Fruit Juices as Perpetrators of Drug Interactions: The Role of Organic Anion–Transporting Polypeptides

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Grapefruit juice is widely recognized to cause important drug interactions via inhibition of CYP3A4, and a wider variety of fruit juices have been shown to inhibit influx transporters in enterocytes known as organic anion–transporting polypeptides (OATPs). Fruit juice coadministration significantly reduces the oral bioavailability of numerous important medicines relying on this anion transporter pathway for absorption. This article reviews the current literature on interactions between clinically used OATP substrates and fruit juice consumption.

Since the initial finding two decades ago, made by chance, that the coadministration of grapefruit juice causes increased felodipine concentrations,[1] drug interactions involving fruit juices have become an increasingly important area of research for both new and established medicines. Although many studies on interactions have focused on the ability of grapefruit juice to significantly increase the systemic exposure of substrates of CYP3A4, a more recently discovered mechanism involving the inhibition of organic anion–transporting polypeptides (OATPs) by the constituents of a wider variety of fruit juices has been shown to result in significantly reduced systemic exposure for several clinically important medicines.

OATPs are membrane transport proteins that facilitate the sodium-independent influx of both endogenous and exogenous compounds.[2,3] OATPs are encoded by solute carrier organic anion–transporting (*SLCO*) genes and include 11 human transporters subdivided into six families, with different OATP subtypes expressed in the apical and basolateral membranes of polarized cells in the gastrointestinal wall, liver, heart, kidney, endothelia, and brain.[3,4] Clinically used OATP substrates include 3-hydroxy-3-methylglutaryl-Coenzyme A reductase inhibitors, angiotensin II receptor antagonists, several β-adrenergic blockers, thyroxine, benzylpenicillin, and the antihistamine fexofenadine.[2]

In the context of orally administered xenobiotics, intestinally expressed OATPs facilitate the uptake of substrate compounds through the gastrointestinal wall, acting in an opposing fashion to efflux transporters in the intestine such as P-glycoprotein (P-gp) (**Figure 1**). In the liver, several OATP subtypes are present in the sinusoidal membrane of hepatocytes, aiding drug uptake into the liver and working in tandem with efflux transporters to eliminate drugs from the body.

In 2002, Dresser *et al.* discovered a 65–75% reduction in the systemic exposure of the OATP substrate fexofenadine when coadministered with grapefruit, orange, or apple juice, as compared with administration with water.[5] In addition, *in vitro* results supported an inhibitory effect of these fruit juices on human OATP1A2 at 5% of normal juice strength.[5] Following these findings, more recent studies have confirmed the findings of Dresser *et al* with fexofenadine and examined the effect of grapefruit, orange, and apple juice on a significant number of other OATP2B1 and OATP1A2 substrates used therapeutically.[6–10]

This concise review aims to critically analyze all studies investigating a potential interaction between clinically used OATP substrates and fruit juices in humans as of June 2012. An overview of the intestinal expression of OATPs, proposed mechanism of fruit juice–drug interactions, and possible active components of fruit juices involved in OATP-mediated drug interactions is also provided. Using the PubMed online database (http://www.ncbi.nlm.nih.gov/pubmed), we searched for the following terms: "grapefruit juice," "orange juice," "apple juice," "OATP," "OATP1A2," "OATP2B1," "OATP1B1," "OATP1B3," "OATP-A," "OATP-B," "OATP-C," "OATP8," "citrus juice," and "fruit juice." In publications identified from this search, reference lists were hand-searched to identify other relevant studies.

OATP EXPRESSION AND INTERACTION MECHANISMS

Studies investigating the expression of OATPs have shown that *SLCO1A2* and *SLCO2B1* mRNA, encoding OATP1A2 and

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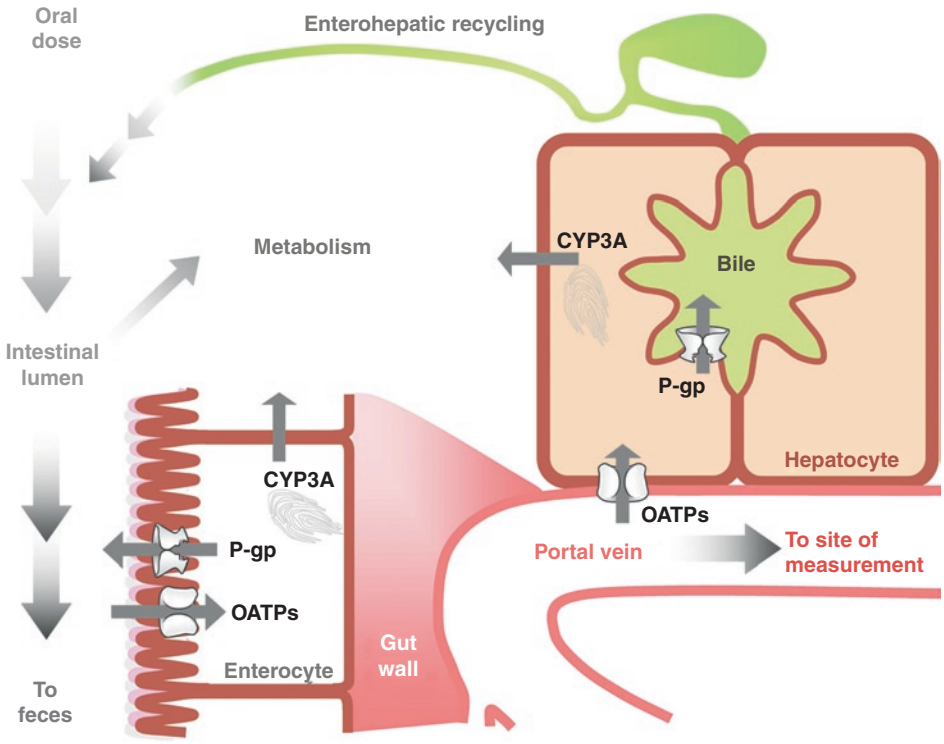

Figure 1 OATPs act in an opposing fashion to P-gp in intestinal enterocytes to increase and decrease absorption of drugs, respectively. In hepatocytes, these transporters work in tandem to eliminate drugs from the body. OATP, organic anion–transporting polypeptide; P-gp, P-glycoprotein. Reprinted from ref. 92.

OATP2B1, respectively, are expressed in the intestine.[6,11–18] In the liver, OATP1B1, OATP1B3, and OATP2B1 are expressed on the sinusoidal membrane of hepatocytes,[2] and OATP1A2 is expressed in cholangiocytes.[19]

The function of OATP1A2 in the intestine has been debated in the literature, as studies have identified relatively low *SLCO1A2* mRNA expression in the intestine;[11,14,15,17] other authors have identified *SLCO1A2* mRNA expression in both the small and large intestine.[6,16,18] It is important to note that *SLCO1A2* mRNA expression may not directly reflect the amount or functional activity of OATP1A2 protein in the intestine;[20] discordant expression of mRNAs and protein has been reported in the liver due to posttranscriptional regulation.[21]

Detection of OATP1A2 protein in the small intestine provides further evidence of OATP1A2 expression in enterocytes. Using immunohistochemical staining of duodenal tissue sections, OATP1A2 protein has been identified in the small intestine and was found to be colocalized with P-gp on the apical membrane of enterocytes.[6] Other investigators have also identified OATP1A2 protein expression in the small intestine.[22]

In regard to OATP2B1, studies have identified higher *SLCO2B1* mRNA concentrations in the intestine than have been observed for *SLCO1A2*.[11,15] Like OATP1A2, OATP2B1 protein is expressed on the apical membrane of enterocytes,[23] thereby facilitating its involvement in the uptake of substrates from the intestine.

Interactions between grapefruit juice and CYP3A4 substrates are known to occur via an irreversible mechanism leading to decreased CYP3A protein levels in enterocytes of the small intestine, without reducing *CYP3A4* mRNA levels.[24] This mechanism results in a prolonged effect of grapefruit juice on CYP3A4 substrates. The magnitude of interaction declines over several days for some medicines.[25]

By contrast, fruit juice interactions with intestinally expressed OATPs appear to be mediated via an inhibition of uptake transport rather than decreased protein expression. Using duodenal biopsies taken 1–2 h after consumption of either water or grapefruit juice, Glaeser *et al.* found that OATP1A2 protein and *SLCO1A2* mRNA levels were not significantly different following grapefruit juice as compared with water; however, exposure of the OATP1A2 substrate fexofenadine was reduced by half when coadministered with grapefruit juice as compared with water, implying an inhibition of transporter function.[6]

Importantly, this study found that the effect of fruit juice OATP inhibition appears to dissipate over a shorter period than has been observed with fruit juice inhibition of CYP3A4. A 52% reduction in fexofenadine area under the plasma concentration–time curve (AUC) was observed when grapefruit juice was ingested concomitantly with fexofenadine; a 38% reduction was observed when grapefruit juice was taken 2 h before the drug, with no effect on fexofenadine AUC observed when grapefruit juice was taken 4 h before drug administration.[6] These results suggest that OATP-mediated fruit juice interactions may potentially be avoided by separating medicine and juice consumption by at least 4 h, however, further studies are needed to confirm these findings with other fruit juices and medicines.

ACTIVE COMPONENTS OF FRUIT JUICES

A number of *in vitro* and clinical studies have evaluated components of fruit juices that may be involved in the inhibition of intestinally expressed OATPs. Grapefruit juice is known to contain several potentially pharmacologically active compounds, including flavonoids (naringin and hesperidin, among others) and furanocoumarins (bergamottin and 6′,7′-dihydroxybergamottin).[26,27] Flavonoids have also been identified in the juice of both sweet (*Citrus sinensis*) and sour (*Citrus aurantium*) oranges, although furanocoumarins such as 6′,7′-dihydroxybergamottin are found only in sour (Seville) oranges,[27–29] which are not usually consumed as juices owing to their poor taste. The primary flavonoids in grapefruit juice and orange juice are naringin and hesperidin, respectively (**Table 1**).[5,27,30] Naringin has been identified in some commercially available orange juice products,[27] although it is not present in pure sweet orange juice,[30] suggesting that these juices may not contain pure orange juice.

A study by Bailey *et al.* examined the *in vitro* effect of naringin and hesperidin on OATP1A2 activity, also investigating the impact of naringin and grapefruit juice furanocoumarins on the pharmacokinetics of the OATP1A2 substrate fexofenadine, in comparison to water and grapefruit juice in healthy volunteers.[7] Both naringin and hesperidin were found to significantly inhibit OATP1A2 transport activity *in vitro* at concentrations far below those found in grapefruit and orange juices (half maximal inhibitory concentration of 3.6 μmol/l and 2.7 μmol/l, respectively), although hesperidin produced only 60% maximal inhibition as compared with complete inhibition observed with naringin.[7] Naringin has also been shown to inhibit OATP2B1 at a concentration of 10 μmol/l;[31] the effect of hesperidin on OATP2B1 is yet to be investigated. Narirutin is present in both grapefruit and orange juices in significant concentrations;[27] however, its effect on OATP1A2 or OATP2B1 is not yet known.

Although certain flavonoids are also known to modulate the efflux transporter P-gp,[32] the half maximal inhibitory concentration of naringin for P-gp is 3000 μmol/l,[5] suggesting that fruit juices are far more potent inhibitors of intestinally expressed OATPs than P-gp.[5,6] Other efflux transporters such as ABCG2, also known as breast cancer–resistance protein, are known to be expressed in the intestine;[33] the aglycone forms of naringin and hesperidin—naringenin and hesperetin—have been shown to significantly inhibit ABCG2 activity.[34]

In *in vivo* studies with fexofenadine, coadministration of a naringin solution reduced fexofenadine exposure by ~25% as compared with administration with water, whereas grapefruit juice reduced exposure by 45%. The authors also tested a particulate fraction of grapefruit juice containing high concentrations of furanocoumarins but very low concentrations of naringin; the particulate fraction did not significantly reduce fexofenadine exposure as compared with coadministration with water.[7] Taken together, these studies suggest that naringin plays an important role in OATP-mediated drug interactions with grapefruit juice, although naringin alone reduced fexofenadine exposure to a lesser extent than grapefruit juice, suggesting that other compounds may also contribute to this interaction. Although less information is available for hesperidin, a recent study in rats identified a similar reduction in the systemic exposure of the OATP1A2 and OATP2B1 substrate celiprolol when administered with orange juice or an equivalent concentration of hesperidin,[35] supporting a role of this flavonoid in drug interactions with orange juices.

The compounds responsible for inhibition of intestinally expressed OATPs in apple (*Malus domestica*) juice have yet to be definitively elucidated. Apple juice is known to contain flavonoids, including quercetin and glycosides of quercetin, epicatechin, phlorizin, and kaempferol.[36–38] Phlorizin has been reported as the primary flavonoid in commercial apple juice, although epicatechin may be present at similar or higher concentrations in other apple juice varieties.[36] Phlorizin significantly inhibits the hepatic transporter OATP1B1,[39] but its effect on OATP1A2 and OATP2B1 have not been evaluated. Although *in vitro* studies have identified quercetin and kaempferol as inhibitors of OATP1A2 and OATP2B1 and epicatechin and glycosides of quercetin as inhibitors of OATP2B1,[40,41] an *in vivo* study with fexofenadine found that high doses of a quercetin preparation led to a 55% increase, rather than a decrease, in fexofenadine AUC, possibly as a result of inhibition of P-gp.[42] The results of this study suggest that quercetin may not play a role in intestinal OATP-mediated drug interactions with apple juice, although these results are also consistent with a potential inhibition of hepatic OATPs by quercetin, which would be expected to result in increased bioavailability. Undoubtedly, further studies are needed to determine possible active constituents in apple juice that may explain the significant drug interactions observed between certain medicines and apple juice products.[5,9]

Table 1 Concentrations of naringin and hesperidin in freshly hand-squeezed and commercially available grapefruit and orange juices

Component		Grapefruit juice (hand-squeezed)	Grapefruit juice (commercial)	Sweet orange juice (hand-squeezed)	Sweet orange juice (commercial)
Naringin (mg/100 ml)	*N*	19	102	ND	9
	Mean ± SD	23.0 ± 12.8	43.5 ± 23.3		2.1 ± 3.0
	Range	4.5–60.2	4.8–119.7		0.2–7.5
Hesperidin (mg/100 ml)	*N*	6	15	44	63
	Mean ± SD	0.9 ± 0.6	2.8 ± 3.9	28.6 ± 11.9	37.5 ± 19.2
	Range	0.3–1.8	0.2–16.4	3.5–55.2	4.5–76.3

N, number of juices tested; ND, not detected.

Adapted from ref. 27.

Differences in juice composition are a significant confounding factor in the assessment of drug interactions with fruit juices. Concentrations of flavonoids in fruit juices may vary based on fruit species, geographic origin, maturity, manufacturing processes, storage conditions, and seasonal variability;[26,43] mixed fruit juice products containing combinations of fruit juices add a further complication. It is likely that these factors contribute to the highly variable concentrations of flavonoids observed in fruit juices (**Table 1**). In addition, these factors contribute to the complexity in interpreting published reports of fruit juice–drug interactions and have implications for clinical recommendations regarding the concomitant administration of fruit juices with selected medicines.

REVIEW FINDINGS AND CLINICAL IMPLICATIONS

A total of 26 studies were identified involving the effect of fruit juice coadministration on clinically used OATP1A2 and OATP2B1 substrates. **Figure 2** summarizes the results of these studies, showing the AUC ratio (and 95% confidence intervals (CI)) of the interacting medicine when administered with fruit juice as compared with administration with water.

OATP substrates with significantly reduced exposure

A number of studies have investigated the interaction between the antihistamine fexofenadine and fruit juices, all identifying significant reductions in fexofenadine systemic exposure when grapefruit, orange, or apple juice is coadministered.[5–7,44–46]

The volume of juice coadministered with fexofenadine varied significantly between studies, with juice volume ingested within 2 h of fexofenadine administration varying between 300 and 900 ml and total juice volume administered varying from 300 to 1920 ml (**Figure 2**). The effect of grapefruit juice volume on fexofenadine pharmacokinetics was investigated directly by Dresser *et al.*[45] Following a single consumption of 300 ml grapefruit juice with fexofenadine, the AUC ratio for juice:water was 0.58 (95% CI 0.43–0.84); when subjects were administered 300 ml grapefruit juice with fexofenadine followed by 150 ml juice every half hour for up to 3 h (total volume of 1200 ml), the AUC ratio of juice:water was significantly lower at 0.36 (95% CI 0.28–0.47).[45] This study suggests that increased juice volume increases the extent of interaction with fexofenadine; however, a significant reduction in fexofenadine exposure remains even with a single consumption of 300 ml grapefruit juice. Other studies have reported similar reductions in fexofenadine exposure with a single glass of grapefruit juice.[6,7]

A more recent study with fexofenadine investigated the interplay of genetic variability in OATP transporters with fruit juice–drug interactions, examining the effect of a single nucleotide polymorphism (c.1457C>T) in the *SLCO2B1* gene, encoding OATP2B1.[46] Although fexofenadine is a substrate for both OATP1A2 and OATP2B1,[6,47] the authors identified genotype-dependent fexofenadine exposure in the absence of fruit juice, with subjects with the *SLCO2B1* c.1457C>T allele achieving significantly lower fexofenadine exposure as compared with subjects without this allele when fexofenadine was coadministered with water (AUC ratio 0.63, 95% CI 0.42–0.94).[46] However, a larger reduction in fexofenadine exposure was observed when coadministered with apple juice (AUC ratio 0.22, 95% CI 0.17–0.28). In addition, apple juice offset the genotype-dependent effect observed with the *SLCO2B1* c.1457C>T allele on fexofenadine pharmacokinetics, supporting an inhibitory effect of apple juice on OATP2B1.[46]

Interaction studies to date have not investigated the potential impact of fruit juice consumption on fexofenadine pharmacodynamics; however, the significant reductions in fexofenadine AUC observed in these studies suggest that fruit juice consumption may reduce the antihistamine activity of fexofenadine. In clinical practice, avoiding fruit juice consumption within 4 h of fexofenadine administration is recommended.

Significant fruit juice–drug interactions have been identified for several β-blockers, including atenolol, celiprolol, and talinolol,[8,10,48–51] all of which are substrates of OATP1A2;[52,53] celiprolol and talinolol are also OATP2B1 substrates.[50,53] The effect of inhibition of intestinally expressed OATPs by fruit juices on systemic exposure differs significantly between these medicines, suggesting that the contribution of intestinal OATP transport to overall intestinal absorption varies; the largest reduction in exposure was observed with celiprolol, with which grapefruit or orange juice coadministration reduced plasma concentrations by >80% on average as compared with water.[8,49,50] A recently published study with apple juice and atenolol found a similarly large reduction in systemic exposure, with 600 ml apple juice consumed over 1.5 h following drug administration causing a mean 58% reduction in atenolol exposure and 1200 ml apple juice consumed over 3 h leading to an average 82% reduction in exposure.[51] These large reductions in bioavailability are likely to affect the therapeutic efficacy of these medicines, suggesting that fruit juice consumption is best avoided within 4 h of drug administration, particularly with celiprolol and atenolol.

Ciprofloxacin is a widely used fluoroquinolone antibiotic, with bacterial killing depending on achieving adequate plasma concentrations; a ciprofloxacin AUC over the minimum inhibitory concentration of the pathogen (AUC/MIC ratio) of >125 has been associated with optimal ciprofloxacin efficacy.[54] One study investigated the effect of a relatively small volume of orange juice (355 ml) on the pharmacokinetics of ciprofloxacin, demonstrating a reduction in ciprofloxacin exposure of ~20%,[55] probably due to inhibition of OATP1A2-mediated uptake.[56] Although only a modest reduction in exposure was observed, it has been reported that a significant number of patients do not achieve optimal AUC/MIC ratios with ciprofloxacin, particularly in the treatment of more resistant pathogens;[57] this interaction may further reduce the probability of achieving adequate AUC/MIC ratios with ciprofloxacin. Lower ciprofloxacin concentrations may also increase the risk of developing bacterial resistance.[58]

The leukotriene receptor antagonist montelukast is used in the treatment of asthma and allergic rhinitis and is a known substrate of OATP2B1.[59–61] Although grapefruit juice did not affect montelukast exposure, a modest decrease in montelukast exposure was observed with orange juice, dependent on *SLCO2B1* genotype.[62] This study suggests that constituents of orange juice may have a small but significant effect on the pharmacokinetics

OATP substrate	Juice volume (ml) within 2h (total)	Reference
Acebutolol	400 (2400)	69
Aliskiren	200 (3000)	68
	200 (3000)	9
	200 (3000)	9
	300 (300)	65
Atenolol	400 (2400)	10
	600 (600)	51
	900 (1200)	51
Celiprolol	400 (2400)	8
	400 (2400)	49
	600 (1800)	50
Ciprofloxacin	355 (355)	55
Fexofenadine	300 (300)	6
	300 (300)	7
	300 (300)	45
	480 (1920)	44
	900 (1200)	45
	900 (1200)	5
	900 (1200)	5
	900 (1200)	5
	900 (1200)	46
Glibenclamide	600 (1800)	70
Levofloxacin	355 (355)	71
Montelukast	240 (240)^	62
	240 (240)^	62
Pitavastatin	250 (3000)	73
Pravastatin	250 (2250)	75
	600 (1800)	74
	700 (800)	79
Repaglinide	300 (300)	76
Talinolol	300 (300)	48
	300 (6000)	48
Thyroxine	600 (1800)	72

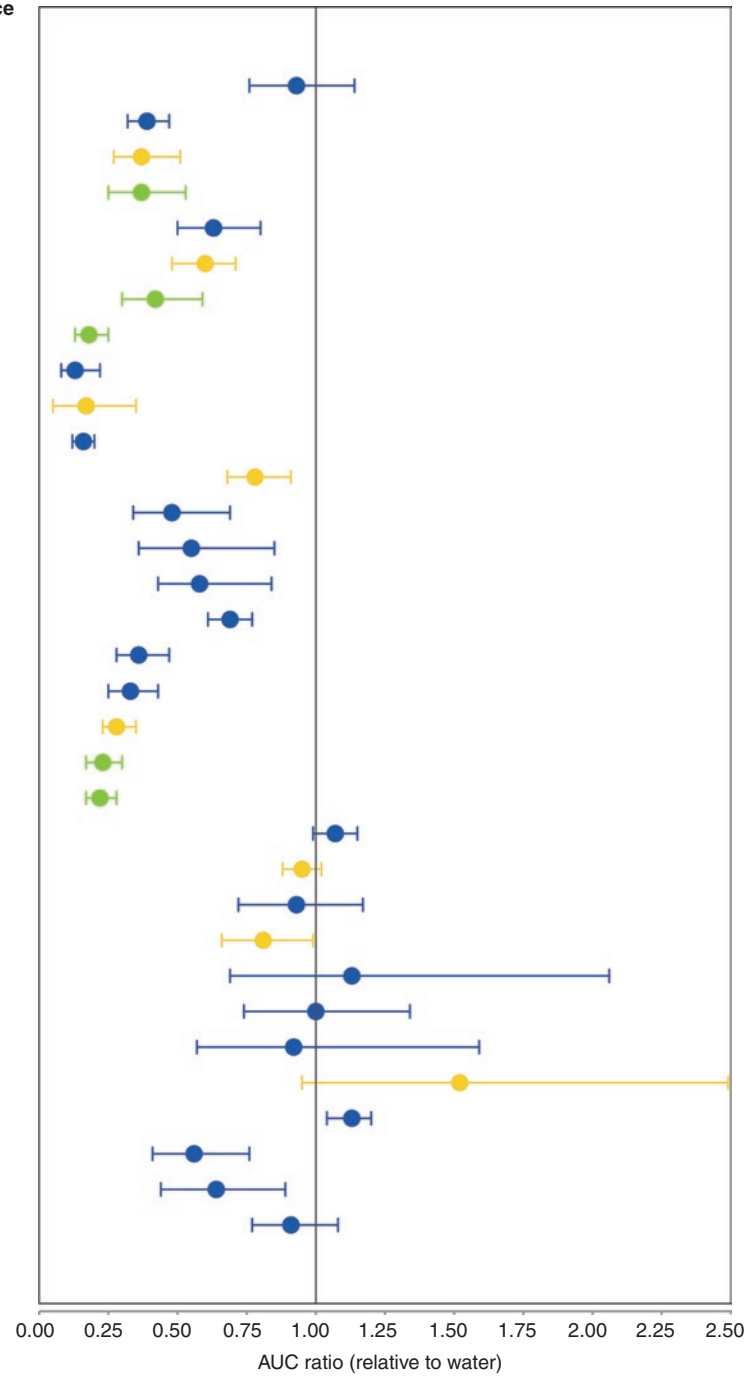

Figure 2 Forest plot showing the ratio of the area under the concentration–time curve (AUC) of OATP substrates when coadministered with fruit juice as compared with administration with water in 26 studies. Interactions involving grapefruit juice are shown in blue, orange juice interactions in yellow, and apple juice interactions in green. Filled circles represent the point estimate for the AUC ratio, with whiskers representing the 95% confidence interval (CI) for the AUC ratio. Juice volume represents volume consumed within 2 h before or after drug administration (total juice volume administered during study). ^Control phase in this study used Gatorade (PepsiCo, Purchase, NY) rather than water. Where 95% CIs for the AUC ratio of juice:water were not reported in the publication, these were calculated using Fieller's theorem.[93,94] OATP, organic anion–transporting polypeptide.

of montelukast, providing the first example of a gene–diet interaction in the context of intestinal OATP-mediated fruit juice–drug interactions.

Aliskiren is the first member of a new class of antihypertensives known as direct renin inhibitors.[63] Aliskiren was initially identified as a substrate of OATP2B1[64] but has recently been shown to be a substrate of OATP1A2;[65] it is known to have poor oral bioavailability of ~2–3% and is primarily excreted unchanged.[66] Furthermore, given that aliskiren is known to exhibit poor permeability across lipid membranes,[64,67] transport via OATP1A2 may be an important mechanism for its absorption.

Three studies have evaluated the effects of grapefruit, orange, and apple juice on aliskiren pharmacokinetics, with two studies involving multiple juice consumptions finding that juice coadministration significantly reduced aliskiren exposure, generally by >60%.[9,68] A third study, involving a single consumption of 300 ml grapefruit juice, found a mean 37% reduction in aliskiren exposure when juice was coadministered.[65] Differences in the juice volume consumed may account for the smaller effect of grapefruit juice in this study as compared with earlier studies, although differences in aliskiren dose and the brand of grapefruit juice ingested may also contribute to the reduced effect of grapefruit juice.[65] Plasma renin activity at 24 h post dose was significantly higher when aliskiren was coadministered with orange or apple juice as compared with water, although no significant changes in blood pressure were observed during juice coadministration.[9] As these studies were based on a single dose of aliskiren, and were conducted in healthy volunteers rather than patients with hypertension, it is possible that ongoing juice consumption may compromise the antihypertensive efficacy of aliskiren. When possible, juice consumption should be avoided within 4 h of aliskiren administration.

OATP substrates without significantly reduced exposure

Studies with acebutolol, glibenclamide, levofloxacin, pitavastatin, pravastatin, repaglinide, and thyroxine have not found a significant effect of fruit juice administration on systemic exposure (**Figure 2**).[69–75,76] (References 76–94 are provided as **Supplementary References** online.) Although these medicines are known to be substrates for one or more OATP subtypes, uptake via intestinally expressed OATP1A2 or OATP2B1 may play only a small role in the gastrointestinal absorption of these medicines. In addition, lipophilic drugs that are extensively metabolized, such as repaglinide and glibenclamide,[77,78] may have a reduced susceptibility to OATP-mediated fruit juice–drug interactions. The slightly increased bioavailability observed with repaglinide is probably the result of decreased intestinal CYP3A4 metabolism due to the furanocoumarin components of grapefruit juice.[76]

The effect of fruit juice consumption on the pharmacokinetics of pravastatin is unclear. Two studies with pravastatin and grapefruit juice did not identify a significant change in exposure,[74,75] but Kobayashi and colleagues identified a significant increase, rather than decrease, in pravastatin exposure when coadministered with orange juice.[79] As pravastatin is known to be a substrate for OATP2B1,[23,47] and its uptake by this transporter is increased at lower pH,[80] a reduction in intestinal pH due to orange juice may partially explain this result; however, the authors of this study found no effect on pravastatin pharmacokinetics in rats when coadministered with acetic acid at an equivalent pH to orange juice.[79] Although species differences in OATPs have been reported and may account for this discrepancy,[53] this finding would also be consistent with a potential inhibition of hepatic uptake transporters (such as OATP1B1) by orange juice; it is clear that further studies are necessary to elucidate the mechanism of increased pravastatin exposure when coadministered with orange juice.

CONFOUNDING FACTORS AND FUTURE DIRECTIONS

The majority of studies investigating intestinal OATP-mediated fruit juice–drug interactions have examined the effect of grapefruit juice, rather than the more commonly consumed orange or apple juice. Where multiple fruit juices have been tested for a specific medicine, orange juice and apple juice have generally reduced drug concentrations to an extent similar to that with grapefruit juice, as observed with aliskiren, fexofenadine, and celiprolol (**Figure 2**). A larger reduction in the systemic exposure of atenolol was observed with the consumption of apple juice than with orange juice, although the larger juice volumes consumed in the apple juice study may explain this difference. Despite this, the furanocoumarins found in grapefruit juice, and implicated in intestinal CYP3A4 inhibition, are absent in orange and apple juice,[28,81] suggesting that substrates for both intestinally expressed OATPs and CYP3A4 may potentially interact differently with orange and apple juice compared with grapefruit juice.

Small sample size is a limitation of many of the studies included in this review. Furthermore, the significant effects of fruit juice coadministration on talinolol and ciprofloxacin have yet to be replicated by other authors. Fruit juice composition, volume, and time of consumption are additional confounding factors in the assessment of fruit juice–drug interactions (**Table 1**). Studies included in this review used a variety of juice consumption schedules in relation to the time of drug administration, ranging from a single consumption of juice at the time of drug administration to multiple consumptions of juice before and/or following drug administration. It is likely that juice consumption within 2 h of drug administration will have the largest effect on intestinal OATP-mediated drug interactions[6] (**Figure 2**).

With respect to juice volume, greater reductions in bioavailability with larger juice volumes were demonstrated with fexofenadine and atenolol;[45,51] however, this effect was not observed with talinolol,[48] suggesting that the effect of juice volume on the extent of interaction may be specific to individual OATP1A2 or OATP2B1 substrates. Regarding juice composition, assessing the concentration of potentially causal active components, such as naringin and hesperidin, in future fruit juice–drug interaction studies may help to minimize this significant confounding factor when comparing fruit juice–drug interaction studies.

The potential effect of fruit juice consumption on OATPs expressed on the sinusoidal membrane of hepatocytes and involved in drug uptake into the liver, such as OATP1B1, OATP1B3, and OATP2B1,[2] is an important area for future study. Mandery and colleagues investigated the effect of naringin and its aglycone, naringenin, on OATP1B1 and OATP1B3, and also estimated possible concentrations of naringenin in portal venous blood to examine the potential physiological relevance of orally administered naringenin as an inhibitor of hepatic OATPs.[82] Both flavonoids inhibited OATP1B1 and OATP1B3, and naringenin concentrations in portal venous blood were estimated between 9.3 and 40.4 μmol/l following a 135 mg dose of naringenin.[82]

These concentrations are significantly below the half maximal inhibitory concentration values determined for naringenin for OATP1B1 and OATP1B3 (101 μmol/l and 81.6 μmol/l, respectively)[82] suggesting that juice consumption may not lead to concentrations capable of significant inhibition of hepatic OATPs; furthermore, the 135 mg dose of naringenin used in these calculations is far higher than the concentrations of naringenin observed in grapefruit juice (median (range) 3.3 (0.4–16.2) mg/100 ml juice).[27] Naringin concentrations in grapefruit juice are ~10-fold higher than naringenin (**Table 1**), and naringin is known to be partially metabolized to naringenin in the large intestine by colonic bacteria,[83–85] however, it is uncertain whether grapefruit juice consumption could result in naringenin concentrations in portal venous blood capable of inhibiting OATP-mediated drug uptake into the liver.

The antiparasitic agent ivermectin and topoisomerase inhibitor etoposide are both known substrates of P-gp[86,87] but have not been investigated as substrates of OATP1A2 or OATP2B1. Fruit juice consumption has been found to reduce the systemic exposure of both ivermectin and etoposide,[88,89] suggesting that intestinal inhibition of OATP-mediated absorption may be a possible mechanism of interaction.

Conversely, several important medicines have been identified as OATP2B1 or OATP1A2 substrates but have not yet been investigated in fruit juice–drug interaction studies. The hydrophilic statin rosuvastatin is known to be a substrate of OATP1B1, OATP1B3, OATP1A2, and OATP2B1.[90] A recent *in vitro* study with rosuvastatin demonstrated a higher contribution of OATP2B1-mediated uptake than was observed due to passive diffusion, with inhibitors of OATP2B1 reducing rosuvastatin uptake in a concentration-dependent manner.[80] These results suggest intestinal OATP2B1 plays a significant role in the oral absorption of rosuvastatin and that OATP inhibition by fruit juices may affect rosuvastatin bioavailability. The β-blocker sotalol and folic acid antagonist methotrexate are known to be OATP1A2 substrates.[52,91] Further studies are needed to examine the potential effect of fruit juice consumption on these medicines, and to investigate the influence of fruit juices other than grapefruit in situations in which intestinal OATP inhibition has been shown to affect drug absorption.

CONCLUSIONS

OATP-mediated fruit juice–drug interactions are a clinically important, rapidly developing area of research that is helping to inform the optimal use of medicines. To date, significant reductions in systemic exposure with fruit juice coadministration have been demonstrated for the β-blocker talinolol and the quinolone antibiotic ciprofloxacin, with reductions in exposure of more than half reported with the antihistamine fexofenadine, the β-blockers celiprolol and atenolol, and the direct renin inhibitor aliskiren. Furthermore, the involvement of the more commonly consumed orange and apple juices in OATP-mediated fruit juice–drug interactions, in addition to grapefruit juice, is likely to substantially increase the prevalence and importance of fruit juice–drug interactions in clinical practice.

SUPPLEMENTARY MATERIAL is linked to the online version of the paper at http://www.nature.com/cpt

AUTHOR CONTRIBUTIONS

A.J.M., M.J.D., and B.D.R. wrote the manuscript and designed the research. M.J.D. performed the research. M.J.D analyzed the research.

CONFLICT OF INTEREST

The authors declared no conflict of interest.

1. Bailey, D.G., Spence, J.D., Edgar, B., Bayliff, C.D. & Arnold, J.M. Ethanol enhances the hemodynamic effects of felodipine. *Clin. Invest. Med.* **12**, 357–362 (1989).
2. Niemi, M. Role of OATP transporters in the disposition of drugs. *Pharmacogenomics* **8**, 787–802 (2007).
3. Giacomini, K.M. *et al*. Membrane transporters in drug development. *Nat. Rev. Drug Discov.* **9**, 215–236 (2010).
4. Koenen, A., Kroemer, H.K., Grube, M. & Meyer zu Schwabedissen, H.E. Current understanding of hepatic and intestinal OATP-mediated drug-drug interactions. *Expert Rev. Clin. Pharmacol.* **4**, 729–742 (2011).
5. Dresser, G.K. *et al*. Fruit juices inhibit organic anion transporting polypeptide-mediated drug uptake to decrease the oral availability of fexofenadine. *Clin. Pharmacol. Ther.* **71**, 11–20 (2002).
6. Glaeser, H. *et al*. Intestinal drug transporter expression and the impact of grapefruit juice in humans. *Clin. Pharmacol. Ther.* **81**, 362–370 (2007).
7. Bailey, D.G., Dresser, G.K., Leake, B.F. & Kim, R.B. Naringin is a major and selective clinical inhibitor of organic anion-transporting polypeptide 1A2 (OATP1A2) in grapefruit juice. *Clin. Pharmacol. Ther.* **81**, 495–502 (2007).
8. Lilja, J.J., Backman, J.T., Laitila, J., Luurila, H. & Neuvonen, P.J. Itraconazole increases but grapefruit juice greatly decreases plasma concentrations of celiprolol. *Clin. Pharmacol. Ther.* **73**, 192–198 (2003).
9. Tapaninen, T., Neuvonen, P.J. & Niemi, M. Orange and apple juice greatly reduce the plasma concentrations of the OATP2B1 substrate aliskiren. *Br. J. Clin. Pharmacol.* **71**, 718–726 (2011).
10. Lilja, J.J., Raaska, K. & Neuvonen, P.J. Effects of orange juice on the pharmacokinetics of atenolol. *Eur. J. Clin. Pharmacol.* **61**, 337–340 (2005).
11. Nishimura, M. & Naito, S. Tissue-specific mRNA expression profiles of human ATP-binding cassette and solute carrier transporter superfamilies. *Drug Metab. Pharmacokinet.* **20**, 452–477 (2005).
12. Englund, G. *et al*. Regional levels of drug transporters along the human intestinal tract: co-expression of ABC and SLC transporters and comparison with Caco-2 cells. *Eur. J. Pharm. Sci.* **29**, 269–277 (2006).
13. Seithel, A., Karlsson, J., Hilgendorf, C., Björquist, A. & Ungell, A.L. Variability in mRNA expression of ABC- and SLC-transporters in human intestinal cells: comparison between human segments and Caco-2 cells. *Eur. J. Pharm. Sci.* **28**, 291–299 (2006).
14. Hilgendorf, C., Ahlin, G., Seithel, A., Artursson, P., Ungell, A.L. & Karlsson, J. Expression of thirty-six drug transporter genes in human intestine, liver, kidney, and organotypic cell lines. *Drug Metab. Dispos.* **35**, 1333–1340 (2007).
15. Meier, Y. *et al*. Regional distribution of solute carrier mRNA expression along the human intestinal tract. *Drug Metab. Dispos.* **35**, 590–594 (2007).
16. Ballestero, M.R., Monte, M.J., Briz, O., Jimenez, F., Gonzalez-San Martin, F. & Marin, J.J. Expression of transporters potentially involved in the targeting of cytostatic bile acid derivatives to colon cancer and polyps. *Biochem. Pharmacol.* **72**, 729–738 (2006).
17. Eechoute, K. *et al*. Environmental and genetic factors affecting transport of imatinib by OATP1A2. *Clin. Pharmacol. Ther.* **89**, 816–820 (2011).
18. Maubon, N. *et al*. Analysis of drug transporter expression in human intestinal Caco-2 cells by real-time PCR. *Fundam. Clin. Pharmacol.* **21**, 659–663 (2007).
19. Lee, W. *et al*. Polymorphisms in human organic anion-transporting polypeptide 1A2 (OATP1A2): implications for altered drug disposition and central nervous system drug entry. *J. Biol. Chem.* **280**, 9610–9617 (2005).
20. Franke, R.M., Scherkenbach, L.A. & Sparreboom, A. Pharmacogenetics of the organic anion transporting polypeptide 1A2. *Pharmacogenomics* **10**, 339–344 (2009).
21. Anderson, L. & Seilhamer, J. A comparison of selected mRNA and protein abundances in human liver. *Electrophoresis* **18**, 533–537 (1997).
22. Su, Y., Zhang, X. & Sinko, P.J. Human organic anion-transporting polypeptide OATP-A (SLC21A3) acts in concert with P-glycoprotein and multidrug

resistance protein 2 in the vectorial transport of Saquinavir in Hep G2 cells. *Mol. Pharm.* **1**, 49–56 (2004).
23. Kobayashi, D., Nozawa, T., Imai, K., Nezu, J., Tsuji, A. & Tamai, I. Involvement of human organic anion transporting polypeptide OATP-B (SLC21A9) in pH-dependent transport across intestinal apical membrane. *J. Pharmacol. Exp. Ther.* **306**, 703–708 (2003).
24. Lown, K.S. *et al*. Grapefruit juice increases felodipine oral availability in humans by decreasing intestinal CYP3A protein expression. *J. Clin. Invest.* **99**, 2545–2553 (1997).
25. Lilja, J.J., Kivistö, K.T. & Neuvonen, P.J. Duration of effect of grapefruit juice on the pharmacokinetics of the CYP3A4 substrate simvastatin. *Clin. Pharmacol. Ther.* **68**, 384–390 (2000).
26. De Castro, W.V., Mertens-Talcott, S., Rubner, A., Butterweck, V. & Derendorf, H. Variation of flavonoids and furanocoumarins in grapefruit juices: a potential source of variability in grapefruit juice-drug interaction studies. *J. Agric. Food Chem.* **54**, 249–255 (2006).
27. Gattuso, G., Barreca, D., Gargiulli, C., Leuzzi, U. & Caristi, C. Flavonoid composition of Citrus juices. *Molecules* **12**, 1641–1673 (2007).
28. Guo, L.Q., Fukuda, K., Ohta, T. & Yamazoe, Y. Role of furanocoumarin derivatives on grapefruit juice-mediated inhibition of human CYP3A activity. *Drug Metab. Dispos.* **28**, 766–771 (2000).
29. Edwards, D.J. *et al*. 6′,7′-Dihydroxybergamottin in grapefruit juice and Seville orange juice: effects on cyclosporine disposition, enterocyte CYP3A4, and P-glycoprotein. *Clin. Pharmacol. Ther.* **65**, 237–244 (1999).
30. Rouseff, R.L., Martin, S.F. & Youtsey, C.O. Quantitative survey of narirutin, naringin, hesperidin, and neohesperidin in citrus. *J. Agric. Food Chem.* **35**, 1027–1030 (1987).
31. Satoh, H. *et al*. Citrus juices inhibit the function of human organic anion-transporting polypeptide OATP-B. *Drug Metab. Dispos.* **33**, 518–523 (2005).
32. Tran, V.H., Marks, D., Duke, R.K., Bebawy, M., Duke, C.C. & Roufogalis, B.D. Modulation of P-glycoprotein-mediated anticancer drug accumulation, cytotoxicity, and ATPase activity by flavonoid interactions. *Nutr. Cancer* **63**, 435–443 (2011).
33. Taipalensuu, J. *et al*. Correlation of gene expression of ten drug efflux proteins of the ATP-binding cassette transporter family in normal human jejunum and in human intestinal epithelial Caco-2 cell monolayers. *J. Pharmacol. Exp. Ther.* **299**, 164–170 (2001).
34. Zhang, S., Yang, X. & Morris, M.E. Flavonoids are inhibitors of breast cancer resistance protein (ABCG2)-mediated transport. *Mol. Pharmacol.* **65**, 1208–1216 (2004).
35. Uesawa, Y. & Mohri, K. Hesperidin in orange juice reduces the absorption of celiprolol in rats. *Biopharm. Drug Dispos.* **29**, 185–188 (2008).
36. Schieber, A., Keller, P. & Carle, R. Determination of phenolic acids and flavonoids of apple and pear by high-performance liquid chromatography. *J. Chromatogr. A* **910**, 265–273 (2001).
37. Van Der Sluis, A.A., Dekker, M., Skrede, G. & Jongen, W.M. Activity and concentration of polyphenolic antioxidants in apple juice. 1. Effect of existing production methods. *J. Agric. Food Chem.* **50**, 7211–7219 (2002).
38. Young, J.F. *et al*. Effect of fruit juice intake on urinary quercetin excretion and biomarkers of antioxidative status. *Am. J. Clin. Nutr.* **69**, 87–94 (1999).
39. Wang, X., Wolkoff, A.W. & Morris, M.E. Flavonoids as a novel class of human organic anion-transporting polypeptide OATP1B1 (OATP-C) modulators. *Drug Metab. Dispos.* **33**, 1666–1672 (2005).
40. Fuchikami, H., Satoh, H., Tsujimoto, M., Ohdo, S., Ohtani, H. & Sawada, Y. Effects of herbal extracts on the function of human organic anion-transporting polypeptide OATP-B. *Drug Metab. Dispos.* **34**, 577–582 (2006).
41. Mandery, K. *et al*. Influence of the flavonoids apigenin, kaempferol, and quercetin on the function of organic anion transporting polypeptides 1A2 and 2B1. *Biochem. Pharmacol.* **80**, 1746–1753 (2010).
42. Kim, K.A., Park, P.W. & Park, J.Y. Short-term effect of quercetin on the pharmacokinetics of fexofenadine, a substrate of P-glycoprotein, in healthy volunteers. *Eur. J. Clin. Pharmacol.* **65**, 609–614 (2009).
43. Robards, K., Li, X., Antolovich, M. & Boyd, S. Characterisation of citrus by chromatographic analysis of flavonoids. *J. Sci. Food Agric.* **75**, 87–101 (1997).
44. Banfield, C., Gupta, S., Marino, M., Lim, J. & Affrime, M. Grapefruit juice reduces the oral bioavailability of fexofenadine but not desloratadine. *Clin. Pharmacokinet.* **41**, 311–318 (2002).
45. Dresser, G.K., Kim, R.B. & Bailey, D.G. Effect of grapefruit juice volume on the reduction of fexofenadine bioavailability: possible role of organic anion transporting polypeptides. *Clin. Pharmacol. Ther.* **77**, 170–177 (2005).
46. Imanaga, J. *et al*. The effects of the *SLCO2B1* c.1457C > T polymorphism and apple juice on the pharmacokinetics of fexofenadine and midazolam in humans. *Pharmacogenet. Genomics* **21**, 84–93 (2011).
47. Nozawa, T., Imai, K., Nezu, J., Tsuji, A. & Tamai, I. Functional characterization of pH-sensitive organic anion transporting polypeptide OATP-B in human. *J. Pharmacol. Exp. Ther.* **308**, 438–445 (2004).
48. Schwarz, U.I. *et al*. Grapefruit juice ingestion significantly reduces talinolol bioavailability. *Clin. Pharmacol. Ther.* **77**, 291–301 (2005).
49. Lilja, J.J., Juntti-Patinen, L. & Neuvonen, P.J. Orange juice substantially reduces the bioavailability of the beta-adrenergic-blocking agent celiprolol. *Clin. Pharmacol. Ther.* **75**, 184–190 (2004).
50. Ieiri, I. *et al*. Microdosing clinical study: pharmacokinetic, pharmacogenomic (*SLCO2B1*), and interaction (grapefruit juice) profiles of celiprolol following the oral microdose and therapeutic dose. *J. Clin. Pharmacol.* **52**, 1078–1089 (2012).
51. Jeon, H.*et al*. Apple juice greatly reduces systemic exposure to atenolol. *Br. J. Clin. Pharmacol.* (2012), e-pub ahead of print 11 May 2012.
52. Kato, Y., Miyazaki, T., Kano, T., Sugiura, T., Kubo, Y. & Tsuji, A. Involvement of influx and efflux transport systems in gastrointestinal absorption of celiprolol. *J. Pharm. Sci.* **98**, 2529–2539 (2009).
53. Shirasaka, Y., Kuraoka, E., Spahn-Langguth, H., Nakanishi, T., Langguth, P. & Tamai, I. Species difference in the effect of grapefruit juice on intestinal absorption of talinolol between human and rat. *J. Pharmacol. Exp. Ther.* **332**, 181–189 (2010).
54. Forrest, A., Nix, D.E., Ballow, C.H., Goss, T.F., Birmingham, M.C. & Schentag, J.J. Pharmacodynamics of intravenous ciprofloxacin in seriously ill patients. *Antimicrob. Agents Chemother.* **37**, 1073–1081 (1993).
55. Neuhofel, A.L., Wilton, J.H., Victory, J.M., Hejmanowsk, L.G. & Amsden, G.W. Lack of bioequivalence of ciprofloxacin when administered with calcium-fortified orange juice: a new twist on an old interaction. *J. Clin. Pharmacol.* **42**, 461–466 (2002).
56. Maeda, T. *et al*. Identification of influx transporter for the quinolone antibacterial agent levofloxacin. *Mol. Pharm.* **4**, 85–94 (2007).
57. Haeseker, M. *et al*. The ciprofloxacin target AUC/MIC ratio is not reached in hospitalized patients with the recommended dosing regimens. *Br. J. Clin. Pharmacol.* (2012), e-pub ahead of print 23 May 2012.
58. Thomas, J.K. *et al*. Pharmacodynamic evaluation of factors associated with the development of bacterial resistance in acutely ill patients during therapy. *Antimicrob. Agents Chemother.* **42**, 521–527 (1998).
59. Mougey, E.B., Feng, H., Castro, M., Irvin, C.G. & Lima, J.J. Absorption of montelukast is transporter mediated: a common variant of OATP2B1 is associated with reduced plasma concentrations and poor response. *Pharmacogenet. Genomics* **19**, 129–138 (2009).
60. Malmstrom, K. *et al*. Oral montelukast, inhaled beclomethasone, and placebo for chronic asthma. A randomized, controlled trial. Montelukast/Beclomethasone Study Group. *Ann. Intern. Med.* **130**, 487–495 (1999).
61. Meltzer, E.O. *et al*. Concomitant montelukast and loratadine as treatment for seasonal allergic rhinitis: a randomized, placebo-controlled clinical trial. *J. Allergy Clin. Immunol.* **105**, 917–922 (2000).
62. Mougey, E.B., Lang, J.E., Wen, X. & Lima, J.J. Effect of citrus juice and *SLCO2B1* genotype on the pharmacokinetics of montelukast. *J. Clin. Pharmacol.* **51**, 751–760 (2011).
63. Gradman, A.H., Schmieder, R.E., Lins, R.L., Nussberger, J., Chiang, Y. & Bedigian, M.P. Aliskiren, a novel orally effective renin inhibitor, provides dose-dependent antihypertensive efficacy and placebo-like tolerability in hypertensive patients. *Circulation* **111**, 1012–1018 (2005).
64. Vaidyanathan, S. *et al*. Pharmacokinetics of the oral direct renin inhibitor aliskiren in combination with digoxin, atorvastatin, and ketoconazole in healthy subjects: the role of P-glycoprotein in the disposition of aliskiren. *J. Clin. Pharmacol.* **48**, 1323–1338 (2008).
65. Rebello, S. *et al*. Intestinal OATP1A2 inhibition as a potential mechanism for the effect of grapefruit juice on aliskiren pharmacokinetics in healthy subjects. *Eur. J. Clin. Pharmacol.* **68**, 697–708 (2012).
66. Vaidyanathan, S., Jarugula, V., Dieterich, H.A., Howard, D. & Dole, W.P. Clinical pharmacokinetics and pharmacodynamics of aliskiren. *Clin. Pharmacokinet.* **47**, 515–531 (2008).
67. Wood, J.M. *et al*. Structure-based design of aliskiren, a novel orally effective renin inhibitor. *Biochem. Biophys. Res. Commun.* **308**, 698–705 (2003).
68. Tapaninen, T., Neuvonen, P.J. & Niemi, M. Grapefruit juice greatly reduces the plasma concentrations of the OATP2B1 and CYP3A4 substrate aliskiren. *Clin. Pharmacol. Ther.* **88**, 339–342 (2010).
69. Lilja, J.J., Raaska, K. & Neuvonen, P.J. Effects of grapefruit juice on the pharmacokinetics of acebutolol. *Br. J. Clin. Pharmacol.* **60**, 659–663 (2005).

70. Lilja, J.J., Niemi, M., Fredrikson, H. & Neuvonen, P.J. Effects of clarithromycin and grapefruit juice on the pharmacokinetics of glibenclamide. *Br. J. Clin. Pharmacol.* **63**, 732–740 (2007).
71. Wallace, A.W., Victory, J.M. & Amsden, G.W. Lack of bioequivalence when levofloxacin and calcium-fortified orange juice are coadministered to healthy volunteers. *J. Clin. Pharmacol.* **43**, 539–544 (2003).
72. Lilja, J.J., Laitinen, K. & Neuvonen, P.J. Effects of grapefruit juice on the absorption of levothyroxine. *Br. J. Clin. Pharmacol.* **60**, 337–341 (2005).
73. Ando, H. *et al*. Effects of grapefruit juice on the pharmacokinetics of pitavastatin and atorvastatin. *Br. J. Clin. Pharmacol.* **60**, 494–497 (2005).
74. Lilja, J.J., Kivistö, K.T. & Neuvonen, P.J. Grapefruit juice increases serum concentrations of atorvastatin and has no effect on pravastatin. *Clin. Pharmacol. Ther.* **66**, 118–127 (1999).
75. Fukazawa, I., Uchida, N., Uchida, E. & Yasuhara, H. Effects of grapefruit juice on pharmacokinetics of atorvastatin and pravastatin in Japanese. *Br. J. Clin. Pharmacol.* **57**, 448–455 (2004).