Inhibition of the organic anion-transporting polypeptide 1B1 by quercetin: an *in vitro* and *in vivo* assessment

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

• OATP1B1 is a liver-specific expression drug transporter and mediates the uptake of a broad range of compounds into hepatocytes. Modulation the activity of OATP1B1 may alter the pharmacokinetics of its substrate drugs, causing potential drug–drug interactions. As a popular dietary supplement in the United States, quercetin has been shown to interact with some drug transporters and efficiently influences their activity, but the effect of quercetin on OATP1B1 activity is not well known.

WHAT THIS STUDY ADDS

• Quercetin inhibits the OATP1B1-mediated transport of E3S and pravastatin *in vitro*, and also has a modest inhibitory influence on the pharmacokinetics of pravastatin in healthy Chinese-Han male volunteers.

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AIM

To investigate the effect of quercetin on organic anion transporting polypeptide 1B1 (OATP1B1) activities *in vitro* and on the pharmacokinetics of pravastatin, a typical substrate for OATP1B1 in healthy Chinese-Han male subjects.

METHODS

Using human embryonic kidney 293 (HEK293) cells stably expressing OATP1B1, we observed the effect of quercetin on OATP1B1-mediated uptake of estrone-3-sulphate (E3S) and pravastatin. The influence of quercetin on the pharmacokinetics of pravastatin was measured in 16 healthy Chinese-Han male volunteers receiving a single dose of pravastatin (40 mg orally) after co-administration of placebo or 500 mg quercetin capsules (once daily orally for 14 days).

RESULTS

Quercetin competitively inhibited OATP1B1-mediated E3S uptake with a K_i value of $17.9 \pm 4.6 \mu\text{M}$ and also inhibited OATP1B1-mediated pravastatin uptake in a concentration dependent manner (IC_{50} , $15.9 \pm 1.4 \mu\text{M}$). In healthy Chinese-Han male subjects, quercetin increased the pravastatin area under the plasma concentration – time curve ($\text{AUC}(0,10 \text{ h})$) and the peak plasma drug concentration (C_{max}) to 24% (95% CI 15, 32%, $P < 0.001$) and 31% (95% CI 20, 42%, $P < 0.001$), respectively. After administration of quercetin, the elimination half-life ($t_{1/2}$) of pravastatin was prolonged by 14% (95% CI 4, 24%, $P = 0.027$), with no change in the time to reach C_{max} (t_{max}). Moreover, quercetin decreased the apparent clearance (CL/F) of pravastatin by 18% (95% CI 75, 89%, $P < 0.001$).

CONCLUSIONS

These findings suggest that quercetin inhibits the OATP1B1-mediated transport of E3S and pravastatin *in vitro* and also has a modest inhibitory influence on the pharmacokinetics of pravastatin in healthy Chinese-Han male volunteers. The effects of quercetin on other OATP1B1 substrate drugs deserve further investigation.

Introduction

Quercetin (3,3',4',5,7-pentahydroxyflavone), is a naturally occurring flavonoid ubiquitously present in fruit and vegetables, such as onion, tea, apples and berries. It exhibits anti-oxidative, anti-inflammatory and vasodilating effects and has been proposed to be a potential anti-cancer agent [1, 2]. Recently, quercetin has been marketed in the United States primarily as a dietary supplement [3]. Accumulating evidence has demonstrated that quercetin interacts with drug transporters and efficiently influences their activity, highlighting its potential for natural product – drug interaction. For example, previous studies have suggested that quercetin is a potent inhibitor of P-glycoprotein, multidrug resistance-associated protein 1/2 (MRP1/2), breast cancer resistance protein (BCRP), organic anion transporting polypeptide 1A2 (OATP1A2), OATP2B1 and organic anion transporter 1/3 (OAT1/3), indicating that it may potentially affect *in vivo* drug disposition by inhibiting different transporter systems [1, 4–6].

OATP1B1, encoded by *SLCO1B1*, is a 691-amino acid glycoprotein with an apparent molecular mass of 84 KDa. OATP1B1 is restrictedly expressed in the basolateral membrane of human hepatocytes and transports a broad range of compounds such as bile acids, conjugated steroids, thyroid hormones, peptides, and drugs including rifampicin and HMG-CoA reductase inhibitors [7–9]. Its apparent liver specific expression and capacity of transporting a large number of structurally different compounds suggest that OATP1B1 plays an important role in hepatic drug uptake and elimination. Thus, modulation of OATP1B1 function can alter the pharmacokinetics of OATP1B1 substrate drugs and potentially lead to adverse effects [10].

Pravastatin, a potent inhibitor of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, is widely used in the treatment of hypercholesterolaemia [11]. As a hydrophilic compound, pravastatin is not significantly metabolized by the cytochrome P450 (CYP450) enzymes. It is taken up by the liver via active transport and mostly excreted unchanged in the bile, with much less excretion in urine [12, 13]. OATP1B1 is thought to be the major transporter involved in the hepatic uptake of pravastatin, and OATP1B1-mediated uptake has been proved to be the rate-limiting step in the hepatic clearance of pravastatin [14–16]. Therefore, pravastatin can be used as a probe drug for evaluating the function of OATP1B1 [17].

In this study, we aim to investigate the potential influence of quercetin on the transport activity of OATP1B1 *in vitro* and assess the impact of quercetin on pravastatin pharmacokinetics in healthy Chinese-Han male volunteers. Considering the frequent consumption of quercetin-containing foods and supplements, the hypothesized interaction with OATP1B1 might be an important determinant of intra-individual variability of drug disposition.

Methods

Materials

Radiolabeled [^3H]-estrone-3-sulphate (E3S) (54.3 Ci mol $^{-1}$, >97% purity) was purchased from PerkinElmer Life Sciences. Pravastatin sodium salt hydrate, rifampicin, quercetin (purity $\geq 98\%$, HPLC), sodium butyrate and Krebs-Henseleit buffer were purchased from Sigma-Aldrich. Polyclonal goat antibody against human OATP1B1 was purchased from Santa Cruz Biotechnology. Blasticidin HCl and LipofectaminTM 2000 reagent were purchased from Invitrogen. Quercetin (purity $\geq 98\%$, HPLC) was purchased from Sigma-Aldrich Chemie GmbH. Quercetin capsules were purchased from GNC (USA; BN: 401922). Human OATP1B1 complementary DNA plasmid was kindly provided by Professor Richard Kim (Division of Clinical Pharmacology, Vanderbilt University, Nashville). Parental human embryonic kidney (HEK) 293 cell line was purchased from the Institute of Basic Medical Sciences, Chinese Academy of Medical Sciences (Beijing, P.R. China).

Cell culture and transfection

HEK293 cells were grown at 37°C in a humidified 5% CO $_2$ atmosphere in Minimum Essential Medium supplemented with 10% FBS (Hyclone, Logan, UT), 100 U ml $^{-1}$ penicillin and 100 $\mu\text{g ml}^{-1}$ streptomycin. OATP1B1 cDNA was transfected into HEK293 cells with LipofectaminTM 2000 according to the manufacturer's instructions. Then, HEK293 cells were selected by 6 $\mu\text{g ml}^{-1}$ blasticidin and colonies were picked up. The colonies which had the highest transport activities were used in the functional analysis (HEK293-OATP1B1). HEK293 cells transfected with vector alone were used to obtain the background activity (HEK293-MOCK). Sodium butyrate (10 mM) was added to the medium 24 h before use to induce the expression of OATP1B1.

Transport assays

HEK293-OATP1B1 and HEK293-MOCK cells were seeded in poly-L-lysine coated 24-well plates at an initial density of 2×10^5 cells/well. Cells were washed twice and pre-incubated with Krebs-Henseleit buffer at 37°C for 15 min, then incubated in 200 μl of Krebs-Henseleit buffer cells containing [^3H]-E3S (50 nM) or pravastatin (50 μM) together with quercetin or the vehicle (0.2% dimethyl sulfoxide) at 37°C for 5 min (E3S) or 10 min (pravastatin). Rifampicin (100 μM) was used as a positive control inhibitor. The uptake reaction was terminated by adding ice-cold Krebs-Henseleit buffer after removal of the incubation buffer. For experiments with [^3H]-E3S as substrate, cells were solubilized in 200 μl of 0.2 N NaOH and kept overnight at 4°C. After adding 50 μl 1 N HCl, aliquots (200 μl) were transferred to scintillation vials. The radioactivity was measured in a liquid scintillation counter (LS6500; Beckman Coulter, Inc., Fullerton, CA) after adding 2 ml of scintillation fluid. The remaining 50 μl of cell lysate was used to determine

the protein concentrations by the method of Lowry *et al.* with bovine serum albumin (BSA) as a standard [18]. For experiments with pravastatin as substrate, cells were solubilized in 250 μ l of 0.2% SDS and kept overnight. Aliquots (200 μ l) were used to determine the amount of intracellular pravastatin by a LC/MS/MS method, and the remaining 50 μ l aliquots of the cell lysis solution were used to determine the protein concentration by the method of Lowry *et al.* [18] with BSA as a standard. The uptake of [3 H]-E3S or pravastatin was expressed as percentage of the control (uptake in HEK293-MOCK cells in the presence of 0.2% dimethyl sulphoxide).

LC/MS/MS assay for pravastatin

Concentrations of pravastatin accumulated in cells were determined using a validated liquid chromatography with tandem mass spectrometric detection (LC/MS/MS) method. Samples were prepared by adding 100 μ l of internal standard solution (200 ng ml $^{-1}$ simvastatin) to 100 μ l of the cell lysates. LC/MS/MS analysis was performed by liquid chromatography-mass spectrometry with the Finnigan LCQ Deca XPplus (Finnigan, San Jose, CA). An Intersil ODS-3 column (5 μ m, 2.1 mm $\times$ 50 mm) and a mobile phase [acetonitrile : 1 mM methanoic acid amine (including 0.1% formic acid) = 3:2] at a flow rate of 0.2 ml min $^{-1}$ were applied. The lower limit of quantification was 0.98 ng ml $^{-1}$. The ion transitions monitored were as follows: m/z 447 to 327 for pravastatin and m/z 441 to 325 for simvastatin. These transitions represent the product ions of the [M+H] $^{+}$ ions. The lower limit of quantification for pravastatin was 0.957 ng ml $^{-1}$. The calibration curves were linear over the range 0.957 to 500 ng ml $^{-1}$ with mean correlation coefficients of 0.9992. The intra- and inter-day coefficients of variation were less than 10%.

Assessment of the effect of quercetin on the pharmacokinetic profile of pravastatin in vivo

Chinese-Han male volunteers were recruited for this study. All subjects underwent past medical history interviews and physical and laboratory examinations (haematology, serum biochemistry, urinalysis and electrocardiography) to confirm that they were healthy. Females were excluded to avoid possible effects of other factors (e.g. menstruation) on drug metabolism. Smokers were not included, nor were subjects deemed alcohol or drug dependent. Other exclusion criteria also included having participated in a clinical study during the past 4 months, taking any prescription or non-prescription drugs, grapefruit juice, herbal dietary supplements, coffee, green tea or any other quercetin-containing products within 2 weeks before study entry and until the study was completed. On the basis of our preliminary observation, we proposed that quercetin would reduce pravastatin AUC(0,10 h) after 14 days pre-treatment by about 20%. Given this effect size, a probability of type 1 error of 0.05 (two-tailed), a power of 0.8,

expected SD of pravastatin AUC(0,10 h) as 101 ng ml $^{-1}$ h, 16 volunteers would be required by power analysis (Graph-Pad, StatMate).

Sixteen healthy Chinese-Han male volunteers (aged 19 to 24 years, weight range 60–80 kg, body mass index range 20–25 kg m $^{-2}$) participated in the study after giving their individual written informed consent. The study was carried out in a two-phase, randomized and crossover manner with a 2 week washout period between phases. In each phase, 16 volunteers received 500 mg quercetin capsules or placebo (identical capsules filled with corn starch), once daily for 14 days. On day 14, after an overnight fast, all subjects received a single oral dose of 40 mg pravastatin (Mevalotin; Daiichi-Sankyo) 1 h after the placebo or quercetin administration. They were given a standardized meal at 4 h after the intake of pravastatin and standardized light meals after 10 h. The quercetin capsules contained quercetin (500 mg) and some other ingredients including cellulose, silicon dioxide, magnesium stearate and gelatin. This study protocol was approved by the Ethics Committee Board of Central South University, Changsha, Hunan, P.R. China (No. CTXY-100008). This clinical trial was registered with the Chinese Clinical Trial Registry (No. ChiCTR-TRC-10001277).

A series of venous blood samples (5 ml) was collected into EDTA-containing tubes before and at 0.25, 0.5, 0.75, 1, 2, 3, 4, 6, 8 and 10 h after pravastatin intake. Plasma pravastatin concentrations were quantified by use of LC/MS/MS analysis as described above. The peak concentration ($C_{\max}$) and time to $C_{\max}$ ($t_{\max}$) were obtained by inspection of the concentration – time data. The area under the concentration – time curve from 0–10 h (AUC(0,10 h)) of pravastatin was calculated by the linear trapezoidal rule. λ_z is the elimination rate constant determined from the terminal slope of the log concentration – time plot and the elimination half-life ($t_{1/2}$) = 0.693/ λ_z . The estimate of oral clearance (CL/F) was calculated as CL/F = Dose (40 mg)/AUC(0,10 h) (ng ml $^{-1}$ h).

Statistical analysis

All data were analyzed with SPSS software for Windows (version 18.0; SPSS, Chicago, IL). *In vitro* data were analyzed using ANOVA followed by a Dunnett's *post hoc* test or by Student's *t*-test. The AUC(0,10 h), $t_{1/2}$ and CL/F values of pravastatin after treatment with placebo and quercetin were analyzed using paired Student's *t*-test. The Wilcoxon signed-rank test was performed for the non-parametric paired two-group comparisons of $C_{\max}$ and $t_{\max}$. The results are expressed as mean $\pm$ SD in the text and tables and, for clarity, as mean $\pm$ SEM in the figures, unless otherwise stated. $P < 0.05$ indicated statistical significance.

Results

In vitro influence of quercetin on the OATP1B1-mediated uptake of E3S

In the present study, we established a HEK293 cell line stably expressing the human OATP1B1. According to a

Figure 1

Characterization of the HEK293-OATP1B1 cell line overexpression human OATP1B1. HEK293-OATP1B1 cells were characterized by real-time PCR (A) and immunoblot analysis (B). Na⁺/K⁺-ATPase, a membrane protein naturally expressed in all HEK293 cells, was used as loading control of surface proteins. The HEK293-OATP1B1 cells showed a significantly higher uptake (about 5.5-fold) of [³H]-E3S compared with HEK293-MOCK cells, rifampicin (100 μM) significantly decreased the OATP1B1 activity (C). ***P* < 0.01 compared with HEK293-MOCK cells. Each value is the mean value ± SEM (*n* = 6)

previous study, the low endogenous transporter expression in HEK293 cells supports its suitability as system for the overexpression of human transport proteins, especially SLC transporters [19]. The HEK293-OATP1B1 cells were characterized by a significantly higher expression of *SLCO1B1* mRNA and OATP1B1 protein compared with HEK293-MOCK cells. The functional characterization showed about 5.5-fold higher uptake of E3S, a well established model substrate of OATP1B1 [20], into HEK293-OATP1B1 cells compared with HEK293-MOCK cells. Rifampicin (100 μM), a known OATP1B1 inhibitor [21], significantly decreased the OATP1B1-mediated [³H]-E3S uptake (Figure 1). Quercetin also substantially decreased OATP1B1 activity and the kinetic study revealed that quercetin inhibited OATP1B1-mediated [³H]E3S uptake in a competitive manner with a *K_i* value of 17.9 ± 4.6 μM (determined from Dixon plots) (Figure 2).

In vitro influence of quercetin on the OATP1B1-mediated uptake of pravastatin

To determine whether quercetin alters the OATP1B1-mediated uptake of pravastatin, we performed a pravastatin uptake and inhibition experiment. We confirmed that pravastatin was transported by OATP1B1 with a substantially higher uptake in HEK293-OATP1B1 cells compared with HEK293-MOCK cells (about 4.1-fold), which was significantly decreased by rifampicin (100 μM). Quercetin

Figure 2

Dixon plot analysis of inhibition of OATP1B1-mediated [³H]-E3S uptake by quercetin in HEK293 cells. Transport of E3S at three different concentrations (solid circles 50 nM, open circles 100 nM, solid triangles 200 nM) was measured for 5 min in the absence or presence of increasing concentrations of quercetin (0–100 μM). The data are expressed as percentage (mean values ± SEM, *n* = 3) relative to the initial radioactivity. The *K_i* values were determined graphically from a Dixon plot

Figure 3

Inhibition of OATP1B1-mediated pravastatin transport by quercetin. Intracellular pravastatin accumulation (50 μM) in HEK293-OATP1B1 cells was about 4.1-fold higher than in HEK293-MOCK cells. Rifampicin (100 μM) was used as a positive control (A). Quercetin inhibited the OATP1B1-mediated uptake of pravastatin (50 μM) in a concentration-dependent manner (B). The data are expressed as percentage of control (uptake without quercetin). All data are presented as means ± SEM ($n = 6$). ** $P < 0.01$ compared with HEK293-MOCK cells

markedly inhibited the OATP1B1-mediated uptake of pravastatin in a concentration-dependent manner with an IC_{50} value of $15.9 \pm 1.4 \mu\text{M}$ (Figure 3).

In vivo influence of quercetin on the pharmacokinetics of pravastatin

The main pharmacokinetic parameters of pravastatin after 14 days of pretreatment by placebo or quercetin are summarized in Table 1. Mean plasma concentration profiles for pravastatin administered in combination with placebo or quercetin are shown in Figure 4. Repeated treatment with quercetin significantly increased C_{max} of pravastatin by 31% (95% CI 20, 42%, $P < 0.001$) and $AUC(0, 10 \text{ h})$ by 24% (95% CI 15, 32%, $P < 0.001$). After quercetin treatment, CL/F of pravastatin significantly decreased by 18% (95% CI 11, 25%, $P < 0.001$) compared with the placebo phase. Meanwhile, quercetin prolonged $t_{1/2}$ by 14% (95% CI 4, 24%, $P = 0.027$) with no significant influence on the t_{max} of pravastatin in comparison with the placebo phase. Intrasybject changes in the $AUC(0, 10 \text{ h})$ of pravastatin are depicted in Figure 5. It

can be seen that only one subject had lower plasma exposure to pravastatin after concomitant administration with quercetin.

Discussion

In this study, we focused on the interactions of quercetin with the human hepatocellular uptake transporter OATP1B1. Firstly, using newly established HEK293 cells with stable expression of human OATP1B1, we examined the effect of quercetin on OATP1B1-mediated [³H]-E3S transport *in vitro*. OATP1B1-mediated uptake of E3S is known to be biphasic with K_m values for high and low affinity components of 67.5 nM and 7 μM, respectively [22]. Because pravastatin mainly shares the high affinity site with E3S on OATP1B1 [23], so we set the concentration of [³H]-E3S at 50 nM (a value lower than 67.6 nM) for estimating the effect of quercetin on the high affinity binding site. Our data showed that quercetin inhibited the OATP1B1-mediated [³H]-E3S uptake in a competitive manner with a K_i value of $17.9 \pm 4.6 \mu\text{M}$. However, Wang *et al.* reported that quercetin did not exhibit a statistically significant influence on OATP1B1-mediated [³H]-DHEAS transport, which is not consistent with our present study [10]. This discrepancy may be possibly explained by differences in the substrates used, because different substrates may interact with different contact points to trigger the subsequent transport process, and this may explain the broad substrate specificity that characterizes many transporters. Then, we investigated the influence of quercetin on OATP1B1-mediated pravastatin transport *in vitro*. Our preliminary study had shown that OATP1B1-mediated pravastatin transport was linear for approximately 10 min, and uptake was saturable with an apparent K_m of 83.9 μM (data not shown), which was consistent with the results of Kameyama *et al.* [24]. Therefore we set the concentration of pravastatin at 50 μM (a value lower than 83.9 μM). The results suggested that quercetin inhibited OATP1B1-mediated pravastatin transport in a dose-dependent fashion.

In order to verify our *in vitro* results, an *in vivo* study was performed to determine whether plasma concentrations of pravastatin and quercetin might be sufficient to cause a drug–drug interaction. Our *in vivo* results showed that co-administration of quercetin had a modest inhibitory effect on pravastatin systemic exposures (AUC).

Pravastatin is a substrate of several drug transporters *in vivo*. Except for OATP1B1, multidrug resistance-associated protein 2 (MRP2, ABCC2) is considered to be the major transporter involved in the biliary excretion of pravastatin [25]. Previous studies indicated that variations in the MRP2 activity mainly alter the liver concentration of pravastatin and have only a minimal effect on the plasma concentration [26]. Other transporters that may play a limited role in the disposition of pravastatin *in vitro* include OATP2B1, OAT3, bile salt export pump (BSEP) and BCRP [27–31],

Table 1

Pharmacokinetic parameters of pravastatin in 16 healthy subjects after a single dose of pravastatin (40 mg) on day 14 after pretreatment with placebo or quercetin for 14 days

Parameter	Placebo phase (control)	Quercetin phase	Percentage of control (95% CI)	P value
AUC(0,10 h) (ng ml ⁻¹ h)	354.6 ± 89.6	437.8 ± 118.6	124% (115, 132%)	<0.001
C _{max} (ng ml ⁻¹)	123.2 ± 48.3	162.0 ± 76.0	131% (120, 142%)	<0.001
t _{max} (h)	0.9 ± 0.4	0.9 ± 0.3	NA	0.065
t _{1/2} (h)	1.6 ± 0.3	1.8 ± 0.3	114% (104, 124%)	0.027
CL/F (l h ⁻¹)	119.5 ± 29.2	97.3 ± 24.7	82% (75, 89%)	<0.001

Data are presented as mean ± SD. t_{max} data are given as medians with range. AUC(0,10 h), area under the plasma drug concentration – time curve from 0 to 10 h; C_{max}, peak plasma drug concentration; t_{max}, time to reach C_{max}; t_{1/2}, elimination half-life; CL/F, apparent clearance; NA not applicable; CI, confidence interval.

Figure 4

Plasma concentration–time profiles of pravastatin in 16 healthy subjects after a single oral dose of 40 mg pravastatin on day 14 after pretreatment with placebo (○) or quercetin (●) for 14 days. Inset depicts the same data on a semilogarithmic scale. Each value is the mean value ± SEM

but their activities are shown not to be associated with differences in pravastatin pharmacokinetics in healthy subjects [32, 33]. Therefore, we concluded that the changes in pravastatin pharmacokinetics in this study were mainly caused by quercetin-mediated inhibition of OATP1B1 activity.

Although quercetin is ubiquitously present in fruit and vegetables, it has been marketed in the United States primarily as a popular dietary supplement due to its beneficial health effects [3]. The recommended daily dosages of supplemental quercetin range from 200 mg to 1200 mg [34]. Therefore, we used a dose of 500 mg daily of quercetin according to the package insert in our study. In this study, we did not examine the pharmacokinetics of quercetin. Human pharmacokinetic studies have demonstrated that after a non-toxic i.v. dose of quercetin, its serum concentration ranges from 1 to 400 µM and after oral administra-

Figure 5

Individual values for AUC(0,10 h) of pravastatin in 16 healthy Chinese-Han male subjects after a single oral dose of 40 mg pravastatin on day 14 after pretreatment with placebo or quercetin for 14 days. Grey circles indicate the mean

tion of 300 mg quercetin supplement, its maximum plasma concentration is 9.72 µM [35, 36]. Furthermore, quercetin has a relatively long plasma half-life of 11–28 h in humans, indicating that repeated dietary intake of quercetin will lead to accumulation in plasma [37]. Comparing these data with the *in vitro* and *in vivo* results obtained in this study, we speculate that our results may have some clinical relevance. Recently, several *in vitro* studies revealed that many metabolites of quercetin, such as 7-O-glucuronosyl quercetin and 3'-O-glucuronide quercetin, also had an inhibitory influence on the function of some drug transporters including MRP1, MRP2 and OAT1 [6, 38]. Further investigation is needed to determine the effects of metabolites of quercetin on OATP1B1 activity.

SLCO1B1 polymorphism, especially c.521T > C, can significantly influence the pharmacokinetics of pravastatin in humans. A number of recent studies suggested that the c.521T > C SNP was strongly associated with decreased activity of OATP1B1 resulting in higher oral bioavailability of pravastatin in different ethnic groups [39]. The SNP

c.388A > G may be also associated with increased activity of OATP1B1 resulting in lower oral bioavailability of pravastatin [40]. The c.521T > C SNP frequency among Chinese individuals (13%) was slightly lower than the frequency observed in Caucasians (14.3%), and the c.388A > G SNP frequency in the Chinese population (79.5%) is much more higher compared with the Caucasian population (30.6%) [41]. Therefore, our results in this study had similar clinical values to Caucasians. However, pravastatin is never given clinically as a single dose and this is a limitation of the present study. Although previous studies had suggested that alteration in OATP1B1 function seemed to have a similar impact on the pharmacokinetics of single and multiple dose pravastatin [16], the effect of quercetin on the pharmacokinetics of multiple dose pravastatin requires further examination.

In conclusion, we showed for the first time that quercetin can inhibit OATP1B1-mediated transport of E3S and pravastatin *in vitro*, and also has a modest inhibitory influence on the pharmacokinetics of pravastatin in healthy Chinese-Han male subjects. The results of this study suggest a new mechanism to account for certain natural product – drug interactions, and may be clinically significant for patients taking quercetin and OATP1B1 substrate drugs simultaneously.

Competing Interests

There are no competing interests to declare.

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