Review
Digestion and bioavailability of bioactive phytochemicals

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Summary   There are many scientific reports on determination of the content and biological activity of compounds found in food. However, these analyses are not sufficient to determine their effect on the human body. During digestion of food ingredients, many changes can modify their structure and this may affect their absorption and bioactivity. Many phenolic aglycones are hydrophilic and can be absorbed through biological membranes by diffusion. However, most polyphenols occur in the glycosidic form, which undoubtedly affects their absorption in the intestine. Oligopeptides are also absorbable via secondary active transport but based on the hydrogen ion gradient or with transporter PepT1. The bioavailability of phytochemicals is determined by their molecular weight or chemical structure and the food matrix. Accordingly, the aim of this work was to present the novel scientific reports related to the influence the many factors on digestibility, bioaccessibility and activity of selected bioactive compounds of plant origin.

Keywords   Bioaccessibility, bioactive peptide, bioactive phytochemicals, bioavailability, digestion, phenolic compounds.

Introduction

Nowadays food of plant origin plays a very important role in the human diet as a valuable source of many bioactive components: phenolic compounds, vitamins or bioactive peptides (Figure 1). These phytochemicals may be beneficial to human health and protect against the negative influence of many pathogenic agents (Del Rio *et al.*, 2013; de Castro & Sato, 2015; Díaz-de-Cerio *et al.*, 2016; Maestri *et al.*, 2016; Marhuenda *et al.*, 2016). Determination of the bioactivity and content of nutritive compounds is not sufficient for prediction of potential *in vivo* effects associated with intensive metabolism that takes place during absorption. Therefore, there is a need to establish the best approach for assessment of the bioavailability and biological effectiveness of entire pool of active phytocomponents.

The bioefficacy of active compounds depends on several factors: food matrix, digestibility, bioaccessibility, solubility, transporters, molecular structures and metabolising enzymes. Investigation of the bioavailability of food constituents is challenging due to the different mechanisms of their absorption and often a complex nature of bioactive compounds.

According to Carbonell-Capella *et al.* (2014), the term 'bioavailability of compounds' includes gastrointestinal digestion (liberation), absorption, tissue distribution, metabolism and excretion, and is determined *in vivo* in animals or humans (LADME). In turn, bioaccessibility is generally determined by an *in vitro* digestion procedure that includes several stages: liberation from the food matrix, conversion during digestion conditions and absorption by the cells of the intestinal epithelium (Figure 2). Bioactivity is defined as a specific effect obtained after exposure to a substance and may be assessed *in vivo*, *ex vivo* or *in vitro*. It includes tissue uptake and the respective physiological (antioxidant, anti-inflammatory, anti-allergic, antibacterial or anticancer) response (Carbonell-Capella *et al.*, 2014).

This review focuses on the digestibility, bioaccessibility and valuable properties of many bioactive compounds of plant origin.

Metabolism of polyphenols

Phenolics are the main group of secondary plant metabolites present in food and beverages. It is a heterogeneous class of phytochemicals, for which the main basis of classification is the presence of at least one aromatic ring in the structure with one or more hydroxyl groups attached (Del Rio *et al.*, 2013). Phenolics vary in their chemical structure and properties, ranging from simple molecules (such as phenolic acids) to highly polymerised molecules, for example

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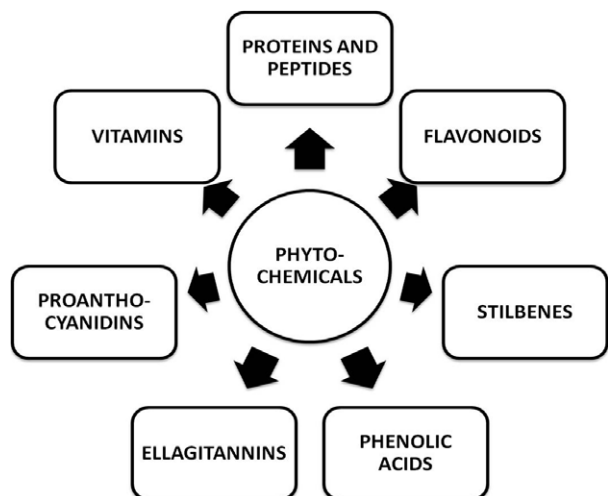e-mail: monika.karas@up.lublin.pl

Figure 1 Profile of bioactive compounds from plant-derived food.

proanthocyanidins (Landete, 2012). Based on their chemical structure, they are divided into two main groups: flavonoids and nonflavonoids. The non-flavonoids include phenolic acids, that is phenols with only one aromatic ring (C6), and stilbenes with a basic skeleton formed by two aromatic rings joined by a methylene bridge (C6-C3-C6). On the other hand, flavonoids are a large group of compounds with a flavane nucleus consisting of two benzene rings linked by an oxygen-containing pyran ring (C6-C3-C6) (D'Archivio *et al.*, 2010).

Phenolic compounds are essential for the quality of plant-derived food products due to their involvement in oxidative stability and organoleptic properties. These phytochemicals possess many biological activities and a documented positive impact on human health. There are data about the antioxidant, anti-allergic, anti-atherosclerotic, anti-inflammatory, antibacterial and antithrombotic properties of phenolic compounds (Del Rio *et al.*, 2013; Chen *et al.*, 2015; Díaz-de-Cerio *et al.*, 2016; Marhuenda *et al.*, 2016). Most of these studies were conducted as *in vitro* experiments. However, the physiological activity of polyphenols does not depend directly on their prevalence in the human diet. Indeed, these compounds are poorly absorbed from the colon or metabolised and are rapidly excreted from the body. Metabolites formed from phenolic compounds in the gastrointestinal tract may differ in their biological activity from the initial materials after transfer of the blood to the target organs (Crozier *et al.*, 2010; Marín *et al.*, 2015). There are differences in the activity of antioxidants, including phenolic compounds, in individual cell organelles, compared to their activity in the whole cell. The cell membrane is a barrier that affects the bioavailability of intracellular antioxidants, which may decrease their
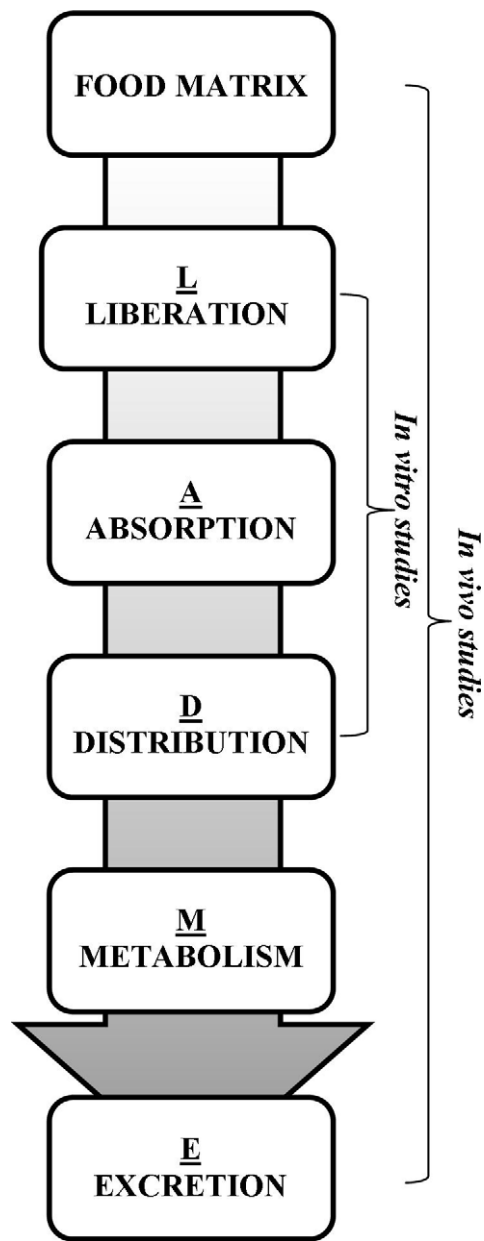

Figure 2 Scheme of steps in bioavailability process of bioactive compounds food matrix.

genotoxic or genoprotective effect (Del Rio *et al.*, 2013). Their relatively low absorption from the gastrointestinal tract limits the biological effect of polyphenol compounds on the organism. It has been shown that the intestinal absorption of polyphenols and their metabolites circulating in the plasma is determined by their chemical structure (D'Archivio *et al.*, 2010). The bioavailability of each class of polyphenols is determined mainly by molecular weight,
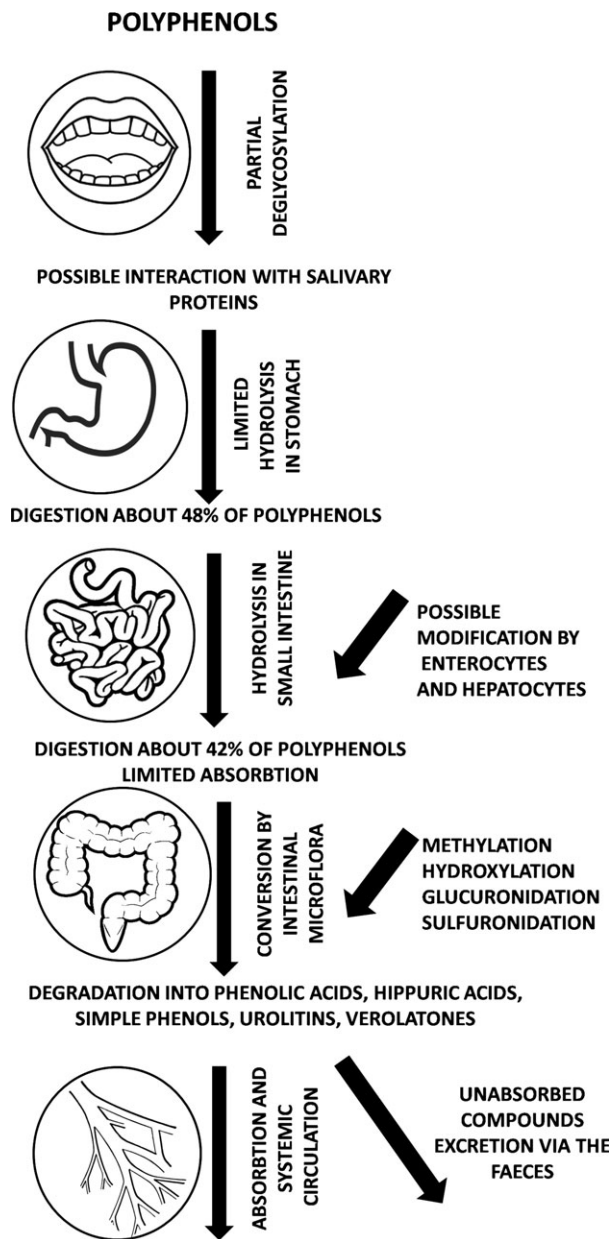
POLYPHENOLS

Figure 3 Scheme of gastrointestinal fate of polyphenols.

glycosylation and esterification. Many phenolic aglycones are hydrophilic in nature and can be absorbed through biological membranes by diffusion (Figure 3). However, most polyphenols contained in fruits and vegetables occur in the glycosidic form, which undoubtedly affects their absorption in the intestine.

Phenolic acids

Phenolic acids (C6–C3) are widespread dietary non-flavonoid compounds. There are two groups: hydroxybenzoic acid derivatives and hydroxycinnamic acid derivatives. Phenolic acids from the hydroxybenzoic family generally occur in low concentrations in food. Only in red fruits, onion, potatoes or black radish, the content of gallic, ellagic, protocatechuic and 4-hydroxybenzoic acids is relatively higher. Bioavailability studies demonstrate that gallic acid is very well absorbed. It has been proved to be absorbed in the stomach, small intestine or both. After consumption of gallic acid in the pure form or in a food source, it occurs in plasma and urine primarily in the 4-O methylated and O-glucuronidated forms (Lafay & Gil-Izquierdo, 2008). Also Quirós-Sauceda *et al.* (2014) reported that gallic and protocatechuic acids were identified in all the digestion phases in an *in vitro* experiment.

Hydroxycinnamic acids, such as caffeic acid, ferulic acid, sinapic acid or p-coumaric acid, are rarely found in the free form in food. Mostly, they are esterified with quinic and tartaric acids or carbohydrate derivatives (D'Archivio *et al.*, 2010). When ingested in the free form, hydroxycinnamates were rapidly absorbed from the stomach or the small intestine and next conjugated by the intestinal and/or hepatic detoxification enzyme (Lafay & Gil-Izquierdo, 2008). In esterified forms, hydroxycinnamic acids are digested in the colon (Olthof *et al.*, 2016). During comparison of the digestion of aleurone-enriched bread and whole-grain bread, Dall'Asta *et al.* (2016) found that caffeic and sinapic acids appeared as the most bioaccessible phenolic acids both in the aleurone-rich and whole-grain bread, while total ferulic acid and p-coumaric acid were less bioaccessible.

Stilbenes

The importance of stilbenes as food ingredients is low. The most famous representative of this group is resveratrol (3,5,4′-trihydroxystilbene), which occurs particularly in grapes, peanuts, berries, pistachios and red wine (Zamora-Ros *et al.*, 2008). *Trans-* and *cis*-isomers of resveratrol have been found in plants, but *trans*-resveratrol is more stable in nature. Cis-isomerisation occurs when resveratrol is exposed to light. At the intestinal level, resveratrol is absorbed by passive diffusion or formation of complexes with membrane transporters, such as integrins. Resveratrol in the bloodstream was found in three different forms: free or as glucuronide or sulphate derivatives. This is the effect of modification of the compound by enterocytes or hepatocytes or both (Burkon & Somoza, 2008).

Flavonoids

Flavonoids are a large group of polyphenols classified according to the oxygenation state of the heterocycle.

Due to structural differences, flavonoid compounds have been divided into six subclasses: flavonols, flavanones, flavan-3-ols, isoflavones, flavones and anthocyanins. Other groups of flavonoids, which have a relatively small share in the diet, comprise dihydroflavones, flavan-3,4-diols and coumarin (Jaganath & Crozier, 2010). To date, more than six thousand different flavonoids have been identified and new compounds from this group are still being discovered. Most flavonoids, except catechins, occur naturally in plants in conjunction with β-glycoside sugars (Ferreyra *et al.*, 2012).

The first step in the flavonoid metabolism is the hydrolysis of the glycosidic bond, which may take place in the lumen of the intestine or in enterocytes directly before absorption; the type of the substituted sugar moiety is an important factor. Polyphenols containing glucose, arabinose and xylose can be substrates of human endogenous cytosolic β-glucosidase. In contrast, the hydrolysis of the rhamnose glycoside bond can catalyse only rhamnosidase produced by the intestinal microflora (Ferreyra *et al.*, 2012). The efficiency of absorption is reduced when bacterial microflora is involved in the metabolism of polyphenols; in this case, aglycones are reduced to simple aromatic acids. Some authors have proved that the metabolites of phenolic compounds formed via the activity of microflora are more active than their precursors (Araújo *et al.*, 2013; Ozdal *et al.*, 2016). Many enzymes, either endogenous or produced by the microflora from the human digestive tract, are involved in the metabolism of polyphenols. These include the aforementioned cytosolic β-glucosidase (EC 3.2.1.21), present in many tissues but predominantly in the liver, and lactase (EC 3.2.1.108), occurring only in the intestine, which may be responsible for the hydrolysis of many polyphenol glycosides, in particular quercetin-3-O-glucoside. During absorption, the phenolic compounds are subject to conjugation in the liver and enterocytes. These processes involve catechol methyltransferase (EC 2.1.1.6) catalysing polyphenol methylation, UDP-glucuronidase (EC 2.4.1.17) responsible for conjugation with glucuronide conjugates and phenol sulfotransferase (EC 2.8.2.1) transforming aglycones in sulphate derivatives. Polyphenolic compounds metabolised in the organism are excreted in the urine or bile (Cao *et al.*, 2015). There are significant differences in the bioavailability, the process of metabolisation and the form of the presence of individual polyphenols in the plasma. For example, the absence of a free hydroxy group at position 5, 7 or 4 protects the compounds from degradation (Ferreyra *et al.*, 2012). An important role is also played by the origin of polyphenolic compounds, that is the source of food, as the compounds may occur in different forms. Isoflavones and gallic acid, followed by catechin, quercetin glycosides and flavanones, are best absorbed in the human digestive tract. The weakest absorption was reported for proanthocyanidins, tea epicatechin gallate and anthocyanins (Fang, 2014). Literature data suggest that anthocyanins are less efficiently absorbed from the gastrointestinal tract than other flavonoids. However, recent studies confirm that anthocyanins can be absorbed in the glycosidic form, not only after hydrolysis by intestinal microflora, as assumed previously (Ozdal *et al.*, 2016). It is also suggested that Na^+-dependent glucose transporters and possibly bilitranslocase (TC 2.A.65.1.1), that is a membrane transporter of organic anions present in gastric mucosal cells, may be involved in the anthocyanin glycosides transport through the gut wall. The presence of mono-, di- and triglycosides of cyanidin, peonidin and delphinidin both in the blood and in the urine may confirm this hypothesis (Zou *et al.*, 2014). Metabolism of anthocyanins leads mainly to the formation of glucuronide and methylated derivatives and, to a lesser extent, UDP-glucuronic transferase, UDP-glucose dehydrogenase, or catechol methyltransferase sulphonated under the influence of enzymes present in the small intestine, liver and kidney. Various concentrations of anthocyanins and their metabolites in urine determined in different studies suggested that the absorption of anthocyanins depends on the chemical structure of the compound, the type and number of substituted sugar moieties, and the acylation type (González-Barrio *et al.*, 2010; Fang, 2014). Bioavailability results of anthocyanins may be discarded for two main reasons: some essential metabolites may be omitted from the analysis (or degraded during storage of the samples intended for analysis) or methods have not been well adapted to the analysis of anthocyanin metabolites. Many researchers indicate that the anthocyanin contents exhibited statistically significant decreases after simulated digestion. This effect does not necessarily indicate a reduction in the amount of the compounds. Structural transformation of anthocyanins, especially under the varied pH conditions of the digestion model, would render them undetectable by the total monomeric and HPLC-based methods employed for the analysis. This conclusion can be confirmed by the analysis of total phenolic contents (Jayawardena *et al.*, 2015). It is well known that various chemical forms of anthocyanins remain in chemical equilibrium, depending on the pH. In most studies, the total anthocyanin content assays involve UV/Vis detection based on the total conversion of all chemical forms of anthocyanins in an acidic cation flavylium colour. It is possible, however, that some forms occurring in an inert environment will not be converted to this embodiment, due to the binding or chemical reactions with other components of the urine or plasma (Fang, 2014).

Methods for determination of bioavailability of anthocyanins include experiments performed in a laboratory (*in vitro*) or humans (*in vivo*). In the last decade, *in vitro* studies concerning anthocyanins absorption were carried out on Caco-2 cells (Kamiloglu *et al.*, 2015). Studies investigating anthocyanin absorption by Caco-2 cells reported very low transport of these compounds. Anthocyanin absorption may be influenced by the aglycone structure (e.g. delphinidin showed lower transport efficacy than malvidin and peonidin), the type of sugar moiety (glucose-based anthocyanins had higher bioavailability than galactose-based anthocyanins), polymeric structure (polymeric anthocyanins show lower absorption than the monomeric compounds) and the presence of other food components (González-Barrio *et al.*, 2010; Fang, 2014). On the other hand, the *in vivo* studies of Czank *et al.* (2013), in which isotopically labelled cyanidin-3-O-glucoside containing three ^{13}C atoms on the A ring and two ^{13}C atoms on the B ring was used, indicate that anthocyanins are more bioavailable than previously observed, and their metabolites are present in blood for ≤48 h after ingestion (relative bioavailability of $12.38 \pm 1.38\%$). The differences between the individual participants of this study are probably a result of a high variation in gastric and intestinal transit times, the amount and composition of microflora, and the ability to excrete metabolites.

The latest studies suggest that anthocyanins could be absorbed through the gastric wall, which is confirmed by detection thereof in the parent forms in plasma (Fernandes *et al.*, 2014). A probable mechanism of anthocyanin absorption is based on binding this molecule to an unidentified protein in the stomach tissue (nonspecific binding or perhaps specific binding to some protein transporter), so thus it could not be quantified as free anthocyanin by HPLC (Fernandes *et al.*, 2014). The anthocyanin fraction that is not absorbed in the stomach reaches the small intestine, where it is converted to carbinol pseudobase. Probably, anthocyanin glycosides are absorbed in the small intestine via the specific glucose transporter GLUT2 (Yoshikawa *et al.*, 2011). Kalt *et al.* (2016) proved that anthocyanin metabolites, mainly aglycones, especially aglycone glucuronides, were present in human urine even 5 days after consumption, which suggests that anthocyanin absorption is greater than previously suggested.

Ellagitannins

Ellagitannins are bioactive polyphenols abundant in some fruits and nuts, such as pomegranates, black raspberries, raspberries, strawberries, walnuts and almonds. Due to their large sizes, these compounds are not adsorbed via intestines. Ellagitannins are stable in the acidic environment of the stomach, but they are degraded in the small intestine to ellagic acid (EA), which is poorly bioavailable. In the large intestine, it may be transformed by colonic microflora to urolithins. Urolithins are much more bioavailable than ellagic acid, but their quantity depends of the number and type of human gut bacteria (Espín *et al.*, 2013). Tomás-Barberán *et al.* (2014) identified three phenotypes of urolithin producers: phenotype 0 (nonurolithin producers), phenotype A (produces only urolithin A) and phenotype B (produces urolithin A, isourolithin A and urolithin B). In their *in vivo* study, González-Sarrías *et al.* (2015) found individual variability in the amount of EA absorbed and the pharmacokinetic profile. Volunteers showed peaks of absorption over 5–24 h. These authors also proved that high intake of free ellagic acid did not enhance ellagic acid bioavailability when it was consumed with a pomegranate extract. García-Villalba *et al.* (2016) identified several urolithin aglycones present in faecal samples and some glucuronide and sulphate conjugates in plasma and urine.

Proanthocyanidins

Proanthocyanidins, also known as condensed tannins, are derivatives of flavonoids, mainly flavon-3-ols, flavan-3,4-diols or both. The most popular are the dimers of (+) catechin and (−) epicatechin. These polymers do not contain sugar moieties. Condensed tannins are widespread in fruits and vegetables, that is grapes, wine, blackberries, cranberries, plums and cherries. They are present mainly in fruit skin and are responsible for the characteristic taste of fruits (Landete, 2012). High molecular weight proanthocyanidins are almost nonresorbable in the digestive tract, which confirms their minimal concentration in the urine (Stoupi *et al.*, 2010). Only a few per cent of condensed tannins are bioaccessible in the small intestine, and the rest is transformed by colonic microbiota in the large intestine into phenylpropionic, phenylacetic, hippuric and benzoic acids with different hydroxylation patterns (Del Rio *et al.*, 2010; Stoupi *et al.*, 2010).

Consumption of plant foods is associated with a simultaneous intake of substantial levels of polyphenols, all of which may be present at various concentrations. Thus, the interaction between the compounds in the intestines can affect the bioavailability of the final products (Bohn, 2016).

Food matrix (interaction between phenolic compounds and other food components)

One of the factors that affect the bioavailability of phenolic compounds described *in vitro* models is the food matrix, which can determine the interactions with

other molecules from food (Rodríguez-Roque *et al.*, 2015). Interactions of polyphenols with proteins, carbohydrates, and lipids are extensively described in the literature (Świeca *et al.*, 2013; Rodríguez-Roque *et al.*, 2015; Velderrain-Rodríguez *et al.*, 2016). Studies of the relationship between polyphenols and proteins in foods have indicated that proteins can bind to polyphenols, and thus, the protein–polyphenol interaction could affect the bioavailability of phenolic compounds. Such an effect was observed in studies concerning *in vitro* bioaccessibility of phenolic compounds (Table 1) from fruit juices with addition of protein from milk or soya milk (Rodríguez-Roque *et al.*, 2014b, 2015). Similarly, investigations performed by Świeca *et al.* (2013) demonstrated a decrease in the bioaccessibility of phenolic compounds and masking of the antioxidant potential of bread enriched with flavonoid-rich onion skin, which resulted from protein–flavonoids interactions. There are also numerous *in vitro* studies concerning the interaction of polyphenols with carbohydrates, especially with dietary fibres. Polyphenols can combine with cell wall components from food by hydrogen and covalent bonds, which determine the bioavailability of phenolic compounds (Jakobek, 2015). Some studies indicated that formation of a polyphenol-dietary fibre complex decreased the bioaccessibility of phenolic compounds (Bouayed *et al.*, 2011); in contrast, in the research conducted by Velderrain-Rodríguez *et al.* (2016), dietary fibre in mango, papaya and pineapple was not a limiting factor in the bioaccessibility of its phenolic compounds during *in vitro* digestion. Therefore, as suggested by Jakobek (2015), the bioavailability of polyphenols in carbohydrate-rich food probably depends on the release of these compounds from complexes, which was determined by various factors, for example the structure of phenolics, complexity of interaction between polyphenols and carbohydrate, and the activity of enzymes. Recently, many studies (Palafox-Carlos *et al.*, 2011; Saura-Calixto, 2011) have indicated possible positive effects of the occurrence of the polyphenol–carbohydrate complex, which could transport dietary phenolics through the gastrointestinal tract; next, they can be released in the large intestine and their bioaccessibility in the colon can be enhanced. Additionally, the results obtained by Sui *et al.* (2016) indicated that anthocyanins fortification of bread may slowing down starch digestion and can decrease glycaemic index of carbohydrate-rich product. Subsequently, the study conducted by Ortega *et al.* (2009) indicates that the fat content in food (such as cocoa samples) increases the bioavailability of some phenolics, especially procyanidins, during duodenal digestion (Table 1). These researchers hypothesise that this effect is probably related to the ability of the fat fraction to interact with certain polyphenolic compounds following better micellisation of the digested phenols. Similarly, in the study performed by Guo *et al.* (2013), dietary fat increased quercetin bioavailability probably by enhancing its micellisation in the small intestine (Table 1).

Additionally, in their studies of the impact of the food matrix on the stability of red cabbage anthocyanins under *in vitro* digestion, Podsędek *et al.* (2014) suggested that other vegetable components protect labile anthocyanins from degradation under simulated digestion.

Besides macromolecules, phenolic compounds may also interact with iron or zinc ions during the *in vitro* digestion. This fact plays a significant role because fruit beverages are often commercially supplemented with some minerals to improve their nutritional or pro-health value. As indicated in the research conducted by Cilla *et al.* (2009), iron addition to fruit

Table 1 Influence of food matrix on the digestibility, bioavailability or bioaccessibility of polyphenols

Source of polyphenols and other molecules	Effects	References
Total phenolic acids and total flavonols from fruit juice (orange, kiwi, pineapple and mango) with milk and soya milk proteins	Decrease the bioaccessibility of phenolic substances	Rodríguez-Roque *et al.* (2015)
Ferulic acid, sinapic acid, quercetin, hesperidin and rutin from blend of fruit juice (orange, kiwi and pineapple) with soya milk proteins	Decrease the bioaccessibility of tested phenolic compounds	Rodríguez-Roque *et al.* (2014b)
TPC, TFC and PAC from blended fruit juice (from orange, kiwi, pineapple and mango) with milk proteins	Decrease the bioaccessibility of phenolic substances	Rodríguez-Roque *et al.* (2014a)
Quercetin and rutin from black and green tea with milk proteins	Decrease the bioavailability of phenolics	Nizamova *et al.* (2011)
Polyphenols from *Artemisia dranculus* L. with soy protein	Increase the bioavailability and bioaccessibility of polyphenols	Ribnicky *et al.* (2014)
Polyphenols from cocoa liquor (more fat content) and cocoa powder (less fat content)	Higher fat content enhanced the digestibility of polyphenols	Ortega *et al.* (2009)
Quercetin from fat-free, low-fat or high-fat muffins	Dietary fat improves quercetin bioavailability	Guo *et al.* (2013)
Phenolic compounds from mango, papaya and pineapple and dietary fibre from fruit	Dietary fibre does not have effect on bioaccessibility of phenolic compounds	Velderrain-Rodríguez *et al.* (2016)

beverages caused a major loss of polyphenols in the gastrointestinal digestion *in vitro*, compared to non-supplemented beverages. On the other hand, other studies (Cilla *et al.*, 2010) concerning fruit beverages with zinc addition with/without iron presence indicated that, after digestion, the zinc-fortified fruit beverages were characterised by lower total soluble extractable phenolic content than beverages without the addition, probably due to chelate formation. However, after digestion, a lower percentage decrease in the phenolic content was observed in the zinc-supplemented beverages in comparison with the control.

Many biological activities of phenolic compounds, especially antioxidant, immunological, anti-inflammatory, antiproliferative and hypoglycaemic effects, are well documented (Chen *et al.*, 2015; Díaz-de-Cerio *et al.*, 2016; Marhuenda *et al.*, 2016). The stability of phenolic compounds as well as the binding or release thereof from various combinations due to *in vitro* digestion may influence the biological activity of these compounds measured after digestion.

Most studies available in the literature relate to the antioxidant activity of the phenolic compounds of food subjected to *in vitro* digestion.

Generally, phenolic compounds from digested food present various complementary modes of antioxidant action. The antioxidant activities measured by the ABTS (2,20-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid) and ORAC (oxygen radical absorbance capacity) methods in fruit extracts (from araticum and jackfruit) after *in vitro* digestion were significantly ($P < 0.05$) higher, but the activity of the papaya extract did not show a significant difference ($P > 0.05$) between digested and undigested extracts (Pavan *et al.*, 2014). Similarly, based on DPPH (1,1-diphenyl-2-picrylhydrazyl) and ORAC results of phenolic extracts from gooseberry after *in vitro* digestion, it was found that the antioxidant activity was higher than that of nondigested extracts (Chiang *et al.*, 2013). The authors hypothesised that the increase in antioxidant activity could also be related to pH and enzymatic interactions that occur during *in vitro* digestion due to gradual release of polyphenols during the digestive process. However, in the study conducted by Marhuenda *et al.* (2016), phenolic compounds from different berries after *in vitro* digestion were characterised by lower antioxidant activity, which was also confirmed by ORAC assays, than these extracts before digestion. It is important to note that the process of *in vitro* digestion in this case included dialysis, but in the previous study, it ended at treatment with intestinal enzymes. Additionally, antioxidant activity after *in vitro* digestion is dependent on the food matrix and the class of phenolic compounds. Similarly, the antioxidant activity (DPPH and superoxide radical scavenging assays) of phenolic compounds from wild blueberry decreased after intestinal digestion (Correa-Betanzo *et al.*, 2014).

In their investigations of the impact of digestion on antioxidant activity of culinary spices such as cinnamon, cloves and nutmeg, Baker *et al.* (2013) observed that digestion altered the antioxidant activity (measured with the ABTS method) of these spices although the changes were not clear. Namely, cooking and digesting of cinnamon and cloves resulted in a decrease in the antioxidant activity of the analysed samples, but there was an opposite effect in the case of nutmeg (Baker *et al.*, 2013). Black tea polyphenols after gastrointestinal digestion led to an increase the ABTS cation radical scavenging capacity (Wu *et al.*, 2015).

Recently, it has become evident that polyphenols may also decrease oxidative stress through indirect antioxidant action such as an anti-inflammatory effect. Thus, the influence of digestion *in vitro* on this activity of the phenolic compounds fraction has also been studied. In some studies, inhibition of pro-inflammatory enzymes such as lipoxygenase (LOX), cyclooxygenase or xanthine oxidase has been reported. In the study performed by Gawlik-Dziki *et al.* (2013), phenolics from *Chenopodium quinoa* leaves exhibiting the lipoxygenase inhibiting activity were characterised by high bioavailability. Furthermore, in the studies of the anti-inflammatory activity of phenols contained in extracts of bread enriched with broccoli sprouts, simulated gastrointestinal digestion increased the capability of LOX inhibition as well as xanthine oxidase inhibition, but absorption *in vitro* resulted in a significant reduction in these activities (Gawlik-Dziki *et al.*, 2014). Additionally, Baker *et al.* (2013) studied the impact of cooking and digestion *in vitro* on the anti-inflammatory activity (specifically the inhibition of cyclooxygenase 2 (COX-2) and the amount of prostaglandin synthesised) of some spices. The capability of COX-2 inhibition was significantly higher in cooked and digested *in vitro* samples in comparison with uncooked and in only cooked cinnamon and nutmeg samples, but not in the case of cloves. Thus, in most cases, digestion *in vitro* released compounds having anti-inflammatory activity, but it may differ depending on the food matrices.

Besides the antioxidant and anti-inflammatory activity of phenolic compounds, the anticancer properties of these compounds have also been reported. Wild blueberry polyphenols in crude extracts and after stomach digestion exhibited significantly higher inhibition of proliferation of human colon cancer cells (HT-29) than these after simulated intestinal digestion (Correa-Betanzo *et al.*, 2014). Similarly, in the study performed by Gawlik-Dziki *et al.* (2014), the effects of extracts from bread enriched with broccoli sprouts on proliferation and motility of human stomach cancer

cells were reduced by gastrointestinal digestion and absorption.

Bioactive peptides

Bioactive peptides, especially derived from plant proteins, exhibit a variety of activities which have benefits to human health, such as anti-inflammatory, antihypertensive, antihypercholesterolaemic, antithrombotic, anticancer, antimicrobial, antioxidative effects, or enhanced absorption of trace minerals (Garcia *et al.*, 2013; Guo *et al.*, 2014; de Castro & Sato, 2015; Maestri *et al.*, 2016) (Table 2). Furthermore, proteins are a rich source of peptides that may turn out to be a good system for delivery and increased bioavailability of various bioactive compounds (Lin *et al.*, 2016). Bioactive peptides can be released from proteins during *in vivo* and *in vitro* digestion, as well as different food technology processes especially fermentation, or germination (López-Barrios *et al.*, 2016). The bioactive properties of many peptides have been defined and their mechanism of action described. Some of them influence the gastrointestinal system, others require absorption and transport into specific sites where they will be active. However, the way in which bioactive peptides are absorbed, distributed and metabolised is currently not well elucidated yet (Foltz *et al.*, 2010).

Various peptides released from the plant proteins have been described to show beneficial effect on health. The bioactive properties of these peptides are related to their molecular size, amino acid composition and hydrophobicity (Udenigwe & Aluko, 2012; Zou *et al.*, 2016).

In the study conducted by Sabbione *et al.* (2016), an amaranth protein isolate was obtained and subjected to simulated gastrointestinal digestion to evaluate its potential antithrombotic activity. The peptide fraction obtained from the amaranth protein hydrolysate exhibited higher antithrombotic activity ($IC_{50} = 0.07 \pm 0.01$ mg mL^{-1}) (IC_{50} – a sample concentration providing 50% inhibition) than protein hydrolysate ($IC_{50} = 0.23 \pm 0.02$ mg mL^{-1}). Additionally, the absorption of the peptide fraction was analysed with an *in vitro* assay of peptide transport through intestinal epithelium. The authors observed that some peptides were able to cross the Caco2-TC7 cell monolayer (Sabbione *et al.*, 2016). Antiplatelet peptides released by simulated gastrointestinal digestion of oat ($IC_{50} = 0.282$ mg mL^{-1}), barley ($IC_{50} = 0.290$ mg mL^{-1}) and buckwheat ($IC_{50} = 0.328$ mg mL^{-1}) flours have been reported by Yu *et al.* (2016).

Antioxidative peptides are widely found in a number of plant foods and are often generated by the digestion process. The effect of heat treatment of chickpea seeds on the antioxidant and fibroblast growth-stimulating activity of peptide fractions released during *in vitro*

gastrointestinal digestion and absorption was investigated by Karaś *et al.* (2015). Chen *et al.* (2013) obtained two novel ACE inhibitory tripeptides (VNP, VWP) from an abundant rice protein hydrolysate and showed their stability against pepsin and chymotrypsin.

A novel octapeptide (PVNNPQIH) with angiotensin I-converting enzyme (ACE) inhibitory activity ($IC_{50} = 206.7 \pm 3.9$ l μм) was identified by Rui *et al.* (2013). The hydrolysate was produced by sequential alcalase and papain digestion of small red bean protein obtained by *in vitro* simulation under gastrointestinal conditions (Rui *et al.*, 2013). Similarly, in the study carried out by Jakubczyk & Baraniak (2014), pea globulins were digested *in vitro* under gastrointestinal conditions and potentially bioaccessible ACE inhibitory peptides were reported (GGSGNY, DLKLP, GSSDNR, MRDLK and HNTPSR). The study by Tapal *et al.* (2016) describes the *ex vivo* digestibility of globulin isolate prepared from oil palm kernel. The hydrolysate, obtained by *in vitro* human gastrointestinal enzyme digestibility of the globulin isolate, exhibited potent ACE inhibitory activity ($IC_{50} = 50$ μg mL^{-1}) and anticancer activity against human colon epithelial cancer HT-29 cells and hepatocarcinoma HepG2 cells (Tapal *et al.*, 2016). Another important group of peptides are anti-inflammatory peptides, as the inflammatory process is fundamental to a number of diseases, for example cancers or cardiovascular disease (Chakrabarti *et al.*, 2014). Dia *et al.* (2014) investigated eight different soya products, which were hydrolysed with pepsin and pancreatin and determinated for their anti-inflammatory properties. The potential of soya protein hydrolysates to inhibit inflammation using lipopolysaccharide-induced macrophages was evaluated in an *in vitro* model. Bioactive peptides generated during the hydrolysis process (RQRK and VIK) inhibited the expression of pro-inflammatory enzymes (nitric oxide synthase, cyclooxygenase-2). Hydrolysates from different soy-milk products inhibited the production of nitric oxide, interleukin-1β, and tumour necrosis factor-α. (Dia *et al.*, 2014). The *in vitro* bioavailability of peptides obtained by enzymatic proteolysis of quinoa was assessed using Caco-2 colon monolayer cells that showed antihypertensive activity. Moreover, these peptides were able to activate the peroxisome proliferator-activated receptor anti-inflammatory transcription factor (Ravisankar *et al.*, 2015).

A number of bioactive peptides and hydrolysates obtained from plant proteins have also been investigated by *in vivo* studies (Chakrabarti *et al.*, 2014). Pyro-glutamyl leucine (bioactive peptide obtained from wheat gluten hydrolysate) was shown to protect against D-galactosamine-induced hepatitis in rats (Sato *et al.*, 2013). Proteins obtained from potato tubers were hydrolysed by autolysis, and rapeseed meal

Table 2 Bioactive peptides derived from food products of plant origin and their bioactive effects

Protein source	Experimental method/Preparation	Bioactive effect	Sequence information when available	References
Amaranth (*Amaranthus L.*)	Simulated gastrointestinal digestion of protein	Antithrombotic activity	Peptide fraction (IC_{50} = 0.07 ± 0.01 mg mL^{-1})	Sabbione *et al.* (2016)
Barley (*Hordeum vulgare* Li) buckwheat (*Fagopyrum esculentum* Moench), oat (*Avena sativa* L.)	Simulated gastrointestinal digestion of protein	Antithrombotic activity	Hydrolysates (IC_{50} = 0.290 mg mL^{-1}), (IC_{50} = 0.328 mg mL^{-1}), (IC_{50} = 0.282 mg mL^{-1})	Yu *et al.* (2016)
Chickpea (*Cicer arietinum*)	Simulated gastrointestinal digestion and absorption	Antioxidant and fibroblast growth-stimulating activity	Peptide fractions	Karaś *et al.* (2015)
Quinoa (*Chenopodium Quinoa*)	Enzymatic proteolysis, *in vitro* bioavailability	ACE inhibitory activity, anti-inflammatory	Peptide fractions	Ravisankar *et al.* (2015)
Palm kernel (*Elaeis guineensis*)	*In vitro* human gastro-intestinal enzyme digestibility	ACE inhibitory activity and anticancer activity	RADVFNPR, KLPLVERIP (IC_{50} = 50 µg mL^{-1})	Tapal *et al.* (2016)
Pea (*Pisum sativum*)	Simulated gastrointestinal digestion of globulin and absorption	ACE inhibitory activity	GGSGNY,MRDLK, DLKLP, GSSDNR, and HNTPSR (IC_{50} = 0.073 mg mL^{-1})	Jakubczyk & Baraniak (2014)
Potato (*Solanum tuberosum* L.) rapeseed (*Brassica napus*)	*In vivo* in Goldblatt rat model of hypertension	ACE inhibitory activity	Peptide fractions	Mäkinen *et al.* (2016)
Rice (*Oryza sativa* L.)	Protein hydrolysate with alcalase, trypsin	ACE inhibitory activity	VNP, VWP *in vitro*, *in vivo* (SH rats 5 mg kg^{-1})	Chen *et al.* (2013)
Small red bean (*Phaseolus vulgaris*)	Sequential digestion of alcalase, papain and by *in vitro* gastrointestinal simulation	ACE inhibitory activity	PVNNPQIH (IC_{50} = 206.7 ± 3.91 M)	Rui *et al.* (2013)
Soya (*Glycine max*)	Hydrolysate with pepsin and pancreatin *in vitro*	Immunomodulatory: decrease in the production of cytokines and inflammatory mediators	RQRK and VIK	Dia *et al.* (2014)
Wheat (*Triticum spp.*)	*Aspergillus oryzae* protease hydrolysis of gluten and fractionation *In vivo* Cell/organism tested in Rat hepatitis	Anti-inflammatory, improved hepatic enzyme profile	Pyro-glutamyl leucine	Sato *et al.* (2013)

proteins were digested with Alcalase. The digestions were followed by ultrafiltration and solid-phase extraction to concentrate the ACE inhibitory peptides. The effects of potato and rapeseed peptides on blood pressure were determined *in vivo* in Goldblatt rat model of hypertension. Cited study shows that proteins obtained from potato and rapeseed can be used in inhibiting the development of hypertension *in vivo* (Mäkinen *et al.*, 2016). The most common methods in investigation of the digestibility and bioaccessibility of active peptides are based on simulated gastrointestinal digestion and absorption (Table 2).

Digestibility, absorption and bioavailability of bioactive peptides

Bioactive peptides may be generated in the body in a free state or they may be supplied with food or generated in the gastrointestinal tract by the digestion of proteins. The first step of protein hydrolysis to peptide fragments in the gastrointestinal tract is digestion in the stomach by pepsin [EC 3.4.23.1], then by trypsin [EC 3.4.21.4] and chymotrypsin [EC 3.4.21.1], which are pancreatic endopeptidases acting in the small intestine, and by exopeptidases such as carboxypeptidase A [EC 3.4.17.1], B [EC 3.4.17.2] and aminopeptidase (Segura-Campos *et al.*, 2011). Smaller fragments of proteins are further digested to free amino acids by proteases and aminopeptidases located on the cells of small intestine epithelium (luminal membrane exactly). In the final stage, amino acids from the carboxyl and amino ends are removed. Therefore, at least 20 various peptidases are disposed on the luminal membrane of the epithelial cells. These enzymes have specific effect on peptide bonds and release free amino acids that can penetrate

epithelial cells via secondary active sodium transporters; therefore, there are 20 types of amino acid transporters. Moreover, peptides composed of two or three amino acids are also absorbable via secondary active transport but based on the hydrogen ion gradient or with transporter PepT1. In epithelial cells, the amino acids from these peptides are split off and can get into bloodstream by diffusion across basolateral membranes. Peptides and free amino acids are transported through the blood to a location or organs where they can exert an effect on body function. The proteins and peptides are hydrolysed mainly in the upper part of the gastrointestinal tract, the small intestine; however, very small proteins involved in endocytosis and exocytosis can cross the epithelium cells undamaged to the intestinal fluid. According to the study, the level of absorption of these compounds is much higher in children than in adults, while infants can absorb antibodies and some immunity compounds from mother's milk (Widmaier *et al.*, 2013).

Moreover, the activity of these compounds depends on their amino acid composition, bioavailability, interactions with other food compounds carried in the bloodstream, resistance to the activity of enzymes and target organs. It is also essential for the physiological activity of native peptides in the interaction with a stable structure and target receptors. Peptides can exhibit local activity or be able to cross the epithelium of the small intestine and, transported by bloodstream, have a strong effect on the site of action. On the other hand, the influence of several different physiological parameters, for example blood pressure, insulin and glucose homoeostasis, plasma cholesterol concentrations and immune function can modulate certain peptide functions (Moller *et al.*, 2008). Small intestine epithelial cells are the main site of absorption of nutritional and bioactive compounds such as peptides. Moreover, after oral administration, peptides can encounter chemical, biochemical and physical barriers by these cells, which may reduce the influence on their physiological functions. The main chemical barrier for bioactive peptides is the low pH environment of the stomach, which is both necessary for the digestion of proteins and, on the other hand, for the successful absorption of amino acids. The same enzymes that hydrolyse proteins can carry out hydrolytic digestion of peptides due to their similarity in the chemical structure and functional groups (Adessi & Soto, 2002). Some bioactive peptides are able to resist the action of proteases, which is related to their characteristic structure. For example, peptide including proline or hydroxyproline and dipeptides or oligopeptides with proline at the C-terminus are not hydrolysed by digestive enzymes (e.g. antihypertensive lactotripeptides Ile-Pro-Pro and Val-Pro-Pro (Boelsma

& Kloek, 2009), KWLPVPQ (Maeno *et al.*, 1996) and LHLPLP (Van Platernik *et al.*, 2006)). The presence of peptidases is a biochemical barrier, but the impermeable gastrointestinal epithelium (Catnach *et al.*, 1994) as well as parameters of the unstirred water/mucous layer, the epithelial membrane of enterocytes (transcellular route), and the tight junctions between the apical ends of epithelial cells (paracellular route) (Yin *et al.*, 2014) represent physical barriers. Peptides can be transported actively or passively depending on their amino acid composition and function. The transepithelial transport of peptides (composed of more than four amino acids) was investigated using human intestinal Caco-2 cell monolayers. The results suggested that the main role in the flux across the epithelial cell layer was played by digestion by cellular peptidases. On the other hand, an intracellular mechanism such as adsorptive transcytosis was suggested as the main factor in the transport of bradykinin and its derivatives, but the paracellular pathway was reported to be involved in transport of the GGYR tetrapeptide (Shimizu *et al.*, 1997). According to the study, antihypertensive peptides such as IF, AF, IPP, and VPP or opioid peptides remain structurally stable during infiltration via the epithelial monolayer and the transport efficiency is dependent on the load, molecular weight and hydrophobic peptides (Sienkiewicz-Szłapka *et al.*, 2009). In order to improve the bioavailability and bioaccessibility of bioactive peptides, studies are carried out on the use of suitable carriers that can protect the peptides against low pH in the stomach and, after dissolution in the gut, allow absorption thereof. Such treatments as chemical modification of peptides (e.g. by glycosylation) and the use of an emulsion or a microencapsulation process can decrease the activity of digestive enzymes, thereby increasing absorption of biopeptides in blood (Renukuntla *et al.*, 2013).

Vitamins

Vitamins are essential organic compounds, necessary for the proper course of cells and tissues life functions, present in very small quantities in natural foods and not synthesised or synthesised in the body in small amounts, which is insufficient to meet the organism demand. They are involved in the metabolism of fats, proteins, and carbohydrates and synthesis of numerous compounds necessary for the proper functioning of the body. Lack of vitamin or deficiency inhibits the mileage of numerous processes and interferes with the functioning of the body (Combs, 2012).

Vitamins can be divided into water-soluble, such as vitamin C, thiamine, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, biotin, pantothenate, and fat-

soluble such as vitamins A, D, E and K. Vitamins have antioxidant capacity, which is expressed in different ways, for example scavenging different free radicals and inhibiting lipid peroxidation (Gliszczyńska-Świgło, 2006).

Extensive research suggested that water-soluble vitamins and vitamin E had antioxidant capacity (Joshi *et al.*, 2001; Terentis *et al.*, 2002; Jung & Kim, 2003; Gliszczyńska-Świgło, 2006). Both, fat-soluble and water-soluble vitamins because of their antioxidant ability are used by human organism to reduce the risk of many diseases, for example cardiovascular or neurological diseases, neuropsychiatric disorders and even cancer (Verhaar *et al.*, 2002; Alpert & Fava, 2003).

Vitamin C (hexuronic lactone l-ascorbic acid) occurs in three different redox states: totally reduced (ascorbate – ASC), partially oxidised (semidehydroascorbate – SDA) and totally oxidised (dehydro-l-ascorbic acid – DHA), and all of them are biologically active. ASC is especially effective in quenching hydroxyl radicals. ASC also protects many compounds in tissues that are highly liable to oxidation, for example folate or cholesterol (Menéndez-Carreño *et al.*, 2008).

B vitamins are included in the list of nutraceuticals because of their potential positive effect on the human organism (Hugenholtz & Smid, 2002). Thiamine (thiamine, vitamin B_1) is important for brain function and cardiovascular function (Misumida *et al.*, 2014; Ortigoza-Escobar *et al.*, 2014). Furthermore, insufficient thiamine availability to tissues is related with increased blood pressure (Zhang *et al.*, 2014). Riboflavin (vitamin B_2) is a precursor of FMN and FAD, which help antioxidant protection, vitamin metabolism (folate, niacin, vitamins A, C, B_6 and B_{12}) and a lot of other purposes (Ramanujam *et al.*, 2011). Nicotinamide (vitamin B_3/vitamin PP) and nicotinic acid are two main water-soluble forms of niacin. Niacin is a precursor of NAD^+ and $NADP^+$ and is essential for normal function of human organism. This vitamin is used for the treatment of hyperlipidaemia (in daily doses approximately several grams) – the use of niacin results in a reduction in low-density lipoprotein (LDL) cholesterol and an increase in high-density lipoprotein (HDL) cholesterol concentrations (Kamanna & Kashyap, 2000). Vitamins B_6 in foods include several compounds that can be metabolically transformed into biologically active pyridoxal-5′-phosphate. Vitamin B_6 takes part in numerous important reactions, for example haeme, neurotransmitter, and amino acid synthesis and vitamin metabolism or metabolic regulation (Choi *et al.*, 2001). Vitamin B_{12} is a cofactor for enzymes that have an important influence on the metabolism of numerous compounds such as amino acids, fatty acids, phospholipids and hormones (Bennett, 2001). Biotin (vitamin H/vitamin B_7) takes part in lipid metabolism, amino acid disintegration and some nuclear functions

(Rodriguez-Meléndez *et al.*, 2001). Pantothenate (vitamin B_5) is important for brain function, growth and regeneration, whereas deficiencies in folate (vitamin B_9) may be risk factors for many disorders, for example cardiovascular and haematological diseases, neurological and neuropsychiatric disorders, and cancer (Zhang *et al.*, 1999; Verhaar *et al.*, 2002).

Fat-soluble vitamins are bioactive as well, and they are essential in many reactions. Vitamin A is very important for vision, immune function and regulation of cell growth. Vitamin A metabolites are relevant elements of the light-detecting complex in the photoreceptor cells of the eye (Saari *et al.*, 2001). Vitamin D is essential for enhancement of the intestinal absorption of calcium and retention of this compound in the body. Furthermore, this vitamin has an effect on the growth of bone and connective tissues and in some cases may even protect against cancer (Norman, 2006; Ricciardi *et al.*, 2015). Vitamin E, as mentioned above, is an antioxidant and as it is a fat-soluble vitamin, it may quench oxygen free radicals in lipid-rich compartments (Wortmann *et al.*, 2013). Vitamin K has several functions, for example, it activates proteins needed for blood coagulation, prevents calcification of arteries and other soft tissues, and supports mineralisation of bone (Vermeer & Theuwissen, 2011).

However, the possible health benefits of vitamins are frequently not fully realised because of their low or changeable bioavailability. The weak bioavailability of vitamins may be caused by several factors, for example low absorption, conversion into an inactive form or low bioaccessibility. Whereas the bioaccessibility of vitamins depends on their chemical structure, source, presence of elements acting synergistically or antagonistically, and the efficiency of the mechanisms of intestinal absorption (McClements & Xiao, 2014; Zou *et al.*, 2015). The solubility of vitamins notably affects its mechanisms of bioavailability. For example, absorption of fat-soluble vitamins from the small intestine is strongly joined to fat absorption (During *et al.*, 2005). Vitamins are absorbed to a varied level, therefore oral consumption does not ensure a beneficial influence on the human organism.

Conclusion

Plants are a valuable source of bioactive compounds, such as polyphenols, vitamins, and peptides derived from proteins, exhibiting many important properties. It can be concluded that bioavailability is a multistage process. Therefore, many factors (not only the content of bioactive phytochemicals, but also their molecular structure and interactions between food matrix compounds) influence on the bioavailability of the aforementioned bioactive compounds of plant origin. Between food compounds, during digestion in

gastrointestinal tract, occurs both synergistic and antagonistic interaction what affect on their bioavailability and bioactivity. For today's dieticians and consumers, very important is health effect of entire pool of phytochemicals included in food, not only individual components. Although there are many research reports on the bioavailability and biological effectiveness of bioactive food components, elucidation of their interactions, metabolism and mechanism of action still needs extensive research.

References

Adessi, C. & Soto, C. (2002). Converting a peptide into a drug: strategies to improve stability and bioavailability. *Current Medicinal Chemistry*, **9**, 963–978.

Alpert, J.E. & Fava, M. (2003). Nutrition and depression: the role of folate. *Nutrition Review*, **55**, 145–149.

Araújo, K.C.F., Costa, E.M.M.B., Pazini, F., Valadares, M.C. & de Oliveira, V. (2013). Bioconversion of quercetin and rutin and the cytotoxicity activities of the transformed products. *Food Chemistry and Toxicology*, **51**, 93–96.

Baker, I., Chohan, M. & Opara, E.I. (2013). Impact of cooking and digestion, *in vitro*, on the antioxidant capacity and anti-inflammatory activity of cinnamon, clove and nutmeg. *Plant Foods for Human Nutrition*, **68**, 364–369.

Bennett, M. (2001). Vitamin B_{12} deficiency, infertility and recurrent fetal loss. *Journal of Reproductive Medicine*, **46**, 209–212.

Boelsma, E. & Kloek, J. (2009). Lactotripeptides and antihypertensive effects: a critical review. *British Journal of Nutrition*, **101**, 776–786.

Bohn, T. (2016). Dietary factors affecting polyphenol bioavailability. *Nutrition Reviews*, **72**, 429–452.

Bouayed, J., Hoffmann, L. & Bohn, T. (2011). Total phenolics, flavonoids, anthocyanins and antioxidant activity following simulated gastro-intestinal digestion and dialysis of apple varieties: bioaccessibility and potential uptake. *Food Chemistry*, **128**, 14–21.

Burkon, A. & Somoza, V. (2008). Quantification of free and protein-bound *trans*-resveratrol metabolites and identification of *trans*-resveratrol-C/O-conjugated diglucuronides – Two novel resveratrol metabolites in human plasma. *Molecular Nutrition & Food Research*, **52**, 549–557.

Cao, H., Chen, X., Jassbi, A.R. & Xiao, J. (2015). Microbial biotransformation of bioactive flavonoids. *Biotechnology Advances*, **33**, 214–223.

Carbonell-Capella, J.M., Buniowska, M., Barba, F.J., Esteve, M.J. & Frígola, A. (2014). Analytical methods for determining bioavailability and bioaccessibility of bioactive compounds from fruits and vegetables: a review. *Comprehensive Reviews in Food Science and Food Safety*, **13**, 155–171.

de Castro, R.J.S. & Sato, H.H. (2015). Biologically active peptides: processes for their generation, purification and identification and applications as natural additives in the food and pharmaceutical industries. *Food Research International*, **74**, 185–198.

Catnach, S.M., Fairclough, P.D. & Hammond, S.M. (1994). Intestinal absorption of peptide drugs: advances in our understanding and clinical implications. *Gut*, **35**, 441–444.

Chakrabarti, S., Jahandideh, F. & Wu, J. (2014). Food-derived bioactive peptides on inflammation and oxidative stress. *BioMed Research International*, **2014**, 1–11.

Chen, J., Liu, S., Ye, R., Cai, G., Ji, B. & Wu, Y. (2013). Angiotensin-I converting enzyme (ACE) inhibitory tripeptides from rice protein hydrolysate: purification and characterization. *Journal of Functional Foods*, **5**, 1684–1692.

Chen, M., Meng, H., Zhao, Y., Chen, F. & Yu, S. (2015). Antioxidant and *in vitro* anticancer activities of phenolics isolated from sugar beet molasses. *BMC Complementary and Alternative Medicine*, **15**, 1–8.

Chiang, C.-J., Kadouh, H. & Zhou, K. (2013). Phenolic compounds and antioxidant properties of gooseberry as affected by *in vitro* digestion. *LWT - Food Science and Technology*, **51**, 417–422.

Choi, S.Y., Bahn, J.H., Lee, B.R. *et al.* (2001). Brain succinic semi-aldehyde dehydrogenase: identification of reactive lysyl residues labeled with pyridoxal-5′-phosphate. *Journal of Neurochemistry*, **76**, 919–925.

Cilla, A., González-Sarrías, A., Tomás-Barberán, F.A., Espín, J.C. & Barberá, R. (2009). Availability of polyphenols in fruit beverages subjected to *in vitro* gastrointestinal digestion and their effects on proliferation, cell-cycle and apoptosis in human colon cancer Caco-2 cells. *Food Chemistry*, **114**, 813–820.

Cilla, A., Lagarda, M.J., Barberá, R. & Romero, F. (2010). Polyphenolic profile and antiproliferative activity of bioaccessible fractions of zinc-fortified fruit beverages in human colon cancer cell lines. *Nutrición Hospitalaria*, **25**, 561–571.

Combs, G.F. Jr. (2012). *The Vitamins: Fundamental Aspects in Nutrition and Health*, 4th ed. Amsterdam: Elsevier.

Correa-Betanzo, J., Allen-Vercoe, E., McDonald, J., Schroeter, K., Corredig, M. & Paliyath, G. (2014). Stability and biological activity of wild blueberry (*Vacciniumangustifolium*) polyphenols during simulated *in vitro* gastrointestinal digestion. *Food Chemistry*, **15**, 522–531.

Crozier, A., Del Rio, D. & Clifford, M.N. (2010). Bioavailability of dietary flavonoids and phenolic compounds. *Molecular Aspects of Medicine*, **31**, 446–467.

Czank, C., Cassidy, A., Zhang, Q. *et al.* (2013). Human metabolism and elimination of the anthocyanin, cyanidin-3-glucoside: a ^{13}C-tracer study. *American Journal of Clinical Nutrition*, **97**, 995–1003.

Dall'Asta, M., Bresciani, L., Calani, L. *et al.* (2016). In vitro bioaccessibility of phenolic acids from a commercial aleurone-enriched bread compared to a whole grain bread. *Nutrients*, **8**, 1–9.

D'Archivio, M., Filesi, C., Varì, R., Scazzocchio, B. & Masella, R. (2010). Bioavailability of the polyphenols: status and controversies. *International Journal of Molecular Sciences*, **11**, 1321–1342.

Del Rio, D., Calani, L., Cordero, C., Salvatore, S., Pellegrini, N. & Brighenti, F. (2010). Bioavailability and catabolism of green tea flavan-3-ols in humans. *Nutrition*, **26**, 1110–1116.

Del Rio, D., Rodriguez-Mateos, A., Spencer, J.P.E., Tognolini, M., Borges, G. & Crozier, A. (2013). Dietary (poly)phenolics in human health: structures, bioavailability, and evidence of protective effects against chronic diseases. *Antioxidants & Redox Signaling*, **18**, 1818–1892.

Dia, V.P., Bringe, N.A. & de Majia, E.G. (2014). Peptides in pepsin-pancreatin hydrolysates from commercially available soy products that inhibit lipopolysaccharide-induced inflammation in macrophages. *Food Chemistry*, **152**, 423–431.

Díaz-de-Cerio, E., Verardo, V., Gómez-Caravaca, A.M., Fernández-Gutiérrez, A. & Segura-Carretero, A. (2016). Exploratory characterization of phenolic compounds with demonstrated anti-diabetic activity in guava leaves at different oxidation states. *International Journal of Molecular Sciences*, **17**, 1–13.

During, A., Dawson, H.D. & Harrison, E.H. (2005). Carotenoid transport is decreased and expression of the lipid transporters SR-BI, NPC1L1, and ABCA1 is down regulated in Caco-2 cells treated with ezetimibe. *Journal of Nutrition*, **135**, 2305–2312.

Espín, J.C., Larrosa, M., García-Conesa, M.T. & Tomás-Barberán, F. (2013). Biological significance of urolithins, the gut microbial ellagic acid-derived metabolites: the evidence so far. *Evidence-Based Complementary and Alternative Medicine*, **2013**(15), Article ID 270418.

Fang, J. (2014). Bioavailability of anthocyanins. *Drug Metabolism Reviews*, **46**, 508–520.

Fernandes, A., Brás, N.F., Mateus, N. & de Freitas, V. (2014). Understanding the molecular mechanism of anthocyanin binding to pectin. *Langmuir*, **30**, 8516–8527.

Ferreyra, F.M.L., Riusand, S.P. & Casati, P. (2012). Flavonoids: biosynthesis, biological functions, and biotechnological applications. *Frontiers of Plant Science*, **3**, 2–15.

Foltz, M., van der Pijl, P.C. & Duchateau, G.S. (2010). Current *in vitro* testing of bioactive peptides is not valuable. *The Journal of Nutrition*, **140**, 117–118.

Garcia, M.C., Puchalska, P., Esteve, C. & Marina, M.L. (2013). Vegetable foods: a cheap source of proteins and peptides with antihypertensive, antioxidant, and other less occurrence bioactivities. *Talanta*, **106**, 328–349.

García-Villalba, R., Espín, J.C. & Tomás-Barberán, F.A. (2016). Chromatographic and spectroscopic characterization of urolithins for their determination in biological samples after the intake of foods containing ellagitannins and ellagic acid. *Journal of Chromatography A*, **1428**, 162–175.

Gawlik-Dziki, U., Świeca, M., Sułkowski, M., Dziki, D., Baraniak, B. & Czyż, J. (2013). Antioxidant and anticancer activities of *Chenopodium quinoa* leaves extracts - in vitro study. *Food and Chemical Toxicology*, **57**, 154–160.

Gawlik-Dziki, U., Świeca, M., Dziki, D. *et al.* (2014). Anticancer and antioxidant activity of bread enriched with broccoli sprouts. *BioMed Research International*, **2014**, 14, Article ID 608053.

Gliszczyńska-Świgło, A. (2006). Antioxidant activity of water soluble vitamins in the TEAC (trolox equivalent antioxidant capacity) and the FRAP (ferric reducing antioxidant power) assays. *Food Chemistry*, **96**, 131–136.

González-Barrio, R., Borges, G., Mullen, W. & Crozier, A. (2010). Bioavailability of anthocyanins and ellagitannins following consumption of raspberries by healthy humans and subjects with an ileostomy. *Journal of Agricultural and Food Chemistry*, **58**, 3933–3939.

González-Sarrías, A., García-Villalba, R., Núñez-Sánchez, M.Á. *et al.* (2015). Identifying the limits for ellagic acid bioavailability: a crossover pharmacokinetic study in healthy volunteers after consumption of pomegranate extracts. *Journal of Functional Foods*, **19**, 225–235.

Guo, Y., Mah, E., Davis, C.G. *et al.* (2013). Dietary fat increases quercetin bioavailability in overweight adults. *Molecular Nutrition and Food Research*, **57**, 896–905.

Guo, L., Harnedy, P.A., Li, B. *et al.* (2014). Food protein-derived chelating peptides: biofunctional ingredients for dietary mineral bioavailability enhancement. *Trends in Food Science & Technology*, **37**, 92–105.

Hugenholtz, J. & Smid, E.J. (2002). Nutraceutical production with food grade microorganisms. *Current Opinion in Biotechnology*, **13**, 497–507.

Jaganath, I.B. & Crozier, A. (2010). Dietary flavonoids and phenolic compounds. In: *Plant Phenolics and Human Health: Biochemistry, Nutrition, and Pharmacology, Vol. 1*. (edited by C.G. Fraga) Pp. 1–50: John Wiley & Sons Inc.

Jakobek, L. (2015). Interactions of polyphenols with carbohydrates, lipids and proteins. *Food Chemistry*, **175**, 556–567.

Jakubczyk, A. & Baraniak, B. (2014). Angiotensin I converting enzyme inhibitory peptides obtained after in vitro hydrolysis of pea (*Pisum sativum* var. Bajka) globulins. *BioMed Research International*, **2014**(8), Article ID 438459.

Jayawardena, N., Watawana, M.I., Jayathilaka, R.T. & Waisundara, V.Y. (2015). The antioxidant and starch hydrolase inhibitory activity of ten spices in an *in vitro* model of digestion: bioaccessibility of anthocyanins and carotenoids. *Evidence-Based Complementary and Alternative Medicine*, **2015**(9), Article ID 764238.

Joshi, R., Adhikari, S., Patro, B.S., Chattopadhyay, S. & Mukherjee, T. (2001). Free radical scavenging behavior of folic acid: evidence for possible antioxidant activity. *Free Radical Biology and Medicine*, **30**, 1390–1399.

Jung, I.L. & Kim, J.G. (2003). Thiamine protects against paraquat-induced damage: scavenging activity of reactive oxygen species. *Environmental Toxicology and Pharmacology*, **15**, 19–26.

Kalt, W., Liu, Y., McDonald, J.E., Vinqvist-Tymchuk, M.R. & Fillmore, S.A.E. (2016). Anthocyanin metabolites are abundant and persistent in human urine. *Journal of Agricultural and Food Chemistry*, **62**, 3926–3934.

Kamanna, V.S. & Kashyap, M.E. (2000). Mechanism of action of niacin on lipoprotein metabolism. *Current Atherosclerosis Reports*, **2**, 36–46.

Kamiloglu, S., Capanoglu, E., Grootaert, C. & Camp, J.V. (2015). Anthocyanin absorption and metabolism by human intestinal caco-2 cells - A review. *International Journal of Molecular Sciences*, **16**, 21555–21574.

Karaś, M., Baraniak, B., Rybczyńska, K., Gmiński, J., Gaweł-Bęben, K. & Jakubczyk, A. (2015). The influence of heat treatment of chickpea seeds on antioxidant and fibroblast growth-stimulating activity of peptide fractions obtained from proteins digested under simulated gastrointestinal conditions. *International Journal of Food Science & Technology*, **50**, 2097–2103.

Lafay, S. & Gil-Izquierdo, A. (2008). Bioavailability of phenolic acids. *Phytochemistry Review*, **7**, 301–311.

Landete, J.M. (2012). Updated knowledge about polyphenols: functions, bioavailability, metabolism, and health. *Critical Review of Food Science and Nutrition*, **52**, 936–948.

Lin, Y., Wang, Y.H., Yang, X.Q., Guo, J. & Wang, J.M. (2016). Corn protein hydrolysate as a novel nano-vehicle: enhanced physicochemical stability and *in vitro* bioaccessibility of vitamin D_3. *LWT-Food Science and Technology*, **72**, 510–517.

López-Barrios, L., Antunes-Ricardo, M. & Gutiérrez-Uribe, J.A. (2016). Changes in antioxidant and antiinflammatory activity of black bean (*Phaseolus vulgaris* L.) protein isolates due to germination and enzymatic digestion. *Food Chemistry*, **203**, 417–424.

Maeno, M., Yamamoto, N. & Takano, T. (1996). Identification of an antihypertensive peptide from casein hydrolysate produced by a proteinase from *Lactobacillus helveticus* CP790. *Journal of Dairy Science*, **79**, 1316–1321.

Maestri, E., Marmiroli, M. & Marmiroli, N. (2016). Bioactive peptides in plant-derived foodstuffs. *Journal of Proteomics*, **147**, 140–155.

Mäkinen, S., Streng, T., Larsen, L.B., Laine, A. & Pihlanto, A. (2016). Angiotensin I-converting enzyme inhibitory and antihypertensive properties of potato and rapeseed protein-derived peptides. *Journal of Functional Foods*, **25**, 160–173.

Marhuenda, J., Alemán, M. D., Gironés-Vilaplana, A. *et al.* (2016). Phenolic composition, antioxidant activity, and *in vitro* availability of four different berries. *Journal of Chemistry*, **2016**, 7 Article ID 5194901.

Marín, L., Miguélez, E.M., Villar, C.J. & Lombó, F. (2015). Bioavailability of dietary polyphenols and gut microbiota metabolism: antimicrobial properties. *BioMed Research International*, **2015** (18), Article ID 905215.

McClements, D.J. & Xiao, H. (2014). Excipient foods: designing food matrices that improve the oral bioavailability of pharmaceuticals and nutraceuticals. *Food & Function*, **5**, 1320–1333.

Menéndez-Carreño, M., Ansorena, D., Milagro, F.I., Campión, J., Martínez, J.A. & Astiasarán, I. (2008). Inhibition of serum cholesterol oxidation by dietary vitamin C and selenium intake in high fat fed rats. *Lipids*, **43**, 383–390.

Misumida, N., Umeda, H. & Iwase, M. (2014). Shoshinberi-beri induced by long-term administration of diuretics: a case report. *Case Reports in Cardiology*, **2014**, 878915.

Moller, N.P., Scholz-Ahrens, K.E., Roos, N. & Schrezenmeir, J. (2008). Bioactive peptides and proteins from foods: indication for health effects. *European Journal of Nutrition*, **47**, 171–182.

Nizamova, A.M., Ziyatdinova, G.K. & Budnikov, G.K. (2011). Electrogenerated bromine as a coulometric reagent for the estimation of the bioavailability of polyphenols. *Journal of Analytical Chemistry*, **66**, 308–316.

Norman, A.W. (2006). Mini review: vitamin D receptor: new assignments for an already busy receptor. *Endocrinology*, **147**, 5542–5548.

Olthof, M.R., Hollman, P.C., Zock, P.L. & Katan, M.B. (2016). Consumption of high doses of chlorogenic acid, present in coffee or black tea increases plasma total homocysteine concentrations in humans. *American Journal of Clinical Nutrition*, **73**, 532–538.

Ortega, N., Reguant, J., Romero, M.P., Macia, A., & Motilva, M.J. (2009). Effect of fat content on the digestibility and bioaccessibility of cocoa polyphenol by an *in vitro* digestion model. *Journal of Agricultural and Food Chemistry*, **57**, 5743–5749.

Ortigoza-Escobar, J.D., Serrano, M., Molero, M. *et al.* (2014). Thiamine transporter-2 deficiency: outcome and treatment monitoring. *Orphanet Journal of Rare Diseases*, **9**, 92.

Ozdal, T., Sela, D. A., Xiao, J., Boyacioglu, D., Chen, F. & Capanoglu, E. (2016). The reciprocal interactions between polyphenols and gut microbiota and effects on bioaccessibility. *Nutrients*, **8**, 78.

Palafox-Carlos, H., Ayala-Zavala, J.F. & González-Aguilar, G.A. (2011). The role of dietary fiber in the bioaccessibility and bioavailability of fruit and vegetable antioxidants. *Journal of Food Science*, **76**, R6–R15.

Pavan, V., Sancho, R.A.S. & Pastore, G.M. (2014). The effect of *in vitro* digestion on the antioxidant activity of fruit extracts (*Carica papaya, Artocarpusheterophillus* and *Annona marcgravii*). *LWT - Food Science and Technology*, **59**, 1247–1251.

Podsędek, A., Redzynia, M., Klewicka, E. & Koziołkiewicz, M. (2014). Matrix effects on the stability and antioxidant activity of red cabbage anthocyanins under simulated gastrointestinal digestion. *BioMed Research International*, **2014**, 1–11.

Quirós-Sauceda, A.E., Palafox-Carlos, H., Sáyago-Ayerdi, S.G. *et al.* (2014). Dietary fibre and phenolic compounds as functional ingredients: interaction and possible effect after ingestion. *Food and Function*, **5**, 1063–1072.

Ramanujam, V.M., Anderson, K.E., Grady, J.J., Nayeem, F. & Lu, L.J. (2011). Riboflavin as an oral tracer for monitoring compliance in clinical research. *The Open Biomarkers Journal*, **2011**, 1–7.

Ravisankar, S., Gutierrez, D.C., Chirinos, R. & Noratto, G. (2015). Quinoa (Chenopodium Quinoa) peptides protect human umbilical vein endothelial cells (HUVEC) against risk markers for cardiovascular disease (CVD). *The FASEB Journal*, **29**, 923–10.

Renukuntla, J., Vadlapudi, A.D., Patel, A., Boddu, S.H. & Mitra, A.K. (2013). Approaches for enhancing oral bioavailability of peptides and proteins. *International Journal of Pharmaceutics*, **15**, 75–93.

Ribnicky, D.M., Roopchand, D.E., Poulev, A. *et al.* (2014). *Artemisia dracunculus* L. polyphenols complexed to soy protein show enhanced bioavailability and hypoglycemic activity in C57BL/6 mice. *Nutrition*, **30**, S4–S10.

Ricciardi, C.J., Bae, J., Esposito, D. *et al.* (2015). 1, 25-Dihydroxyvitamin D_3/vitamin D receptor suppresses brown adipocyte differentiation and mitochondrial respiration. *European Journal of Nutrition*, **54**, 1001–1012.

Rodriguez-Meléndez, R., Cano, S., Mendez, S.T. & Velazquez, A. (2001). Biotin regulates the genetic expression of holocarboxylasesynthetase and mitochondrial carboxylases in rats. *Journal of Nutrition*, **131**, 1909–1913.

Rodríguez-Roque, M.J., Rojas-Graü, M.A., Elez-Martínez, P. & Martín-Belloso, O. (2014a). *In vitro* bioaccessibility of health related compounds as affected by the formulation of fruit juice- and milk-based beverages. *Food Research International*, **62**, 771–778.

Rodríguez-Roque, M.J., Rojas-Graü, M.A., Elez-Martínez, P. & Martín-Belloso, O. (2014b). *In vitro* bioaccessibility of health-related compounds from a blended fruit juice-soymilk beverage: influence of the food matrix. *Journal of Functional Foods*, **7**, 161–169.

Rodríguez-Roque, M.J., de Ancos, B., Sánchez-Moreno, C., Cano, M.P., Elez-Martínez, P. & Martín-Belloso, O. (2015). Impact of food matrix and processing on the *in vitro* bioaccessibility of vitamin C, phenolic compounds, and hydrophilic antioxidant activity from fruit juice-based beverages. *Journal of Functional Foods*, **14**, 33–43.

Rui, X., Boye, J.I., Simpson, B.K. & Prasher, S.O. (2013). Purification and characterization of angiotensin I-converting enzyme inhibitory peptides of small red bean (*Phaseolus vulgaris*) hydrolysates. *Journal of Functional Foods*, **5**, 1116–1124.

Saari, J.C., Nawrot, M., Kennedy, B.N. *et al.* (2001). Visual cycle impairment in cellular retinaldehyde binding protein (CRALBP) knockout mice results in delayed dark adaptation. *Neuron*, **29**, 739–748.

Sabbione, A.C., Nardo, A.E., Añón, M.C. & Scilingo, A. (2016). Amaranth peptides with antithrombotic activity released by simulated gastrointestinal digestion. *Journal of Functional Foods*, **20**, 204–214.

Sato, K., Egashira, Y., Ono, S. *et al.* (2013). Identification of a hepatoprotective peptide in wheat gluten hydrolysate against D-galactosamine-induced acute hepatitis in rats. *Journal of Agricultural and Food Chemistry*, **61**, 6304–6310.

Saura-Calixto, F. (2011). Dietary fiber as a carrier of dietary antioxidants: an essential physiological function. *Journal of Agricultural and Food Chemistry*, **59**, 43–49.

Segura-Campos, M., Chel-Guerrero, L., Betancur-Ancona, D. & Hernandez-Escalante, V.M. (2011). Bioavailability of bioactive peptides. *Food Review International*, **27**, 213–226.

Shimizu, M., Tsunogai, M. & Arai, S. (1997). Transepithelial transport of oligopeptides in the human intestinal cell, Caco-2. *Peptides*, **18**, 681–687.

Sienkiewicz-Szłapka, E., Jarmołowska, B., Krawczuk, S., Kostyra, E., Kostyra, H. & Bielikowicz, K. (2009). Transport of bovine milk-derived opioid peptides across a Caco-2 monolayer. *International Dairy Journal*, **19**, 252–257.

Stoupi, S., Williamson, G., Drynan, J.W., Barron, D. & Clifford, M.N. (2010). A comparison of the *in vitro* biotransformation of (–)-epicatechin and procyanidin B2 by human faecal microbiota. *Molecular Nutrition and Food Research*, **54**, 747–759.

Sui, X., Zhang, Y. & Zhou, W. (2016). Bread fortified with anthocyanin-rich extract from black rice as nutraceutical sources: its quality attributes and *in vitro* digestibility. *Food Chemistry*, **196**, 910–916.

Świeca, M., Gawlik-Dziki, U., Dziki, D., Baraniak, B. & Czyż, J. (2013). The influence of protein–flavonoid interactions on protein digestibility *in vitro* and the antioxidant quality of breads enriched with onion skin. *Food Chemistry*, **141**, 451–458.

Tapal, A., Vegarud, G.E., Sreedhara, A., Hegde, P., Inamdar, S. & Tiku, P.K. (2016). *In vitro* human gastro-intestinal enzyme digestibility of globulin isolate from oil palm (*Elaeisguineensis* var. tenera) kernel meal and the bioactivity of the digest. *RSC Advances*, **6**, 20219–20229.

Terentis, A.C., Thomas, S.R., Burr, J.A., Liebler, D.C. & Stocker, R. (2002). Vitamin E oxidation in human atherosclerotic lesions. *Circulation Research*, **90**, 333–339.

Tomás-Barberán, F.A., García-Villalba, R., González-Sarrías, A., Selma, M.V. & Espín, J.C. (2014). Ellagic acid metabolism by human gut microbiota: consistent observation of three urolithin phenotypes in intervention trials, independent of food source, age, and health status. *Journal of Agricultural and Food Chemistry*, **62**, 6535–6538.

Udenigwe, C. & Aluko, R. (2012). Food protein-derived bioactive peptides: production, processing, and potential health benefits. *Journal of Food Science*, **77**, 11–24.

Van Platernik, C., Janssen, H.-G., Horsten, R. & Haverkamp, J. (2006). Quantification of ACE inhibiting peptides in human plasma using high performance liquid chromatography-mass spectrometry. *Journal of Chromatography B*, **830**, 151–157.

Velderrain-Rodríguez, G., Ouirós-Sauceda, A., Mercado-Mercado, G., et al. (2016). Effect of dietary fiber on the bioaccessibility of phenolic compounds of mango, papaya and pineapple fruits by an

in vitro digestion model. *Food Science and Technology*, **36**, 188–194.

Verhaar, M.C., Stroes, E. & Rabelink, T.J. (2002). Folates and cardiovascular disease. *Arteriosclerosis Thrombosis and Vascular Biology*, **22**, 6–13.

Vermeer, C. & Theuwissen, E. (2011). Vitamin K, osteoporosis and degenerative diseases of ageing. *Menopause International*, **17**, 19–23.

Widmaier, E., Strang, K. & Raff, H. (2013). *Vander's Human Physiology: The Mechanisms of Body Function*, 13th ed. P. 562 McGraw-Hill Science Engineering Math.

Wortmann, M., Schneider, M., Pircher, J. *et al*. (2013). Combined deficiency in glutathione peroxidase 4 and vitamin E causes multi-organ thrombus formation and early death in mice. *Circulation Research*, **113**, 408–417.

Wu, Z., Teng, J., Huang, L., Xia, N. & Wei, B. (2015). Stability, antioxidant activity and *in vitro* bile acid-binding of green, black and dark tea polyphenols during simulated *in vitro* gastrointestinal digestion. *RSC Advances*, **5**, 92089–92095.

Yin, N., Brimble, M.A., Harris, P.W. & Wen, J. (2014). Enhancing the oral bioavailability of peptide drugs by using chemical modification and other approaches. *Medicinal Chemistry*, **4**, 763–769.

Yoshikawa, T., Inoue, R., Matsumoto, M., Yajima, T., Ushida, K. & Iwanaga, T. (2011). Comparative expression of hexose transporters (SGLT1, GLUT1, GLUT2 and GLUT5) throughout the mouse gastrointestinal tract. *Histochemistry and Cell Biology*, **135**, 183–194.

Yu, G., Wang, F., Zhang, B. & Fan, J. (2016). *In vitro* inhibition of platelet aggregation by peptides derived from oat (*Avena sativa* L.), highland barley (*Hordeum vulgare* Linn. var. nudum Hook. f.), and

buckwheat (*Fagopyrum esculentum Moench*) proteins. *Food Chemistry*, **194**, 577–586.

Zamora-Ros, R., Andres-Lacueva, C., Lamuela-Raventós, R.M. *et al*. (2008). Concentrations of resveratrol and derivatives in foods and estimation of dietary intake in a Spanish population: European Prospective Investigation into Cancer and Nutrition (EPIC)-Spain cohort. *British Journal of Nutrition*, **100**, 188–196.

Zanotti, I., Dall'Asta, M., Mena, P. *et al*. (2014). Atheroprotective effects of (poly)phenols: a focus on cell cholesterol metabolism. *Food and Function*, **6**, 13–31.

Zhang, S., Hunter, D.J., Hankinson, S.E. *et al*. (1999). A prospective study of folate intake and the risk of breast cancer. *Journal of the American Medical Association*, **281**, 1632–1637.

Zhang, K., Huentelman, M.J., Rao, F., Sun, E.I., Corneveaux, J.J. & Schork, A.J. (2014). International consortium for blood pressure genome-wide association studies, genetic implication of a novel thiamine transporter in human hypertension. *Journal of the American College of Cardiology*, **63**, 1542–1555.

Zou, T.B., Feng, D., Song, G., Li, H.W., Tang, H.W. & Ling, W.H. (2014). The role of sodium-dependent glucose transporter 1 and glucose transporter 2 in the absorption of cyanidin-3-*O*-β-glucoside in Caco-2 cells. *Nutrients*, **6**, 4165–4177.

Zou, L., Liu, W., Liu, C., Xiao, H. & McClements, D.J. (2015). Designing excipient emulsions to increase nutraceutical bioavailability: emulsifier type influences curcumin stability and bioaccessibility by altering gastrointestinal fate. *Food & Function*, **6**, 2475–2486.

Zou, T.B., He, T.P., Li, H.B., Tang, H.W. & Xia, E.Q. (2016). The structure-activity relationship of the antioxidant peptides from natural proteins. *Molecules*, **21**, 72.