Eur J Clin Pharmacol (2005) 61: 743–747
DOI 10.1007/s00228-005-0989-3

PHARMACOGENETICS

Kyoung-Ah Kim · Sook-Young Lee · Pil-Whan Park
Jong-Myung Ha · Ji-Young Park

Genetic polymorphisms and linkage disequilibrium of sulfotransferase *SULT1A1* and *SULT1A2* in a Korean population: comparison of other ethnic groups

Received: 29 June 2005 / Accepted: 28 July 2005 / Published online: 26 August 2005

Abstract *Aims*: To determine the allele frequencies of sulfotransferases (SULTs) 1A1 and 1A2 and their linkage disequilibrium in a Korean population and compare them with those of other ethnic groups. *Methods*: Genotypes of the *SULT1A1*1, *2*, and **3* and *SULT1A2*1, *2*, and **3* allelic variants were determined in 234 Korean subjects using polymerase chain reaction (PCR)–restriction fragment length polymorphism (RFLP) methods. *Results*: Allele frequencies for *SULT1A1*1* and **2* were 0.876 [95% confidence interval (CI), 0.843–0.905] and 0.124 (95% CI, 0.096–0.157), respectively. Similarly, those for *SULT1A2*1* and **2* were 0.885 (95% CI, 0.852–0.912) and 0.115 (95% CI, 0.088–0.150), respectively. However, no subject with *SULT1A1*3* or *SULT1A2*3* was detected. These genotype distributions are similar to those of Asian populations including the Chinese and Japanese, but quite different from other ethnic groups such as African-Americans and Caucasians. The expected allelic frequencies of *SULT1A1* and *SULT1A2* at Hardy–Weinberg equilibrium are quite similar to the observed distributions in the population. *SULT1A1*2* and *SULT1A2*2*, the most common variant alleles of these two genes, are strongly and positively linked in the Korean population ($D' = 0.8919$, $\chi^2 = 343.24$, $P = 0.0034$). *Conclusions*: *SULT1A1*2* and *SULT1A2*2* are the major allelic variants in the Korean population, whereas the *SULT1A1*3* and *SULT1A2*3* alleles were not found. *SULT1A1*2* and *SULT1A2*2* are strongly linked.

Keywords Sulfotransferase · Genetic polymorphism · Linkage disequilibrium · *SULT1A1* · *SULT1A2*

Abbreviations *SULT*: Sulfotranseferase · *D'*: Constant for linkage disequilibrium

K.-A. Kim · S.-Y. Lee · J.-Y. Park (✉)
Department of Pharmacology, Gil Medical Center,
Gachon Medical School, 1198 Kuwol-dong, Namdong-gu,
Incheon, 405-760, Korea
E-mail: jypark@gachon.ac.kr
Tel.: +82-32-4602151
Fax: +82-32-4225105

P.-W. Park
Department of Laboratory Medicine, Gil Medical Center,
Gachon Medical School, 1198 Kuwol-dong, Namdong-gu,
Incheon, 405-760, Korea

J.-M. Ha
Department of Biotechnology and Bioscience,
Silla University, Busan, Korea

Introduction

Cytosolic sulfotransferases (SULTs) play an important role in the phase II metabolism of several xenobiotics. Sulfation can result in either inactivation or the metabolic activation of sulfate acceptor substrates [1, 2]. Functional polymorphisms of the SULT1As may influence individual susceptibility to adverse drug reactions involving drug substrates and to chemical carcinogenesis [3–6]. The human SULT1A subfamily contains several members, which share over 92% identity at the amino acid level [7–9]. SULT1A1 is an important enzyme in xenobiotic metabolism because it has broad substrate specificity with a high affinity for many compounds and it is expressed widely in the body, particularly in the liver, brain, jejunum, and platelets [10–12]. Importantly, *SULT1A1* is subject to a common functional polymorphism [13, 14], with the variant allele (*SULT1A1*2*, allele frequency 32% in Caucasians) encoding an allozyme with a single amino acid change (Arg213His) that is uniformly associated with low SULT activity and low thermal stability [15]. Another single amino acid change, Met223Val, in *SULT1A1*3* is also commonly detected in several ethnic groups, especially African-Americans [14, 15]. Like *SULT1A1*, *SULT1A2* is highly polymorphic, and several polymorphisms have been detected in humans [9, 14, 16]. *SULT1A2*2* differs from *SULT1A2*1* at two amino acid residues (Ile7Thr and Asn235Thr) [15]. The change at codon 235 appears to be

particularly important functionally. Threonine in that position drastically reduces the affinity for the substrate 4-nitrophenol [15–17].

It has been reported that the gene *SULT1A2* is usually in linkage disequilibrium with *SULT1A1* and that the high-activity alloenzymes (*SULT1A1*1* and *SULT1A2*1*) are associated in several ethnic groups [18, 19]. Moreover, the genes for the low-affinity alloenzymes (*SULT1A1*2* and *SULT1A2*2*) are also associated.

In this study, we determined the allele frequencies of *SULT1A1* and *SULT1A2* using polymerase chain reaction (PCR)-based restriction fragment length polymorphism (RFLP) assays designed to detect common variant alleles of *SULT1A1* and *SULT1A2*. We then assessed the linkage disequilibrium between these two genes in a Korean population. Finally, we compared the results with those for other ethnic populations.

Materials and methods

Study population

After written informed consent was obtained from participants, blood samples were collected from 234 healthy Korean subjects. Genomic DNA was isolated from peripheral blood using the QIAamp Blood Kit (Qiagen, Chatsworth, CA, USA). All the samples were collected under conditions that had been reviewed and approved by the Institutional Review Board of Gil Medical Center, Gachon Medical School, Incheon, Korea.

Identification of *SULT1A1* and *SULT1A2* polymorphisms

The presence of alleles of both *SULT1A1*2* or *1A1*3* and *SULT1A2*2* or *1A2*3* was determined by PCR-RFLP analysis as previously described, with a slight modification [20]. *Hha*I, *Hsp*92II, *Bst*EII, and *Bst*U1 were used as restriction enzymes for *SULT1A1*2* or *SULT1A1*3* and *SULT1A2*2* or *SULT1A2*3*, respectively.

Statistical analysis

Statistical analysis was performed with SPSS version 12.0 (SPSS, IL, USA), using a chi-square (χ^2) test. Linkage analysis was performed with the Estimating Haplotype Frequency Test using the EH program (http://linkage.rockefeller.edu/soft/) described by Terwilliger and Ott [21] with the population-based sampling method. D' values, a standard measure of linkage disequilibrium, were calculated as D/D_{max}, where D is the coefficient of linkage disequilibrium, indicating the amount of disequilibrium ($-0.25 < D < 0.25$) [22]. Because D depends on allele frequencies, the standardized value D' is preferred [23]. D' values can vary from $+1$ when alleles at the two loci studied are maximally positively associated (that is, always occurring together in the same haplotype) to -1 when the alleles are maximally negatively associated (that is, never occurring together in the same haplotype).

Results

The *SULT1A1* and *SULT1A2* genotypes were determined in 234 healthy Korean subjects (195 males, 39 females). The observed allelic frequencies of *SULT1A1*1* and *SULT1A1*2* were 0.876 and 0.124, respectively. However, we found no subject carrying the *SULT1A1*3* allele in this population. The allelic frequencies of *SULT1A2*1* and *SULT1A2*2* were 0.885 and 0.115, respectively, but no subject carried the *SULT1A2*3* allele (Table 1). The expected allelic frequencies of *SULT1A1* and *SULT1A2* estimated at Hardy–Weinberg equilibrium were quite similar to the observed distributions in the population ($\chi^2 = 4.475$, $P = 0.107$ for *SULT1A1*, $\chi^2 = 3.378$, $P = 0.185$ for *SULT1A2*). There were no significant gender differences in allele frequencies for SULT1A1 or SULT1A2 ($\chi^2 = 0.0269$, $P = 0.870$ for *SULT1A1*, $\chi^2 = 0.0031$, $P = 0.956$ for *SULT1A2*).

We estimated the allele frequencies of *SULT1A1* and *SULT1A2* to determine whether the two alleles mapped together or independently. The results showed that the observed subject numbers were quite similar to the expected numbers, assuming that the two genes are associated (Table 2). The statistical hypothesis that these alleles are independently distributed was rejected ($\chi^2 = 159.2$, $P < 0.001$), indicating that the two genes are strongly linked. Furthermore, the calculated value of D', the coefficient of linkage disequilibrium, showed a strong positive linkage between *SULT1A1*2* and *SULT1A2*2* ($D' = 0.8919$, $\chi^2 = 343.24$, $P = 0.0034$) in this population.

Discussion

In this study we compared the frequencies of all six common *SULT1A1* and *SULT1A2* alleles that occur in the Korean population with the frequencies of those same alleles in other ethnic groups, and we observed striking ethnic differences compared with African-American and Caucasian populations [19]. However, the allele frequencies for *SULT1A1* and *SULT1A2* were similar to those of Chinese and Japanese populations [19]. We could not find any allele of *SULT1A1*3* or *SULT1A2*3* in this population. Similarly, other Asian ethnic groups, including the Chinese and Japanese, had very low frequencies of these alleles, whereas African-Americans had higher frequencies (0.23 for *SULT1A1* and 0.11 for *SULT1A2*) [19]. Even though we found no subject with the allele of *SULT1A1*3* or *SULT1A2*3* in this population, the small sample size might have limited the detection of these alleles. Therefore, the study should be conducted with larger populations.

Table 1 Allele frequencies of *SULT1A1* and *SULT1A2* determined in the present study, compared with those from other studies (data in parentheses represent 95% confidence intervals)

Study population	Number of subjects	Allele frequency						References
		*SULT1A1*1*	*SULT1A1*2*	*SULT1A1*3*	*SULT1A2*1*	*SULT1A2*2*	*SULT1A2*3*	
Koreans	234	0.876 (0.843–0.905)	0.124 (0.096–0.157)	0	0.885 (0.852–0.912)	0.115 (0.088–0.150)	0	Present study
Caucasians (Germany)	300	0.635 (0.581–0.689)	0.365 (0.314–0.423)	0	0.626 (0.571–0.680)	0.374 (0.321–0.430)	–	[15]
Caucasians (USA)	245	0.656 (0.596–0.714)	0.332 (0.275–0.392)	0.012 (0.003–0.037)	0.507 (0.444–0.568)	0.389 (0.329–0.450)	0.104 (0.070–0.147)	[25]
African-Americans	70	0.477 (0.360–0.587)	0.294 (0.205–0.417)	0.229 (0.145–0.341)	0.637 (0.526–0.746)	0.249 (0.157–0.356)	0.114 (0.057–0.213)	[25]
Japanese	143	0.832 (0.762–0.885)	0.168 (0.115–0.239)	0	–	–	–	[11]
Chinese	290	0.914 (0.876–0.942)	0.080 (0.053–0.117)	0.006 (0.000–0.027)	0.924 (0.888–0.950)	0.076 (0.050–0.113)	0	[25]

In this study, the allele frequencies of *SULT1A2*1* and **2* were very similar to those of the *SULT1A1*1* and **2* alleles. Because the expected allele frequencies of *SULT1A1* and *SULT1A2* estimated at Hardy–Weinberg equilibrium was quite similar to the observed distribution, we assessed whether *SULT1A1* and *SULT1A2* are associated by estimating their haplotype frequencies [24]. The frequencies of the alleles *SULT1A1*1* (0.876) and *1A2*1* (0.885) and of the haplotype *SULT1A1*1*/*1A2*1* (0.752) were comparable, suggesting that nearly all *SULT1A1*1* alleles (97.1%) are associated with *SULT1A2*1* and, conversely, that most *SULT1A1*2* alleles (84.5%) are associated with *SULT1A2*2*. On the other hand, the frequencies of the other two haplotypes, *SULT1A1*1*/*1A2*2* and *SULT1A1*2*/*1A2*1*, were very low in the Korean population (3.9 and 2.1%, respectively). The association between *SULT1A1* and *SULT1A2* is consistent with previous data reported [19]. Furthermore, the calculated D' value is close to $+1.0$ ($D' = 0.8919$), indicating that the two genes are strongly and positively linked. This value is similar to that for the Chinese population but higher than those for the Caucasian and African-American populations [19]. The *SULT1A* genes, including *SULT1A1* and *SULT1A2*, are located in the chromosomal region 16p11.2–12.1 [25]. The encoded proteins have the same number of amino acid residues (295 amino acids) and 93–96% identity in the amino acid sequences, but they differ substantially in their substrate specificity and regulation [26]. In addition, a previous study in which the *SULT1A1* gene was cloned suggested that *SULT1A1* and *SULT1A2* are approximately 45 kb apart and oriented head-to-tail [25]. These findings may explain the strong positive linkage disequilibrium of the two genes. However, until now, there has been no clear demonstration of this association at a mechanistic level.

SULT1A1 and *SULT1A2* are the focus of intense research in the fields of cancer, drug metabolism, and pharmacogenetics because genetic variation may predispose carriers to lung cancers [27], protect against colorectal cancers [4], and affect the age of onset of breast cancer [5]. In the case of drug metabolism, SULT1A1 and SULT1A2 are involved in the sulfation of various drugs and chemicals [28, 29]. Rossi et al. [30] have shown that the sulfation rate for 4-nitrophenol, a substrate of SULT1A, by isoenzyme SULT1A1*2/*2 was significant lower than that by that of SULT1A1*1/*1 or *1/*2. Therefore, it would be important to determine the difference in biochemical properties based on the genetic polymorphisms of *SULT1A1* and *SULT1A2* in humans. As mentioned above, the allele frequencies in this population are very close to those observed in other Asian populations but are fairly different from those observed in Caucasians and African-Americans. Therefore, sulfation of certain substance may be clinically significantly different between these populations and thus affect the occurrence of sulfation-related toxicities and diseases. However, little is known of

Table 2 Observed distributions of the combined *SULT1A1* and *SULT1A2* genotypes: comparison of observed numbers with the expected numbers, assuming an independent distribution of the alleles or assuming linkage

Genotype	Observed number	Expected % (no.) assuming independence	Expected % (no.) with association
*1A1*1/*1–1A2*1/*1*	171	77.5 (181)	86.5 (202)
*1A1*1/*2–1A2*1/*1*	9	10.1 (24)	1.1 (3)
*1A1*1/*1–1A2*1/*2*	5	11.0 (26)	1.9 (4)
*1A1*1/*2–1A2*1/*2*	49	1.4 (3)	10.4 (25)
Total	234	100 (234)	100 (234)

the roles of the genetic polymorphisms of these two genes, and future research should be directed to that end.

In summary, we have demonstrated that *SULT1A1*2* and *SULT1A2*2* are major allelic variants but that *SULT1A1*3* and *SULT1A2*3* are rare in the Korean population. Furthermore, *SULT1A1* and *SULT1A2* are strongly and positively linked in this population.

Acknowledgments This work was supported by Korea Research Foundation Grant R0-2004-000-11952-0.

References

1. Otterness DM, Weinshilboum R (1994) Human dehydroepiandrosterone sulfotransferase: molecular cloning of cDNA and genomic DNA. Chem Biol Interact 92:145–159
2. Falany CN (1997) Enzymology of human cytosolic sulfotransferases. FASEB J 11:206–216
3. Nebert DW (1997) Polymorphisms in drug-metabolizing enzymes: what is their clinical relevance and why do they exist? Am J Hum Genet 60:265–271
4. Bamber DE, Fryer AA, Strange RC, Elder JB, Deakin M, Rajagopal R, Fawole A, Gilissen RAHJ, Campbell FC, Coughtrie MWH (2001) Phenol sulphotransferase SULT1A1*1 genotype is associated with reduced risk of colorectal cancer. Pharmacogenetics 11:679–685
5. Seth P, Lunetta KL, Bell DW, Gray H, Nasser SM, Rhei E, Kaelin CM, Iglehart DJ, Marks JR, Garber JE, Haber DA, Polyak K (2000) Phenol sulfotransferases: hormonal regulation, polymorphism, and age of onset of breast cancer. Cancer Res 60:6859–6863
6. Nowell S, Sweeney C, Winters M, Stone A, Lang NP, Hutchins LF, Kadlubar FF, Ambrosone CB (2002) Association between sulfotransferase 1A1 genotype and survival of breast cancer patients receiving tamoxifen therapy. J Natl Cancer Inst 94:1635–1640
7. Wilborn TW, Comer KA, Dooley TP, Reardon IM, Heinrikson RL, Falany CN (1993) Sequence analysis and expression of the cDNA for the phenol-sulfating form of human liver phenol sulfotransferase. Mol Pharmacol 43:70–77
8. Zhu X, Veronese ME, Sansom LN, McManus ME (1993) Molecular characterisation of a human aryl sulfotransferase cDNA. Biochem Biophys Res Commun 192:671–676
9. Ozawa S, Nagata K, Shimada M, Ueda M, Tsuzuki T, Yamazoe Y, Kato R (1995) Primary structures and properties of two related forms of aryl sulfotransferases in human liver. Pharmacogenetics 5:S135–S140
10. Hart RF, Renskers KJ, Nelson EB, Roth JA (1979) Localization and characterization of phenol sulfotransferase in human platelets. Life Sci 24:125–130
11. Rein G, Glover V, Sandler M (1982) Multiple forms of phenolsulphotransferase in human tissues: selective inhibition by dichloronitrophenol. Biochem Pharmacol 31:1893–1897
12. Young WF Jr, Okazaki H, Laws ER Jr, Weinshilboum RM (1984) Human brain phenol sulfotransferase: biochemical properties and regional localization. J Neurochem 43:706–715
13. Coughtrie MWH, Gilissen RAHJ, Shek B, Strange RC, Fryer AA, Jones PW, Bamber DE (1999) Phenol sulphotransferase SULT1A1 polymorphism: molecular diagnosis and allele frequencies in Caucasian and African populations. Biochem J 337:45–49
14. Raftogianis RB, Wood TC, Otterness DM, Van Loon JA, Weinshilboum RM (1997) Phenol sulfotransferase pharmacogenetics in humans: association of common SULT1A1 alleles with TS PST phenotype. Biochem Biophys Res Commun 239:298–304
15. Raftogianis RB, Wood TC, Weinshilboum RM (1999) Human phenol sulfotransferases SULT1A2 and SULT1A1. Genetic polymorphisms, allozyme properties, and human liver genotype–phenotype correlations. Biochem Pharmacol 58:605–616
16. Zhu X, Veronese ME, Iocco P, McManus ME (1996) cDNA cloning and expression of a new form of human aryl sulfotransferase. Int J Biochem Cell Biol 28:565–571
17. Meinl W, Meerman JH, Glatt H (2002) Differential activation of promutagens by alloenzymes of human sulfotransferase 1A2 expressed in *Salmonella typhimurium*. Pharmacogenetics 12:677–689
18. Engelke CEH, Meinl W, Boeing H, Glatt H (2000) Association between functional genetic polymorphisms of human sulfotransferases 1A1 and 1A2. Pharmacogenetics 10:163–169
19. Carlini EJ, Raftogianis RB, Wood TC, Jin F, Zheng W, Rebbeck TR, Weinshilboum RM (2001) Sulfation pharmacogenetics: SULT1A1 and SULT1A2 allele frequencies in Caucasian, Chinese and African-American subjects. Pharmacogenetics 11:57–68
20. Peng CT, Chen JC, Yeh KT, Wang YF, Hou MF, Lee TP, Shih MC, Chang JY, Chang JG (2003) The relationship among the polymorphisms of SULT1A1, 1A2 and different types of cancers in Taiwanese. Int J Mol Med 11:85–89
21. Terwilliger JD, Ott J (1993) A novel polylocus method for linkage analysis using the lod-score or affected sib-pair method. Genet Epidemiol 10:477–482
22. Falconer DS, Mackay TFC (1989) Introduction to quantitative genetics, 4th edn. Longman Group, Harlow
23. Hartl DL, Clark AG (1989) Principles of population genetics, 2nd edn. Sinauer Associates, Sunderland, MA
24. Nagata K, Yamazoe Y(2000) Pharmacogenetics of sulfotransferase. Annu Rev Pharmacol Toxicol 40:159–176
25. Raftogianis RB, Her C, Weinshilboum RM (1996) Human phenol sulfotransferase pharmacogenetics: STP1 gene cloning and structural characterization. Pharmacogenetics 6:473–487
26. Honma W, Kamiyama Y, Yoshinari K, Sasano H, Shimada M, Nagata K, Yamazoe Y (2001) Enzymatic characterization and interspecies difference of phenol sulfotransferases, ST1A forms. Drug Metab Dispos 29:274–281
27. Wang Y, Spitz MR, Tsou AM, Zhang K, Makan N, Wu X (2002) Sulfotransferase (SULT) 1A1 polymorphism as a predisposition factor for lung cancer: a case-control analysis. Lung Cancer 35:137–142

28. Levy RH, Thummel KE, Trager WF, Hanstein PD, Eichelbaum M (2000) Sulfotransferase and methyltransferase. In: Metabolic drug interactions, 1st edn. Lippincott Williams & Wilkins, Philadelphia, pp 191–204
29. Kauffman FC (2004) Sulfonation in pharmacology and toxicology. Drug Metab Rev 36:823–843
30. Rossi AM, Maggini V, Fredianelli E, Di Bello D, Pietrabissa A, Mosca F, Barale R, Pacifici GM (2004) Phenotype–genotype relationships of SULT1A1 in human liver and variations in the IC50 of the SULT1A1 inhibitor quercetin. Int J Clin Pharmacol Ther 42:561–567