REVIEW

Resveratrol bioavailability and toxicity in humans

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Numerous data are now available on the beneficial properties of the polyphenolic compound resveratrol including its anti-inflammatory and antitumor effects. However, few studies have been performed with resveratrol in humans, and the results of these studies appear fragmentary and sometimes contradictory due to variations in conditions of administration, protocols and methods of assessment. This review article presents the results of recent studies investigating the pharmacokinetics, bioavailability, and toxicity of resveratrol in humans. Resveratrol is well absorbed, rapidly metabolized, mainly into sulfo and glucuronides conjugates which are eliminated in urine. Resveratrol seems to be well tolerated and no marked toxicity was reported. These data are important in the context of human efficacy studies, and they provide further support for the use of resveratrol as a pharmacological drug in human medicine.

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1 Introduction

Since its discovery, resveratrol, a polyphenol present in several plants including grapes, has been shown to have a number of physiological properties that could be useful in human medicine. Scientists have been aware of resveratrol for several decades and have regained interest in this molecule at the beginning of the 1990s when it was first reported to be present in red wine [1], leading to speculation that resveratrol might contribute at least in part to the ''French paradox'' [2]. This paradox results from the observation that mortality from cardiovascular disease is relatively low in France, despite high consumption of saturated fats, perhaps in relation to moderate consumption of red wine (*i.e.* 2–3 glasses/day). Because resveratrol is present in red wine and consumption of red wine protects against cardiovascular disease [3], it was hypothesized that resveratrol could be a protective agent against cardiovascular disease, even though wine contains several other polyphenolic compounds (*e.g.* quercetin, catechin), tannins and alcohol that might participate in cardiovascular protection. Numerous studies have now been performed and resveratrol, at least *in vitro*, has many properties that might support this hypothesis. Thus, resveratrol exhibits direct antioxidant activity, an anti-inflammatory effect, and inhibits the effects of vascular cell adhesion molecule expression. Resveratrol also possesses antitumor activity [4, 5], particularly against colorectal cancer cells [6], and could be a candidate to slow the progression of Alzheimer's disease [7]. Recently, resveratrol has been shown to mimic caloric restriction [8], and extend the lifespan of a number of species from yeasts [9] to mice [8]. In Western populations, where obesity is difficult to control, resveratrol is commercially available as a dietary supplement with aggressive marketing and represents a potential life-long medicine.

Today, numerous data are available on the beneficial properties of resveratrol. However, confusion exists due to several pieces of contradictory information, essentially because *in vitro* and animal experimental approaches are often presented as directly transposable to humans. As a contribution to explain how resveratrol could exert a biolo-

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Abbreviations: po, *peros*; **iv,** intravenous

gical effect in healthy subjects and/or during illness, this review presents recent data on the bioavailability, pharmacokinetics and toxicity of resveratrol in humans.

2 Resveratrol administration

Experimental studies have used heterogeneous conditions for resveratrol administration in human cohorts, and it appears that the presence of the matrix (*e.g.* in alcohol, or other polyphenolic compounds in wine) or fed/fasting conditions may result in discrepancy between data.

2.1 Resveratrol naturally present in wine or grape juice

Since the main dietary source of resveratrol is red wine, wine ingestion represents a way of administering resveratrol in natural conditions. Wine is mostly absorbed during a meal. *trans*-resveratrol bioavailability seems to be independent of the lipid content [10]. One of the pitfalls of wine administration is the great qualitative and quantitative variation in resveratrol content depending on the wine type [11–13]. Because early research focused on the *trans*-isomer, often only *trans*-resveratrol content is measured in beverages. However, *cis*-resveratrol and the glucose-bound derivative termed piceid have been reported to be the major forms in wine [14–16] and should therefore be taken into consideration as a source of resveratrol [15, 17, 18]. Indeed, *cis* metabolites can be recovered in plasma and urine [17–20], and piceid has been shown to be a potential natural form of resveratrol administered orally [15].

Some studies have attempted to evaluate resveratrol bioavailability by measuring intake of resveratrol contained in wine [10, 17, 18], or using metabolites as biomarkers of wine consumption [19, 20]. However, wine intake involves the concomitant absorption of other polyphenols and substances such as alcohol that may interact with the action of resveratrol. The specific effects of resveratrol are then difficult to identify.

2.2 Resveratrol as non-conjugated compound

Oral intake is the major route of resveratrol administration. To our knowledge, only one study has used intravenous (iv) bolus administration [21]. Because resveratrol is a lipophilic molecule, early administrations were performed by dissolving the pure compound in various alcoholic solutions such as white wine [22–24], whisky [25], diluted ethanol [21], ethanol in low fat milk [15] or in non-alcoholic liquids such as fruit or vegetable juices [24]. When studied, no matrix effect was reported [24]. Resveratrol has also been administered in capsule form [26–30]. In a two-way cross-over study performed in 24 healthy volunteers, the rate of absorption of a single dose of 400 mg *trans*-resveratrol appeared to be significantly delayed by the presence of food compared with fasting conditions, without modification of total absorption reflected by the area under the plasma concentration–time curve [28]. Piceid administration might be an alternative to pure compound intake, as suggest by Burkon *and* Somoza [15]. These authors suggested that piceid might be enzymatically hydrolyzed in the colon or inside enterocytes, resulting in the formation of *trans*-resveratrol. Piceid may then represent an alternative soluble form for resveratrol administration.

3 Measurement of resveratrol and metabolites

3.1 Free resveratrol measurement

The measurement of free resveratrol (Fig. 1) in human plasma was first described in 2001 by Soleas *et al.* using GC

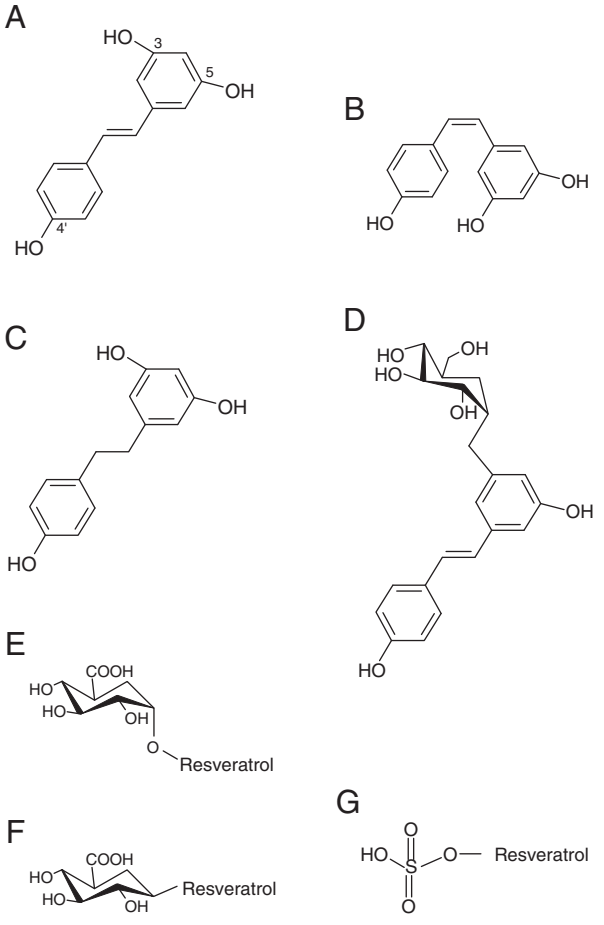

Figure 1. Chemical structure of (A) *trans*-resveratrol, (B) *cis*-resveratrol, (C) dihydroresveratrol, (D) *trans*-piceid, (E) resveratrol-*O*-glucuronide, (F) resveratrol-*C*-glucuronide, and (G) resveratrol–sulfate.

analysis followed by MS detection [22, 23]. These authors reported a limit of detection of 0.01 ng/mL, and a limit of quantification of approximately 0.1 ng/mL. Since 2004, preference has been given to HPLC methods with very satisfactory analytical performances [10, 15, 21, 25, 29, 31] (Table 1). The aim of the development of these methods is to lower the limit of detection at which the presence of resveratrol and its metabolites can be detected and as well as lowering quantification limit by which to assess very small quantities of the molecule. Ideally, all the resveratrol metabolites should be identified and linked only to the administered resveratrol.

Although resveratrol exists naturally as both *cis*- and *trans*-isomers, most studies have used *trans*-resveratrol for administration due to lack of stability of the *cis* isomer, which is not commercially available for this reason. On the other hand, *trans*-resveratrol has been often reported to be the major natural form, even though the *cis* isomer is also present in wine [11]. *cis*-isomerization occurs when the trans isoform is exposed to artificial ultraviolet or natural daylight [32], requiring strict preanalytical precautions. In addition, HPLC assays can distinguish the two isomers since *cis*-resveratrol exhibits a distinct λ_{max} and retention time [32]. Therefore, resveratrol concentrations may be underestimated when only the *trans*-resveratrol peak is taken into account by the analytical method.

3.2 Metabolite identification

Metabolite identification has progressed with improvements in analytical methods. In early studies [10, 21, 23–25], total conjugated metabolites were measured indirectly. *trans*-resveratrol glucuronides and sulfates were determined in humans by enzymatic treatment of samples with glucuronidase or arylsulfatase, resulting in the formation of free *trans*-resveratrol which could then be measured. In subsequent studies, HPLC with MS/MS enabled the identification of different metabolites and the position of hydroxyl substitution. Later, the availability of a metabolite standard enabled the measurement of resveratrol derivatives. Interestingly, the nature and quantity of metabolites may differ between subjects, resulting in high inter-individual variability [10, 18]. Several resveratrol metabolites have been identified in human plasma or urine. Glucuronides and sulfates are the most frequently reported metabolites and may bind one or two hydroxyl residues or a carbon in position 2 [15]. Glucuronide–sulfate metabolites have also been described [26, 27]. As specified by Walle *et al.* [21], the poor chromatic behavior of sulfate conjugates may explain why these conjugates are detected less frequently compared with glucuronides. However, in most recent studies, or when high doses of resveratrol or piceid are administered, sulfates may be the main metabolite [15, 18, 27]. After oral intake of 1 g resveratrol, Boocock *et al.* identified two monosulfate conjugates, one disulfate, two monoglucuronides, and, interestingly, one glucuronide–sulfate [26]. The authors emphasized that their LC-MS/MS methodology was not optimized to detect dihydroresveratrol and its conjugates proposed by Walle *et al.* [21]. Recently, Burkon and Somoza identified up to seven *trans*-resveratrol metabolites (included two new diglucuronides) in plasma and urine from nine healthy volunteers after administration of 85.5 mg of piceid (corresponding to 50 mg of free resveratrol) [15].

cis-metabolites have also been detected in several studies [17–20]. In urine samples collected from five volunteers after wine intake, the main metabolites were *cis*-resveratrol-4′-sulfate, *cis*-resveratrol-3-*O*-glucuronide, and *cis*-resveratrol-4′-*O*-glucuronide [18]. However, no information is available to indicate whether these metabolites are the result of *cis*-resveratrol metabolism or isomerization of *trans*-sulfate or glucuronide conjugates.

The identification and detection of other metabolites remains complex. Walle *et al.* reported that hydrogenation of the aliphatic double bond may occur in both sulfate and glucuronic metabolites and lead to the loss of 306 nm absorbance that characterizes resveratrol [21].

3.3 Evaluation of free and bound forms of resveratrol in plasma

To determine total plasma resveratrol or metabolite concentrations, it is also necessary to take into account LDL- and protein-bound fractions. Burkon and Somoza reported that, *in vitro*, more than 90% of free *trans*-resveratrol is bound to human plasma lipoproteins in a non-covalent manner [15]. Furthermore, *trans*-resveratrol-3-sulfates, *trans*-resveratrol-disulfates and *trans*-resveratrol-diglucuronides are also significantly non-covalently bound to proteins (33.9, 43.6 and 46%, respectively) in humans [15]. This notion of a bound fraction of resveratrol and its metabolites is reinforced by a study that focused on the binding of resveratrol to LDL. Indeed, resveratrol and its metabolites were recovered in the LDL fraction of healthy volunteers after consumption of 250 mL of Merlot wine [17].

4 Pharmacokinetics of resveratrol in humans

4.1 ^{14}C-labeled resveratrol kinetics

To study bioavailability thoroughly, Walle *et al.* [21] administered ^{14}C-labeled resveratrol orally (25 mg) or iv (0.2 mg) to six and five healthy subjects, respectively. Measurement of total radioactivity demonstrated high absorption (at least 70%) after oral intake of resveratrol, without distinguishing resveratrol from its metabolites. Most of the radioactivity was recovered in urine (53.4–84.9% and 42.3–83.2% after oral and iv administration, respectively) but highly variable radioactivity was present in feces after oral intake (0.3–38.1%) and iv administration (0.6–22.7%). This latter

Table 1. Compilation of the published studies that reported pharmacokinetic characteristics of resveratrol in humans

Ref.	Administered form	Number of patients	Doses	Plasma concentration (C_{max})	Plasma peak	Assessment and/or identification	Main metabolites in plasma	Total recovery of Resv and metabolites
[22]	Pure *trans*-Resv in white wine	2	25 mg po	1.5–4 μg/L	30 min	GC-MS		
[23]	Pure *trans*-Resv in white wine	10	25 mg po	7.1 μg/L	30 min	GC-MS	*trans*-Resv conjugates	25% in urine
[24]	Pure *trans*-Resv in white wine/juice	4	25 mg po	7–8 μg/L	30 min	GC-MS	*trans*-Resv conjugates	16.5% in urine
[25]	Pure *trans*-Resv in diluted whisky; grape juice	3	0.03, 0.5 and 1 mg/kg po	ND	ND	HPLC; LC-MS; LC-MS/MS	Glucuronides	26–52% in urine
[30]	Pure Resv	7	1 g po			HPLC; LC-MS	Glucuronides,	
[21]	^{14}C-labeled *trans*-Resv in diluted ethanol	6	100 mg po		ND	Radioactivity; LC-MS	Sulfates,	70.5–97.6% (po),
			25 mg po	< 5 ng/mL			glucuronides,	
			0.2 mg iv	30.7–16.4 ng/mL			dihydroresv metabolites	53.5–91.2% (iv) urine and feces
[10]	*trans*-Resv in red wine associated with different meals	25	0.25 mg	ND		HPLC; LC-MS; LC-MS/MS		
			0.48 mg	1–6 ng/mL or ND	30 min			
			1.92 mg	ND or NQ				
[17]	Resv and piceid *cis–trans* isomers in red wine	11	5.4 mg of total Resv			HPLC; LC-MS/MS	*trans*- and *cis*-glucuronides and glucosides	
[19]	Resv in sparkling or red wine	20, 52	0.357, 0.398 and 2.56 mg total Resv	ND	ND	LC-MS/MS	*trans*- and *cis*-glucuronides	
[18]	Resv and piceid *cis–trans* isomers in red wine	11	5.4 mg total Resv	Low concentration in LDL		HPLC; LC-MS/MS	*trans*- and *cis*-glucuronides, glucosides and sulfates	
[26]	Resv capsules	4	1 g po			HPLC; LC-MS/MS	Mono- and disulfates, monoglucuronides and glucuronide–sulfate	
[27]	Resv capsules	40	0.5 g po	73 ng/mL	< 1 h	HPLC; LC-MS/MS	Mono- and disulfates, monoglucuronides and glucuronide–sulfate	Up to 22% of dose in urine. Present in feces
			1 g po	147 ng/mL	< 1 h			
			2.5 g po	268 ng/mL	$1 < T_{max} < 1.5$ h			
			5 g po	534 ng/mL	1.5 h			
[15]	*trans*-piceid 85.5 mg/70 kg ethanol/low fat milk	9	Corresponding to 50 mg/70 kg po	ND		HPLC; LC-MS/MS	Mono- and disulfates, mono- and diglucuronides	13.6–35.7% in urine

Table 1. Continued

Ref.	Administered form	Number of patients	Doses	Plasma concentration (C_{max})	Plasma peak	Assessment and/or identification	Main metabolites in plasma	Total recovery of Resv and metabolites
[28]	*trans*-Resv capsules in fed or fasting conditions	24	400 mg po	Fed 47.3 ng/mL	30 min	LC-MS		
				Fasting 42.2 ng/mL	2 h			
[20]	Resv in red wine (PREDIMED trial)	1000	ND			LC-MS/MS	*trans*- and *cis*-glucuronides, glucosides and sulfates	
[29]	*trans*-Resv capsules	40	25 mg	1.48–3.83 ng/mL		LC-MS		
			50 mg	6.59–7.39 ng/mL				
			100 mg	21.4–23.1 ng/mL	0.8–1.54 h			
			150 mg	24.8–63.8 ng/mL				
			po six times/day for 48 h					

ND: not dosable; NQ: not quantifiable; po: *per os;* Resv: resveratrol.

result supports the occurrence of the enterohepatic cycle. Overall recovery in urine and feces was 71–98% after oral and 54–91% after iv administration. The maximum plasma concentration was observed 1 h after oral intake and a second peak was present after 6 h, probably as a result of enteric recirculation of conjugate metabolites by reabsorption after intestinal hydrolysis. There was no second peak in any subject after the iv dose. However, the presence of an enterohepatic cycle has been previously described in rats after a resveratrol iv injection but at a very high dose (15 mg/kg) [33]. Moreover, iv bolus leads to the distribution of resveratrol in the whole body, and probably its binding by proteins and tissues before reaching the liver and being included in the enterohepatic cycle. After oral intake, however, the resveratrol and its metabolites first pass through the liver before reaching the rest of the body. Taken together, these points may explain the absence of evidence of an enterohepatic cycle in this study after resveratrol iv injection. With iv administration, a rapid fall in plasma radioactivity was noted after 1 h. However, the half-life of plasma total radioactivity was 10 h after both routes of administration, and plasma concentrations decreased in parallel in an exponential manner long after administration.

4.2 Plasma concentrations

Several studies have reported peripheral blood resveratrol levels after oral ingestion of different quantities of the compound; the analytical methods used were able to measure free resveratrol. The values obtained are listed in Table 1; the main conclusions are as follows: (i) when resveratrol is administered through consumption of wine or juices, the free form is either not detectable at all or present in very low concentrations in plasma. In a series of experiments performed by Vitaglione *et al.* [10] on 25 subjects, the total amounts of *trans*-resveratrol ingested through red wine (300 or 600 mL) ranged from 0.25–1.92 mg. Resveratrol was not detected in any form in sera of 14/25 subjects tested (56%). Among the positive serum samples (11 subjects), the quantities of free resveratrol detected were very low (a few ng/mL only, or below the limit of quantification). (ii) When resveratrol was administered at a dose of approximately 25 mg [21–24, 29], the plasma concentration of the free form ranged from 1–5 ng/mL; administration of higher doses (up to 5 g) led to values of free resveratrol of up to 530 ng/mL [27]. (iii) The maximum peak plasma concentration was reached in the first 30 min after low dose intake. Administration of higher doses, piceid form or fasting status seemed to delay the peak to 1.5 or 2 h.

4.3 Dose-escalation pharmacokinetics

In 2007, Boocock *et al.* reported the first phase I dose-escalation pharmacokinetics study [27]. Bioavailability of

resveratrol was investigated in a population of 40 healthy volunteers. The doses administered in this study are interesting: since resveratrol intake is generally around 25 mg, an approximate dose provided by wine consumption (containing up to 5.8 mg/L [34]), Boocock *et al.* administered a single dose of resveratrol ranging from 0.5 to 5 g. Their results confirmed that free resveratrol is absorbed rapidly (peak plasma concentration between 0.83 and 1.5 h after ingestion) at a relatively low mean plasma concentration (from 73 ng/mL (= 0.3 μmol/L) to 539 ng/mL (= 2.4 μmol/L) for a 0.5 and 5 g resveratrol intake, respectively). A light rebound was observed after 5–6 h supporting the occurrence of the enterohepatic cycle. The corresponding concentrations of the three main metabolites exceeded those of free resveratrol by approximately 20-fold. The major metabolites were resveratrol-3-*O*-sulfate (with a maximum concentration of 1135–4294 ng/mL (3.7–14 μmol/L) and monoglucuronides. The plasma half-lives of resveratrol and its three main conjugates were similar (between 2.9 and 11.5 h). In this study, resveratrol exhibited a mean apparent whole body clearance of 2235–4930 L/h and a mean volume of distribution of 9128–22 226 L, which is in accordance with its low bioavailability and lipophilic status. In urine, within 24 h post-dose, excretion rates were highest during the initial 4 h collection period. In feces, traces of resveratrol metabolites were detected, consistent with enterohepatic recirculation. This study showed that even after high-dose *trans*-resveratrol administration, only a small amount of the free form is present in plasma. Interestingly, the assay method was reported to be efficient but, although recovery was reproducible, the authors highlighted the fact that resveratrol concentrations were probable underestimated as the efficiency of extraction in plasma and urine was only 60–70% [26].

4.4 Multiple-dose pharmacokinetics

Almeida *et al.* [29] performed a double-blind, randomized, placebo-controlled study to investigate the multiple-dose pharmacokinetics of *trans*-resveratrol. Four groups of eight healthy volunteers (and eight in the placebo group) were studied over 48 h. In each group, patients received 25, 50, 100 or 150 mg of pure *trans*-resveratrol six times/day (every 4 h), for 2 days (13 doses in total). As in most previous studies, peak plasma concentrations were reached 0.8–1.5 h post-dose. The half-life of *trans*-resveratrol increased from 1–3 h after a single dose to 2–5 h after repeated administration. Plasma concentrations rose with increasing doses after the first intake in a non-proportional manner (dose: 150 mg/25 mg = 6; C_{max} 150 mg/C_{max} 25 mg = 25; AUC 150 mg/AUC 25 mg = 39). C_{max} was < 20 ng/mL in plasma after an initial dose of 150 mg but reached > 40 ng/mL after the last dose (13th intake). In addition, the authors assessed residual resveratrol plasma concentrations (C_{min}) immediately before intake. C_{min} values were below the lower limit of detection (< 0.5 ng/mL) before most of the 25 mg doses, and approximately 1, 3, and < 10 ng/mL after 50, 100 and 150 mg, respectively. Interestingly, circadian variations were reported for the first time in humans: C_{min} levels were highest in the morning and tended to decrease throughout the day, being the lowest during the night.

5 Elimination

5.1 Resveratrol in urine and feces

As it is highly lipophilic, resveratrol is metabolized in the same way as hydrophilic compounds and is eliminated by the kidneys. No difference in type of metabolites has been observed between plasma and urine. Resveratrol and/or its metabolites are also present in feces, probably due to the enterohepatic cycle (see Section 4.1).

5.2 Urine metabolites as markers of moderate wine consumption

Zamora-Ros *et al.* [19] recently proposed resveratrol urine metabolites as biomarkers of moderate wine consumption. In two open, prospective, crossover clinical studies, these authors observed a significant increase in total resveratrol urine metabolites after 28 days of moderate wine consumption. They identified mainly *trans*- and *cis*-resveratrol-3-*O*-glucuronides in urine, and no free resveratrol, piceid, or sulfoconjugates were detected. Resveratrol and its metabolites were undetectable in plasma. Interestingly, in a study of 52 patients, total resveratrol metabolites appeared to be increased in urine of moderate daily consumers compared to intermittent consumers; this reinforces the hypothesis of a cumulative effect of repeated resveratrol intake. This longitudinal study, using the PREDIMED cohort, included over 1000 patients [20] and allowed the authors to conclude that urinary resveratrol metabolites may be considered as nutritional biomarkers of wine consumption, although noticeable inter-individual variability was observed.

6 Toxicity

6.1 *In vivo* animal studies

Although this review focuses on human studies, toxicological data from animal studies should also be mentioned. In a study in rats, Crowell *et al.* [35] administered 0.3, 1 and 3 g/kg/day *trans*-resveratrol for 4 wk (corresponding to 21, 70, and 210 g/day, respectively, in a human weighing 70 kg). Only two of the 40 rats receiving the highest doses died due to the treatment. Most of the adverse events occurred with the higher dose and consisted mainly of nephrotoxicity.

No histological effect on the liver was observed and no adverse event was observed in animals treated with 0.3 g/kg/day. In addition, the results of this study did not confirm previous observations of a mild increase in serum liver aspartate aminotransferase enzyme levels and brain and testicular weight after a 20 mg/kg/day intake for 4 weeks [36]. In a recent study conducted on high-purity *trans*-resveratrol, Williams *et al.* have studied numerous toxicity models *in vivo* and *in vitro*. Low and high doses of resveratrol, up to 750 mg/kg/day for 3 months, were investigated *in vivo* in rabbits and rats. The authors concluded that resveratrol is well tolerated and non-toxic and has no effect on reproductive capacity in male or female rats and no embryo-fetal toxicity [37]

6.2 Data in humans

The adverse effects in humans have been investigated in several studies after high-dose resveratrol intake [27–29], representing a total of 104 patients (including placebo). The highest doses were 5 g/70 kg for a single intake and 0.9 g/day for iterative administration, corresponding, respectively, to approximately 1/40 and 1/200 of the dose reported to cause nephrotoxicity and 1/4 and 1/20 of the highest dose reported to be safe in rats [35]. No serious adverse event was detected in any of these studies. Adverse events were mild and only lasted a few days. After a single administration of 400 mg of resveratrol, Vaz-da-Silva *et al.* [28] reported three events (blood electrolyte changes, nasopharyngitis and erythematous rash) in 3/24 patients, possibly related to treatment. In the other single-dose administration pharmacokinetic study [27], 2/40 patients receiving 1 g resveratrol exhibited one or more minor biological adverse event, consisting of a small increase in blood bilirubin or alanine amino transferase level. In the multiple-dose study, 40 volunteers received one dose of resveratrol (25, 50 100, 150 mg, or placebo) every 4 h for 48 h. The most frequent adverse event was frontal headache (three cases). The other adverse events appeared only once: headache, myalgia of the lower extremities, somnolence (25 mg group), epididymitis (100 mg group), and dizziness and occipital headache (150 mg group), without any clear relation to the administered dose [29].

7 Comments

The studies described in this review have significantly contributed to our knowledge on human resveratrol bioavailability, pharmacokinetics and toxicology. Although differences in resveratrol administration, doses and assay methods make data from different studies difficult to compare, these data nevertheless provide important information and raise some interesting questions.

First, resveratrol seems to be well tolerated. However, no information is available on long-term administration. For use in chronic diseases such as diabetes, colorectal cancer or Alzheimer's disease, or for prevention of cardiovascular disease and anti-aging antioxidative care, resveratrol administration would occur over several months/years at doses which are not yet known. Although resveratrol is considered to be a food supplement and a relatively safe natural medication, further investigations are required to determine its long-term effects.

Second, resveratrol is rapidly absorbed and metabolized, mainly as sulfo- and glucuro-conjugates which are excreted in urine. Similar results have been observed in studies performed in rodents [31, 33, 38–40]. This high metabolic rate probably allows the transport, the distribution and the excretion of resveratrol. In rodents, the gut epithelium has been shown to be highly implicated in the metabolic process, resulting in polar resveratrol compounds which then need specific transporters to cross cell membranes. A recent study suggests that several ATP-binding cassette transporters may be involved in the tissue distribution and subsequent elimination of resveratrol from the body [41]. However, concentrations of free *trans*-resveratrol are very low in plasma and several authors have raised doubts about its efficiency [10, 24]. A lot of questions remain unanswered; the answers depend on the availability of both specific and sensitive assays for metabolite identification and quantification. Several points have to be discussed in order to validate resveratrol activity: (i) *trans*-resveratrol is bound by LDL [17] and albumin [15]. When assessing plasma resveratrol concentrations, the bound part must be taken into account as a potential reserve of resveratrol; (ii) The *cis*-isoform of resveratrol, which is also an active form, is often overlooked; (iii) Conjugated metabolites may contribute to resveratrol activity. Although information about resveratrol metabolite activity is sparse, other conjugated polyphenols chemically similar to resveratrol have been shown to exhibit biological activity [42–44]. On the other hand, the occurrence of deconjugation at certain sites leading to the parent aglycon should not be overlooked [45]; (iv) In all of the studies reviewed here, several resveratrol derivatives were reported but total recovery in urine was only about 25% (52% in one case) [25]. These results are far from the 70–98% recovery of radioactivity in urine and feces after 14C-labeled resveratrol oral intake. In the later case, the identity of the radioactive moiety was not taken into consideration in the recovery calculation and the parent molecule and all metabolites are presumed to participate in radioactivity recovery. This result suggests that all resveratrol metabolites have not yet been identified. Among them, piceatannol, a monohydroxylated derivative of resveratrol, has physiological effects which may contribute to the action of resveratrol [46]; (v) There is great inter-individual variability in metabolism. What is the fate of the 2–30% that is not recovered in urine and feces [25]? Because of the high volume of distribution of resveratrol, a large part of the molecule may be bound by cell membranes

or lipophilic tissue. As suggested by Soleas *et al.* [22], free resveratrol levels in serum could be seriously underestimated because of large amounts potentially contained in the cellular fraction. The effects of resveratrol may not therefore be a result of the ''visible'' plasma fraction but rather of the resveratrol cellular fraction which is not assessed.

However, at the moment, several pathways are under investigation to improve resveratrol bioavailability or effects. One strategy is to modify the resveratrol structural determinants such as number and position of the hydroxyl groups, intramolecular hydrogen bonding, stereoisomerism and double bond [47–51]. Moreover, as discussed above, effects of metabolites or resveratrol conjugates as piceid need to be most carefully examined. Some authors also propose the use of resveratrol oligomers [52, 53]. At last, several teams are exploring various galenic forms as calcium-pectinated beads [54] or polymeric micelles [55].

All the studies reviewed here concern healthy patients and the resulting information is necessary to move forward. Several clinical projects have been yet recorded on clinicaltrial.gov concerning resveratrol effect in cancer, metabolic syndrome and Alzheimer's disease. The resulting data will allow to appreciate the resveratrol efficacy in human pathology.

8 Concluding remarks

In conclusion, the pharmacological properties of resveratrol in the field of antitumor therapy or in relation to its anti-inflammatory/antioxidant effects support the use of this agent as a complementary nutritional/pharmacological biomolecule. Numerous data are now available on its bioavailability and toxicity in humans. However, these data are still patchy and many areas warrant further investigation and clarification. In particular, the exact nature of the endogenous metabolites, including their biological properties, and the precise distribution of the molecule within tissues and cells remain to be determined precisely.

The authors have declared no conflict of interest.

9 References

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