# BIOAVAILABILITY OF FLAVONOIDS AND POTENTIAL BIOACTIVE FORMS *IN VIVO*

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**SUMMARY**

Flavonoids are powerful antioxidants *in vitro*, but their overall functions *in vivo* have yet to be clarified, whether antioxidant, anti-inflammatory, enzyme inhibitor or inducer, or some other role. The reducing properties of flavonoids might also contribute to redox regulation in cells independently of their antioxidant properties. However, in order to understand their bioactivity *in vivo*, it is necessary to understand the factors influencing the absorption of flavonoids by the gastrointestinal tract, the nature of the conjugates and metabolites in the circulation and how this influences their antioxidant activities.

**KEY WORDS**

flavonoid, antioxidant activity, bioavailability, flavonoid metabolism, gastrointestinal absorption

## 1. INTRODUCTION

Fruit, vegetables, beverages and grains are rich in the phenolic families of antioxidant phytochemicals, the flavonoids and phenolics. Flavonoids represent the single, most widely occurring group of phenolic phytochemicals /1,2/. Recent work is beginning to highlight the potential health beneficial properties of the flavonoid and polyphenolic components of the diet, known to be powerful antioxidants *in vitro* /3-6/ and also believed to be capable of acting in redox-sensitive signalling cascades. Flavonoids are classified according to the oxidation level of their central C ring (Fig. 1). These variations define the four distinct families: anthocyanidin, flavone or flavonol (i.e. 3-hydroxyflavone), flavanone and proanthocyanidin oligomers of the monomeric flavan-3-ols. Most flavonoids are glycosylated in their natural forms with the exception of the catechin family. All fruit, vegetables, beverages and grains are rich in a variety of phenolic families, in particular, the (epi)catechin flavanol constituents and their oligomers in teas, red wine, apples and chocolate, the anthocyanins in berry fruit, the flavanones characteristic of citrus fruit and the flavone/flavonols in a number of fruit and vegetables (Fig. 1). In

addition, the fifth family of hydroxycinnamates, the simple phenolics, are major components of all fruit and some vegetables.

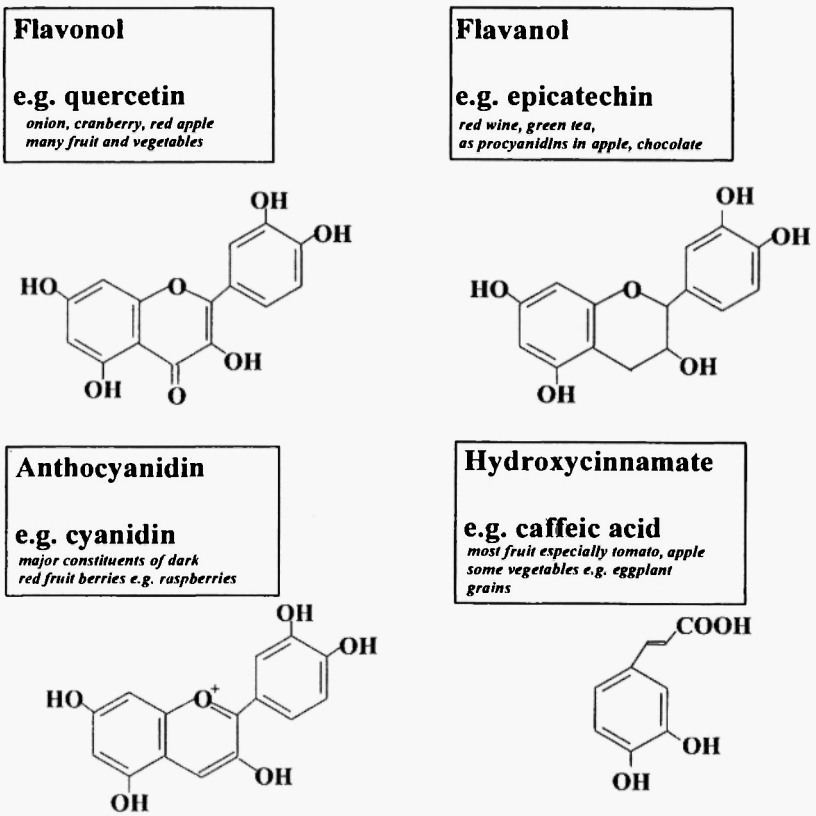


**Fig. 1:** The structural formulae of some flavonoid classes and hydroxycinnamates.

Little is known about the conjugated or metabolised forms of the dietary phenolics that might be responsible for their bioactivity *in vivo*. In order to exploit the proposed health beneficial effects of flavonoids, their roles in disease prevention and their possible applications to the functional food industry, we need to understand their physiological mechanisms of action and bioactivities *in vivo*. To do this more information is required on their bioavailability, their uptake into cells, metabolism and excretion, and the factors influencing the extent of their absorption. Furthermore the modifications which these compounds undergo in the gastrointestinal tract require elucidation.

Epidemiological studies associate flavonoid intake with reduction in risk of coronary heart disease and myocardial infarction, especially from sources such as apples, onions, teas and red wine /7-14/. Recent model studies also suggest that flavanol-rich tea extracts prevent corneal neovascularisation /15/ and decrease the susceptibility to arthritis in animal models of inflammatory polyarthritis /16/. Furthermore, long-term dietary supplementation with flavonoid-rich foods has shown retardation of the onset of age-related neuronal signal transduction and cognitive behavioural deficits in animals /17/.

A hypothesis can be readily formulated from *in vitro* studies about how flavonoids might act as antioxidants *in vivo*. However, without knowledge of the forms in which flavonoids and their metabolites circulate, it is not possible to translate *in vitro* observations into the reality of cells and tissues *in vivo*. In order to understand the potential for flavonoids to function as antioxidants *in vivo* we need to examine their bioavailability and interactions in the gastrointestinal tract. Furthermore, the influence of conjugation and metabolism needs to be assessed.

## 2. ANTIOXIDANT PROPERTIES OF FLAVONOIDS

Flavonoids are reported to function as antioxidant agents by scavenging reactive oxygen species (ROS) /3,18,19/, reactive nitrogen species (RNS) /4-6,20/, and in some instances chelating transition metal ions /21,22/, in a structure-dependent manner. Flavonoids are antioxidants by virtue of the H-donating properties of their phenolic hydroxyl groups attached to ring structures, dependent on their number and arrrangement. Their ability to act as antioxidants by donating an electron to an oxidant critically depends on the reduction potentials of their radicals /23/ and the accessibility of the radical. For example, flavonoids are ideal scavengers of peroxyl radicals due to their favourable reduction potentials relative to alkyl peroxyl radicals, and thus, in principle, are effective inhibitors of lipid peroxidation. The major structural characteristics which underlie their reducing properties are a catechol (3′,4′-dihydroxy) structure in the B-ring; a 2,3 double bond and a 3-hydroxyl group in the C ring may also contribute, the involvement of the 3-OH group in the planarity of the structure explaining the difference between quercetin and luteolin. Indeed, in the absence of conjugation across the phenyl rings, the

reduction potential of the flavonoid radical matches that of the ring with the lower reduction potential.

Many studies *in vitro* have clearly demonstrated the potent peroxyl radical scavenging abilities of flavonoids in inhibiting lipid peroxidation and oxidation of low density lipoproteins (LDL) /24-27/. However, it is not clear that the forms of flavonoid applied (mainly the aglycone) are relevant to their circulating forms *in vivo*. Since oxidation of LDL is implicated in the pathogenesis of coronary heart disease, a number of researchers have undertaken investigations examining the activity of dietary agents rich in flavonoids in inhibiting LDL oxidation *ex vivo*. The outcomes /28-32/ are ambiguous, protection in some instances, no effect in others, demonstrating that the situation is not clear cut /28-32/.

There have been few studies on the ability of flavonoids and phenolic acids to scavenge reactive nitrogen species. NO is such a species produced by the action of nitric oxide synthase (NOS) in endothelial cells, neurons, etc. At sites of inflammation iNOS is also induced and further NO synthesis activated. Concomitant production of nitric oxide and superoxide radical at such sites of chronic inflammation induces the production of peroxynitrite. Peroxynitrite is a toxic oxidising and nitrating species, produced by rapid interaction of superoxide radical and nitric oxide /33/. Phenolic compounds are especially susceptible to peroxynitrite-dependent reactions, e.g. the conversion of tyrosine to 3-nitrotyrosine has been postulated as a marker of peroxynitrite-dependent reaction in tissues /33/. For example, 3-nitrotyrosine has been detected in several disease states which are associated with increased NO production, including atherosclerosis, rheumatoid arthritis, inflammatory lung disorders, septic shock and gastritis. It is unclear whether nitrotyrosine contributes to the onset of these diseases or is formed as a consequence of the disease process due to the generation of RNS. Nitrated proteins are immunogenic and nitration can alter their function and stability, thus interfering with cell signalling pathways, cytoskeletal structures and repair mechanisms. Tyrosine nitration has been suggested to initiate the onset of the apoptotic process. Flavonoids, being polyphenolic, are ideal candidates for scavenging RNS so as to prevent peroxynitrite-dependent protein nitration. Recent research has shown that the activity of flavonoids to inhibit peroxynitrite-dependent nitration of tyrosine is structure dependent. Thus, catechol-containing phenolics

inhibit by scavenging peroxynitrite through electron donation /4,15/ whereas monohydroxycinnamates and flavonoids with monophenolic B-rings intercept the reaction between tyrosine and peroxynitrite via the anticipated mechanism of competitive nitration /5/.

Another potential source of RNS derives from dietary nitrite which can also be produced from dietary nitrate by the action of nitrate reductases. Nitrate reacts with the acidic gastric juice to produce nitrous acid, which decomposes to oxides of nitrogen. Nitrous acid and its products have been shown to nitrosate amines, deaminate DNA bases and nitrate aromatic compounds including tyrosine /34/. Several flavonoids and phenolic compounds are powerful inhibitors of nitrous acid-dependent nitration and DNA deamination *in vitro* /34/, and this role could also be significant *in vivo*. Thus flavonoids from fruit, vegetables, chocolate, tea and other beverages might provide a gastro-protective effect when high levels of reactive nitrogen species are generated.

Flavonoids and phenolic compounds can also interact with transition metal ions to form chelates, especially those with catechol groups, and 4-keto, 3-hydroxy,5-hydroxy structures. /21,22/. These chelates might be stable, or redox cycling might take place in the case of catechol B-rings leading to the reduction of iron or copper to a more pro-oxidant form and the oxidised quinone.

## 3. ABSORPTION

There is still much research required to understand whether polyphenols are absorbed *in vivo* in the natural form as glycosides or as aglycones after deglycosylation in the small intestine, the extent to which they may be conjugated as glucuronides, sulphates or methylated compounds in the small intestine, or whether they enter the colon to be metabolised through degradation by gut microflora (Fig. 2), and how this relates to their precise chemical structures. Absorbed flavonoids are also expected to be subject to various reactions in the liver, the chief organ for the metabolism of dietary flavonols /35/, where the capacities of flavonoid biotransformations, such as catechol-*O*-methyl transferase or various transferases catalysing conjugations with sulphate and glucuronic acid, are particularly active.

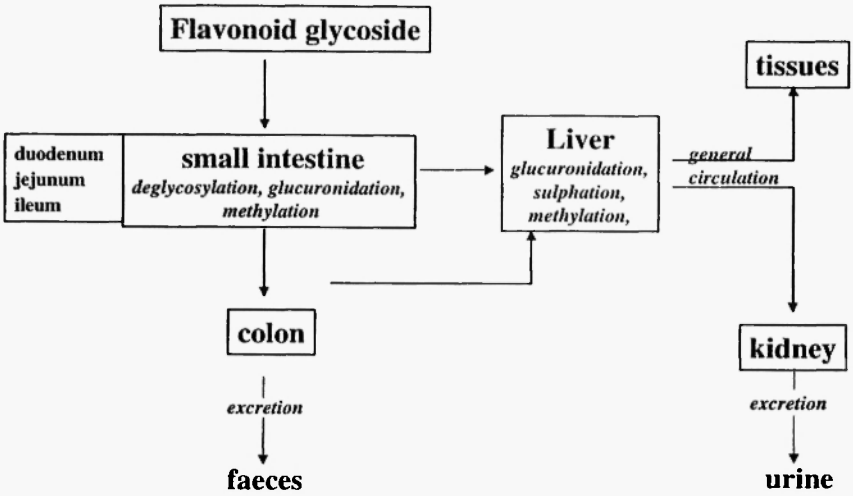


**Fig. 2:** Proposed *in vivo* route and metabolism of flavonoid glycosides.

## 4. INTERACTIONS IN THE SMALL INTESTINE

The understanding of the biotransformation of flavonoids in the small intestine and the liver and that of their absorbed colonic metabolites is crucial for their bioactivity. The most common routes of metabolism are likely to be oxidation of phenolic hydroxyl groups, hydroxylation, reduction or hydrolysis or deglycosylation (Phase I metabolism), followed by conjugation reactions (Phase II metabolism), or several competing pathways. Conjugation with glucuronic acid, sulphate, or *O*-methylation might occur. Conjugation reactions with glucuronic acid or sulphate are reported to be the most common final step in the metabolic pathway of flavonoids.

As mentioned earlier, it was assumed that flavonoid glycosides could not be absorbed from the small intestine and cleavage of the β-glycosidic linkage would not occur until compounds reach the microflora of the large intestine /36/. However, earlier proposals that only flavonoids in the aglycone form could be absorbed by the small intestine were consistent with the findings of Wahlgren *et al.* /37/, applying intestinal epithelial Caco-2 cells as a model of human intestine. These researchers examined the comparative transepithelial flux of the quercetin aglycone itself, in comparison with two of the major dietary forms, the 4′-monoglucoside and the 3,4′-diglucoside.

Facile absorption of quercetin itself occurred through human intestinal epithelium but the results did not support an active transport process for the quercetin glucosides. This contrasts with other studies in which quercetin and its 3-rhamnoglucoside, rutin, have been observed to move rapidly into Caco-2 cells /38/.

The presence of glycosidases in the small intestine has been demonstrated from several studies /39-41/. Certain monoglucosides were rapidly deglycosylated by cell-free extracts of human small intestine, the hydrolytic mechanism being attributed to a broad specificity β-glucosidase. This was not the case with diglycosides, such as rutin (quercetin rhamnoglucoside) and naringenin-7 rhamnoglucoside nor the 3-monoglucosides of quercetin and kaempferol /39/. Glucosidase enzymes have also been isolated from rat small intestinal mucosa of the duodenum, jejunum and ileum and reacted with the quercetin monoglucosides, quercetin-3-glucoside, quercetin-4′-glucoside and quercetin-7-glucoside, as well as with the diglucoside quercetin-3-rutinoside /40/. The enzyme activity of the jejunum was highest for all the glycosides tested and quercetin-4′-glucoside was the richest substrate, whereas rutin was a very poor substrate. This demonstrates that dietary flavonoid monoglucosides can be hydrolyzed and liberated as aglycones in the jejunum and that this may occur at different rates depending on the substrate.

In support of this our recent studies have shown that deglycosylation of monoglucosylated flavonoids can occur in the jejunal and ileal sections of the small intestine, followed by conjugation reactions such as glucuronidation /41/ and methylation of catechol-containing structures /42/. Indeed, a large number of conjugating enzymes are reported in the enterocytes of the small intestine, including UDP-glucuronyl transferases and catechol-*O*-methyl transferases /43/. Investigating a range of flavonoids and their common glycosides, glucuronidation occurs on absorption across the enterocytes of the jejunum, and this occurs after first being deglycosylated in the case of administered natural flavonoid glycosides /41/. The combined levels of glucuronide and parent compound detected vary widely and seem to be structure dependent.

In contrast, absorption of quercetin-3-glucoside via the small intestine did not fit this pattern /41/, the major component on uptake being the administered compound itself, with only a small proportion hydrolysed or glucuronidated. Consistent with these observations,

evidence is available from human studies challenging the concept that ingested flavonoid glycosides are first hydrolysed by the digestive microflora before being absorbed /44/. Indeed, evidence has been provided for preferential uptake of specific flavonol glycosides from the human small intestine compared to the aglycone. Quercetin glucosides and quercetin rhamnoglucoside (rutin) have been detected in humans /45,46/ and dietary glycosides of quercetin from onions have been shown to be absorbed more efficiently than free purified aglycone in healthy ileostomy patients /47/. Different dietary glycosides (from onions and apples) show different rates of absorption and bioavailability /46/. For example, rutin was found to be absorbed more slowly and less extensively than quercetin-3-monoglucoside by the small intestine, suggesting the possibility of degradation of rutin by glycosidases in the colon /48-51/.

Comparison of the absorption of quercetin and rutin in rats fed supplemented diets /48/ led to the conclusion that rutin was absorbed more slowly than quercetin since, according to the authors, it must be hydrolysed by the cecal microflora, whereas quercetin was absorbed from the small intestine. Thus quercetin appeared in detectable amounts in the blood circulation from the quercetin diet prior to the quercetin from the hydrolysed rutin source. Conjugated derivatives of quercetin, and its methylated forms isorhamnetin and tamarixetin, were recovered in plasma from rats receiving the experimental diets after the first meal but, after 10 days, no traces of tamarixetin were detected. Low rates of elimination of quercetin metabolites were observed and high plasma concentrations sustained through a continued supply of quercetin or rutin in the diet. Earlier studies of this group /49/ had established that 80% of circulating plasma quercetin is present as the *O*-methylated derivative, isorhamnetin, the flavonol metabolites circulating as conjugated forms.

Urinary elimination studies comparing oral with intravenous administration of flavonoids in rats over 24 h *in vivo* showed very limited recoveries of quercetin, its metabolites (especially tamarixetin and isorhamnetin, the methylated derivatives) and conjugates. This suggests that the majority is excreted in the bile and that extensive metabolism occurs even when bypassing the gut /52/. These findings are consistent with those in humans showing the recovery of quercetin conjugates, mainly as isorhamnetin and quercetin glucuronides and sulfates (in human plasma after ingestion of quercetin glycosides from

the diet), although circulating levels increased by only 0.2-0.3 μM /50/.

Several flavonoids with 3′,4′-dihydroxylation in the B-ring are excreted in mammalian species as conjugates of their 3′-*O*-methyl ethers. Glucuronide and sulphate conjugates of these methyl ethers are major urinary metabolites in man /53/. Identification of epicatechin metabolites and their fates in rats and humans reveal that the most predominant metabolites in plasma and urine are conjugates of epicatechin and 3′-*O*-methyl epicatechin /54,55/. Early studies of Hackett *et al.* /53/ following the quantitative disposition of 3-*O*-methyl-(+)-[$^{14}$C-U]-catechin in man on oral administration showed that the major urinary metabolites were glucuronides of 3,3′-*O*-dimethyl-(+)-catechin, 3-*O*-methyl-(+)-catechin glucuronide and sulphate (15.8%, 11.4% and 10.6% dose, respectively). No evidence for demethylation was obtained. Excretion of the unchanged compound accounted for an average of ~0.3% of the 2 g single dose. 3-*O*-Methyl-(+)-catechin was detected and measured in plasma with peak levels (11-18 μg/ml) attained within 2 h of administration.

## 5. STRUCTURAL DEPENDENCE OF ABSORPTION AND EXCRETION

Recent studies *in vitro* have shown that phenolics with a catechol structure in the B-ring are more potent antioxidants /3/ but also may be more extensively metabolised, in general, than those with less highly oxidisable arrangements of the phenolic hydroxyl groups /41,42,52, 56/. Thus, for example, whereas oral absorption of dietary quercetin and catechin in humans and rats is reported to be low and variable in relation to the amount ingested /52,54,55,57/, the less oxidisable structures of naringenin and ferulic acid are apparently more bioavailable from a variety of sources /58-61/. It must, however, be borne in mind that ferulic acid can arise from the metabolism of caffeoyl ester.

In contrast, urinary excretion of the phenolics and the conjugates of the aglycones of the non-catechol-containing phenolics ferulic acid and naringenin-7-glucoside gave comparable recoveries after oral versus intravenous administration to rats. About 15% of the original dose was recovered in the form of feruloyl glucuronide and naringenin glucuronide, showing more extensive absorption and less extensive metabolism, suggesting minimal cleavage or decomposition in the gastrointestinal tract /52,56/. These findings are supported by human

studies /58-61/ and substantiate the notion that the catechol-containing flavanols seem to be more extensively metabolised on absorption than non-catechol structures which are more extensively absorbed *in vivo*. These findings point to the suggestion that the most bioavailable phenolics are those with moderate antioxidant properties, whereas those with greater reducing properties (lower reduction potentials or more oxidisable, i.e. most potent antioxidants) appear to be less extensively absorbed in their native or aglycone forms or as conjugates and may be more likely to be modified prior to reaching the circulation.

## 6. METABOLISM

It is assumed that the major component of ingested flavonoids (>80%) may be the consequences of degradation by the colonic microflora rather than being absorbed via the small intestine. Bacterial enzymes have the potential to catalyse several reactions including hydrolysis, dehydroxylation, hydroxylation, *O*-methylation, ring cleavage and decarboxylation. Current knowledge of flavonoid metabolism emanates from the pioneering studies of Booth *et al.* in the 1950s /62-64/ and the continued work of Hackett *et al.* /53/ that demonstrated the catabolism and scission of the flavonoid rings in animal studies. Evidence also suggests that the enzymes responsible for the initial ring fission of specific flavonoids and demethylation and dehydroxylation of the resulting phenolic acids are, to a great extent, enzymes of intestinal microorganisms. Fission of the flavonoid structure can occur as shown in Figure 3 /65-67/ and this depends on their hydroxylation pattern. Absence of hydroxyl groups in the B-ring prevents ring fission. Chrysin, the simplest flavone with an unsubstituted B-ring, is generally resistant to scission of the heterocyclic ring system. Free 5,7-dihydroxylation in the presence of *O*-methyl substitution in these positions increases the resistance to ring fission. In contrast, a monohydroxy-substituted 4′-position in the B-ring favours ring cleavage, apigenin (4′-hydroxychrysin) forming *p*-hydroxyphenylpropionic acid, *p*-hydroxybenzoic acid (leading to the potential for hydroxyhippuric acid formation in the liver), *p*-coumaric acid (and some uncleaved glucuronide of apigenin).

The aglycones of the most widespread dietary flavonoids, catechin and quercetin, have a 5,7,3′,4′-hydroxylation pattern that would, on

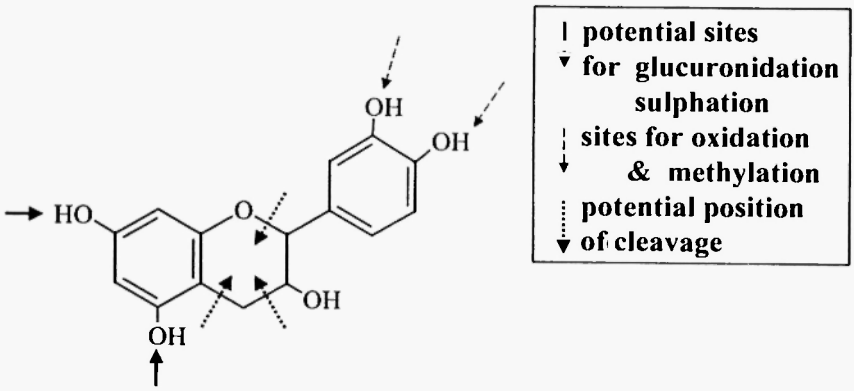


**Fig. 3:** Potential metabolic sites on the flavanol molecule.

this basis alone, enhance ring opening after hydrolysis. Furthermore, collectively all studies have shown the dependence of the pathway of ring scission on the type and extent of oxidation of the carbon atoms of the heterocyclic ring /65,68,69/. The metabolism and degradation pathways for quercetin and its cleavage products have been derived from a plethora of investigations, including studies on the metabolic fate of $^{14}$C-labelled quercetin in animals. Urinary and biliary metabolites are the glucuronide and sulphate conjugates of [$^{14}$C]-quercetin, 3′-*O*-methyl-[$^{14}$C]-quercetin (isorhamnetin) and 4′-O-methyl-[$^{14}$C]-quercetin (tamaraxetin). Previous studies /70/ did not detect measurable plasma or urinary concentrations of quercetin in man after oral administration, and it was deduced that the flavonoid was extensively degraded by microorganisms in the gut. Other studies of oral administration of [$^{14}$C]-quercetin to rats resulted in urinary excretion of the following metabolites believed to derive from the B-ring, not the A-ring: 3,4-dihydrophenylacetic acid, 3-hydroxyphenylacetic acid, 4-hydroxy,3-methoxyphenylacetic acid (and their conjugates) /68/. Many studies on the oral administration of quercetin or its most common glycoside, rutin, in rats and humans, have shown almost complete degradation by intestinal bacteria, yielding phenolic acids by fission of the C-ring /69/. Metabolites in humans were identified as 3,4-dihydroxyphenylacetic acid, 3-hydroxy phenylacetic acid, homovanillic acid and 3,4-dihydroxytoluene, accounting for over 50% of the dose administered /62,71-74/. Other human studies have detected low levels of quercetin glycosides /45,46/, and in some cases the aglycone. Others suggest that degradation to benzoic acid derivatives

rather than phenylacetic acid derivatives is a predominant route of metabolism /75/. This may be accounted for by the metabolism of different flavonoid structures or differences in individual gut flora or dietary considerations /76/.

The major metabolites arising from the oral administration of flavones/flavonones (lacking the 3-hydroxyl group) to animals were phenylacetic acids and hydroxyphenylpropionic acids, derived from cleavage of the B-ring. In contrast, administered flavanols such as catechin/epicatechin (with 3-OH group but lacking C-4 carbonyl group) degraded to phenyl γ-valerolactones as well as phenylpropionic acids derived from fission of the A-ring (reviewed in /69,77/). Thus a variety of cleavage and scission products might be excreted in the urine. Phenylpropionic acids have also been suggested to undergo β-oxidation to benzoic acids, which might form hippuric acids on interaction with glycine in the liver. Another important aspect of biotransformation derives from model studies using rat liver microsomes that investigated the *in vitro* metabolism of flavonoids. The conclusions were that the B-ring was the key structural component undergoing biotransformation. Thus, flavonoids lacking hydroxyl groups in the B-ring or with a lone B-ring 4′-hydroxyl group were hydroxylated by microsomal cytochrome P450 enzymes to the corresponding catechol structure, and flavonoids with a methoxy group in the 4′-position were demethylated to the corresponding hydroxylated compound /65/.

This is consistent with previous *in vivo* studies showing the oxidation of chrysin to apigenin and the hydroxylation of naringenin to eriodictyol (from detection of eriodictyol glucuronide). Flavone was also hydroxylated to several metabolites, including 4′-hydroxyflavone and 3′,4′-dihydroxyflavone, which were excreted following flavone administration in animals /69/. Certain compounds, although possessing similar carbon skeletons to the readily metabolised flavonoids, have been found to be resistant to ring scission. A structural modification which appears to decrease susceptibility to ring scission includes the absence of a 5-OH group; for example, apigenin (4′,5,7-trihydoxyisoflavone) but not (4′,7-dihydroxyflavone) undergoes cleavage. 5-Methoxyquercetin also apparently resists ring scission in rats /69/.

Other specific metabolites have been defined in relation to oral administration of a variety of phenolics. Hippuric acid, as mentioned

above, is the product of glycine conjugation with benzoic acid /78/, a physiologically important mechanism for the elimination and detoxification of aromatic carboxylic acids, quinic acid and catecholamines /79/. Benzoic acid is primarily derived from plant phenolics and aromatic amino acids through the action of intestinal bacteria and thus high levels of hippuric acid are expected in the urine of individuals consuming diets rich in fruit and vegetables /80/ and black tea /81/. It has been suggested that less urinary elimination of hippuric acid can be accounted for by 3-hydroxyphenylpropionic acid, due to dietary influences on enzymes involved in glycine conjugation processes /76/. Other hydroxyhippuric acids, such as 4-hydroxy-, 3-methoxy-, 4-methoxy-, 3-hydroxy- and 2-hydroxyhippuric acid, seem to be possible and likely.

## 7. CONCLUSIONS

Until flavonoid absorption and bioavailability are clearly understood, and the nature of their circulating metabolites identified, the question of the bioactivities of flavonoids as antioxidants *in vivo* will not be resolved. Furthermore, the concentrations reaching the tissues and the way the structural modifications influence the antioxidant properties require elucidation. For example, methylation or glucuronidation of catechins on one of the hydroxyl moieties that contributes to the catechol group will modulate the antioxidant properties.

Concerning a role for dietary flavonoids as antioxidants *in vivo* it is necessary to understand the chemical nature of the absorbed forms circulating *in vivo* and how the multiplicity of research findings *in vitro* reflect the bioactivity of flavonoids *in vivo*. Only when we gain adequate information on the circulating forms can we begin to understand the targeting to the tissues, whether flavonoids cross the blood-brain barrier, for example, and in what forms. (Interestingly, there are receptors for glucuronides of morphine at the blood-brain barrier.) Flavonoids are powerful antioxidants *in vitro*, but their overall function *in vivo* has yet to be clarified, whether antioxidant, anti-inflammatory, enzyme inhibitor, enzyme inducer, signal transducer, inhibitor of cell division, or some other role. It should also be emphasised that the reducing properties of flavonoids may contribute to redox regulation in cells, independently of their antioxidant

properties, which thus may protect against cell ageing, for example, by working together with the intracellular reductant network.

To gain understanding of these issues the factors influencing the absorption of flavonoids in the gastrointestinal tract need to be addressed, namely the questions of deglycosylation before absorption, conjugation in the small intestine through glucuronidation, sulphation or methylation, etc., metabolism and degradation in the colon to smaller phenolic molecules. The forms in which they circulate *in vivo* will influence their polarity and, thus, their localization and bioactivities *in vivo*. Finally, if antioxidant activities are important, the elucidation is required of how such properties *in vitro* relate to the potential for conjugates and metabolites *in vivo* to act as antioxidants. The absorbed flavonoid components may function as an antioxidant in the aqueous phase (like vitamin C) or in the lipophilic milieu (as vitamin E). This will depend on their polarity properties on uptake, how they are metabolised on absorption, and their resulting structural forms in the circulation. Little is known about the relationship between intake (mainly in the glycosidic form, except for catechins) and uptake in the circulation.

Interest is currently focusing on the influence of dietary components on gastrointestinal metabolism and transport of drugs, especially ignited more recently by the finding that flavanone components of grapefruit juice can increase the bioavailability of certain drugs by reducing intestinal metabolism. In particular, the effects of naringin on the activity of CYP3A4 on drug absorption via interactions with intestinal P-glycoprotein are being explored /82/. It has also been proposed that the efficacy of flavonoids in terms of health benefits may relate to their influence, or that of their intestinal metabolites, on gut microflora. Such studies have reported beneficial effects on *Helicobacter pylori* /83,84/.

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