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Current Medicinal Chemistry, 2016, *23*, ???-????

Modulation of Expression and Activity of ABC Transporters by the Phytoestrogen Genistein. Impact on Drug Disposition

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Abstract: ATP binding cassette (ABC) transporters are involved in drug absorption, distribution and elimination. They also mediate multidrug resistance in cancer cells. Isoflavones, such as genistein (GNT), belong to a class of naturally-occurring compounds found at high concentrations in commonly consumed soya based-foods and dietary supplements. GNT and its metabolites interact with ABC transporters as substrates, inhibitors and/or modulators of their expression. This review compiles information about regulation of ABC transporters by GNT with special emphasis on the three major groups of ABC transporters involved in excretion of endo- and xenobiotics as follows: P-glycoprotein (MDR1, ABCB1), a group of multidrug resistance associated proteins (MRPs, ABCC subfamily) and ABCG2 (BCRP), an ABC half-transporter. The impact of these regulations on potential GNT-drug interactions is further considered.

Keywords: ATP binding cassette transporters, breast cancer resistance protein, genistein, multidrug resistance-associated proteins, nutrient-drug interactions, P-glycoprotein, xenobiotic transport.

1. INTRODUCTION

1.1. ABC Transporters: General Considerations

Transporters belonging to the ATP binding cassette family (ABC) are multispanning transmembrane proteins present in almost all the cells of the human body. Structurally, they exhibit 2 cytoplasmatic nucleotide-binding domains, which use the energy of ATP hydrolysis to transport substrates against a concentration gradient across the plasma membrane (Fig. **1**) [1]. ABC transporters exhibit a wide variety of physiological roles. Among them, P-glycoprotein (MDR1, ABCB1), the multidrug resistance associated proteins (MRPs, all belonging to the ABCC subfamily) and the breast cancer resistance protein (BCRP, ABCG2), an ABC half-transporter, play a significant role in the depuration of endo- and xenobiotics. For instance, many compounds resulting from cell catabolism and exogenous substances like therapeutic drugs and their metabolites are extruded from the cell by ABC transporters [2, 3]. As a consequence, ABC transporters regulate the absorption and excretion of therapeutic drugs, thus affecting their pharmacokinetics and frequently underlying adverse drug reactions and drug-drug interactions.

What follows next is a description of each of the above referred transporters with special emphasis on their function as well as a section about their regulatory mechanisms. In the sake of clarity, capital letters are used throughout for transporter denomination regardless the species studied.

1.2. P-glycoprotein (MDR1/ABCB1)

1.2.1. General Features

P-glycoprotein (P-gp), also known as ABCB1 or MDR1, was first identified by Juliano and Ling in 1976 in the plasma membrane of Chinese hamster ovary cells resistant to colchicine [4]. Later, it was found in human cancer cells, in association with the presence of a multidrug resistant phenotype, conferring to this family of transporters the denomination of multidrug resis-

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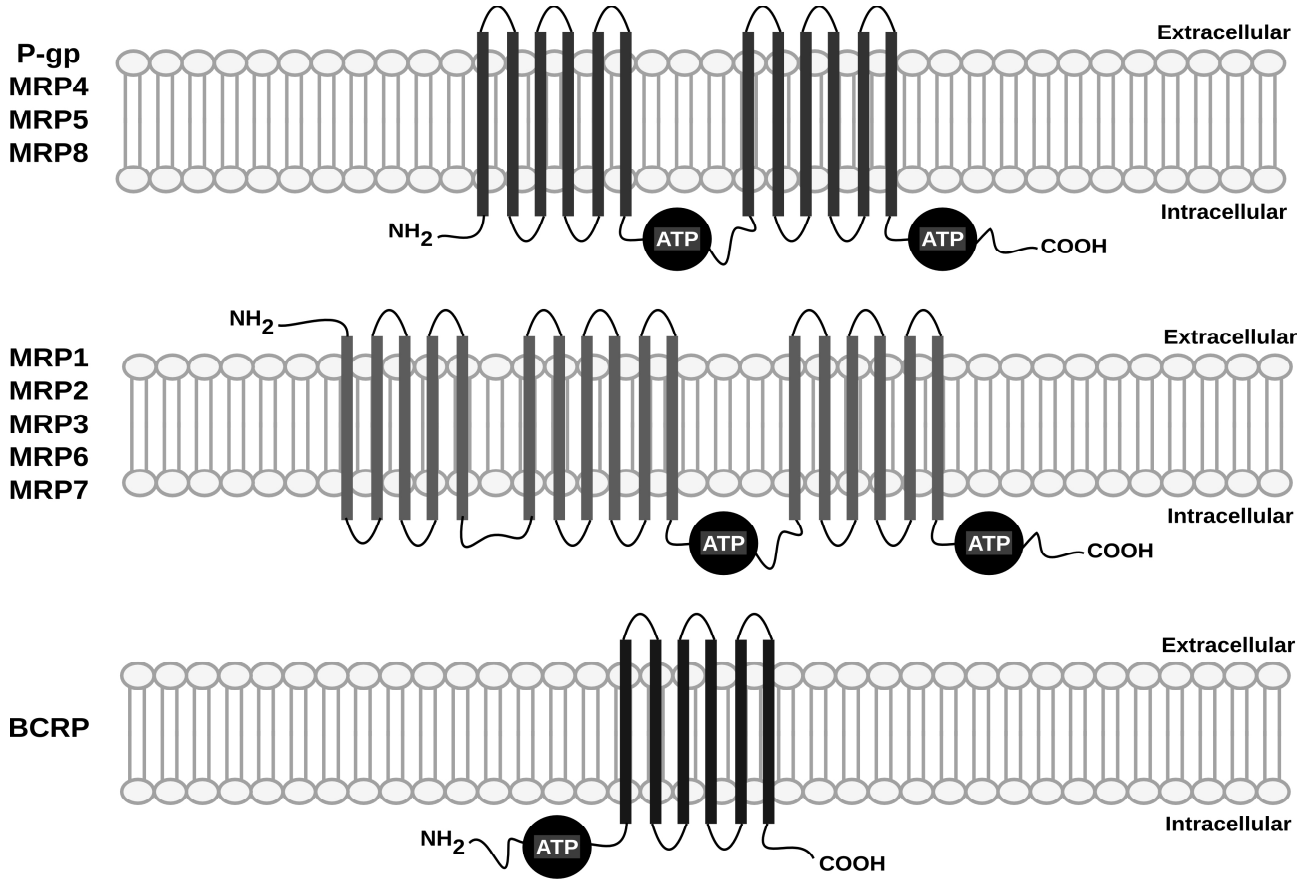

Fig. (1). General structure of pharmacologically relevant ABC transporters. Depicted are the transmembrane- and nucleotide binding domains of the P-glycoprotein (P-gp), the multidrug resistance associated proteins 1-8 (MRP1-8) and the breast cancer resistance protein (BCRP). Due to its localization in intracellular membrane compartments and its scarce relevance on drug efflux, MRP9 is not depicted in the illustration.

tance proteins (MDR) [5]. This was confirmed by immunocytochemical studies showing high expression in epithelial solid tumors including colon [6], kidney [7] and breast [8]. P-gp is also expressed constitutively in a wide range of normal tissues such as kidney, liver, small intestine, colon, adrenal glands and lung, and to a lower extent in prostate, skin, spleen, heart, skeletal muscle, stomach and ovary [9]. Also, it is expressed in brain [10-12], choroid plexus [13], peripheral blood mononuclear cells and placenta [14].

1.2.2. Physiological Role of P-gp

The physiological role of P-gp was first studied using P-gp knock-out mice. Noteworthy, these mice did not show evident physiological alterations. However, when exposed to certain toxins, they were significantly more affected than wild-type mice, suggesting a major role of P-gp in xenobiotic efflux [15, 16]. In this regard, the localization of P-gp in the intestinal brush border membrane plays a role as an intrinsic barrier limiting the uptake of xenobiotics from the intestinal lumen [17]. In addition, its expression in the liver canalicular membrane and in the apical membrane of renal tubular cells indicates a key role in the vectorial efflux of xenobiotics to bile and urine, respectively, thus contributing to systemic drug clearance [9, 18-20]. Similarly, P-gp localization in the luminal side of endothelial cells of the blood-brain barrier limits the penetration of potential harmful substances into the central nervous system [21].

Several therapeutic drugs, environmental toxicants and diet compounds have been characterized as P-gp substrates. Characterization of major compounds transported by P-gp and their chemical properties conditioning transporter-compound interaction were extensively reviewed by Schinkel and Jonker [22] and Klaassen and Aleksunes [23]. In the case of therapeutic agents transported by P-gp, the interference in the absorption may be judged as undesirable, in opposition to the beneficial function of the transporter in limiting the absorption of toxins. In addition, hormones and other lipophilic endobiotics have been reported to be trans-

ported by P-gp [24, 25]. However, taking into account that P-gp knock-out mice do not exhibit physiological alterations [15, 16], a minor role of this transporter in disposition of endobiotics may be inferred.

1.3. MRP Family

1.3.1. General Features

Human multidrug resistance-associated proteins (MRPs) comprise a group of 9 transporters with conserved structure but heterogeneous substrate specificity, localization and function. All MRPs exhibit the same core structure displayed by P-gp. MRP1, MRP2, MRP3, MRP6 and MRP7 have an additional N-terminal domain with 5 transmembrane segments, while MRP4, MRP5, MRP8 and MRP9 lack this extra domain (Fig. **1**) [26-28].

MRP1/ABCC1 was the first member of this family to be described; it was detected in a mutant lung cancer cell line that exhibited enhanced resistance towards chemotherapeutic agents with respect to the parental cell line [29]. It was also detected in normal tissues like kidney [30], intestine [31], blood-brain barrier [32], lung, testis and peripheral blood mononuclear cells [29, 33]. The normal liver exhibits low MRP1 expression [27]. However, a liver canalicular (apical) isoform of MRP1 was later found. It was named MRP2/ABCC2 [34] and its expression was not restricted to the liver but was found also in kidney [35], small intestine [36] and blood-brain barrier [37]. Moreover, MRP2 expression was also detected in tumors of different origins and usually associated to a poorer disease prognosis [38, 39]. MRP2 deficiency is responsible for the biliary secretion impairment observed in patients with Dubin-Johnson Syndrome [40]. A basolateral isoform, MRP3/ABCC3, was later identified in the sinusoidal membrane of hepatocytes [41], where it mediates the vectorial transport of substrates from the hepatocyte to the sinusoidal blood [41-43]. It was also detected in kidney, intestine, pancreas, lung, gallbladder [44, 45], as well as in different tumor entities where its expression was associated with enhanced chemoresistance [46, 47]. MRP4/ABCC4 and MRP5/ABCC5 are more ubiquitously expressed, although at variable levels. They both exhibit basolateral localization in polarized epithelia, except for MRP4, which is expressed at the apical membrane of the proximal tubule cells [48]. MRP6/ABCC6 was found to be relevant in the pathogenesis of pseudoxanthoma elasticum, an inheritable disorder characterized by mutations leading to a dysfunctional transporter [49]. MRP7/ABCC10 has been associated to resistance to taxanes in many tumoral entities [50]. MRP8/ABCC11 was identified as a cyclic nucleotide efflux transporter and a resistance factor for nucleotide analogs [51]. MRP9/ABCC12 expression was described in mouse testis and in breast cancer cells. Experimental evidence indicates that MRP9 is predominantly localized in intracellular compartments [52-54]. Little is known about the substrates and functions of this particular transporter.

1.3.2. Physiological Role of MRPs

MRPs mediate the efflux of endo- and xenobiotics, including therapeutic drugs. Substrate specificity of each MRP within this subfamily is highly variable, (reviewed in Schinkel and Jonker [22] and Klaassen and Aleksunes [23]). An insight into physiological roles of MRP transporters was gained with the use of knock-out mice. In this respect, $MRP1^{-/-}$ mice were viable and fertile, thus arguing against a major role of this protein in growth and development. However, these mice exhibited increased tissue damage in bone marrow and seminiferous tubules upon exposure to etoposide, confirming MRP1 role in protection against xenobiotic toxicity [55]. Besides, MRP1 plays a key role in the acquired immune response, for example, by promoting the migration of dendritic cells from peripheral tissues to the lymph nodes [56]. A protective role of MRP1, as well as of MRP2, against oxidative stress was also demonstrated [57, 58]. MRP2 and MRP3 act coordinately to mediate efflux of organic anions from polarized epithelia through the apical and basolateral route, respectively. For instance, MRP2, highly expressed in the liver, mediates the biliary secretion of endobiotics like bilirubin glucuronides and hormone conjugates. Conversely, MRP3, which presents a low basal expression, is highly inducible in human and rat liver and mediates their basolateral efflux. MRP3 is usually induced under conditions of MRP2 malfunction, thus providing an alternative pathway for the elimination of MRP2 substrates [41]. Additionally, MRP2 contributes to the intestinal chemical barrier function limiting the uptake of potentially toxic compounds into the enterocyte [59]. A similar role of MRP2 was also described in the capillaries of brain endothelial cells and in the choroid plexus, limiting the entrance of toxic compounds into the central nervous system [60, 61]. In the kidney, MRP2 mediates the secretion of organic anions to the urine [35, 62]. MRP4, MRP5 and MRP8 role as cyclic nucleotide transporters indicates a participation in physiological processes involving these molecules as signal mediators such as cell migration, cell death [63], thrombus formation [64] and sperm capacitation [65]. MRP6 has been suggested to mediate ATP release by

hepatocytes. This process appears to be impaired in the inheritable disorder pseudoxanthoma elasticum [66, 67].

1.4. Breast Cancer Resistance Protein (BCRP/ ABCG2)

1.4.1. General Features

Breast Cancer Resistance Protein (BCRP/ABCG2) was first cloned by Doyle *et al.* from the human breast cancer cell line MCF-7 [68]. The same year, Allikemets *et al*. [69] and Rabindran *et al*. [70] identified the transporter in human placenta and colon carcinoma cells, respectively. Overexpression of the cloned BCRP cDNA in cell lines demonstrated that the transporter was able to confer resistance to chemotherapeutic drugs such as mitoxantrone, doxorubicin and daunorubicin in an energy-dependent way [68]. BCRP consists of 6 transmembrane domains (Fig. **1**). However, in human cells the transporter is likely to exist and function as a homodimer and oligomer [71-74]. BCRP is expressed in the canalicular membrane of hepatocytes, in the brush border membrane of enterocytes and in the apical membrane of renal proximal tubules. In addition, BCRP is expressed in brain capillary endothelial cells, thus contributing to the blood-brain barrier. A similar role was described in blood-placental and blood-testis barriers and in the breast [75]. BCRP expression was also described in immature myeloid and leukemia stem cells, suggesting a role in multidrug resistance in blood cancer therapy [75].

1.4.2. Physiological Role of BCRP

One of the main physiological roles of BCRP consists in the hepatobiliary excretion of endo- and xenobiotics. In the intestine, it functions as a defense barrier, limiting the absorption of dietary carcinogens, therapeutic drugs and toxins [76]. In kidney proximal tubular cells, it is involved in the urate elimination and in the secretion of different compounds through the apical membrane. Moreover, BCRP transports several drugs administered in cancer chemotherapy. In this regard, BCRP expression has been associated with chemotherapy efficacy and cancer prognosis [77, 78]. More detailed information on BCRP substrates is available in the reviews by Schinkel and Jonker [22] and Klaassen and Aleksunes [23].

BCRP expression in lactating mammary glands is associated with the active secretion of clinically and toxicologically relevant substrates into the milk. This BCRP function results in an increased exposure of sucklings to noxes substrates of this transporter but also provides a pathway for the secretion into milk of vitamins such as riboflavin [79, 80].

1.5. Modulation of ABC Transporters

ABC transporters are subjected to a tight regulation at different levels. Transcriptional modulation represents the mechanism most frequently described. *In silico* studies scanning promoters and enhancers of ABC transporters have identified several putative transcription factor binding sites. The mechanism of action for several of these factors was further validated in biological assays [81-85]. A list of most important transcription factors is provided in Table **1.A**. Concerning ABC regulation by xenobiotics, it must be highlighted the role of the nuclear receptor PXR (NR1I2, Pregnane X Receptor), which is activated by a wide range of therapeutic drugs, environmental toxicants and micronutrients [86, 87]. PXR was already described to modulate the transcription of P-gp [88], MRP2 [84], MRP3 [89] and BCRP [90]. In addition to regulation by transcription factors, epigenetic mechanisms have been also described to modify the expression of ABC transporter mRNAs [91].

In addition to transcriptional modulation, ABC transporters have been reported to be regulated at post-transcriptional- (*e.g.* due to an altered mRNA half-life), translational (*e.g.* due to changes in regulatory microRNA expression) and post-translational levels (*e.g.* ubiquitination, changes in the glycosylation status, acute changes in transporter localization). Major mechanisms belonging to these categories are listed in Table **1** (**B** to **D**).

2. GENISTEIN

2.1. General Features

Genistein (4′,5,7-trihydroxyisoflavone, GNT) is a phytoestrogen belonging to the family of isoflavones usually ingested as a dietary supplement or as component of vegetables such as soybeans, fava beans and lupins. It is also found in derived products like soybean oil, tofu, miso, tempeh, soy infant and cereal formulations. GNT can be present in two different forms: as glycoside (*i.e.* genistin or genistein-7-O-β-D-glucoside) and as aglycone (*i.e.* genistein) (Fig. **2**). The major form present in soybeans and other plants is the glycoside [130]. On the contrary, in manufactured food such as miso and tempeh, the aglycone is present in a greater proportion due to the hydrolysis of the glycoside by microbial β-glucosidases during the fermentation process [131].

Table 1. Mechanisms involved in ABC transporter regulation.

Below are presented the molecular mediators and regulatory pathways reported to modify ABC transporter expression at transcriptional-**(A)**, post-transcriptional-**(B)**, translational-**(C)** and post-translational levels **(D)**. Corresponding references are provided for each listed mechanism.

A. Transcriptional mechanisms	
Transcription factor or regulatory mechanism	**Reference**
Activator protein 1 (AP-1)	[92, 93]
Aryl hydrocarbon receptor (AhR)	[83, 94]
Changes in histone acetylation and methylation patterns	[91]
Changes in promoter methylation patterns	[95, 96]
Constitutive androstane receptor (CAR)	[97, 98]
Estrogen receptors α and β (ERα, ERβ)	[92, 93, 99-102]
Farnesoid X Receptor (FXR)	[84]
Hypoxia-inducible factor 1α (HIF-1α)	[103]
Nuclear factor erythroid 2-related factor 2 (Nrf2)	[83, 104-106]
Nuclear factor κB (NFκB)	[107]
Peroxisome proliferator activated receptor α (PPARα)	[83, 108]
Pregnane X Receptor (PXR)	[86-89, 98, 109-111]
Progesterone receptor B (PRB)	[112]
B. Post-transcriptional mechanisms	
Regulatory mechanism	**Reference**
mRNA stabilization	[113-115]
C. Translational mechanisms	
Regulatory mechanism	**Reference**
Translation control by microRNAs	[116-124]
Use of alternative open reading frames	[125]
D. Post-translational mechanisms	
Regulatory mechanism	**Reference**
Control of transporter ubiquitination	[126]
Transporter glycosylation	[127, 128]
Acute changes in transporter localization	[129]

2.2. GNT Metabolism and Disposition

After its ingestion, GNT glucoside is hydrolyzed to GNT by epithelial and microbial β-glucosidases in the oral cavity [132] and small intestine [133]. Then, the aglycone form is absorbed by the intestinal epithelium by passive diffusion. In humans, soy aglycones are the biologically active form of isoflavones and are absorbed faster and in higher amounts than their glucosides [134-136]. Inside the enterocyte, GNT is metabolized to its glucuronide derivative by UDP-glucuronosyltransferases (UGTs; UGT1A1, 1A8, 1A9, 1A10) and in less proportion to sulfate conjugates by sulfotransferases (SULTs, SULT1A1*2, 1E, 2E1) [137]. Conjugated metabolites are then transported by apical BCRP [138-140] and MRP2 [141] back into the intestinal lumen or alternatively by basolateral MRP3 and MRP4 [142] to reach the portal circulation. A fraction of consumed GNT may reach the liver intact, as aglycone, and undergo intrahepatic conjugation. The conjugated metabolites may ultimately be excreted into the bile by canalicular MRP2 and BCRP or into the sinusoids by basolateral MRP3 and MRP4. Strong evidence concerning MRP2 role in conjugated GNT efflux was provided by Krumpochova *et al.*, [143], who demonstrated that GNT-glucuronide is more abundant in urine from *MRP2*$^{-/-}$ mice than in urine from wild type mice. These data fit those of Jäger *et al.* [144], describing an impaired biliary excretion of GNT-glucuronide in MRP2 deficient rats. Similarly, Kato *et al.*, [145] demonstrated that following oral administration the plasma concentrations of total GNT and total daidzein, another soy isoflavone, mainly as sulfoglucuronide conjugates, were higher in MRP2-deficient Eisai hyperbilirubinemic (EHBR) than in normal rats [145]. Under this condition, a higher basolateral excretion of GNT conjugates by MRP3 and MRP4 is expected, leading to increased plasma and urine concentration. Finally, GNT metabolites reaching the small intestine through the bile duct are deconjugated by microbial enzymes and further reabsorbed or alternatively excreted in feces. GNT deconjugation may also occur in blood, thus leading to increasing aglycone concentration [146]. All these pathways are illustrated in Fig. (**3**).

Interestingly, many phase II biotransformation systems that metabolize GNT are expressed in the same tissues expressing ABC transporters. This reinforces the assumption of a coordinated function between phase II enzymes and the apical export pump MRP2 in many epithelial cells [147]. For example, in the rat intestine, conjugation of endo and xenobiotics by UGT

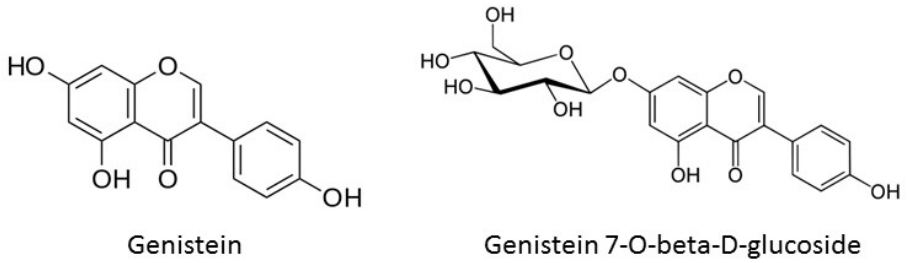

Fig. (2). Chemical structures of genistein (GNT, aglycone) and its glycoside form (Genistin or Genistein 7-O-beta-D-glucoside).

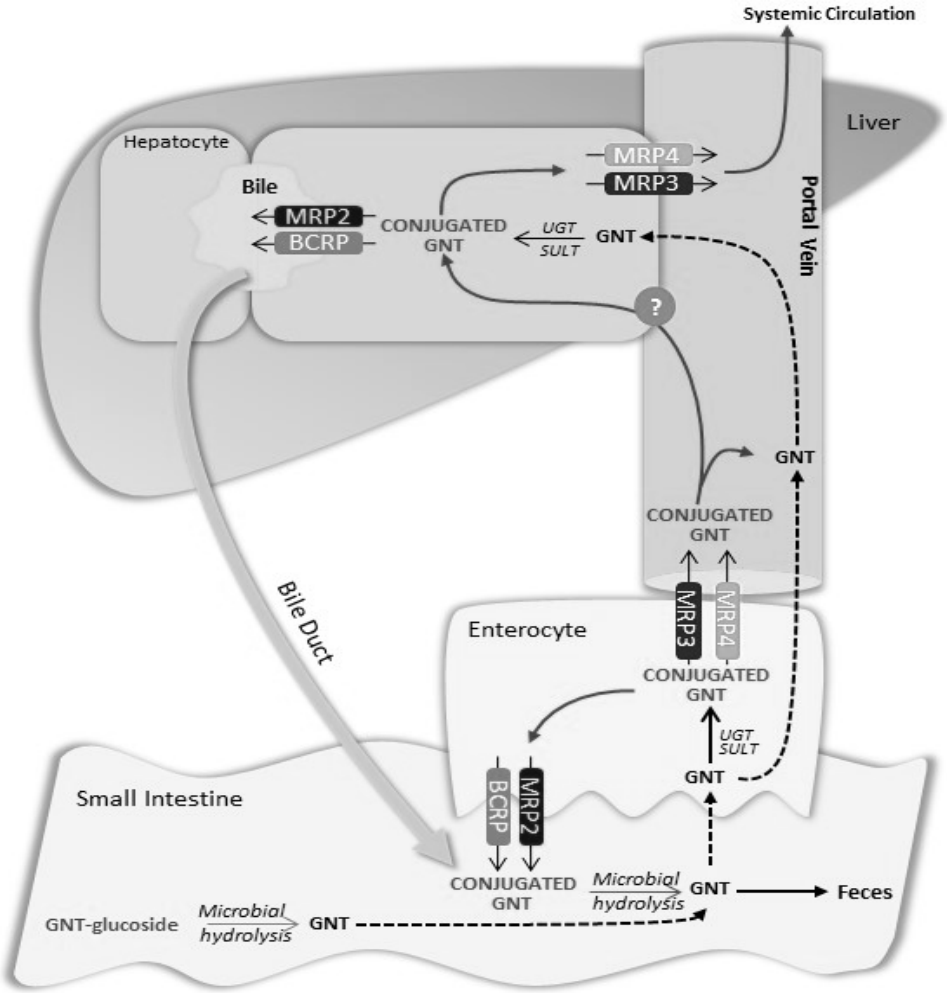

Fig. (3). GNT metabolism and enterohepatic recirculation. This is a schematic representation of the major enzymes and efflux transporters involved in GNT hepatic and intestinal metabolism and disposition. Dashed arrows represent passive diffusion of GNT. Since there is no data available about basolateral efflux transporters involved in conjugated-GNT extrusion *in vivo,* it is assumed that MRP3 and MRP4 are involved in conjugated-GNT basolateral extrusion as was described in HeLa cells [142]. While conjugated estrogens are uptaken by the hepatocytes through organic anion transporter polypeptides (OATPs), it is still unknown if this transporter is involved in conjugated-GNT hepatic uptake. GNT: genistein, UGTs: UDP-glucuronosyltransferases, SULTs: sulfotransferases.

and SULTs takes place predominantly in the proximal portion with a decrease observed further down the intestinal tract. The same distribution along the small intestine has been reported for intestinal MRP2 [147]. This co-localization of biotransformation and transporter systems represents a useful strategy to improve the systemic clearance of potentially harmful substances. In addition, it suggests the occurrence of common regulatory mechanisms.

2.3. GNT Effects

GNT displays estrogenic activity related to its structural similarity to 17β-estradiol, the major physiological estrogen. GNT consumption either with the diet or as hormone replacement supplements is believed to alleviate menopausal symptoms and even to prevent hormone associated malignancies. Nevertheless, evidence in this regard is highly controversial [148]. It is postulated that GNT inhibits the growth of tumor cells by interfering with the tyrosine kinase activity of growth factor receptors and cytoplasmic tyrosine kinases and by inhibiting DNA topoisomerase as well. Similarly, Davis *et al*. investigated the inhibitory effect of GNT on growth of cells derived from pancreatic cancer and demonstrated that GNT can modulate specific genes involved in cell growth inhibition and apoptosis, such as the cyclin-dependent kinase inhibitor 1 (p21) [149]. In this regard, a study by Zander *et al.* [150] evaluated the effects of a GNT enriched diet on overall survival of mice harboring *BRCA1* and *p53* mutations leading to the development of spontaneous breast cancer. Effects were evaluated in BCRP deficient mice, with expected higher GNT bioavailability due to reduced GNT clearance, and in mice with normal BCRP activity. Results showed no difference in overall survival between groups. However, the authors did not measure GNT plasma concentrations. If GNT bioavailability in BCRP knock-out mice was, as expected, higher than in wild type mice, these results would suggest a minor relevance of GNT exposure in breast cancer outcome, at least in malignancies with a genetic background similar to that in the mice studied.

In human cell lines, GNT exhibited both proliferative and antiproliferative effects depending on the concentration. For instance, in the breast cancer cell line MCF-7 (estrogen receptor positive), cell growth is stimulated at low concentrations of GNT (10^{-5}- 10^{-8} M) while it is inhibited at high concentrations (10^{-5} - 10^{-4} M) [151]. Considering that GNT plasma concentrations after the ingestion of GNT enriched supplements do not exceed 10^{-6} M [152, 153], this evidence, although speculative, would support stimulatory effect on cancer cell proliferation under such conditions. Importantly, most of the cell lines used in *in vitro* studies exhibit reduced activity of phase II biotransformation systems [154] and are usually exposed to the aglycone form of GNT. These conditions may not reproduce the situation *in vivo,* where more efficient conjugation systems may result in much lower exposition to unconjugated GNT. Whether such conditions are compatible with the low range of concentrations used *in vitro* and found to stimulate cancer cell proliferation [151] is far from being demonstrated.

GNT was also found to exhibit antioxidant activity [155]. It was demonstrated that oral administration of GNT reduces lipid peroxidation in liver and oxidative stress induced by vitamin E deficiency in hamsters [156]. Similarly, Gonzalez-Granillo *et al*. described an effect on lipid homeostasis in mice. The authors demonstrated an increase in ABCG5 and ABCG8 mRNA levels as well as increased total fecal bile acid excretion and insulin sensitivity [157].

3. REGULATION OF ABC TRANSPORTERS BY GNT

GNT is ligand of nuclear receptors such as ER-α and ER-β [158]. In addition, PXR activation by GNT and induction of human CYP3A4 and murine CYP3A11 through this pathway were observed [159]. Furthermore, GNT also interacts with a variety of transcription factors that modulate gene expression and/or protein localization such as aryl hydrocarbon receptor, the nuclear factor erythroid 2-related factor 2 (Nrf2) and the epidermal growth factor receptor [160-162]. Considering that these transcriptions factors have been already shown to regulate ABC transporters (Table **1.A**), an important role of GNT in regulating endo- and xenobiotic transport can be expected.

Several authors studied the effects of GNT on ABC transporters in different experimental models, at different incubation times and using different concentrations of GNT. In view of the significant amount of information available, we first performed a classification of these effects according to whether they resulted from sustained or short treatments. These conditions may be tentatively associated to prolonged or sporadic ingestion of GNT, respectively. Accordingly, Tables **2** and **3** summarize these long- and short-term effects, respectively. It is evident from Table **2** that GNT exerts an overall increase in expression of the transporters studied, except for BCRP, where mRNA downregulation was observed. The mechanisms of induction are unlikely universal and mostly involve transcriptional or translational regulations, as mentioned in Table **2**. Short-term treatments resulted in variable effects depending on the transporter studied. In view of the complexity of all these long- and short-term regulations, we next describe in more detail the most relevant effects for each of the drug transporters currently reviewed.

3.1. P-gp Regulation

It has been shown that GNT (200 μM) exerts an acute inhibition of P-gp activity in tumor cells [163]. P-

Table 2. Long-term regulation of ABC transporters by GNT.

Transporter	Model	Treatment	Effect	Mechanism	Reference
P-gp*	Caco-2 cells	GNT 1 μM, 48 h	↑ mRNA ↑ protein ↑ activity	ER-β and SP-1 dependent	[99]
P-gp*	HepG2 cells	GNT 1 μM, 48 h	↑ protein ↑ activity	Translational regulation	[167]
P-gp*	HepG2 cells	GNT 10 μM, 48 h	↑ mRNA ↑ protein ↑ activity	PXR dependent	[167]
MRP2*	Caco-2 cells	GNT 1 μM, 48 h	↑ protein ↑ activity	Translational regulation, ER-β dependent	[99]
MRP2*	Rat small intestine	Standardized soy extract (100 mg/kg p.o., 10 days)	↑ mRNA	-	[168]
MRP2*	HepG2 cells	GNT 1 μM, 48 h	↑ protein ↑ activity	Decrease in hsa-miR-379 expression	[167]
MRP2*	HepG2 cells	GNT 10 μM, 48 h	↑ mRNA ↑ protein ↑ activity	PXR dependent	[167]
MRP2	HepG2-C3 cells	GNT 50 μM, 10-24 h	↑ mRNA ↑ protein	Increase in PXR-RXR-α binding to MRP2 promoter	[169]
MRP2	HepG2 cells	GNT 22 and 70 μM, 24 h	↓ mRNA = Protein	-	[170]
MRP4	HepG2 cells	GNT 70 μM, 24 h	↓ mRNA	-	[170]
MRP2*	Rat liver	Standardized soy extract (100 mg/kg p.o., 10 days)	↑ mRNA	-	[168]
MRP1	Rat liver	Standardized soy extract (100 mg/kg p.o., 10 days)	↑ mRNA	-	[168]
BCRP	Gastric cancer cell line MGC-803	GNT 15 μM, 24 h	↓ mRNA	-	[178]

**In vivo* reports harboring potential clinical relevance as well as *in vitro* studies using pharmacologically relevant concentrations are indicated with * next to the modulated transporter.

gp transport inhibition could be due to ATP depletion by GNT. However, this phenomenon in tumor cells was described for GNT concentrations higher than 400 μM [164], thus ruling out this mechanism as the cause of the inhibition of P-gp-mediated transport. Several studies focused on the inhibition of tyrosine kinase activity by GNT as a potential cause of P-gp inhibition. Germann *et al*. speculated that as Rhodamine-123 (R123) efflux was inhibited by GNT and rapidly reversed (2 min), changes in phosphorylation status are unlikely involved. Also, P-gp did not show phosphorylation of tyrosine residues in an *in vivo* experiment [165]. Moreover, the potent tyrosine kinase inhibitor lavendustin A had no effect on R123 accumulation in BALB/c-3T3-1000 cells, further supporting a mechanism of P-gp inhibition independent of tyrosine kinase inhibition [163]. The exact underlying mechanism remains unknown. In contrast, Okura *et al*. found in LS180 cells that 100 μM GNT increases R123 efflux after a 1-h incubation [166]. Similarly, Jäger *et al*. demonstrated that GNT increases R123 efflux in isolated and perfused rat liver. This occurs due to competitive inhibition of MRP2 by GNT glucuronide that affects transport of R123-glucuronide, also pumped by MRP2. As a result, the accumulation of R123-glucuronide may inhibit glucuronidation and as a consequence more R123 is available to be excreted by P-gp [144].

Table 3. Short-term regulation of ABC transporters by GNT.

Transporter	Model	Treatment	Effect	Mechanism	Reference
P-gp	Breast cancer cell line MCF-7/MDR1	GNT 200 μM, 1 h	↓ activity (↑R123 and daunorubicin accumulation)	Independent of P-gp phosphorylation status	[163, 165]
P-gp	LS180V cells	GNT 100 μM, 1 h	↑ activity (↑ R123 efflux)	Independent of P-gp expression levels	[166]
P-gp*	Isolated and perfused rat liver	GNT 20 μM, within 5 min of administration	↑ activity (↑ R123 efflux)	Increase in intracellular availability of R123 efflux due to MRP2 inhibition	[144]
MRP1	Human small cell lung carcinoma cell line GLC4/ADR	GNT 200 μM, 1 h	↓ activity (↑ daunorubicin accumulation)	Competitive inhibition	[164, 171, 172]
MRP1	Rat brain microglia cell line MLS-9	GNT 200 μM, 2 h	↓ activity (↑ vincristine accumulation)	-	[173]
MRP1	Pancreatic cancer cell line Panc-1	GNT 100 μM, 2 h	↓ activity (↑ daunorubicin and vinblastine accumulation)	-	[174]
MRP1	Human erythrocytes	GNT (different concentrations), 15 min	↓ activity (↓ BCPCF) efflux). IC_{50}: 80 μM	-	[175]
MRP2*	Isolated and perfused rat liver	GNT 20 μM, within 5 min of administration	↓ activity (↓ bilirubin and BSP efflux)	Competitive inhibition	[144]
MRP4	Rat brain microglia cell line MLS-9	GNT 200 μM, 1 h	↓ activity (↓PMEA efflux)	-	[176]
MRP5	Rat brain microglia cell line MLS-9	GNT 200 μM, 1 h	↓ activity (↓ PMEA efflux)	-	[178]
BCRP	Breast cancer cell line MCF-7 MX100	GNT (different concentrations, 30 min)	↓ activity (↑mitoxantrone accumulation). IC_{50}: 14.9 μM	-	[180]
BCRP	Human leukemic cell line K562/BCRP	GNT 30 and 100 μM, 30 min	↓ activity (↑ topotecan accumulation)	Competitive inhibition	[137]
BCRP	Caco-2	GNT (different concentrations, 30 min)	↑ activity (↓ BODIPY-prazosin uptake)	-	[179]

**In vivo* or *ex vivo* reports harboring potential clinical relevance are indicated with * next to the modulated transporter

Regarding the effects observed after sustained treatments, Okura *et al*. found that 100 μM GNT did not change P-gp mRNA or protein levels in LS180 cells [166]. In contrast, we demonstrated that GNT increased P-gp protein levels at 1 μM concentration in Caco-2 [99] and at 1 and 10 μM concentrations in HepG2 cells [167]. The reason for the discrepancy could rely on the different GNT concentrations or cell lines used. Accordingly, R123 accumulation is decreased in Caco-2 cells after GNT treatment. Increased P-gp protein levels in this experimental model can explain this observation. HepG2 cells also showed increased P-gp activity when they were incubated with GNT 1 or 10 μM and calcein-AM was used as P-gp model substrate [167].

The molecular mechanism underlying P-gp induction by GNT depends on the experimental model and on the concentration of GNT involved. At 1 μM of GNT, P-gp is translationally regulated in HepG2 cells [167] while in Caco-2 cells P-gp is transcriptionally regulated, being this induction prevented by co-administration of the estrogen receptor antagonist ICI182/780. ER-β and SP-1 are probably involved [99]. At a higher concentration of GNT (10 μM), P-gp is transcriptionally regulated in HepG2 cells through the nuclear receptor PXR, whereas in Caco-2 cells P-gp protein expression remained unchanged under similar treatment conditions [99, 167].

3.2. MRP Regulation

MRPs can also undergo either a long-term or a short-term regulation by GNT. There is *in vitro* evidence showing MRP2 long-term regulation by GNT at concentrations similar to those achieved in plasma of patients consuming a soy-rich diet or hormonal supplements containing the isoflavone. Arias *et al*. described indeed an induction at the protein level by 1 μM GNT without changes in the mRNA in the human intestinal cell line Caco-2 [99]. This resulted in an enhanced efflux of the MRP model substrate DNP-SG and increased chemoresistance against 1-chloro-2,4-dinitrobenzene, cytotoxic compound precursor of DNP-SG. The authors demonstrated participation of the estrogen receptor β (ER-β), as the inducing effect was prevented by incubation with the antagonist ICI182/780. However, the absence of a modulation at the mRNA level suggests a non-classical mechanism, presently unknown. The effect of GNT on intestinal expression of MRPs *in vivo* was assessed in rats treated with a standardized soy extract [168]. Expression was evaluated for MRP1 and MRP2 at the mRNA level, showing a significant induction only for MRP2.

MRP2 regulation by GNT was also described in HepG2 cells [167]. In this case, GNT induced MRP2 expression at the protein level both at 1 and 10 μM GNT. Both inductions correlated with an enhanced activity, as determined assessing DNP-SG efflux. However, the underlying molecular mechanism differed between these two concentrations. A translational mechanism based on the microRNA hsa-miR379 down-regulation was demonstrated for GNT 1 μM. Conversely, a PXR-dependent transcriptional mechanism was found for GNT 10 μM. This latter regulation was supported by a previous study demonstrating retinoid X receptor α (RXR-α) (*i.e.* PXR partner) binding to MRP2 promoter during incubation with GNT 50 μM [169]. In another work with HepG2 cells, Hanet *et al*. evaluated MRP2 expression at the mRNA and protein level after GNT treatment, though for different treatment times and GNT concentrations. HepG2 cells were incubated with 2.2, 7.0, 22 and 70 μM GNT for 24 h and showed a decrease in the mRNA expression at the two higher concentrations without changes at the lower ones. Protein expression was assessed only for 70 μM and no changes were observed [170]. The absence of a typical concentration-response curve (*i.e.* higher concentration implies stronger induction) was previously described for GNT and other estrogens [99] and could rely on concentration dependent mechanisms, occurring simultaneously but with opposite effects on the transporter expression. Whereas inducing mechanisms prevail at lower concentrations, inhibiting mechanisms become activated at higher concentrations and counterbalance the induction. MRP3 expression was also assessed in both studies. However, no changes were observed, clearly indicating target gene specific regulation mechanisms. MRP4 regulation by GNT was assessed by Hanet *et al.*, although only at the mRNA level. In this case, GNT (70 μM) decreased MRP4 expression after 24 h [170]. Data on transporter regulation after *in vivo* administration of GNT are scarce and limited to one study performed in rats given soy extract showing an induction at the mRNA level of hepatic MRP1 and MRP2 [168]. As stated above, it is important to consider discrepancies in phase II metabolism between cell line models and intact animals, with concomitant differences in exposition to GNT aglycone *versus* GNT conjugates, which could result in erroneous extrapolation of the *in vitro* studies to the *in vivo* situation.

In addition to the regulation of the expression, GNT modulates the activity of different MRPs without changes in the expression (short-term regulation). No long after the first description of MRP1 by Cole *et al*. [29], it was reported that GNT prevents the reduced intracellular accumulation of daunorubicin displayed by non-P-gp multidrug resistant non-small cell lung cancer *in vitro*, suggesting an interaction with other transporters, different from P-gp [171]. The interaction with the unknown transporter was later proved to be based on a competitive mechanism [164]. Confirmation that GNT indeed interacts with MRP1, was finally made as GNT modulates its ATPase activity, resulting in daunorubicin efflux inhibition [172]. Later on, it was demonstrated that GNT also inhibits vincristine efflux in a MRP1 overexpressing microglia cell line [173] as well as daunorubicin and vinblastine efflux in pancreatic carcinoma cells [174]. Similarly, GNT inhibited

the efflux of the MRP1 substrate BCPCF-AM in human erythrocytes [175]. It should be noted that most of these acute effects were described for GNT concentrations up to 200 μM, almost 20-fold higher than the highest GNT levels achieved in plasma of subjects taking hormonal or nutritional supplements containing GNT, and thus may lack clinical relevance.

Similar results were obtained for short term regulation of MRP2, first studied in isolated perfused liver. In this model, GNT inhibited the secretion of bilirubin-glucuronides and bromosulphthalein-glutathione into bile. Considering that MRP2 plays a major role in the canalicular efflux of both substrates, an inhibition of this transporter by GNT can be suggested. The simultaneous detection of GNT and its metabolites in bile of Wistar rats reinforces a concept of a competitive mechanism [144]. It is not clear, however, whether other metabolites of GNT, apart from glucuronides could also exert the inhibitory effects described above. Apart from MRP1 and MRP2, MRP4 and MRP5 have been demonstrated to be inhibited by GNT, as suggested by an *in vitro* study using microglia cells expressing both transporters [176]. No data is available for the activity modulation of other MRPs by GNT.

3.3. BCRP Regulation

Ebert *et al*. studied the effect of GNT (1-50 μM) on BCRP mRNA levels in Caco-2 cells and reported no alteration after 48 h of incubation [177]. We confirmed this finding and reported no change in BCRP protein expression in Caco-2 cells (GNT 0.1-10 μM, 48 h) [100]. BCRP protein expression also remained unchanged in K562 cells after five days of exposure to GNT (1-3 μM) [138]. Conversely, in cancer gastric cells (MGC-803), mRNA levels of BCRP decreased after 15 μM of GNT for 24 h [178]. Thus, the effect of GNT on BCRP regulation seems to be tissue-specific.

Regarding the short-term regulation of BCRP by GNT, a study by Imai *et al.,* demonstrated an inhibitory action on BCRP-overexpressing cell lines [138]. In contrast, Schexnayder and Stratford reported that GNT exposure slightly stimulated BCRP activity in Caco-2 cells [179]. This controversy could be a consequence of the different cells and expression systems used. Unfortunately no studies are available on the effect of GNT in animal models preventing from making any conclusion regarding its potential role in BCRP mediated elimination of xenobiotics, either after occasional or prolonged exposition.

4. GNT-DRUG INTERACTIONS

According to the evidence presented here, it is clear that the alteration in the expression and activity of ABC transporters by GNT or associated dietary constituents could modify the *in vivo* disposition of therapeutic agents, consequently affecting their pharmacokinetics and therapeutic efficacy.

Until now, most of the GNT-drug interactions were observed after administration of GNT at high, single, doses; such is the case for paclitaxel and irinotecan hydrochloride. As many anticancer drugs, paclitaxel is a substrate for P-gp [181], MRP2 [182] and BCRP [183] and its metabolism in humans is mainly catalyzed by CYP3A4 and CYP2C8 [184]. Oral administration of GNT (10 mg/kg) to rats, 30 min before a single dose of paclitaxel (30 mg/kg, p.o.) increased the AUC and Cmax, decreased total plasma clearance and increased the bioavailability of the anticancer agent. GNT also increased the AUC and reduced the total clearance of intravenously administered paclitaxel (3 mg/kg). These observations could be attributed to the already reported acute inhibition of P-gp and MRP2 by GNT [144, 185]. In addition, Yokooji *et al*. reported an inhibition of biliary and intestinal MRP2-mediated secretion of irinotecan hydrochloride (CPT-11, 10 mg/kg i.v.) and its metabolites by GNT (54 mg/kg i.v, 10 min before CPT-11 administration) in rats [186].

GNT and the phytoestrogen daidzein are both known inhibitors of BCRP [138, 180]. Considering that both GNT and daidzein are transported by BCRP [140], a competitive inhibition seems feasible. Enrofloxacin and its derivative ciprofloxacin, both BCRP substrates, are antibacterial drugs used in veterinary and human medicine, respectively [187, 188]. BCRP is present in the mammary epithelia, increases during lactation and mediates the secretion of endo- and xenobiotics into the milk [79, 187, 189]. The effect of GNT on enrofloxacin milk concentration was evaluated infusing enrofloxacin i.v. alone (2.5 mg/kg) or together with GNT as BCRP inhibitor (0.8 mg/kg, i.m.) in lactating sheep. Plasma pharmacokinetic parameters for enrofloxacin (AUC and Cmax) did not differ between animals that received GNT and controls. However, GNT decreased the content of enrofloxacin in milk demonstrating a lower transfer of drugs that are BCRP substrates and reducing exposure of their pups [187]. Nitrofurantoin (NFT), another recognized BCRP substrate, is an antibacterial agent prescribed to treat urinary tract infections even in lactating women [190-192]. A decreased NFT secretion into milk was ob-

served after a single oral dose to lactating sheep (20 mg/kg) in animals fed for 15 days with a diet containing isoflavones in comparison to animals that received isoflavone-free diet. NFT secretion was further reduced when animals were acutely treated with GNT and daidzein (20 mg/kg, p.o., 5 min before NFT administration) [193]. Additionally, Merino *et al.* co-administered NFT (20 mg/kg) along with a mixture of GNT (100 mg/kg) and daidzein (100 mg/kg) to $BCRP^{-/-}$ and wild-type mice. The authors observed a significantly decrease in BCRP-mediated NFT secretion into milk and into bile in wild-type mice, whereas no effect of GNT was observed in the knock-out mice [194]. Furthermore, the synthetic fluoroquinolone danofloxacin (DAN) is used in veterinary medicine as an antibacterial agent [195]. DAN is a substrate for BCRP and is actively secreted into milk where it reaches concentrations up to 10-fold higher than in plasma [196, 197]. Pérez *et al.* observed that DAN concentrations in milk as well as the AUC (0-24 h) milk/plasma ratio were decreased in sheep fed with soy-enriched forage for 15 days in comparison with those fed with standard diet [198]. All these results proved that secretion of BCRP substrates into milk can be reduced by isoflavones present in the diet. In humans, mammary BCRP inhibition by GNT could result in a reduced exposure of infants if the mother is medicated with drugs that are BCRP substrates. Although this effect could be beneficial to avoid exposure of the suckling babies, it may also reduce the efficiency of treatments directed to newborns through the milk.

In contrast to the observations described above, sustained administration of GNT results in a decrease of the bioavailability of co-administered drugs, agreeing well with *in vivo* and *in vitro* reports showing an induction of drug transporters by the phytoestrogen. An *in vivo* study administering 1 g of GNT to 18 healthy volunteers for 14 consecutive days showed decreased systemic exposure to the P-gp substrate talinolol, suggesting that GNT would be a potent inducer of this transporter [199]. In addition, pharmacokinetics of imatinib, a drug approved for treatment of chronic myelogenous leukemia (CML) and other myeloproliferative diseases, was also reported to be modified by GNT [200]. Indeed, administration of GNT to rats (50 or 100 mg/kg/day for consecutive 15 days) reduced imatinib exposure, as concluded after Cmax and AUC examination [201]. The authors suggested an increased drug metabolism after exposition to GNT as demonstrated *in vitro* in V79 cells treated with the phytoestrogen [202]. In addition, as imatinib is a P-gp substrate [203-205] and this transporter expression can be induced by GNT [99], enhanced drug efflux could be expected. In line with this assumption, Alves *et al*., recently described that Reversin 205, used as a P-gp inhibitor, decreased the IC_{50} for imatinib in two resistant CML cell lines [206].

CONCLUSION

The interaction between GNT and the ABC transporters occurs at several levels, either as substrate, inhibitor or modulator of their expression. Relevant consequences are induction or repression of gene transcription, translational modulations or direct interactions with the ABC transporter structures resulting in stimulation or inhibition of transport activity.

During the last decades it was proposed that GNT ingestion may result in cytoprotection and reduction of the incidence of specific tumors. Taken together, the data collected in this review lead to the conclusion that sustained exposition to low doses of GNT, usually associated with intake of dietary supplements or rich-soy diet, results in cytoprotection. This action is due in part to positive modulation of ABC transporters involved in extrusion of toxic substrates, namely dietary contaminants, pollutants, etc. On the other hand, at high doses and short periods of exposure, GNT behaves as a competitive inhibitor of these same ABC transporters. The consequence is an increased cytotoxicity of their substrates.

In the case of substrates of the ABC transporters of therapeutic application, it would be expected that GNT alters their disposition and pharmacokinetics and consequently their therapeutic efficacy. Sustained exposure to GNT, resulting in induction of the expression of ABC transporters, would result in increased drug clearance. Data supporting this assumption was reported in this review. Contrarily, at high doses of GNT and short periods of exposure, increased intracellular accumulation of ABC substrates is observed, most likely as a consequence of competitive inhibition.

This review compiled information about regulation of ABC transporters by GNT with special emphasis on P-gp, MRPs and BCRP. Many of the data arise from studies using concentrations of GNT in the range found in plasma from Asian population or after administration of GNT-enriched formulations. However, it should be noted that the activity of phase II enzymes in cell lines is low if compared to normal tissues, which is expected to result in different exposition to GNT aglycone respect to their conjugates. Since most of the studies reviewed here rely on *in vitro* models, extrapolation of the data and conclusions to humans must be cau-

tiously done. Further investigations *in vivo* in experimental animals or pharmacokinetics studies in humans will help to predict and overcome GNT-drug interactions, ultimately leading to improve the guidance for drug prescription.

LIST OF ABBREVIATIONS

AP-1	=	activating protein 1
AUC	=	area under the curve
BCPCF	=	2,7'-bis-(carboxypropyl)-5(6)-carboxyfluorescein
BSP	=	bromsulphthalein
C/EBP-α	=	CCAAT/enhancer-binding protein alpha
C/EBP-β	=	CCAAT/enhancer-binding protein beta
HNF-1	=	hepatocyte nuclear factor 1
HNF-3β	=	hepatocyte nuclear factor 3 beta
PMEA	=	9-(2-Phophonylmethoxyethyl)adenine
SP-1	=	specificity protein 1.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGEMENTS

Juan Pablo Rigalli, Nadia Ciriaci, Viviana Catania and María Laura Ruiz conceived, designed, and drafted the manuscript. Aldo Domingo Mottino carefully revised the manuscript. All authors read and approved the final work.

This review study was supported by grants from Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) [grant number PIP 2012-0240 (to A.D.M.)], and from Fondo para la Investigación Científica y Tecnológica (FONCyT) [grant number PICT 0360 (to V.A.C.) and grant number PICT 2013-1573 (to M.L.R.)].

REFERENCES

[1] Choudhuri, S.; Klaassen, C.D. Structure, function, expression, genomic organization, and single nucleotide polymorphisms of human ABCB1 (MDR1), ABCC (MRP), and ABCG2 (BCRP) efflux transporters. *Int. J. Toxicol.*, **2006**, *25*(4), 231-259.

[2] Glavinas, H.; Krajcsi, P.; Cserepes, J.; Sarkadi, B. The role of ABC transportersin drug resistance, metabolism and toxicity. *Curr. Drug Deliv.*, **2004**, *1*(1), 27-42.

[3] Beringer, P.M.; Slaughter, R.L. Transporters and their impact on drug disposition. *Ann. Pharmacother.*, **2005**, *39*(6), 1097-1108.

[4] Juliano, R.L.; Ling, V. A surface glycoprotein modulating drug permeability in Chinese hamster ovary cell mutants. *Biochim. Biophys. Acta*, **1976**, *455*, 152-162.

[5] Schinkel, A.H.; Borst, P. Multidrug resistance mediated by P-glycoproteins. *Semin. Cancer Biol.*, **1991**, *2*(4), 213-226.

[6] Cordon-Cardo, C.; O'Brien, J.P.; Boccia, J.; Casals, D.; Bertino, J.R.; Melamed, M.R. Expression of the multidrug resistance gene product (P-glycoprotein) in human normal and tumor tissues. *J. Histochem. Cytochem.*, **1990**, *38*(9), 1277-1287.

[7] Fojo, A.T.; Ueda, K.; Slamon, D.J.; Poplack, D.G.; Gottesman, M.M.; Pastan, I. Expression of a multidrug-resistance gene in human tumors and tissues. *Proc. Natl. Acad. Sci. USA*, **1987**, *84*(1), 265-269.

[8] Merkel, D.E.; Fuqua, S.A.; McGuire, W.L. P-glycoprotein in breast cancer. *Cancer Treat. Res.*, **1989**, *48*, 97-105.

[9] Gatmaitan, Z.C.; Arias, I.M. Structure and function of P-glycoprotein in normal liver and small intestine. *Adv. Pharmacol.*, **1993**, *24*, 77-97.

[10] Begley, D.J.; Lechardeur, D.; Chen, ZD.; Rollinson, C.; Bardoul, M.; Roux, F.; Scherman, D.; Abbott, N.J. Functional expression of P-glycoprotein in an immortalised cell line of rat brain endothelial cells, RBE4. *J. Neurochem.*, **1996**, 67, 988-995.

[11] Lee, C.H. Induction of P-glycoprotein mRNA transcripts by cycloheximide in animal tissues: evidence that class I Pgp is transcriptionally regulated whereas class II Pgp is post-transcriptionally regulated. *Mol. Cell Biochem.*, **2001**, *216*, 103-110.

[12] Tsuji, A.; Terasaki, T.; Takabatake, Y.; Tenda, Y.; Tamai, I.; Yamashima, T.; Moritani, S.; Tsuruo, T.; Yamashita, J. P-glycoprotein as the drug efflux pump in primary cultured bovine brain capillary endothelial cells. *Life Sci.*, **1992**, *51*, 1427-1437.

[13] Rao, V.V.; Dahlheimer, J.L.; Bardget, M.E.; Snyder, A.Z.; Finch, R.A.; Sartorelli, AC.; Piwnica-Worms, D. Choroid plexus epithelial expression of MDR1 P glycoprotein and multidrug resistance-associated protein contribute to the blood-cerebrospinal-fluid drug-permeability barrier. *Proc. Natl. Acad. Sci. USA*, **1999**, *96*, 3900-3905.

[14] Ushigome, F.; Takanaga, H.; Matsuo, H.; Yanai, S.; Tsukimori, K.; Nakano, H.; Uchiumi, T.; Nakamura, T.; Kuwano, M.; Ohtani, H.; Sawada, Y. Human placental transport of vinblastine, vincristine, digoxin and progesterone: contribution of P-glycoprotein. *Eur. J. Pharmacol.*, **2000**, *408*, 1-10.

[15] Smit, J.J.; Schinkel, A.H.; Mol, C.A.; Majoor, D.; Mooi, W.J.; Jongsma, A.P.; Lincke, C.R.; Borst, P. Tissue distribution of the human MDR3 P-glycoprotein. *Lab. Invest.*, **1994**, 71(5), 638-649. Erratum in: *Lab. Invest.*, **1995**, *72*(3).

[16] Schinkel, A.H.; Wagenaar, E.; Mol, C.A.; van Deemter, L. P-glycoprotein in the blood-brain barrier of mice influences the brain penetration and pharmacological activity of many drugs. *J. Clin. Invest.,* **1996***, 97*, 2517-2524.

[17] Chan, L.M.; Lowes, S.; Hirst, B.H. The ABCs of drug transport in intestine and liver: efflux proteins limiting drug absorption and bioavailability. *Eur J Pharm Sci.,* **2004**, *21*(1), 25-51.

[18] Thiebaut, F.; Tsuruo, T.; Hamada, H.; Gottesman, M.M.; Pastan, I.; Willingham, M.C. Cellular localization of the multidrug-resistance gene product P-glycoprotein in normal human tissues. *Proc. Natl. Acad. Sci. USA*, **1987**, *84*, 7735-7738.

[19] Terao, T.; Hisanaga, E.; Sai, Y.; Tamai, I.; Tsuji, A. Active secretion of drugs from the small intestinal epithelium in rats by P-glycoprotein functioning as an absorption barrier. *J. Pharm. Pharmacol.*, **1996**, *48*, 1083-1089.

[20] Tsuji, A.; Tamai, I. Carrier-mediated intestinal transport of drugs. *Pharm. Res.*, **1996**, *13*, 963-977.

[21] Cordon-Cardo, C.; O'Brien, J.P.; Casals, D.; Rittman-Grauer, L.; Biedler J.L.; Melamed, M.R.; Bertino, J.R. Multidrug-resistance gene (P-glycoprotein) is expressed by endothelial cells at blood-brain barrier sites. *Proc. Natl. Acad. Sci. USA*, **1989**, *86*(2), 695-698.

[22] Schinkel, A.H.; Jonker, J,W. Mammalian drug efflux transporters of the ATP binding cassette (ABC) family: an overview. *Adv. Drug Deliv. Rev.,* **2003**, *55*(1), 3-29.

[23] Klaassen, C.D.; Aleksunes, L.M. Xenobiotic, bile acid, and cholesterol transporters: function and regulation. *Pharmacol. Rev*., **2010**, *62*(1), 1-96.

[24] Ernest, S.; Bello-Reuss, E. Secretion of platelet-activating factor is mediated by MDR1 P-glycoprotein in cultured human mesangial cells. *J. Am. Soc. Nephrol.,* **1999**, *10*(11), 2306-2313.

[25] Masuda, M.; Nakai, E.; Mizutani, T. Study of oxidized lipids as endogenous substrates of P-gp (ABCB1). *Drug Metab. Lett.,* **2008**, *2*(4), 238-244.

[26] Bakos, E.; Evers, R.; Calenda, G.; Tusnády, G.E.; Szakács, G.; Váradi, A.; Sarkadi, B. Characterization of the amino-terminal regions in the human multidrug resistance protein (MRP1). *J. Cell Sci*., **2000**, *113*(24), 4451-4461.

[27] Borst, P.; Evers, R.; Kool, M.; Wijnholds, J. A family of drug transporters: the multidrug resistance-associated proteins. *J. Natl. Cancer Inst*., **2000**, *92*(16), 1295-1302.

[28] Kruh, G.D.; Belinsky, M.G. The MRP family of drug efflux pumps. *Oncogene*, **2003**, *22*(47), 7537-7552.

[29] Cole, S.P.; Bhardwaj, G.; Gerlach, J.H.; Mackie, J.E.; Grant, C.E.; Almquist, K.C.; Stewart, A.J.; Kurz, E.U.; Duncan, A.M.; Deeley, R.G. Overexpression of a transporter gene in a multidrug-resistant human lung cancer cell line. *Science*, **1992**, 258(5088), 1650-1654. Erratum in: *Science*, **1993**, *260*(5110), 879.

[30] Valente, R.C.; Capella, L.S.; Nascimento, C.R.; Lopes, A.G.; Capella, M.A. Modulation of multidrug resistance protein (MRP1/ABCC1) expression: a novel physiological role for ouabain. *Cell Biol. Toxicol.*, **2007**, *23*(6), 421-427.

[31] Berggren, S.; Gall, C.; Wollnitz, N.; Ekelund, M.; Karlbom, U.; Hoogstraate, J.; Schrenk, D.; Lennernäs, H. Gene and protein expression of P-glycoprotein, MRP1, MRP2, and CYP3A4 in the small and large human intestine. *Mol. Pharm.*, **2007**, *4*(2), 252-257.

[32] Nies, A.T.; Jedlitschky, G.; König, J.; Herold-Mende, C.; Steiner, H.H.; Schmitt, H.P.; Keppler, D. Expression and immunolocalization of the multidrug resistance proteins, MRP1-MRP6 (ABCC1-ABCC6), in human brain. *Neuroscience*, **2004**, *129*(2), 349-360.

[33] Albermann, N.; Schmitz-Winnenthal, F.H.; Z'graggen, K.; Volk, C.; Hoffmann, M.M.; Haefeli, W.E.; Weiss, J. Expression of the drug transporters MDR1/ABCB1, MRP1/ABCC1, MRP2/ABCC2, BCRP/ABCG2, and PXR in peripheral blood mononuclear cells and their relationship with the expression in intestine and liver. *Biochem. Pharmacol*., **2005**, *70*(6), 949-958.

[34] Büchler, M.; König, J.; Brom, M.; Kartenbeck, J.; Spring, H.; Horie, T.; Keppler, D. cDNA cloning of the hepatocyte canalicular isoform of the multidrug resistance protein, cMrp, reveals a novel conjugate export pump deficient in hyperbilirubinemic mutant rats. *J. Biol. Chem.*, **1996**, *271*(25), 15091-15098.

[35] Schaub, TP.; Kartenbeck, J.; König, J.; Vogel, O.; Witzgall, R.; Kriz, W.; Keppler, D. Expression of the conjugate export pump encoded by the mrp2 gene in the apical membrane of kidney proximal tubules. *J. Am. Soc. Nephrol*., **1997**, *8*(8), 1213-1221.

[36] Mottino, A.D.; Hoffman, T.; Jennes, L.; Vore, M. Expression and localization of multidrug resistant protein mrp2 in rat small intestine. *J. Pharmacol. Exp. Ther.*, **2000**, *293*(3), 717-723.

[37] Potschka, H.; Fedrowitz, M.; Löscher, W. Multidrug resistance protein MRP2 contributes to blood-brain barrier function and restricts antiepileptic drug activity. *J. Pharmacol. Exp. Ther*., **2003**, *306*(1), 124-131.

[38] Korita, P.V.; Wakai, T.; Shirai, Y.; Matsuda, Y.; Sakata, J.; Takamura, M.; Yano, M.; Sanpei, A.; Aoyagi, Y.; Hatakeyama, K.; Ajioka, Y. Multidrug resistance-associated protein 2 determines the efficacy of cisplatin in patients with hepatocellular carcinoma. *Oncol. Rep*., **2010**, *23*(4), 965-972.

[39] Yamasaki, M.; Makino, T.; Masuzawa, T.; Kurokawa, Y.; Miyata, H.; Takiguchi, S.; Nakajima, K.; Fujiwara, Y.; Matsuura, N.; Mori, M.; Doki, Y. Role of multidrug resistance protein 2 (MRP2) in chemoresistance and clinical outcome in oesophageal squamous cell carcinoma. *Br. J. Cancer*, **2011**, *104*(4), 707-713.

[40] Toh, S.; Wada, M.; Uchiumi, T.; Inokuchi, A.; Makino, Y.; Horie, Y.; Adachi, Y.; Sakisaka, S.; Kuwano, M. Genomic structure of the canalicular multispecific organic anion-transporter gene (MRP2/cMOAT) and mutations in the ATP-binding-cassette region in Dubin-Johnson syndrome. *Am. J. Hum. Genet.*, **1999**, *64*(3), 739-746.

[41] König, J.; Rost, D.; Cui, Y.; Keppler, D. Characterization of the human multidrug resistance protein isoform MRP3 localized to the basolateral hepatocyte membrane. *Hepatology*, **1999**, 29(4), 1156-1163.

[42] Zelcer, N.; van de Wetering, K.; Hillebrand, M.; Sarton, E.; Kuil, A.; Wielinga, P.R.; Tephly, T.; Dahan, A.; Beijnen, J.H.; Borst, P. Mice lacking multidrug resistance protein 3 show altered morphine pharmacokinetics and morphine-6-glucuronide antinociception. *Proc. Natl. Acad. Sci. USA*, **2005**, *102*(20), 7274-7279.

[43] Kuroda, M.; Kobayashi, Y.; Tanaka, Y.; Itani, T.; Mifuji, R.; Araki, J.; Kaito, M.; Adachi, Y. Increased hepatic and renal expressions of multidrug resistance-associated protein 3 in Eisai hyperbilirubinuria rats. *J. Gastroenterol. Hepatol.,* **2004**,*19*(2), 146-153.

[44] Rost, D.; König, J.; Weiss, G.; Klar, E.; Stremmel, W.; Keppler, D. Expression and localization of the multidrug resistance proteins MRP2 and MRP3 in human gallbladder epithelia. *Gastroenterology*, **2001**, *121*(5), 1203-1208.

[45] Borst, P.; Zelcer, N.; van de Wetering, K. MRP2 and 3 in health and disease. *Cancer Lett*., **2006**, *234*(1), 51-61.

[46] Zollner, G.; Wagner, M.; Fickert, P.; Silbert, D.; Fuchsbichler, A.; Zatloukal, K.; Denk, H.; Trauner, M. Hepatobiliary transporter expression in human hepatocellular carcinoma. *Liver Int*., **2005**, *25*(2), 367-379.

[47] Benderra, Z.; Faussat, A.M.; Sayada, L.; Perrot, J.Y.; Tang, R.; Chaoui, D.; Morjani, H.; Marzac, C.; Marie, J.P.; Legrand, O. MRP3, BCRP, and P-glycoprotein activities are prognostic factors in adult acute myeloid leukemia. *Clin. Cancer Res*., **2005**, *11*(21), 7764-7772.

[48] Borst, P.; de Wolf, C.; van de Wetering, K. Multidrug resistance-associated proteins 3, 4, and 5. *Pflugers Arch*., **2007**, *453*(5), 661-673.

[49] Bergen, A.A.; Plomp, A.S.; Schuurman, E.J.; Terry, S.; Breuning, M.; Dauwerse, H.; Swart, J.; Kool, M.; van Soest, S.; Baas, F.; ten Brink, J.B.; de Jong, P.T. Mutations in ABCC6 cause pseudoxanthoma elasticum. *Nat. Genet.,* **2000**, *25*(2), 228-231.

[50] Domanitskaya, N.; Wangari-Talbot, J.; Jacobs, J.; Peiffer, E.; Mahdaviyeh, Y.; Paulose, C.; Malofeeva, E.; Foster, K.; Cai, K.Q.; Zhou, Y.; Egleston, B.; Hopper-Borge, E. Abcc10 status affects mammary tumour growth, metastasis, and docetaxel treatment response. *Br. J. Cancer,* **2014**, *111*(4), 696-707.

[51] Guo, Y.; Kotova, E.; Chen, Z.S.; Lee, K.; Hopper-Borge, E.; Belinsky, M.G.; Kruh, G.D. MRP8, ATP-binding cassette C11 (ABCC11), is a cyclic nucleotide efflux pump and a resistance factor for fluoropyrimidines 2',3'-dideoxycytidine and 9'-(2'-phosphonylmethoxyethyl) adenine. *J. Biol. Chem.,* **2003**, *278*(32), 29509-29514.

[52] Bera, T.K.; Iavarone, C.; Kumar, V.; Lee, S.; Lee, B.; Pastan, I. MRP9, an unusual truncated member of the ABC transporter superfamily, is highly expressed in breast cancer. *Proc. Natl. Acad. Sci. USA*, **2002**, *99*(10), 6997-7002.

[53] Shimizu, H.; Taniguchi, H.; Hippo, Y.; Hayashizaki, Y.; Aburatani, H.; Ishikawa T. Characterization of the mouse Abcc12 gene and its transcript encoding an ATP-binding cassette transporter, an orthologue of human ABCC12. *Gene*, **2003**, *310*, 317-328.

[54] Ono, N.; Van der Heijden, I.; Scheffer, G.L.; Van de Wetering, K.; Van Deemter, E.; De Haas, M.; Boerke, A.; Gadella, B.M.; De Rooij, D.G.; Neefjes, J.J.; Groothuis, T.A.; Oomen, L.; Brocks, L.; Ishikawa, T.; Borst, P. Multidrug resistance-associated protein 9 (ABCC12) is present in mouse and boar sperm. *Biochem. J.,* **2007**, *406*(1), 31-40.

[55] Wijnholds, J.; Evers, R.; van Leusden, M.R.; Mol, C.A.; Zaman, G.J.; Mayer, U.; Beijnen, J.H.; van der Valk, M.; Krimpenfort, P.; Borst, P. Increased sensitivity to anticancer drugs and decreased inflammatory response in mice lacking the multidrug resistance-associated protein. *Nat. Med.* **1997**, *3*(11), 1275-1279.

[56] Robbiani, D.F.; Finch, R.A.; Jäger, D.; Muller, W.A.; Sartorelli, A.C.; Randolph, G.J. The leukotriene C(4) transporter MRP1 regulates CCL19 (MIP-3beta, ELC)-dependent mobilization of dendritic cells to lymph nodes. *Cell*, **2000**, *103*(5), 757-768.

[57] Ji, B.; Ito, K.; Suzuki, H.; Sugiyama, Y.; Horie, T. Multidrug resistance-associated protein2 (MRP2) plays an important role in the biliary excretion of glutathione conjugates of 4-hydroxynonenal. *Free Radic. Biol. Med.,* **2002**, *33*(3), 370-378.

[58] Takahashi, K.; Tatsunami, R.; Sato, K.; Tampo, Y. Multidrug resistance associated protein 1 together with glutathione plays a protective role against 4-hydroxy-2-nonenal-induced oxidative stress in bovine aortic endothelial cells. *Biol. Pharm. Bull.,* **2012**, 35(8), 1269-1274.

[59] Arana, M.R.; Tocchetti, G.N.; Domizi, P.; Arias, A.; Rigalli, J.P.; Ruiz, M.L.; Luquita, M.G.; Banchio, C.; Mottino A.D.; Villanueva, S.S. Coordinated induction of GST and MRP2 by cAMP in Caco-2 cells: Role of protein kinase A signaling pathway and toxicological relevance. *Toxicol. Appl. Pharmacol.,* **2015**, *287*(2), 178-190.

[60] Choudhuri, S.; Cherrington, N.J.; Li, N.; Klaassen, C.D. Constitutive expression of various xenobiotic and endobiotic transporter mRNAs in the choroid plexus of rats. *Drug Metab. Dispos.*, **2003**, *31*(11), 1337-1345.

[61] Bauer, B.; Hartz, A.M.; Lucking, J.R.; Yang, X.; Pollack, G.M.; Miller, D.S. Coordinated nuclear receptor regulation of the efflux transporter, Mrp2, and the phase-II metabolizing enzyme, GSTpi, at the blood-brain barrier. *J. Cereb. Blood Flow Metab.,* **2008**, *28*(6), 1222-1234.

[62] Van Aubel, R.A.; Peters, J.G.; Masereeuw, R.; Van Os, C.H.; Russel, F.G. Multidrug resistance protein mrp2 mediates ATP-dependent transport of classic renal organic anion p-aminohippurate. *Am. J. Physiol. Renal Physiol.*, **2000**, *279*(4), F713-F717.

[63] Tagami, M.; Kusuhara, S.; Imai, H.; Uemura, A.; Honda, S.; Tsukahara, Y.; Negi, A. MRP4 knockdown enhances migration, suppresses apoptosis, and produces aggregated morphology in human retinal vascular endothelial cells. *Biochem. Biophys. Res. Commun.,* **2010**, *400*(4), 593-598.

[64] Lien, L.M.; Chen, Z.C.; Chung, C.L.; Yen, T.L.; Chiu, H.C.; Chou, D.S.; Huang, S.Y.; Sheu, J.R.; Lu, W.J.; Lin, K.H. Multidrug resistance protein 4 (MRP4/ABCC4) regulates thrombus formation *in vitro* and *in vivo*. *Eur. J. Pharmacol.,* **2014**, *737*, 159-167.

[65] Osycka-Salut, C.; Diez, F.; Burdet, J.; Gervasi, M.G.; Franchi, A.; Bianciotti, L.G.; Davio, C.; Perez-Martinez, S. Cyclic AMP efflux, *via* MRPs and A1 adenosine receptors, is critical for bovine sperm capacitation. *Mol. Hum. Reprod.,* **2014**, *20*(1), 89-99.

[66] Jansen, R.S.; Küçükosmanoglu, A.; de Haas, M.; Sapthu, S.; Otero, J.A.; Hegman, *I.E.*; Bergen, A.A.; Gorgels, T.G.; Borst, P.; van de Wetering, K. ABCC6 prevents ectopic mineralization seen in pseudoxanthoma elasticum by inducing cellular nucleotide release. *Proc. Natl. Acad. Sci. USA,* **2013**, *110*(50), 20206-20211.

[67] Jansen, R.S.; Duijst, S.; Mahakena, S.; Sommer, D.; Szeri, F.; Váradi, A.; Plomp, A.; Bergen, A. A.; Oude Elferink, R.P.; Borst, P.; van de Wetering, K. ABCC6-mediated ATP secretion by the liver is the main source of the mineralization inhibitor inorganic pyrophosphate in the systemic circulation-brief report. *Arterioscler. Thromb. Vasc. Biol.*, **2014**, *34*(9), 1985-1989.

[68] Doyle, L.A.; Yang, W.; Abruzzo, L.V.; Krogmann, T.; Gao, Y.; Rishi, A.K.; Ross, D.D. A multidrug resistance transporter from human MCF-7 breast cancer cells. *Proc. Natl. Acad. Sci. U. S. A.,* **1998**, *95*(26),15665-15670. Erratum in: *Proc. Natl. Acad. Sci. USA*,**1999**, *96*(5), 2569.

[69] Allikmets, R.; Schriml, L. M.; Hutchinson, A.; Romano-Spica, V.; Dean, M. A human placenta-specific ATPbinding cassette gene (ABCP) on chromosome 4q22 that is involved in multidrug resistance. *Cancer Res.*, **1998**, *58*, 5337-5339.

[70] Rabindran, S. K.; He, H.; Singh, M.; Brown, E.; Collins, K. I.; Annable, T.; Greenberger, L. M. Reversal of a novel multidrug resistance mechanism in human colon carcinoma cells by fumitremorgin C. *Cancer Res.*, **1998**, *58*, 5850-5858.

[71] Henriksen, U.; Fog, J.U.; Litman, T.; Gether, U. Identification of intra- and intermolecular disulfide bridges in the multidrug resistance transporter ABCG2. *J. Biol. Chem.*, **2005**, *280*, 36926-36934.

[72] Kage, K.; Fujita, T.; Sugimoto, Y. Role of Cys-603 in dimer/oligomer formation of the breast cancer resistance protein BCRP/ABCG2. *Cancer Sci.*, **2005**, 96, 866-872.

[73] Ni, Z.; Mark, M.E.; Cai, X.; Mao, Q. Fluorescence resonance energy transfer (FRET) analysis demonstrates dimer/oligomer formation of the human breast cancer resistance protein (BCRP/ABCG2) in intact cells. *Int. J. Biochem. Mol. Biol.*, **2010**, *1*, 1-11.

[74] Xu, J.; Peng, H.; Chen, Q.; Liu, Y.; Dong, Z.; Zhang, J.T. Oligomerization domain of the multidrug resistance-associated transporter ABCG2 and its dominant inhibitory activity. *Cancer Res.*, **2007**, 67, 4373-4381.

[75] Bruhn, O.; Cascorbi, I. Polymorphisms of the drug transporters ABCB1, ABCG2, ABCC2 and ABCC3 and their impact on drug bioavailability and clinical relevance. *Expert Opin. Drug Metab. Toxicol.*, **2014**, 10(10), 1337-1354.

[76] Durmus, S.; Hendrikx, J.J.; Schinkel, A.H. Apical ABC transporters and cancer chemotherapeutic drug disposition. *Adv. Cancer Res.,* **2015**, *125*, 1-41.

[77] Mao, Q.; Unadkat, J.D. Role of the breast cancer resistance protein (BCRP/ABCG2) in drug transport--an update. *AAPS J.*, **2015**, *17*(1), 65-82.

[78] Kim, B.; Fatayer, H.; Hanby, A.M.; Horgan, K.; Perry, S.L.; Valleley, E.M.; Verghese, E.T.; Williams, B.J.; Thorne, J.L.; Hughes, T.A. Neoadjuvant chemotherapy induces expression levels of breast cancer resistance protein that pre-

dict disease-free survival in breast cancer. *PLoS One*, **2013**, *8*(5), e62766.

[79] Jonker, J.W.; Merino, G.; Musters, S.; van Herwaarden, A.E.; Bolscher, E.; Wagenaar, E.; Mesman, E.; Dale, T.C.; Schinkel, A.H. The breast cancer resistance protein BCRP (ABCG2) concentrates drugs and carcinogenic xenotoxins into milk. *Nat. Med.*, **2005**, *11*(2), 127-129.

[80] van Herwaarden, A.E.; Wagenaar, E.; Merino, G.; Jonker, J.W.; Rosing, H.; Beijnen, J.H.; Schinkel, A.H. Multidrug transporter ABCG2/breast cancer resistance protein secretes riboflavin (vitamin B2) into milk. *Mol. Cell Biol.*, **2007**, *27*(4), 1247-1253.

[81] Tanaka, T.; Uchiumi, T.; Hinoshita, E.; Inokuchi, A.; Toh, S.; Wada, M.; Takano, H.; Kohno, K.; Kuwano, M. The human multidrug resistance protein 2 gene: functional characterization of the 5'-flanking region and expression in hepatic cells. *Hepatology*, **1999**, *30*(6), 1507-1512.

[82] Stöckel, B.; König, J.; Nies, A.T.; Cui, Y.; Brom, M.; Keppler, D. Characterization of the 5'-flanking region of the human multidrug resistance protein 2 (MRP2) gene and its regulation in comparison with the multidrug resistance protein 3 (MRP3) gene. *Eur. J. Biochem.,* **2000**, *267*(5), 1347-1358.

[83] Johnson, D.R.; Klaassen, C.D. Regulation of rat multidrug resistance protein 2 by classes of prototypical microsomal enzyme inducers that activate distinct transcription pathways. *Toxicol. Sci.*, **2002**, *67*(2), 182-189.

[84] Kast, H.R.; Goodwin, B.; Tarr, P.T.; Jones, S.A.; Anisfeld, A.M.; Stoltz, C.M.; Tontonoz, P.; Kliewer, S.; Willson, T.M.; Edwards, P.A. Regulation of multidrug resistance-associated protein 2 (ABCC2) by the nuclear receptors pregnane X receptor, farnesoid X-activated receptor, and constitutive androstane receptor. *J. Biol. Chem.,* **2002**, *277*(4), 2908-2915.

[85] Bailey-Dell, K.J.; Hassel, B.; Doyle, L.A.; Ross, D.D. Promoter characterization and genomic organization of the human breast cancer resistance protein (ATP-binding cassette transporter G2) gene. *Biochim. Biophys. Acta*, **2001**, *1520*, 234-241.

[86] di Masi, A.; De Marinis, E.; Ascenzi, P.; Marino, M. Nuclear receptors CAR and PXR: Molecular, functional, and biomedical aspects. *Mol. Aspects Med.,* **2009**, *30*(5), 297-343.

[87] Köhle, C.; Bock, K.W. Coordinate regulation of human drug-metabolizing enzymes, and conjugate transporters by the Ah receptor, pregnane X receptor and constitutive androstane receptor. *Biochem. Pharmacol.,* **2009**, *77*(4), 689-699.

[88] Geick, A.; Eichelbaum, M.; Burk, O. Nuclear receptor response elements mediate induction of intestinal MDR1 by rifampin. *J. Biol. Chem.*, **2001**, *276*, 14581-14587.

[89] Jiang, H.; Chen, K.; He, J.; Pan, F.; Li, J.; Chen, J.; Chen, W.; Liang, H. Association of pregnane X receptor with multidrug resistance-related protein 3 and its role in human colon cancer chemoresistance. *J. Gastrointest. Surg*., **2009**, *13*(10), 1831-1838.

[90] Qiao, E.Q.; Yang, H.J. Effect of pregnane X receptor expression on drug resistance in breast cancer. *Oncol. Lett.,* **2014**, *7*(4), 1191-1196.

[91] Baker, E.K.; Johnstone, R.W.; Zalcberg, J.R.; El-Osta, A. Epigenetic changes to the MDR1 locus in response to chemotherapeutic drugs. *Oncogene*, **2005**, *24*, 8061-8075.

[92] Ruiz, M.L.; Rigalli, J.P.; Arias, A.; Villanueva, S.; Banchio, C.; Vore, M.; Mottino, A.D.; Catania, V.A. Induction of hepatic multidrug resistance-associated protein 3 by ethynylestradiol is independent of cholestasis and mediated by estrogen receptor. *Drug Metab. Dispos.,* **2013**, *41*(2), 275-280.

[93] Ruiz, M.L.; Rigalli, J.P.; Arias, A.; Villanueva, S.S.; Banchio, C.; Vore, M.; Mottino, A.D.; Catania, V.A. Estrogen receptor-α mediates human multidrug resistance associated protein 3 induction by 17α-ethynylestradiol. Role of activator protein-1. *Biochem. Pharmacol.,* **2013**, *86*(3), 401-409.

[94] To, K.K.; Robey, R.; Zhan, Z.; Bangiolo, L.; Bates, S.E. Upregulation of ABCG2 by romidepsin via the aryl hydrocarbon receptor pathway. *Mol. Cancer Res.*, **2011**, *9*, 516-527.

[95] To, K.K.; Zhan, Z.; Bates, S.E. Aberrant promoter methylation of the ABCG2 gene in renal carcinoma. *Mol. Cell Biol*., **2006**, *26*, 8572-8585.

[96] Turner, J.G.; Gump, J.L.; Zhang, C.; Cook, J.M.; Marchion, D.; Hazlehurst, L.; Munster, P.; Schell, M.J.; Dalton, W.S.; Sullivan, D.M. ABCG2 expression, function, and promoter methylation in human multiple myeloma. *Blood*, **2006**, *108*, 3881-3889.

[97] Burk, O.; Arnold, K.A.; Geick, A.; Tegude, H.; Eichelbaum, M. A role for constitutive androstane receptor in the regulation of human intestinal MDR1 expression. *Biol. Chem.*, **2005**, *386*, 503-513.

[98] Cerveny, L.; Svecova, L.; Anzenbacherova, E.; Vrzal, R.; Staud, F.; Dvorak, Z.; Ulrichova, J.; Anzenbacher, P.; Pavek, P. Valproic acid induces CYP3A4 and MDR1 gene expression by activation of constitutive androstane receptor and pregnane X receptor pathways. *Drug Metab. Dispos.,* **2007***, 35*, 1032-1041.

[99] Arias, A.; Rigalli, J.P.; Villanueva, S.S.; Ruiz, M.L.; Luquita, M.G.; Perdomo, V.G.; Vore, M.; Catania, V.A.; Mottino, A.D. Regulation of expression and activity of multidrug resistance proteins MRP2 and MDR1 by estrogenic compounds in Caco-2 cells. Role in prevention of xenobiotic-induced cytotoxicity. *Toxicology*, **2014**, *320*, 46-55.

[100] Ee, P.L.; Kamalakaran, S.; Tonetti, D.; He, X.; Ross, D.D.; Beck, W.T. Identification of a novel estrogen response element in the breast cancer resistance protein (ABCG2) gene. *Cancer Res.,* **2004**, *64*, 1247-1251.

[101] Yasuda, S.; Itagaki, S.; Hirano, T.; Iseki, K. Expression level of ABCG2 in the placenta decreases from the mid stage to the end of gestation. *Biosci. Biotechnol. Biochem.*, **2005**, *69*, 1871-1876.

[102] Li, W.; Jia, M.; Qin, X.; Hu, J.; Zhang, X.; Zhou, G. Harmful effect of ERbeta on BCRP-mediated drug resistance and cell proliferation in ERalpha/PR-negative breast cancer. *FEBS J*., **2013**, *280*, 6128-6140

[103] Krishnamurthy, P.; Ross, D.D.; Nakanishi, T.; Bailey-Dell, K.; Zhou, S.; Mercer, K.E.; Sarkadi B., Sorrentino, B.P.; Schuetz, J.D. The stem cell marker Bcrp/ABCG2 enhances hypoxic cell survival through interactions with heme. *J. Biol. Chem*., **2004**, *279*, 24218-24225.

[104] Vollrath, V.; Wielandt, A.M.; Iruretagoyena, M.; Chianale, J. Role of Nrf2 in the regulation of the Mrp2 (ABCC2) gene. *Biochem. J.,* **2006**, *395*(3), 599-609.

[105] Ji, L.; Li, H.; Gao, P.; Shang, G.; Zhang, D.D.; Zhang, N.; Jiang, T. Nrf2 pathway regulates multidrug-resistance-associated protein 1 in small cell lung cancer. *PLoS One*, **2013**, *8*(5), e63404.

[106] Ghanem, C.I.; Rudraiah, S.; Bataille, A.M.; Vigo, M.B.; Goedken, M.J.; Manautou, J.E. Role of nuclear factor-erythroid 2-related factor 2 (Nrf2) in the transcriptional regulation of brain ABC transporters during acute acetaminophen (APAP) intoxication in mice. *Biochem. Pharmacol*., **2015**, *94*(3), 203-211.

[107] Natarajan, K.; Xie, Y.; Baer, M.R.; Ross, D.D. Role of breast cancer resistance protein (BCRP/ABCG2) in cancer drug resistance. *Biochem. Pharmacol*., **2012**, *83*, 1084-1103.

[108] Hirai, T.; Fukui, Y.; Motojima, K. PPARalpha agonists positively and negatively regulate the expression of several nutrient/drug transporters in mouse small intestine. *Biol. Pharm. Bull.,* **2007**, *30*(11), 2185-2190.

[109] Kauffmann, H.M.; Pfannschmidt, S.; Zöller, H,; Benz, A.; Vorderstemann, B.; Webster, J.I.; Schrenk, D. Influence of redox-active compounds and PXR-activators on human MRP1 and MRP2 gene expression. *Toxicology,* **2002**, *171*(2-3), 137-146.

[110] Rigalli, J.P.; Perdomo, V.G.; Luquita, M.G.; Villanueva, S.S.; Arias, A.; Theile, D.; Weiss, J.; Mottino, A.D.; Ruiz, M.L.; Catania, V.A. Regulation of biotransformation systems and ABC transporters by benznidazole in HepG2 cells: involvement of pregnane X-receptor. *PLoS Negl. Trop. Dis.,* **2012**, *6*(12), e1951.

[111] Rigalli, J.P.; Ruiz, M.L.; Perdomo, V.G.; Villanueva, S.S.; Mottino, A.D.; Catania V.A. Pregnane X receptor mediates the induction of P-glycoprotein by spironolactone in HepG2 cells. *Toxicology,* **2011***, 285*, 18-24.

[112] Wang, H.; Lee, E.W.; Zhou, L.; Leung, P.C.; Ross, D.D.; Unadkat, J.D.; Mao, Q. Progesterone receptor (PR) isoforms PRA and PRB differentially regulate expression of the breast cancer resistance protein in human placental choriocarcinoma BeWo cells. *Mol. Pharmacol.*, **2008**, *73*, 845-854.

[113] Lee, C.H.; Bradley, G.; Ling, V. Increased P-glycoprotein messenger RNA stability in rat liver tumors *in vivo*. *J. Cell Physiol.,* **1998**, *177*(1), 1-12.

[114] Yague, E.; Armesilla, A.L.; Harrison, G.; Elliott, J.; Sardini, A.; Higgins, C.F.; Raguz, S. P-glycoprotein (MDR1) expression in leukemic cells is regulated at two distinct steps, mRNA stabilization and translational initiation. *J. Biol. Chem.,* **2003**, *278*(12), 10344-10352.

[115] Ménez, C.; Mselli-Lakhal, L.; Foucaud-Vignault, M.; Balaguer, P.; Alvinerie, M.; Lespine, A. Ivermectin induces P-glycoprotein expression and function through mRNA stabilization in murine hepatocyte cell line. *Biochem. Pharmacol.,* **2012**, *83*(2), 269-278.

[116] To, K.K.; Zhan, Z.; Litman, T.; Bates, S.E. Regulation of ABCG2 expression at the 3′ untranslated region of its mRNA through modulation of transcript stability and protein translation by a putative microRNA in the S1 colon cancer cell line. *Mol. Cell Biol.*, **2008**, *28*, 5147-5161.

[117] Pan, Y.Z.; Morris, M.E.; Yu, A.M. MicroRNA-328 negatively regulates the expression of breast cancer resistance protein (BCRP/ABCG2) in human cancer cells. *Mol. Pharmacol*., **2009**, *75*, 1374-1379.

[118] To, K.K.; Robey, R.W.; Knutsen, T.; Zhan, Z.; Ried, T.; Bates, S.E. Escape from hsa-miR-519c enables drug-resistant cells to maintain high expression of ABCG2. *Mol. Cancer Ther.,* **2009**, *8*, 2959-2968.

[119] Liang, Z.; Wu, H.; Xia, J.; Li, Y.; Zhang, Y.; Huang, K.; Wagar, N.; Yoon, Y.; Cho, H.T.; Scala,S.; Shim, H. Involvement of miR-326 in chemotherapy resistance of breast cancer through modulating expression of multidrug resistance-associated protein 1. *Biochem. Pharmacol*., **2010**, *79*(6), 817-824.

[120] Wang, F.; Xue, X.; Wei, J.; An, Y.; Yao, J.; Cai, H.; Wu, J.; Dai, C.; Qian, Z.; Xu, Z.; Miao, Y. hsa-miR-520h downregulates ABCG2 in pancreatic cancer cells to inhibit migration, invasion, and side populations. *Br. J. Cancer,* **2010**, *103*, 567-574.

[121] Haenisch, S.; Laechelt, S.; Bruckmueller, H.; Werk, A.; Noack, A.; Bruhn, O.; Remmler, C.; Cascorbi, I. Down-regulation of ATP-binding cassette C2 protein expression in HepG2 cells after rifampicin treatment is mediated by microRNA-379. *Mol. Pharmacol.,* **2011**, *80*(2), 314-320.

[122] Molina-Pinelo, S.; Gutiérrez, G.; Pastor, M.D.; Hergueta, M.; Moreno-Bueno, G.; García-Carbonero, R.; Nogal, A.; Suárez, R.; Salinas, A.; Pozo-Rodríguez, F.; Lopez-Rios, F.; Agulló-Ortuño, M.T.; Ferrer, I.; Perpiñá, A.; Palacios, J.; Carnero, A.; Paz-Ares, L. MicroRNA-dependent regulation of transcription in non-small cell lung cancer. *PLoS One*, **2014**, 9(3), e90524.

[123] Markova, S.M.; Kroetz, D.L. ABCC4 is regulated by microRNA-124a and microRNA-506. *Biochem. Pharmacol*., **2014**, *87*(3), 515-522.

[124] Wu, Q.; Yang, Z.; Xia, L.; Nie, Y.; Wu, K.; Shi, Y.; Fan, D. Methylation of miR-129-5p CpG island modulates multidrug resistance in gastric cancer by targeting ABC transporters. *Oncotarget,* **2014**, *5*(22), 11552-11563.

[125] Zhang, Y.; Li, W.; Vore, M. Translational regulation of rat multidrug resistance-associated protein 2 expression is mediated by upstream open reading frames in the 5' untranslated region. *Mol. Pharmacol*., **2007**, *71*(1), 377-383.

[126] Hayashi, H.; Mizuno, T.; Horikawa, R.; Nagasaka, H.; Yabuki, T.; Takikawa, H.; Sugiyama, Y. 4-Phenylbutyrate modulates ubiquitination of hepatocanalicular MRP2 and reduces serum total bilirubin concentration. *J. Hepatol.,* **2012**, *56*(5), 1136-1144.

[127] Sereš, M.; Cholujová, D.; Bubenčíkova, T.; Breier, A.; Sulová, Z. Tunicamycin depresses P-glycoprotein glycosylation without an effect on its membrane localization and drug efflux activity in L1210 cells. *Int. J. Mol. Sci*., **2011**, *12*(11), 7772-7784.

[128] Wojtowicz, K.; Januchowski, R.; Nowicki, M.; Zabel, M. Inhibition of protein glycosylation reverses the MDR phenotype of cancer cell lines. *Biomed. Pharmacother.,* **2015**, *74*, 49-56.

[129] Roma, M.G.; Crocenzi, F.A.; Mottino, A.D. Dynamic localization of hepatocellular transporters in health and disease. *World J. Gastroenterol.,* **2008**, *14*(44), 6786-6801.

[130] Song, T.; Barua, K.; Buseman, G.; Murphy, P.A. Soy isoflavone analysis: quality control and a new internal standard. *Am. J. Clin. Nutr.,* **1998**, *68*(6),1474S-1479S.

[131] Barnes, S. The biochemistry, chemistry and physiology of the isoflavones in soybeans and their food products. *Lymphat. Res. Biol.,* **2010**, *8*(1), 89-98.

[132] Walle, T.; Browning, A.M.; Steed, L.L.; Reed, S.G.; Walle, U.K. Flavonoid glucosides are hydrolyzed and thus activated in the oral cavity in humans. *J. Nutr.,* **2005**, *135*(1), 48-52.

[133] Day, A.J.; DuPont ,M.S.; Ridley, S.; Rhodes, M.; Rhodes, M.J.; Morgan, M.R.; Williamson, G. Deglycosylation of flavonoid and isoflavonoid glycosides by human small intestine and liver beta-glucosidase activity. *FEBS Lett*., **1998**, *436*(1), 71-75.

[134] Izumi, T.; Piskula, M.K.; Osawa, S.; Obata, A.; Tobe, K.; Saito, M.; Kataoka, S.; Kubota, Y.; Kikuchi, M. Soy isoflavone aglycones are absorbed faster and in higher amounts than their glucosides in humans. *J. Nutr.,* **2000**, *130*(7), 1695- 1699.

[135] Morito, K.; Hirose,T.; Kinjo, J.; Hirakawa, T.; Okawa, M.; Nohara, T.; Ogawa, S.; Inoue, S.; Muramatsu, M.; Masamune, Y. Interaction of phytoestrogens with estrogen receptors alpha and beta. *Biol. Pharm. Bull.,* **2001**, *24*(4), 351-356.

[136] Akiyama, T.; Ishida, J.; Nakagawa, S.; Ogawara, H.; Watanabe, S.; Itoh, N.; Shibuya, M.; Fukami, Y. Genistein, a specific inhibitor of tyrosine-specific protein kinases. *J. Biol. Chem.,* **1987**, *262*(12), 5592-5595.

[137] Bircsak, K.M.; Aleksunes, L.M. Interaction of Isoflavones with the BCRP/ABCG2 Drug Transporter. *Curr. Drug Metab.,* **2015**, *16*(2), 124-140.

[138] Imai, Y.; Tsukahara, S.; Asada, S.; Sugimoto, Y. Phytoestrogens/flavonoids reverse breast cancer resistance protein/ABCG2-mediated multidrug resistance. *Cancer Res.,* **2004**, *64*(12), 4346-4352.

[139] Enokizono, J.; Kusuhara, H.; Sugiyama, Y. Effect of breast cancer resistance protein (Bcrp/Abcg2) on the disposition of phytoestrogens. *Mol. Pharmacol.*, **2007**, *72*(4), 967-975.

[140] van de Wetering, K.; Sapthu, S. ABCG2 functions as a general phytoestrogen sulfate transporter *in vivo*. *FASEB J.*, **2012**, *26*(10), 4014-4024.

[141] Walle, U.K.; French, K.L.; Walgren, R.A.; Walle, T. Transport of genistein-7-glucoside by human intestinal CACO-2 cells: potential role for MRP2. *Res. Commun. Mol. Pathol. Pharmacol.,* **1999**, *103*(1), 45-56.

[142] Zhang, X.; Dong, D.; Wang, H.; Ma, Z.; Wang, Y.; Wu, B. Stable knock-down of efflux transporters leads to reduced glucuronidation in UGT1A1-overexpressing HeLa cells: the evidence for glucuronidation-transport interplay. *Mol. Pharm*., **2015**, *12*(4), 1268-1278.

[143] Krumpochova, P.; Sapthu, S.; Brouwers, J.F.; de Haas, M.; de Vos, R.; Borst, P.; van de Wetering, K. Transportomics: screening for substrates of ABC transporters in body fluids using vesicular transport assays. *FASEB J.*, **2012**, *26*(2), 738-747.

[144] Jäger, W.; Winter, O.; Halper, B.; Salamon, A.; Sartori, M.; Gajdzik, L.; Hamilton, G.; Theyer, G.; Graf, J.; Thalhammer, T. Modulation of liver canalicular transport processes by the tyrosine-kinase inhibitor genistein: implications of genistein metabolism in the rat. *Hepatology*, **1997**, *26*(6), 1467-1476.

[145] Kato, K.; Kusuhara, H.; Kumagai, Y.; Ieiri, I.; Mori, H.; Ito, S.; Nakai, Y.; Maeda, K.; Sugiyama, Y. Association of multidrug resistance-associated protein 2 single nucleotide polymorphism rs12762549 with the basal plasma levels of phase II metabolites of isoflavonoids in healthy Japanese individuals. *Pharmacogenet. Genomics*, **2012**, *22*(5), 344-354.

[146] Yang, Z.; Zhu, W.; Gao, S.; Yin, T.; Jiang, W.; Hu, M. Breast cancer resistance protein (ABCG2) determines distribution of genistein phase II metabolites: reevaluation of the roles of ABCG2 in the disposition of genistein. *Drug Metab. Dispos.*, **2012**, *40*(10), 1883-1893.

[147] Catania, V.A.; Sánchez Pozzi, E.J.; Luquita, M.G.; Ruiz, M.L.; Villanueva, S.S.; Jones, B.; Mottino, A.D. Coregulation of expression of phase II metabolizing enzymes and multidrug resistance-associated protein 2. *Ann. Hepatol.,* **2004**, *3*(1), 11-7.

[148] Sirtori, C.R.; Arnoldi, A.; Johnson, S.K. Phytoestrogens: end of a tale? *Ann. Med*., **2005**, *37*(6), 423-438.

[149] Davis, J.N.; Singh, B.; Bhuiyan, M.; Sarkar, F.H. Genistein-induced upregulation of p21WAF1, downregulation of cyclin B, and induction of apoptosis in prostate cancer cells. *Nutr. Cancer*, **1998**, *32*(3), 123-131.

[150] Zander, S.A.; Kersbergen, A.; Sol, W.; Gonggrijp, M.; van de Wetering, K.; Jonkers, J.; Borst, P.; Rottenberg, S. Lack of ABCG2 shortens latency of BRCA1-deficient mammary tumors and this is not affected by genistein or resveratrol. *Cancer Prev. Res.,* **2012**, *5*(8), 1053-1060.

[151] Murkies, A.L.; Wilcox, G.; Davis, S.R. Clinical review 92: Phytoestrogens. *J. Clin. Endocrinol. Metab.,* **1998**, *83*(2), 297-303.

[152] Cao, Y.; Calafat, A.M.; Doerge, D.R.; Umbach, D.M.; Bernbaum, J.C.; Twaddle, N.C.; Ye, X.; Rogan, W.J. Isoflavones in urine, saliva, and blood of infants: data from a pilot study on the estrogenic activity of soy formula. *J. Expo. Sci. Environ. Epidemiol.,* **2009**, *19*(2), 223-234.

[153] Salleh, N.; Helmy, M.M.; Fadila, K.N.; Yeong, S.O. Isoflavone genistein induces fluid secretion and morphological changes in the uteri of post-pubertal rats. *Int. J. Med. Sci.,* **2013**, *10*(6), 665-675.

[154] Donato, M.T.; Lahoz, A.; Castell, J.V.; Gómez-Lechón, M.J. Cell lines: a tool for *in vitro* drug metabolism studies. *Curr. Drug Metab.,* **2008**, *9*(1), 1-11.

[155] Ganai, A.A.; Farooqi, H. Bioactivity of genistein: A review of *in vitro* and *in vivo* studies. *Biomed. Pharmacother*., **2015**, *76*, 30-38.

[156] Fang, Y.C.; Chen, B.H.; Huang, R.F.; Lu, Y.F. Effect of genistein supplementation on tissue genistein and lipid peroxidation of serum, liver and low-density lipoprotein in hamsters. *J. Nutr. Biochem.,* **2004**, *15*(3), 142-148.

[157] González-Granillo, M.; Steffensen, K.R.; Granados, O.; Torres, N.; Korach-André, M.; Ortíz, V.; Aguilar-Salinas, C.; Jakobsson, T.; Díaz-Villaseñor, A.; Loza-Valdes, A.; Hernandez-Pando, R.; Gustafsson, J.A.; Tovar, A.R. Soy protein isoflavones differentially regulate liver X receptor isoforms to modulate lipid metabolism and cholesterol transport in the liver and intestine in mice. *Diabetologia*, **2012**, *55*(9), 2469-2478.

[158] Li, Y.; Luh, C.J.; Burns, K.A.; Arao, Y.; Jiang, Z.; Teng, C.T.; Tice, R.R.; Korach, K.S. Endocrine-disrupting chemicals (EDCs): *in vitro* mechanisms of estrogenic activation and differential effects on ER target genes. *Environ. Health Perspect.,* **2013**, *121*(4), 459-466.

[159] Li, Y.; Ross-Viola, J.S.; Shay, N.F.; Moore, D.D., Ricketts, M.L. Human CYP3A4 and murine Cyp3A11 are regulated by equol and genistein via the pregnane X receptor in a species-specific manner. *J. Nutr.*, **2009**, *139*(5), 898-904.

[160] Froyen, E.B.; Steinberg, F.M. Soy isoflavones increase quinone reductase in hepa-1c1c7 cells *via* estrogen receptor beta and nuclear factor erythroid 2-related factor 2 binding to the antioxidant response element. *J. Nutr. Biochem.,* **2011**, *22*(9), 843-848.

[161] Yan, G.R.; Xiao, C.L.; He, G.W.; Yin, X.F.; Chen, N.P.; Cao, Y.; He, Q.Y. Global phosphoproteomic effects of natural tyrosine kinase inhibitor, genistein, on signaling pathways. *Proteomics*, **2010**, *10*(5), 976-986.

[162] Zhang, S.; Qin, C.; Safe, S.H. Flavonoids as aryl hydrocarbon receptor agonists/antagonists: effects of structure and cell context. *Environ. Health Perspect*., **2003**, *111*(16), 1877-1882.

[163] Castro, A.F.; Altenberg, G.A. Inhibition of drug transport by genistein in multidrug-resistant cells expressing P-glycoprotein. *Biochem. Pharmacol.,* **1997**, *53*(1), 89-93.

[164] Versantvoort, C.H.; Broxterman, H.J.; Lankelma. J.; Feller. N.; Pinedo, H.M. Competitive inhibition by genistein and ATP dependence of daunorubicin transport in intact MRP overexpressing human small cell lung cancer cells. *Biochem. Pharmacol*., **1994**, *48*(6), 1129-1136.

[165] Germann, U.A.; Chambers, T.C.; Ambudkar, S.V.; Pastan, I.; Gottesman, M.M. Effects of phosphorylation of P-glycoprotein on multidrug resistance. *J. Bioenerg. Biomembr.,* **1995**, *27*(1), 53-61.

[166] Okura, T.; Ibe, M.; Umegaki, K.; Shinozuka, K.; Yamada, S. Effects of dietary ingredients on function and expression of P-glycoprotein in human intestinal epithelial cells. *Biol. Pharm. Bull.*, **2010**, *33*(2), 255-259.

[167] Rigalli, J.P.; Ciriaci, N.; Arias, A.; Ceballos, M.P.; Villanueva, S.S.; Luquita, M.G.; Mottino, A.D.; Ghanem, C.I.; Catania, V.A.; Ruiz, M.L. Regulation of multidrug resistance proteins by genistein in a hepatocarcinoma cell line: impact on sorafenib cytotoxicity. *PLoS One*, **2015**, *10*(3), e0119502.

[168] Bogacz, A.; Mikołajczak, P.L.; Mikołajczak, P.L.; Rakowska-Mrozikiewicz, B.; Grześkowiak, E.; Wolski, H.; Czerny, B.; Mrozikiewicz, P.M. The influence of soybean extract on the expression level of selected drug transporters,

transcription factors and cytochrome P450 genes encoding phase I drug-metabolizing enzymes. *Ginekol. Pol*., **2014**, *85*(5), 348-353.

[169] Kim, J.H.; Chen, C.; Tony Kong, A.N. Resveratrol inhibits genistein-induced multi-drug resistance protein 2 (MRP2) expression in HepG2 cells. *Arch. Biochem. Biophys*., **2011**, *512*(2), 160-166.

[170] Hanet, N.; Lancon, A.; Delmas, D.; Jannin, B.; Chagnon, M.C.; Cherkaoui-Malki, M.; Latruffe, N.; Artur, Y.; Heydel J.M. Effects of endocrine disruptors on genes associated with 17beta-estradiol metabolism and excretion. *Steroids*, **2008**, *73*(12), 1242-1251.

[171] Versantvoort, C.H.; Schuurhuis, G.J.; Pinedo, H.M.; Eekman, C.A.; Kuiper, C.M.; Lankelma, J.; Broxterman, H.J. Genistein modulates the decreased drug accumulation in non-P-glycoprotein mediated multidrug resistant tumour cells. *Br. J. Cancer,* **1993**, *68*(5), 939-946.

[172] Hooijberg, J.H.; Broxterman, H.J.; Heijn, M.; Fles, D.L.; Lankelma, J.; Pinedo, H.M. Modulation by (iso)flavonoids of the ATPase activity of the multidrug resistance protein. *FEBS Lett*., **1997**, *413*(2), 344-348.

[173] Dallas, S.; Zhu, X.; Baruchel, S.; Schlichter, L.; Bendayan, R. Functional expression of the multidrug resistance protein 1 in microglia. *J. Pharmacol. Exp. Ther*., **2003**, *307*(1), 282-290.

[174] Nguyen, H.; Zhang, S.; Morris, M.E. Effect of flavonoids on MRP1-mediated transport in Panc-1 cells. *J. Pharm. Sci.,* **2003**, *92*(2), 250-257.

[175] Lania-Pietrzak, B.; Michalak, K.; Hendrich, A.B.; Mosiadz, D.; Grynkiewicz, G.; Motohashi, N.; Shirataki, Y. Modulation of MRP1 protein transport by plant, and synthetically modified flavonoids. *Life Sci*., **2005**, *77*(15), 1879-1891.

[176] Dallas, S.; Schlichter, L.; Bendayan, R. Multidrug resistance protein (MRP) 4- and MRP 5-mediated efflux of 9-(2-phosphonylmethoxyethyl)adenine by microglia. *J. Pharmacol. Exp. Ther.,* **2004**, *309*(3), 1221-1229.

[177] Ebert, B.; Seidel, A.; Lampen, A. Phytochemicals induce breast cancer resistance protein in Caco-2 cells and enhance the transport of benzo[a]pyrene-3-sulfate. *Toxicol. Sci*., **2007**, *96*(2), 227-236.

[178] Huang, W.; Wan, C.; Luo, Q.; Huang, Z.; Luo, Q. Genistein-inhibited cancer stem cell-like properties and reduced chemoresistance of gastric cancer. *Int. J. Mol. Sci*., **2014**, *15*, 3432-3443.

[179] Schexnayder, C.; Stratford, R.E. Genistein and Glyceollin Effects on ABCC2 (MRP2) and ABCG2 (BCRP) in Caco-2 Cells. *Int. J. Environ. Res. Public Health*, **2015**, *13*(1), pii: E17.

[180] Zhang, S.; Yang, X.; Morris, M.E. Combined effects of multiple flavonoids on breast cancer resistance protein (ABCG2)-mediated transport. *Pharm. Res*., **2004**, *21*(7), 1263-1273.

[181] Sparreboom, A.; van Asperen, J.; Mayer, U.; Schinkel, A.H.; Smit, J.W.; Meijer, D.K.; Borst, P.; Nooijen, W.J.; Beijnen, J.H.; van Tellingen, O. Limited oral bioavailability and active epithelial excretion of paclitaxel (Taxol) caused by P-glycoprotein in the intestine. *Proc. Natl. Acad. Sci. USA*., **1997**, *94*(5), 2031-2035.

[182] Huisman, M.T.; Chhatta, A.A.; van Tellingen, O.; Beijnen, J.H.; Schinkel, A.H. MRP2 (ABCC2) transports taxanes and confers paclitaxel resistance and both processes are stimulated by probenecid. *Int. J. Cancer*, **2005**, *116*(5), 824-829.

[183] Doyle, L.; Ross, D.D. Multidrug resistance mediated by the breast cáncer resistance protein BCRP (ABCG2). *Oncogene*, **2003**, *22*(47), 7340-7358.

[184] Sonnichsen, D.S.; Liu, Q.; Schuetz, *E.G.*; Schuetz, J.D.; Pappo, A.; Relling, M.V. Variability in human cytochrome P450 paclitaxel metabolism. J. *Pharmacol. Exp. Ther*., **1995**, *275*(2), 566-575.

[185] Li, X.; Choi, J.S. Effect of genistein on the pharmacokinetics of paclitaxel administered orally or intravenously in rats. *Int. J. Pharm*., **2007**, *337*(1-2), 188-193.

[186] Yokooji, T.; Kawabe, Y.; Mori, N.; Murakami, T. Effect of genistein, a natural soy isoflavone, on the pharmacokinetics and intestinal toxicity of irinotecan hydrochloride in rats. *J. Pharm. Pharmacol*., **2013**, *65*(2), 280-291.

[187] Pulido, M.M.; Molina, A.J.; Merino, G.; Mendoza, G.; Prieto, J.G.; Alvarez, A.I. Interaction of enrofloxacin with breast cancer resistance protein (BCRP/ABCG2): influence of flavonoids and role in milk secretion in sheep. *J. Vet. Pharmacol. Ther.*, **2006**, *29*(4), 279-287.

[188] Haslam, I.S.; Wright, J.A.; O'Reilly, D.A.; Sherlock, D.J.; Coleman, T.; Simmons, N.L. Intestinal ciprofloxacin efflux: the role of breast cancer resistance protein (ABCG2). *Drug Metab. Dispos.*, **2011**, *39*(12), 2321-2328.

[189] Alvarez, A.I.; Merino, G.; Molina, A.J.; Pulido, M.M.; McKellar, Q.A.; Prieto, J.G. Role of ABC transporters in veterinary drug research and parasite resistance. *Curr. Drug Deliv.*, **2006**, *3*(2), 199-206.

[190] Gerk, P.M.; Oo, C.Y.; Paxton, E.W.; Moscow, J.A.; McNamara, P.J. Interactions between cimetidine, nitrofurantoin, and probenecid active transport into rat milk. *J. Pharmacol. Exp. Ther.,* **2001**, *296*(1), 175-180.

[191] Merino, G.; Jonker, J.W.; Wagenaar, E.; van Herwaarden, A.E.; Schinkel, A.H. The breast cancer resistance protein (BCRP/ABCG2) affects pharmacokinetics, hepatobiliary excretion, and milk secretion of the antibiotic Nitrofurantoin. *Mol. Pharmacol*., **2005**, *67*(5), 1758-1764.

[192] Wang, L.; Leggas, M.; Goswami, M.; Empey, P.E.; McNamara, P.J. N- (4-[2-(1, 2, 3, 4-tetrahydro-6, 7-dimethoxy-2-isoquinolinyl)ethyl]- phenyl)-9, 10-dihydro-5-methoxy-9-oxo-4-acridine carboxamide (GF120918) as a chemical ATP-binding cassette transporter family G member 2 (Abcg2) knockout model to study nitrofurantoin transfer into milk. *Drug Metab. Dispos.,* **2008**, *36*(12), 2591-2596.

[193] Pérez, M.; Real, R.; Mendoza, G.; Merino, G.; Prieto, J.G.; Alvarez, A.I. Milk secretion of nitrofurantoin, as a specific BCRP/ABCG2 substrate, in assaf sheep: modulation by isoflavones. *J. Vet. Pharmacol. Ther*., **2009**, *32*(5), 498-502.

[194] Merino, G.; Perez, M.; Real, R.; Egido, E.; Prieto, J.G.; Alvarez, A.I. *In vivo* inhibition of BCRP/ABCG2 mediated transport of nitrofurantoin by the isoflavones genistein and daidzein: a comparative study in Bcrp1 (-/-) mice. *Pharm. Res*., **2010**, 27(10), 2098-2105.

[195] Poutrel, B.; Stegemann, M.R.; Roy, O.; Pothier, F.; Tilt, N.; Payne-Johnson, M. Evaluation of the efficacy of systemic danofloxacin in the treatment of induced acute Escherichia coli bovine mastitis. *J. Dairy Res.*, **2008**, *75*(3), 310-318.

[196] Shem-Tov, M.; Ziv, G.; Glickman, A.; Saran, A. Pharmacokinetics and penetration of danofloxacin from the blood into the milk of ewes. *Vet. Res*., **1997**, 28(6), 571-579.

[197] Escudero, E.; Carceles, C.M.; Fernandez-Varon, E.; Marin, P.; Benchaoui, H. Pharmacokinetics of danofloxacin 18% in lactating sheep and goats. *J. Vet. Pharmacol. Ther.*, **2007**, *30*(6), 572-577.

[198] Pérez, M.; Otero, J.A.; Barrera, B.; Prieto, J.G.; Merino, G.; Alvarez, A.I. Inhibition of ABCG2/BCRP transporter by soy isoflavones genistein and daidzein: effect on plasma and milk levels of danofloxacin in sheep. *Vet. J*., **2013**, 196(2), 203-208.

[199] Xiao, C.Q.; Chen, R.; Lin, J.; Wang, G.; Chen, Y.; Tan, Z.R.; Zhou, H.H. Effect of genistein on the activities of cytochrome P450 3A and P-glycoprotein in Chinese healthy participants. *Xenobiotica*, **2012**, 42(2), 173-178.

[200] Frazer, R.; Irvine, A.E.; McMullin, M.F. Chronic myeloid leukaemia in the 21st century. *Ulster Med. J*., **2007**, *76*(1), 8-17.

[201] Wang, Z.; Wang, L.; Xia, M.M.; Sun, W.; Huang, C.K.; Cui, X.; Hu, G.X.; Lian, Q.Q.; Wang, Z.S. Pharmacokinetics interaction between imatinib and genistein in rats. *Biomed. Res. Int.*, **2015**, *2015*, 368976.

[202] Scott, L.M.; Durant, P.; Leone-Kabler, S.; Wood, C.E.; Register, T.C.; Townsend, A.; Cline, J.M. Effects of prior oral contraceptive use and soy isoflavonoids on estrogen-metabolizing cytochrome P450 enzymes. *J. Steroid Biochem. Mol. Biol.,* **2008**, *112*(4-5), 179-185.

[203] Dai, H.; Marbach, P.; Lemaire, M.; Hayes, M.; Elmquist, W.F. Distribution of STI-571 to the brain is limited by P-glycoprotein-mediated efflux. *J. Pharmacol. Exp. Ther*., **2003**, *304*(3), 1085-1092.

[204] Mahon, F.X.; Belloc, F.; Lagarde, V.; Chollet, C.; Moreau-Gaudry, F.; Reiffers, J.; Goldman, J.M.; Melo, J.V. MDR1 gene overexpression confers resistance to imatinib mesylate in leukemia cell line models. *Blood*, **2003**, *1*(6), 2368-2373.

[205] Thomas, J.; Wang, L.; Clark, R.E.; Pirmohamed, M. Active transport of imatinib into and out of cells: implications for drug resistance. *Blood*, **2004**, *104*(12), 3739-3745.

[206] Alves, R.; Fonseca, A.R.; Gonçalves, A.C.; Ferreira-Teixeira, M.; Lima, J.; Abrantes, A.M.; Alves, V.; Rodrigues-Santos, P.; Jorge, L.; Matoso, E.; Carreira, I.M.; Botelho, M.F. Sarmento-Ribeiro, A.B. Drug transporters play a key role in the complex process of Imatinib resistance *in vitro*. *Leuk. Res*., **2015**, *39*(3), 355-360.

Received: January 04, 2016 Revised: March 15, 2016 Accepted: March 28, 2016