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Genetic polymorphisms of organic cation transporter 1 (OCT1) and responses to metformin therapy in individuals with type 2 diabetes

A systematic review

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Abstract

Background: Metformin is one of the most commonly used drugs for the treatment of type 2 diabetes mellitus (T2DM). Despite its widespread use, there are considerable interindividual variations in metformin response, with about 35% of patients failing to achieve initial glycemic control. These variabilities that reflect phenotypic differences in drug disposition and action may indeed be due to polymorphisms in genes that regulate pharmacokinetics and pharmacodynamics of metformin. Moreover, interethnic differences in drug responses in some cases correspond to substantial differences in the frequencies of the associated pharmacogenomics risk allele.

Aim: This study aims to highlight and summarize the overall effects of organic cation transporter 1(OCT1) polymorphisms on therapeutic responses to metformin and to evaluate the potential role of such polymorphisms in interethnic differences in metformin therapy.

Methods: We conducted a systematic review according to the Preferred Reporting Items for Systematic Review and Meta-Analysis (PRISMA) guidelines. We searched for PubMed/MEDLINE, Embase, and CINAHL, relevant studies reporting the effects of OCT1 polymorphisms on metformin therapy in T2DM individuals. Data were extracted on study design, population characteristics, relevant polymorphisms, measure of genetic association, and outcomes. The presence of gastrointestinal side effects, glycated hemoglobin A1 (HbA1c) levels, fasting plasma glucose (FPG), and postprandial plasma glucose (PPG) concentrations after treatment with metformin were chosen as measures of the metformin responses. This systematic review protocol was registered with the International Prospective Register of Systematic Reviews (PROSPERO).

Results: According to the data extracted, a total of 34 OCT1 polymorphisms were identified in 10 ethnic groups. Significant differences in the frequencies of common alleles were observed among these groups. *Met408Val (rs628031)* variant was the most extensively explored with metformin responses. Although some genotypes and alleles have been associated with deleterious effects on metformin response, others indeed, exhibited positive effects.

Conclusion: Genetic effects of OCT1 polymorphisms on metformin responses were population specific. Further investigations in other populations are required to set ethnicity-specific reference for metformin responses and to obtain a solid basis to design personalized therapeutic approaches for T2DM treatment.

Abbreviations: AMPK = adenosine monophosphate activated protein kinase, BMI = body mass index, FPG = fasting plasma glucose, HbA1c = glycosylated hemoglobin, LKB1 = liver kinase B1, OCT1 = organic cation transporter 1, OCTs = organic cation transporters, OR = odds ratio, PPG = postprandial plasma glucose, SLC22A1 = solute carrier family 22 member 1, SNP = single nucleotide polymorphism, T2DM = type 2 diabetes mellitus, WHO = World Health Organization.

Keywords: genetic polymorphisms, glycemic response, metformin, OCT1, type 2 diabetes mellitus

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Data availability statement: All data are all contained in supporting information files

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1. Introduction

Metformin is the first-choice oral anti-hyperglycemic drug for use as monotherapy in individuals with newly diagnosed type 2 diabetes mellitus (T2DM).[1,2] It has several beneficial effects on cardiovascular risk factors, cancer, and polycystic ovary syndrome.[3,4] Moreover, metformin specifically reduces hepatic gluconeogenesis without increasing insulin secretion, inducing weight gain or risk of hypoglycemia.[1,5] The precise molecular mechanisms of metformin action are not well understood. It was initially suggested that a key action of metformin was to activate AMP-activated protein kinase (AMPK) through a decrease in hepatic energy status (i.e., increasing AMP: ADP and/or ADP/ATP concentration ratio) or through an upstream modulator, liver kinase B1 (LKB1), thereby leading to a reduction in gluconeogenic gene transcription.[6] However, recent investigations in conditional AMPK knockout mice demonstrated that metformin inhibits hepatic gluconeogenesis in an LKB1- and AMPK-independent manner via a decrease in the hepatic energy state.[7] Emerging evidence also indicates that inhibition of mitochondrial glycerophosphate dehydrogenase (mGPD), a critical enzyme in the glycerophosphate shuttle, could be the primary mechanism of metformin-induced inhibition of gluconeogenesis.[8] This preferential action of metformin in hepatocytes is due to the predominant expression of organic cation transporters 1 (OCT1) that are responsible of hepatic uptake of metformin. OCT1 belongs to the Solute Carrier family *(SLC22A)* and is localized in the sinusoidal membrane of rat and human hepatocytes.[9] Other reported locations of human OCT1 include the lateral membrane of intestinal epithelial cells,[10] the luminal (apical) membrane of ciliated cells in the lung, and of tubule epithelial cells in the kidney.[11] The human *SLC22A1* gene encoding OCT1 consists of 11 exons, has been mapped to chromosome 6q26 and spans about 37 kb. OCT1 is highly polymorphic in ethnically diverse populations and mediate differences in transporter function.[12] This helps provide a possible mechanism to account for interindividual variations in the metformin responses.[13] Moreover, carriers with loss of function OCT1 polymorphisms displayed decreased hepatic metformin exposure after intravenous injection of ^{11}C metformin.[14] Many studies have identified genetic polymorphisms in the *SLC22A1* gene among different populations groups but there are still contradictory reports on the effects of OCT1 polymorphisms on metformin-related therapeutic responses.[15] In light of this, it is crucial to obtain a greater understanding of the influence of OCT1 polymorphisms in the context of variable responses elicited by metformin treatment. This systematic review therefore summarizes the overall effects of OCT1 polymorphisms on metformin-related therapeutic responses and also evaluates its potential role in terms of interethnic differences in this instance.

2. Methods

2.1. Literature search strategy

We searched the following electronic databases PubMed/MEDLINE, Embase, and CINAHL from January 1990 to July 2017, to identify studies reporting on the effects of OCT1 variants on metformin responses in T2DM individuals. The search strategy based on the combination of relevant terms was designed by a librarian. The main search strategy conducted in PubMed/Medline was as follows: "((((Diabetes [MeSH Terms]) OR type 2 diabetes)) AND (((((((Genetic [MeSH Terms]) OR genetic markers) OR genetic polymorphism) OR Single nucleotide polymorphism) OR Polymorphism) OR variant) OR gene) OR allele)) AND (((solute carrier family 22 organic cation transporter, member 1 [MeSH Terms]) OR Organic Cation Transporter 1) OR OCT 1)". This search strategy was adapted when searching other databases. The search was performed independently by 2 investigators (EPMM and MGF) who identified articles in sequential fashion (titles, abstracts, and then full texts). In addition, references cited in the selected articles and published reviews were manually searched in order to identify any additional relevant studies.

2.2. Study selection

We included genetic association studies that reported data on the genetic effects of OCT1 polymorphisms on levels of HbA1c, fasting plasma glucose (FPG) and post-prandial plasma glucose (PPG), and also on gastrointestinal side effects in T2DM individuals. Two review authors (EPMM and MGF) independently assessed eligibility for inclusion in the review based on the inclusion and exclusion criteria. Any disagreements between the 2 review authors were resolved by consensus or consulting a third review author if necessary. Here we excluded animal studies, review articles, meta-analyses, case reports, editorials, and comments. In addition, 2 articles written in Russian were also excluded. A PRISMA (Preferred Reporting Items for Systematic Review and Meta-Analysis) flow diagram was used to document the process of literature selection and reasons for exclusion (Fig. 1).

2.3. Data extraction, assessment, and synthesis

Two reviewers (EPMM and MGF) independently extracted data using a preconceived data extraction sheet. Data were collected on the first author name, year of publication, geographical location (population where the study was performed), study design, sample size, participants' characteristics (mean or median age, age range, and proportion of males), duration of treatment with metformin monotherapy, relevant OCT1 polymorphisms, minor allelic frequencies in each population with Hardy Weinberg equilibrium if available, and primary outcome measurements (measure of metformin response after treatment with metformin). Disagreements were settled by consensus among the authors. The STREGA (Strengthening the Reporting of Genetic Association Studies) statement was used to assess the reporting quality of included studies.[16] Briefly, we assessed the reporting quality of all included studies in accordance with the following criteria: title and abstract, study design, selection criteria and basic characteristics of study participants, duration of metformin treatment, genotyping methods and its reliability, statistical method, accuracy and the outcome data on association between gene variants and the metformin responses. As the metrics used for assessment of genetic effects of OCT1 polymorphisms and study designs in the metformin response were not sufficiently similar, a narrative synthesis of the findings from the included studies was provided. This study is based on published data; therefore, ethical approval is not a requirement. This systematic review protocol was registered with the International Prospective Register of Systematic Reviews (PROSPERO) on the December 7, 2017. Trial registration number: CRD42017079978.

3. Results

3.1. Study characteristics

Our initial search identified 4186 records: 1719 from PubMed/MEDLINE, 1907 from Embase, 546 from CINAHL, and 14 from

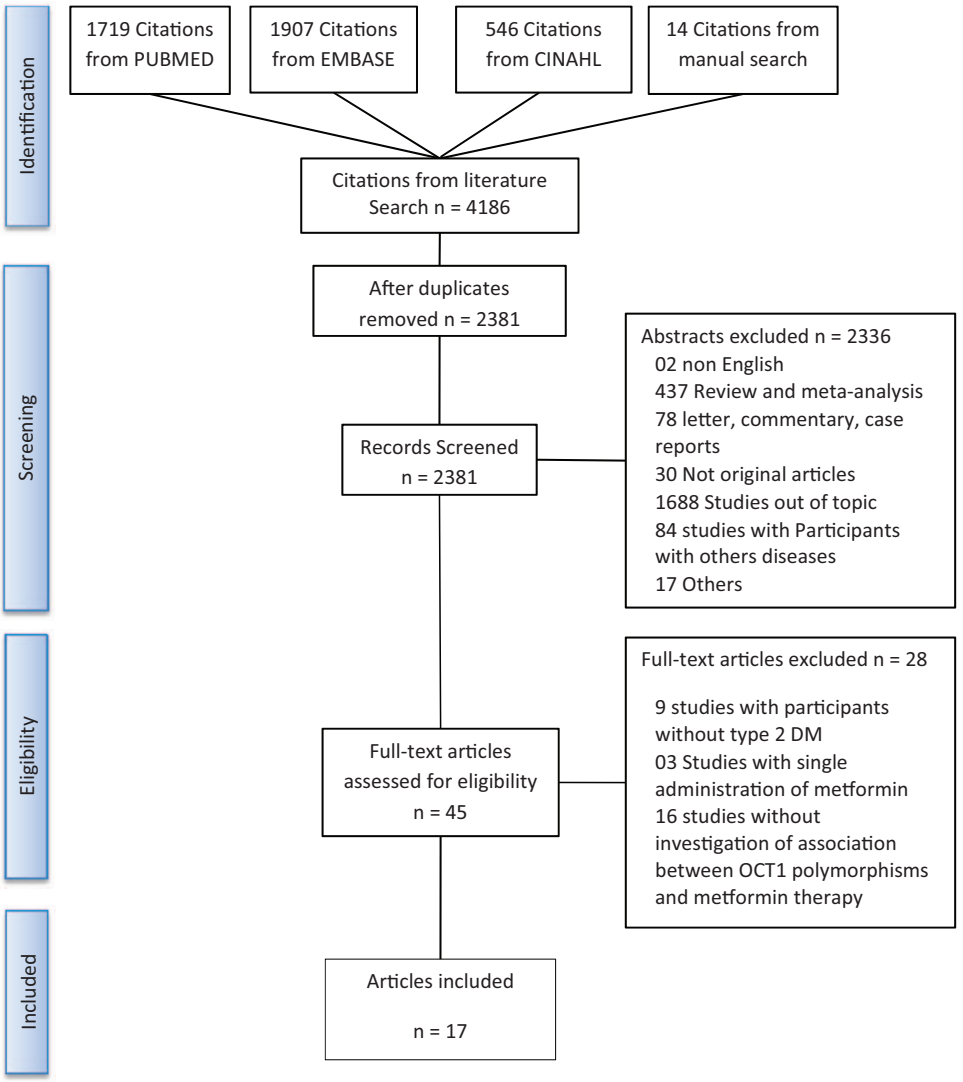

Figure 1. PRISMA flow diagram, describing the process of literature selection and reasons for exclusion. A total of 4186 records have been identified: 1719 from PubMed/MEDLINE, 1907 from Embase, 546 from CINAHL and 14 from manually searching the reference list of other articles. After screening of the titles and abstracts, 45 studies were found potentially eligible and their full texts were downloaded for further screenings. Of these 45 studies, we excluded 28 articles and the remaining 17 articles were included in the review.

manually searching the reference list of other articles. After screening of the titles and abstracts, 45 studies were found potentially eligible and their full texts were downloaded for further screenings. Of these 45 studies, we excluded 28 articles and the remaining 17 articles were included in the review (Fig. 1).

The majority of included studies were concentrated in Asia (47%), followed by Europe (29.4%). There were no study reported for the African continent. Of these eligible studies, 3 were conducted for each of the Caucasian and Indian population, 2 were conducted for each of the Chinese, Japanese and Scottish populations, while only 1 study was conducted for each of the Latvian, Danish, and Iranian populations. In 2 studies, the population was not specified. Most studies, but not all, reported sufficient details about selection criteria and basic characteristics for participants. The number of participants ranged from 33 to 2216, the proportion of men from 13.6% to 62.7% and the age of participants was above 29 years. Study participants were diagnosed with T2DM and treated with metformin monotherapy for at least 3 months and here the diagnosis was based on World Health Organization (WHO) criteria, HbA1c levels, FBG, and PPG. The response to metformin in individuals diagnosed with T2DM was categorized as responders and nonresponders, respectively. Nonresponders constituted patients whose HbA1c levels declined by <1% after 3 months treatment or who experienced gastrointestinal side effects, while responders were cases where HbA1c levels decreased by more than 1%. Measures of association were used to assess the effects of OCT1 polymorphisms in terms of the metformin response and included: differences in HbA1c, FPG, PPG levels among the various genotyping groups before and after metformin therapy, and odds ratios that were often adjusted for age, gender, and use of co-medications.

3.2. Genetic effects of OCT1 polymorphisms

Over 34 polymorphisms were investigated in 10 different populations with a minimum of 1 and a maximum of 12 per study. Significant differences in the frequencies of common variants have been observed among ethnic groups (Table 1).

3.2.1. rs628031 (1222A>G). The polymorphism *rs628031 (1222A>G)* was the most genotyped and its frequency has been found to range from 15% to 80% (median, 36%). Japanese population has the highest frequency (80%) while; *rs628031* is present only in 40% in Caucasians.[17,18] Association of *rs628031* with the glycemic response to metformin was assessed in 7 studies.[17–23] Here a study investigating *rs628031* in 277 Han Chinese participants found a significant reduction in HbA1c levels ($P<.02$) in individuals carrying the A/A genotype compared to those with the heterozygous genotype (A/G).[20] However, the A allele of A/A genotype was significantly associated with metformin side effects in a study conducted with 246 Latvian participants ($P=.02$).[22] By contrast, investigation of *rs628031* in Iranian,[19] Indian,[21] Caucasian,[18] and Japanese[17,23] populations showed that there were no association between this polymorphism and metformin responses.

3.2.2. rs122083571 (181C>T) and rs72552763 (1258_1260delATG). Polymorphisms *rs122083571 (181 C>T)* and *rs72552763 (1258_1260del ATG)* were assessed in 6 studies[13,22,24–28] and their frequencies varying from 67% to 89% and 18% to 28%, respectively. In 1 study with 92 participants, from unspecified population, the *rs122083571* and *rs72552763* polymorphisms were significantly associated with gastrointestinal side effects (OR=2.31, 95% CI [1.07–5.01], $P=.034$).[24] In addition, the investigation of *rs122083571* and *rs72552763* in 2216 participants from the GoDART Study found that such individuals (and receiving treatment with known OCT1 inhibitors) were over 4 times more likely to develop intolerance to metformin (OR=4.13, 95% CI 2.09–8.16, $P<.001$).[25] However, there was no association for such polymorphisms and metformin responses in Latvian and Danish populations.[22,27]

3.2.3. rs622342 (1386A>C). The association of *rs622342 (1386A>C)* with metformin responses was investigated in 5 studies that involved 3 populations (South Indian, Danish, and Caucasian)[18,27,29–31] with the frequency ranging from 5% to 37%. A study conducted in a Caucasian population found a positive significant association between *rs622342* and HbA1c levels ($P=.005$). This translated to an average of 0.28% lower decrease in HbA1c levels for each minor C allele.[31] By contrast, the major allele A of *rs622342* displayed a 5.6× greater chance of responding to metformin treatment in a South Indian population.[29] There was no significant relationship with HbA1c levels in a Danish population and also for 2 other studies involving a Caucasian population.[27,30]

3.2.4. rs2297374 (+43C>T). The polymorphism *rs2297374 (+43C>T)* and metformin response showed no significant association in Indian populations.[21] However, Han Chinese Shanghai individuals with the *rs 2297374 C/T* genotype exhibited significantly greater reductions in their FPG ($P=.002$) and HbA1c ($P=.039$) levels following metformin treatment versus homozygous *rs2297374* genotypes (C/C) and (T/T).[20] The frequency of *rs2297374* in these populations is close to 40%.

3.2.5. rs4646272 (−43T>G). The *rs4646272 (-43T>G)* polymorphism was assessed in 3 studies[17,21,31] with a frequency ranging from 20% to 67%. Here the one conducted with 66 Japanese subjects showed that *rs4646272* is a negative predictor of metformin efficacy.[17] However, the other studies showed no association.

3.2.6. rs34130495 (17857 G>A). Two studies assessed the association between *rs34130495 (17857 G>A)* and metformin responses.[25,27] Here a study investigating *rs34130495* in 371 Danish subjects found a significant association with absolute decreases in Hb1Ac levels.[27]

3.2.7. rs2282143 (1022C>T). *rs2282143 (1022C>T)* has been assessed in Indian population with a frequency of 20%. It has been demonstrated to impair OCT1 function in this population.[21]

3.2.8. rs1867351 (156T>C). The effect of *rs1867351 (156T>C)* on the glycemic response to metformin was investigated in Han Chinese and Indian populations with a frequency of 50% and 27%, respectively. No significant effect of the minor C allele has been shown while, the T/T genotype of *rs1867351* exhibited a greater reduction in PPG and HbA1c levels ($P=.020$) in Han Chinese population.[20]

3.2.9. rs594709 (597 A>G). The *rs594709 (597 A>G)* polymorphism was associated with the metformin treatment response in the Chinese population. Here GG genotype displayed a higher increase in FINS ($P=.015$) and a greater decrease in HOMA-IS ($P=.001$) and QUICKI ($P=.002$) than A allele carriers.[32]

3.2.10. rs200684404 (350C>T), 289C>A, and 616C>T. *rs200684404 (350C>T), 289C>A* and *616C>T* polymorphisms were investigated in the Japanese population and revealed a significant reduction in metformin uptake.[17]

3.2.11. Others polymorphisms. The association of other OCT1 polymorphisms, including *rs34104736*, *rs2297373*, *rs622591*, *rs2197296*, *rs4709400*, *rs461473*, *rs1443844*, *rs9457843,* and *rs6937722* was also investigated. However, no significant relationships were found between such variants and the metformin response.[18,23]

4. Discussion

Metformin is the most widely used first-line pharmacotherapy for T2DM. However, the interindividual variations in metformin efficacy ranging from improvement in HbA1c levels (by up to 4%) to a worse outcome with, estimates of ∼35% failure rate with treatment clearly suggest that this treatment modality is affected by individual genetic imprints.[33–35] Although several studies have now identified a plethora of OCT1 genetic variants that underlie such interindividual differences,[36,37] no systematic review has thus far been conducted (as far as we are aware) to assess its impact. Most studies investigating the effects of OCT1 polymorphisms in the context of therapeutic responses to metformin for T2DM, essentially focused on European, Asian and Caucasian populations without consideration for other population group. Of note, Seitz et al[37] performed a global scale population analysis of OCT1 variants and identified 85 variants in 52 worldwide population groups that included sub-Saharan Africa, the Middle East and North Africa, Central Asia, East Asia and Oceania, Europe, and America. Although OCT1 polymorphisms have also been identified in Africans Americans and African populations such as the Xhosas (South Africa), Luhyas

Table 1

Characteristics of genetic association studies conducted.

Population	Reference	Study design	Total sample (Case/Control)	Age (Mean/ range), years	%Men	Duration of treatment	Polymorphism	MAF: (case/control) or population group		(*P*)* with HWE (i.a)	Reported outcome
Chinese	Di Xiao et al[32]	Case control	449 (53/214/182)	29–73	51	3 months	*rs 594709*	0.286	0.268	*P*=.555	Minor allele G is associated with higher increase of FINS (*P*=.015), decrease in HOMA-IS (*P*=.001) and QUICKI (*P*=.002).
Iranian	Shokri et al[19]	Cross-sectional	140 (77/63)	35–71	13.57	6 months	*rs628031*	0.317	0.331	–	No significant effect in metformin response (OR=0.45, [95% CI 0.64–1.76], *P*=.45)
Not specify	Dujic et al [24]	Prospective observational cohort	92 (43/49)	51.48±8.91	–	6 months	*rs12208357*		–	–	Significant association with common metformin induced gastrointestinal side effect. *P*=.034
							rs72552763		–	–	
GoDARTs Scotland	Dujic et al[25]	Observational cohort	2216 (251/1915)	57.80±10.52	56.72	6 months	*rs12208357* *rs 55918055* *rs34130495* *rs72552763* *rs34059508*		–	–	Significant associated between intolerance to metformin and the presence of 2 reduced-function allele (*P*<.001)
South Indian	Umamaheswaran et al[29]	Observational cohort	122 (29/93)	31–60	39	3 months	*rs 622342*	0.156	0.362	–	Association of A allele with therapeutic efficacy of metformin and beneficial effects on HbA1c, FPG and PPG.
Han Chinese	Zhou et al[20]	Single-center prospective cohort	277 (153/124)	45–68	–	3 months	*rs1867351*	0.62	0.50	*P*=.44 *P*=.53	Association of T/T genotype with greater reductions in the HbA1c levels (*P*=.02).
							rs4709400	0.532	0.70	*P*=.45 *P*=.88	Association of G/G genotype with greater reductions in FPG levels (*P*=.046)
							rs628031	0.262	0.10	*P*=.49 P<0.01	Association of G/G genotype with greater reductions in FPG level (*P*<.01), and A/A genotype exhibited significantly greater reductions in HbA1c level (*P*<.02)
							rs2297374	0.343	0.40	*P*=.53 *P*=.43	Association of C/T genotype with greater reductions in FPG (*P*=.002) and HbA1c (*P*=.039) level.
Not specified	Maahroz et al[13]	Cross-sectional	108 (59/49)	43–63	19.4	3 months	*rs 72552763*	0.188	0.288	*P*=.088	No significant change in the ability of metformin to lower HbA1c (OR: 0.57, [95% CI 0.298–1.09], *P*=.088). More effective in reducing FPG.
Indian	D Sur et al[21]	Cross-sectional	50	46±12.72	48	–	*rs 683369*		0.63	*P*=.0002	Unchanged function of OCT1
							rs 2282143		0.20	*P*=.008	Probably damaging function of OCT1
							rs628031		0.43	–	Unchanged function of OCT1
							rs1867351		0.27	*P*=.091	Not provided
							rs4646272		0.2	–	Not provided
							rs3737088		0.17	–	Not provided
							rs2282142		0.18	–	Not provided
							rs2297374		0.37	–	Not provided
South Indian Tamalian	Koshy et al[26]	Cross-sectional	60	35–55	–	–	*rs1220833571*		0.89	–	C allele is associated with poor glycemic control and significantly higher FPG, insulin and blood metformin level ($p < 0.05$)

(*continued*)

Table 1

(continued).

Population	Reference	Study design	Total sample (Case/Control)	Age (Mean/ range), years	%Men	Duration of treatment	Polymorphism	MAF: (case/control) or population group	(*P*)* with HWE (i.a)	Reported outcome
Caucasian	Tkáč et al[30]	Cross-sectional	148	57.5±0.9	49	6 months	*rs622342*	0.05	*P*=.95	No significant relationship with HbA1c (*P*=.95)
Latvian	Tarasova et al[22]	Case control	246 (53/193)	50–69	30.1	≥3 months	*rs628031*	0.39	*P*=.785	Significant association with gastrointestinal side effects of metformin (OR=0.389, [95% CI 0.186–0.815], *P*=.012)
							rs 34059508	0.04	P=1	Not provided
							rs 72552763	0.18	P=1	Not provided
							rs 36056065	0.39	*P*=.686	Significant association with side effects of metformin (OR=0.405, [95% CI 0.226–0.724], *P*=.002).
Danish	Christensen et al[27]	Randomized controlled trial	371	52–62	62.7	9 months	*rs 122083571*	0.082	–	No significant effect in metformin response
							rs 34104736	0.0	–	No significant effect in metformin response
							rs34130495	0.044	–	Significant association with absolute decrease in Hb1Ac
							rs72552763	0.175	–	No significant effect in metformin response
							rs 461473	0.114	–	Significant association with initial decrease in Hb1Ac
							rs 622342	0.38	–	No significant effect in metformin response
Caucasian	Becker et al[31]	Prospective cohort	98	70–83	39	–	*rs 622342*	0.37	*P*=.40	A allele associated with decrease of Hb1Ac
Japanese	Chen et al[17]	Cross-sectional	66	46–71	48.5	–	*rs4646272*	0.34		Negative predictor of efficacy of metformin
							rs 200684404	0.023	–	Significant association with reduced uptake of metformin
							616C>T	0.008	–	Significant association with reduced uptake of metformin
							rs 628031	0.15	–	No significant effect in metformin response
							289C> A	0.017	–	Significant Association with reduced uptake of metformin
Scotland	Zhou et al[28]	Cross-sectional	1531	46–68	56.14	At least 6 months	*rs 122083571*	0.67	*P*>.05	No significant reduction in the ability of metformin to lower HbA1c (*P*=.47)
							rs 72552763	0.198	*P*>.05	No significant reduction in the ability of metformin to lower HbA1c (*P*=.92)
Caucasian	Becker et al[18]	Cross-sectional	102	70–83	39	–	*rs3798174*	0.05	*P*=.56	No significant effect in metformin response (*P*=.49)
							rs6937722	0.06	*P*=.49	No significant effect in metformin response (*P*=.40)
							rs3798168	0.02	*P*=.80	No significant effect in metformin response
							rs628031	0.40	*P*=.39	No significant effect in metformin response (*P*=.47)
							rs9457843	0.16	*P*=.63	No significant effect in metformin response (*P*=.40)
							rs3798167	0.19	*P*=.31	No significant effect in metformin response (*P*=.20)

(*continued*)

Table 1
(continued).

Population	Reference	Study design	Total sample (Case/Control)	Age (Mean/ range), years	%Men	Duration of treatment	Polymorphism	MAF: (case/control) or population group	(*P*)* with HWE (i.a)	Reported outcome
							rs2197296	0.26	*P*=.11	No significant effect in metformin response (*P*=.61)
							rs622342	0.37	*P*=.72	Association with glucose-lowering effect of metformin (*P*<.05)
							rs1443844	0.45	*P*=.60	No significant effect in metformin response (*P*=.18)
							rs2297374	0.41	*P*=.99	No significant effect in metformin response (*P*=.15)
							rs1564348	0.17	*P*=.55	No significant effect in metformin response (*P*=.71)
							rs622591	0.18	*P*=.81	No significant effect in metformin response (*P*=.19)
Japanese	Shikata et al[23]	Cross-sectional	33 (9/24)	29–73	27.3	>1 month	*rs2297373*	0.00 0.02	–	Not provided
							rs 200684404	0.06 0.00	–	Not provided
							rs683369	0.17 0.13	–	Not provided
							rs2282143	0.11 0.19	–	Not provided
							rs628031	0.72 0.81	–	Positive predictor for metformin efficacy
							rs1867351	0.56 0.42	–	Not provided
							243C>T	0.06 0.00	–	Not provided
							rs4646272	0.67 0.58	–	Negative predictor for metformin efficacy
							+26C>T	0.00 0.02	–	Not provided
							rs622591	0.61 0.54	–	Not provided

Reported outcome: genetic effect of polymorphism in the study population
FINS = fasting insulin, FPG = fasting plasma glucose, HWE (i.a) = Hardy Weinberg Equilibrium if applicable, HOMA IS = homeostatic model assessment of insulin sensitivity, MAF = minor allele frequency, PPG = postprandrial plasma glucose, QUICKI = quantitative insulin sensitivity check index.
(*P*)*: *P* value

(Kenya), and the Yorubas (Nigeria), their effects on metformin responses remain unknown.[38,39] In light of this, there is a robust need to complete association studies between OCT1 polymorphisms and therapeutic responses to metformin, by evaluating a comprehensive and representative dataset.

The common polymorphism *rs628031 (A>G)* causes a missense mutation in exon 7 that consists of an amino acid substitution of methionine to valine at position 408 *(Met408Val)* in the OCT1 protein.[18] Its frequency varies across ethnic population. *Met408Val* tends to lower OCT1 mRNA expression in enterocytes leading to decreased intestinal metformin uptake and hence its accumulation.[40] Here the local increase of metformin concentrations within intestinal tissues is proposed as a putative mechanism for gastrointestinal side effects.[41] This deleterious effect of *Met408Val* has been reported in Latvian population.[25] However, *Met408Val* has been also characterized as a variant that lacks strong effects (did not cause >50% decrease in OCT1 activity).[42,43] In agreement with this, no significant effects of *Met408Val* against metformin response have been found in Iranian,[19] Indian,[21] Caucasian,[18] and Japanese[17,23] populations. Surprisingly, G/G and A/A genotypes of *Met408Val* exhibited significant reduction of FPG and HbA1c in Han Chinese,[20] and have been revealed to be a positive predictor for metformin efficacy in Japanese population.[23]

Numerous studies reported the deleterious effect of *rs122083571* polymorphism.[24–26] The *181C>T* polymorphism at *rs122083571* consisting of an amino acid substitution (arginine to cysteine at position 61 *(Arg61Cys)*, is known to induce a robust substrate-wide loss of OCT1 activity, leading to decrease in OCT1-mediated uptake by more than 70% for all substrates tested (including metformin). Indeed, the *rs122083571* polymorphism is responsible for the retention of OCT1 proteins in the endoplasmic reticulum thus leading decreased sarcolemma protein expression.[37]

The *rs72552763* polymorphism constitutes a 3 bp deletion at position 420 *(Met420del)* and is the most common functional OCT1 variant. *Met420del* does not change OCT1 membrane localization and the exact mechanism how it affects OCT1 function remains unknown.[44] Although it is associated with gastrointestinal side effects in Asian and Caucasian populations, no significant effects were reported for European populations.[22,25,27] Functional modifications by *Met420del* appear substrate dependent and in combination with other OCT1 variants. For example, *Met420del* does not affect the uptake of MPP^+ (1-methyl-4-phenyl pyridinium). By contrast, it causes a robust decrease in metformin uptake (>60%) together with more than 80% reduction in tropisetron uptake.[37] If the *Met420del* manifests in combination with *Cys88Arg* or *Gly465Arg*, the encoded OCT1 will be inactive regardless of the substrate used.[45,46]

The *rs622342 (1386 A>C)* variant (in intron between exons 8 and 9) does not elicit strong effects on OCT1 function.[18] The A allele of *rs622342* variant has rather been associated beneficial effects on HbA1c in Caucasian population.[31] The study performed by Umamaheswaran et al[29] strengthened this positive effect of A allele on HbA1c in Indian population.

The *32870 G>A* polymorphism at *rs34059508* consisting of the amino acid substitution glycine to arginine at codon 465 *(Gly465Arg)*, leads to the impairment of OCT1 localization and complete inactivation of OCT1. The exact mechanisms leading to this impairment remain unclear.[44] Dujic et al[25] observed metformin intolerance in carriers of both *rs34059508* and another OCT1 reduced function.

The amino acid substitution of glycine to serine at codon 401 *(Gly401Ser)*, resulting from the *17857 G>A* polymorphism at *rs34130495*, causes a strong substrate-independent loss of OCT1 activity. The *Gly401Ser* variant apparently causes a general impairment of the transport process without affecting OCT1 membrane localization.[37] Surprisingly, in a study conducted in a Danish population, *Gly401Ser* was associated with a significant, absolute decrease in HbA1c levels.[27]

Cys88Arg (rs55918055) in exon 1 is a rare loss of function polymorphism that causes improper membrane localization of OCT1 in the cytoplasmic membrane. It has been associated with intolerance to metformin in a Scottish population.[25]*Cys88Arg* is located in the large extracellular loop that contains the transporter regulatory and substrate recognition domains. It is generally observed in combination with the *Met420del* variant. Substitution of Cysteine 88 by arginine destroys a cysteine residue known to build disulfide bonds. Indeed, several cysteine residues within the large extracellular loop between transmembrane helices 1 and 2 are involved in building intramolecular disulfide bonds essential for the oligomerization and targeting of OCT1 to the plasma membrane.[45–47]

The *1022C>T* polymorphism at *rs2282143* consisting of the amino acid substitution proline to leucine at codon 341. Pro341Leu, can either elicit no effects or instead decrease OCT1 activity (<50%).[37] The replacement of a rigid proline with leucine (which contains a relatively flexible side chain) could possibly change the local structure of OCT1.[24] This is in agreement with findings of the Indian population in which *rs2282143* has been reported to probably damage OCT1 function.[21]

The true effect of the intronic variant *rs4646272 (-43T>G)* and *rs2297374 (+43C>T)* remains unknown. *rs4646272 (-43T>G)* was considered as negative predictor of metformin efficacy in a Japanese population[17] while *rs4646272 (-43T>G)* exhibited a greater reduction of FPG and HbA1c in Han Chinese population.[20]

Other rare variants known to cause OCT1 loss of function, that is, *Pro117Leu (rs200684404)*, *Gln97Lys (289C>A)*, *Arg206Cys (616C>T)*, and *Ser189Leu (rs34104736)*, were also reported and associated with significantly lowered metformin uptake.[17,21,23,27] For Gln97Lys, the replacement of the polar glutamate with a stronger positively charged lysine at 97 could increase the repulsion of cationic substrates. By contrast, the reduced activity of the *Arg206Cys* can be explained by decreased export of the OCT1 from the endoplasmic reticulum to the plasma membrane.[26]

This systematic review demonstrated that the potential role of OCT1 polymorphisms in metformin therapeutic responses is population specific and some of them exhibited positive effects on metformin efficacy. The controversial findings related to these polymorphisms may be attributable to differences in the frequency of associated genetic variants and/or population differences that could be genetic or environnemental.[48] Indeed, environmental factors like chemicals and radiation exposure, lifestyle factors like diet, drinking, smoking, exercises, and physiological factors like age, sex can also work alone or in combination to influence drug responses.[49,50] Advancing age for example is characterized by physiological changes affecting different organ systems and their implications for pharmacokinetics and pharmacodynamics of drugs. Although very few data exist in literature about the effect of biguanides particularly metformin in aging patients, no evidence indicated that metformin should be denied "a priori" to aging T2DM patients.[51,52] However, age-dependent downregulation of OCT1 has been shown in mice brain microvessels of mice.[53]

Moreover, the way a person responds to a drug (this includes both positive and negative reactions) is a complex trait that is influenced by many different genes. Knowing all of the genes involved in drug response and understanding an individual's genetic make-up, can help to develop genetic tests that could predict a person's response to a particular drug and create personalized drugs with greater efficacy and safety.[54,55]

All studies included in this systematic review were rigorously assessed for their reporting quality—a key criterion for inclusion for analyses here completed. Different study designs have been used in included studies but, most of them had a cross-sectional design. As such, a causal relationship between genetic variants of OCT1 and responses to metformin cannot be inferred from these studies. For instance, confounding variables such as duration of treatment, metformin dosing might affect metformin responses leading to inconsistent results among studies. In terms of limitations, our investigation was conducted on a relatively small number of primary studies and more research work should help to obtain a wider range of relevant data to generate more conclusive findings. In addition, all studies surveyed did not always report key methodological information, for example, testing of the Hardy Weinberg Equilibrium and the sample size/power calculation. Some studies also reported a relatively small sample size meaning that the study population may not cover the entire spectrum of OCT1 variants. Thus larger sample sizes will enable a firmer correlation between OCT1 genetic variants and metformin responses. For example, we found that 2 studies included more than 1500 participants, 4 studies more than 200 participants, while the rest included less than 200 participants. Discordance of the type of study population and methods used for assessing the genetic effects of OCT1 variants may all have contributed to less powerful conclusions being derived by the current study. Finally, we were not able to pool data collected for a meta-analysis due to the methodological heterogeneity observed.

Great efforts are made to understand the effects of OCT1 genetic polymorphisms on interindividual variability in relation to metformin's clinical efficacy. However, some questions remain unanswered, e.g. the relationship between OCT1 variants and lactic acidosis. This is a crucial issue as lactic acidosis is a rare but potentially fatal metabolic consequence of metformin therapy.[56] For example, metformin-induced lactic acidosis is associated with an elevation in plasma metformin concentrations in patients with severe renal impairments and is considered as a contraindication of this drug. However, such adverse effects also occur in patients without well-known risk factors.[57] Thus a hypothesis emerges that OCT1 polymorphisms that decrease metformin uptake and cause its accumulation in circulation may induce lactic acidosis. However, further studies are required to investigate this intriguing notion.

In summary, this systematic review focused on the genetic effects of OCT1 polymorphisms on metformin treatment in T2DM patients. Our study shows evidence for a contribution of some OCT1 polymorphisms to variability in response to metformin with T2DM. Thus such associations remain unresolved and we suggest that further association studies be completed on defined populations with relatively large sample sizes as this should reveal significant insights into this vital clinical issue.

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All authors had full access to the data

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References

[1] Inzucchi SE, Bergenstal RM, Buse JB, et al. Management of hyperglycemia in type 2 diabetes: a patient-centered approach. Diabetes Care 2012;35:1364–79.

[2] Graham GG, Punt J, Arora M, et al. Clinical pharmacokinetics of metformin. Clin Pharmacokinet 2011;50:81–98.

[3] Foretz M, Guigas B, Bertrand L, et al. Metformin: from mechanisms of action to therapies. Cell Metab 2014;20:953–66.

[4] Nyane NA, Tlaila TB, Malefane TG, et al. Metformin-like antidiabetic, cardio-protective and non-glycemic effects of naringenin: molecular and pharmacological insights. Eur J Pharmacol 2017;803:103–11.

[5] UK Prospective Diabetes Study (UKPDS) GroupEffect of intensive blood glucose control with metformin on complications in overweight patients with type 2 diabetes (UKPDS 34). Lancet 1998;352:854–65.

[6] Pawlyk AC, Giacomini KM, McKeon C, et al. Metformin pharmacogenomics: current status and future directions. Diabetes 2014;63:2590–9.

[7] Foretz M, Hébrard S, Leclerc J, et al. Metformin inhibits hepatic gluconeogenesis in mice independently of the LKB1/AMPK pathway via a decrease in hepatic energy state. J Clin Invest 2010;120:2355–69.

[8] Hur KY, Lee MS. New mechanisms of metformin action: focusing on mitochondria and the gut. J Diabetes Investig 2015;6:600–9.

[9] Kimura N, Masuda S, Tanihara Y, et al. Metformin is a superior substrate for renal organic cation transporter OCT2 rather than hepatic OCT1. Drug Metab Pharmacokinet 2005;20:379–86.

[10] Koehler MR, Wissinger B, Gorboulev V, et al. The two human organic cation transporter genes SLC22A1 and SLC22A2 are located on chromosome 6q26. Cytogenet Cell Genet 1997;79:189–200.

[11] Jonker JW, Wagenaar E, Van Eijl S, et al. Deficiency in the organic cation transporters 1 and 2 (Oct1 and Oct2 [Slc22a1 and Slc22a2]) in mice abolishes the renal secretion of organic cations. Mol Cell Biol 2003;23:7902–8.

[12] Wang D-S, Jonker JW, Kato Y, et al. Involvement of organic cation transporter 1 in hepatic and intestinal distribution of metformin. J Pharmacol Exp Ther 2002;302:510–5.

[13] Mahrooz A, Parsanasab H, Hashemi-Soteh MB, et al. The role of clinical response to metformin in patients newly diagnosed with type 2 diabetes: a monotherapy study. Clin Exp Med 2015;15:159–65.

[14] Sundelin EIO, Gormsen LC, Jensen JB, et al. Genetic polymorphisms in organic cation transporter 1 attenuates hepatic metformin exposure in humans. Clin Pharmacol Ther 2017;102:841–8.

[15] Gong L, Goswami S, Giacomini KM, et al. Metformin pathways: pharmacokinetics and pharmacodynamics. Pharmacogenet Genomics 2012;22:820–7.

[16] Moher D, Liberati A, Tetzlaff J, et al. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. PLoS Med 2009;6:e1000097.

[17] Chen L, Takizawa M, Chen E, et al. Genetic polymorphisms in organic cation transporter 1 (OCT1) in Chinese and Japanese populations exhibit altered function. J Pharmacol Exp Ther 2010;335:42–50.

[18] Becker ML, Visser LE, van Schaik RH, et al. Genetic variation in the organic cation transporter 1 is associated with metformin response in patients with diabetes mellitus. Pharmacogenomics J 2009;9:242–7.

[19] Shokri F, Ghaedi H, Fard SG, et al. Impact of ATM and SLC22A1 polymorphisms on therapeutic response to metformin in Iranian diabetic patients. Int J Mol Cell Med 2016;5:1–7.

[20] Zhou Y, Ye W, Wang Y, et al. Genetic variants of OCT1 influence glycemic response to metformin in Han Chinese patients with type-2 diabetes mellitus in Shanghai. Int J Clin Exp Pathol 2015;8:9533–42.
[21] Dipanshu S. A Tale of Genetic Variation in the Human Slc22a1 Gene Encoding Oct1 Among Type 2 Diabetes Mellitus Population Groups of West Bengal, India. IMPACT Int J Res Applied, Nat Soc Sci 2014;2:97–106.
[22] Tarasova L, Kalnina I, Geldnere K, et al. Association of genetic variation in the organic cation transporters OCT1, OCT2 and multidrug and toxin extrusion 1 transporter protein genes with the gastrointestinal side effects and lower BMI in metformin-treated type 2 diabetes patients. Pharmacogenet Genomics 2012;22:659–966.
[23] Shikata E, Yamamoto R, Takane H, et al. Human organic cation transporter (OCT1 and OCT2) gene polymorphisms and therapeutic effects of metformin. J Hum Genet 2007;52:117–22.
[24] Dujic T, Causevic A, Bego T, et al. Organic cation transporter 1 variants and gastrointestinal side effects of metformin in patients with Type 2 diabetes. Diabet Med 2016;33:511–4.
[25] Dujic T, Zhou K, Donnelly LA, et al. Association of organic cation transporter 1 with intolerance to metformin in type 2 diabetes: a GoDARTS study. Diabetes 2015;64:1786–93.
[26] Koshy M, Sethupathy S, Annamalai PT, et al. Association of oct1 gene polymorphism with glycemic status and serum metformin levels in type ii diabetes mellitus patients. Int J Phar Sci Res 2013;4:1940–5.
[27] Christensen MMH, Højlund K, Hother-Nielsen , et al. Steady-state pharmacokinetics of metformin is independent of the OCT1 genotype in healthy volunteers. Eur J Clin Pharmacol 2015;71:691–7.
[28] Zhou K, Donnelly L, Kimber CH, et al. Organic cation transporter 1 and glycemic response to metformin: a GoDARTS study. Diabetes 2009; 58:1434–9.
[29] Umamaheswaran G, Praveen RG, Damodaran SE, et al. Influence of SLC22A1 rs622342 genetic polymorphism on metformin response in South Indian type 2 diabetes mellitus patients. Clin Exp Med 2015;15:511–7.
[30] Tkáč I, Klimčáková L, Javorský M, et al. Pharmacogenomic association between a variant in SLC47A1 gene and therapeutic response to metformin in type 2 diabetes. Diabetes, Obes Metab 2013;15:189–91.
[31] Becker ML, Visser LE, Van Schaik RH, et al. Interaction between polymorphisms in the OCT1 and MATE1 transporter and metformin response. Pharmacogenet Genomics 2010;20:38–44.
[32] Xiao D, Guo Y, Li X, et al. The impacts of SLC22A1 rs594709 and SLC47A1 rs2289669 polymorphisms on metformin therapeutic efficacy in Chinese type 2 diabetes patients. Int J Endocrinol 2016;2016: 4350712.
[33] Cook MN, Girman CJ, Stein PP, et al. Initial monotherapy with either metformin or sulphonylureas often fails to achieve or maintain current glycaemic goals in patients with Type 2 diabetes in UK primary care. Diabet Med 2007;24:350–8.
[34] Florez JC. Does metformin work for everyone? A genome-wide association study for metformin response. Curr Diab Rep 2011; 11:467–9.
[35] Van Leeuwen N, Swen JJ, Guchelaar HJ, et al. The role of pharmacogenetics in drug disposition and response of oral glucose-lowering drugs. Clin Pharmacokinet 2013;52:833–54.
[36] Dujic T, Zhou K, Yee SW, et al. Variants in pharmacokinetic transporters and glycemic response to metformin: a metgen meta-analysis. Clin Pharmacol Ther 2017;101:763–72.
[37] Seitz T, Stalmann R, Dalila N, et al. Global genetic analyses reveal strong inter-ethnic variability in the loss of activity of the organic cation transporter OCT1. Genome Med 2015;7:1–23.
[38] Shu Y, Leabman MK, Feng B, et al. Evolutionary conservation predicts function of variants of the human organic cation transporter, OCT1. Proc Natl Acad Sci U S A 2003;100:5902–7.
[39] Du Plessis M, Pearce B, Jacobs C, et al. Genetic polymorphisms of the organic cation transporter 1 gene (SLC22A1) within the Cape Admixed population of South Africa. Mol Biol Rep 2015;42:665–72.
[40] Semiz S, Dujic T, Causevic A. Pharmacogenetics and personalized treatment of type 2 diabetes. Biochem Medica 2013;23:154–71.
[41] Wang DS, Kusuhara H, Kato Y, et al. Involvement of organic cation transporter 1 in the lactic acidosis caused by metformin. Mol Pharmacol 2003;63:844–8.
[42] Sakata T, Anzai N, Shin HJ, et al. Novel single nucleotide polymorphisms of organic cation transporter 1 (SLC22A1) affecting transport functions. Biochem Biophys Res Commun 2004;313:789–93.
[43] Shu Y, Sheardown SA, Brown C, et al. Effect of genetic variation in the organic cation transporter 1 (OCT1) on metformin action. J Clin Invest 2007;117:1422–31.
[44] Koepsell H, Lips K, Volk C. Polyspecific organic cation transporters: structure, function, physiological roles, and biopharmaceutical implications. Pharm Res 2007;24:1227–51.
[45] Tzvetkov MV, Saadatmand AR, Lötsch J, et al. Genetically polymorphic OCT1: another piece in the puzzle of the variable pharmacokinetics and pharmacodynamics of the opioidergic drug tramadol. Clin Pharmacol Ther 2011;90:143–50.
[46] Arimany-Nardi C, Koepsell H, Pastor-Anglada M. Role of SLC22A1 polymorphic variants in drug disposition, therapeutic responses, and drug–drug interactions. Pharmacogenomics J 2015;15:473–87.
[47] Jonker JW, Schinkel AH. Pharmacological and physiological functions of the polyspecific organic cation transporters: OCT1, 2, and 3 (SLC22A1-3). J Pharmacol Exp Ther 2004;308:2–9.
[48] Bachtiar M, Lee CGL. Genetics of population differences in drug response. Curr Genet Med Rep 2013;1:162–70.
[49] Meyer UA, Zanger UM, Schwab M. Omics and drug response. Annu Rev Pharmacol Toxicol 2013;53:475–502.
[50] Akhondzadeh S. Personalized medicine: a tailor made medicine. Avicenna J Med Biotechnol 2014;6:191.
[51] Gregorio F, Ambrosi F, Filipponi P, et al. Is metformin safe enough for ageing type 2 diabetic patients? Diabetes Metab 1996;22:43–50.
[52] Gregorio F, Manfrini S, Testa I, et al. Metformin treatment in elderly type II diabetic patients. Arch Gerontol Geriatr 1996;22:261–70.
[53] Wu KC, Lu YH, Peng YH, et al. Decreased expression of organic cation transporters, Oct1 and Oct2, in brain microvessels and its implication to MPTP-induced dopaminergic toxicity in aged mice. J Cereb Blood Flow Metab 2015;35:37–47.
[54] Fiers W, Contreras R, Duerinck F, et al. Complete nucleotide sequence of bacteriophage MS2 RNA: primary and secondary structure of the replicase gene. Nature 1976;260:500–7.
[55] Aneesh TP, Sonal SM, Asha J, et al. Pharmacogenomics: the right drug to the right person. J Clin Med Res 2009;1:191–4.
[56] Lalau JD, Race JM. Metformin and lactic acidosis in diabetic humans. Diabetes Obes Metab 2000;2:131–7.
[57] Nies AT, Koepsell H, Winter S, et al. Expression of organic cation transporters OCT1 (SLC22A1) and OCT3 (SLC22A3) is affected by genetic factors and cholestasis in human liver. Hepatology 2009;50:1227–40.