect of grape seed proanthocyanidins (GSPs) on cadmium-induced renal toxicity (Nazima et al., 2015a). These compounds enhanced SOD and GSH peroxidase (GPx) activity by reducing malondialdehyde (MDA) content (Zhou et al., 2014). Another interesting and recent study has evaluated the protective effect of grape seed proanthocyanidins (GSPs) significantly improved total antioxidant capacity, SOD, GSH, GPx and catalase (CAT) and reduced the MDA levels. Proanthocyanidins therefore effectively antagonize oxidative damage and enhance antioxidant capacity, but the antagonistic effect may be extensively metabolized by gut microbiota significantly improved cognitive impairments by blocking endoplasmic reticulum stress and NF -κB-mediated inflammatory pathways in rats (Liu et al., 2016). A meta-analysis of experimental studies regarding the efficacy of proanthocyanidins against oxidative damage has recently been performed (Li et al., 2015a). Compared with the control group, proanthocyanidins significantly improved total antioxidant capacity, SOD, GSH, GPx and catalase (CAT) and reduced the MDA levels. Proanthocyanidins therefore effectively antagonize oxidative damage and enhance antioxidant capacity, but the antagonistic effect may be which the indexes are estimated. The authors observed, in fact, significant differences in the effects of proanthocyanidins in relation to thependencies of administration (gavage vs. feeding) and dependent on the method by which the indexes are estimated. The authors observed, in fact, significant differences in the effects of proanthocyanidins in relation to themode of administration (gavage vs. feeding) and dependent on the biological matrix analysed (Li et al., 2015a). At the present, there is only limited clinical evidence available about the antioxidant properties of hydrolysable tannins and proanthocyanidins. Three pilot studies on human volunteers have recently been carried out using a French oak wood (Quercus robur L.) extract rich in roburin (a generic tannin), known with the brand name of Robuvit®. The first examined the pharmacokinetics and effects at transcriptome level of this formulation in healthy volunteers and showed that Robuvit® modulated different genes associated with oxidative stress, cell cycle and spliceosome pathways (Natella et al., 2014). The second study, which analysed the effect of 1 month intake of Robuvit® (300 mg·day−1) on oxidative stress markers in the plasma of healthy volunteers, demonstrated a decrease of plasma lipid peroxides, an increased SOD and CAT activity and total antioxidant capacity (Horvathova et al., 2014). The third study, which was the most recent, showed Robuvit® to have no significant influence on GSH levels and paraoxonase (an antioxidant enzyme) activity, although homocysteine and cysteine levels decreased significantly (Dešková et al., 2015). Finally, a double-blind crossover trial was performed on 18 volunteers to evaluate the effects of procyanidin-rich chocolate on fecal free radical production and antioxidant activity. Subjects were fed a low-polyphenol diet and chocolate for two 4 week washout period. The authors observed a decreased free radical production in fecal water with respect to control group, suggesting that chocolate proanthocyanidins may be effective in rebalancing redox status (Sieniawska, 2015). Many studies, carried out to highlight the mechanisms of action of proanthocyanidins, have identified several molecular targets potentially useful for the prevention and treatment of cancer. It should be stated, however, that knowledge in this area is still at an embryonic stage, and further studies are needed to better clarify the anti-cancer properties of these compounds. Proanthocyanidins have been extensively studied on a wide range of cancer cells. GSPs induced p53-dependent apoptosis on JB6 C141 mouse skin epidermal cells, through involvement of Bax/Bcl-2 proteins and caspase-3 activation (Gollucke et al., 2013). Furthermore, inhibition of the phosphorylation of proteins belonging to the MAPK family and activation of NF-κB and its related genes were observed in normal human epidermal keratinocytes (Gollucke et al., 2013). The same molecular mechanisms were found to be involved in the inhibition of cell growth and in the down-regulation of MMP-2 and −9 in both androgen-insensitive (DU145) and androgen-sensitive human prostate carcinoma cells (LNCaP) (Gollucke et al., 2013) on which GSPs also seem able to regulate androgen receptor-mediated transcriptome activity (Park et al., 2011). The down-regulation of these MMPs , involved in tumour development and metastasis, was also observed in pancreatic carcinoma cells (Chung et al., 2012). The inhibition of cell viability and proliferation and the pro-apoptotic activity of GSPs were also observed on mouse mammary tumour 4T1 and colorectal cancer HT29 and LoVo cell lines (Sieniawska, 2015). GSPs interfered with normal regulation of cell-cycle progression inducing an up-regulation of Cip1/p21 and Kip1/p27 protein levels together with a down-regulation of cyclins (D1, D2 and E) and cyclin-dependent kinases (CDK2, CDK4 and CDK6) leading to G0/G1 phase arrest (Prasad and Katiyar, 2014). Concentration-dependent effects of GSPs

were observed on breast and lung cancer and gastric adenocarcinoma cells (Gollucke et al., 2013). Besides GSPs, grapes and pine bark procyanidin-rich fractions (with the highest percentage of galloylation and mean degree of polymerization) also effectively inhibit cell proliferation, arresting the cell cycle in G2 phase and inducing apoptosis, as observed in HT29 human colorectal cancer cells (Ouédraogo et al., 2011). In vitro anti-proliferative and pro-apoptotic effects were also observed with several berry procyanidin-rich extracts on human oral (KB, CAL-27), breast (MCF-7), colon (HT-29, HCT116) and prostate (LNCaP) cancer cells (Nile and Park, 2014; Skrovankova et al., 2015). The in vitro anti-cancer properties of proanthocyanidins have been confirmed by several animal studies. Dietary feeding of GSPs was found to inhibit UVB-induced photocarcinogenesis in SKH-1 hairless mice. This event seems to be correlated with the ability of these compounds to decrease UVB-induced immune suppression in mice, by enhancing the production of IL-12 and reducing the expression of IL-10 (Gollucke et al., 2013; Sieniawska, 2015). A study performed in immunocompetent Balb/c mice fed with GSPs showed tumour-growth inhibition with an increase in the survival period and inhibition of metastatic status (Gollucke et al., 2013). Similar results were observed in other in vivo cancer models (Gollucke et al., 2013). Moreover, it has been demonstrated that GSPs inhibit the spontaneous development of prostate cancer in male TRAMP mice (Sieniawska, 2015). Although a number of experimental studies have been published, few clinical trials on the anti-cancer properties of proanthocyanidins are available, to date. A double-blind, placebo-controlled, randomized phase II trial was planned to elucidate the effects of supplementation with GSP extract (300 mg day−1 for 6 months) in patients affected by radiation-induced breast induration. At 12 months post-randomization, ≥50% reduction in cancer area (cm2) of breast induration in 29.5% of GSPs extract treated patients was reported. No significant difference between treatment and control groups in terms of external assessments of tissue hardness, breast appearance or patient self-assessments of breast hardness, pain or tenderness was recorded (Sieniawska, 2015). Proanthocyanidins have also been evaluated for their potential to attenuate the adverse effects of cancer radiotherapy. A randomized, double-blind, placebo-controlled, pilot study was conducted using standardized cranberry capsules, containing 72 mg of proanthocyanidins, for the prevention and treatment of radiation cystitis in prostate cancer patients. The authors observed a statistically significant decrease in cystitis incidence in treated patients (65%) compared with control group (90%); severe cystitis was reported in 30% of treated patients and 45% in the control group. Moreover, the incidence of pain and burning was significantly lower in treated group. The results demonstrate that patients who receive radiation therapy for prostate cancer, particularly those on low hydration regimens or with baseline urinary symptoms, may benefit from cranberry supplementation (Hamilton et al., 2015). The antimicrobial properties of proanthocyanidins and HTs present in many medicinal plants and foods are well documented. These phytochemicals seem to affect bacterial growth via several mechanisms such as inhibition of extracellular enzymes, deprivation of microbial essential substrates (for example by complexing metal ions), disintegration of bacterial outer membrane with cytoplasm leak or by direct action on microbial metabolism. Furthermore, these compounds can interfere with microbial cell wall polypeptides reacting with sulfhydryl groups, often leading to loss of function of the membrane proteins (Marín et al., 2015).

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Proanthocyanidins and hydrolysable tannins: occurrence, dietary intake and pharmacological effects. British Journal of Pharmacology, 174: 1244–1262. doi: 10.1111/bph.13630. [... full reference list continues ...]