SHORT REPORT

CYP2C19*2 status in patients with Stevens-Johnson syndrome and toxic epidermal necrolysis

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Purpose: Genetic polymorphisms have been linked to an increased predisposition to developing certain diseases. For example, patients of Han-Chinese descent carrying the HLA-B*1502 allele are at an increased risk of developing Stevens-Johnson syndrome and toxic epidermal necrolysis (SJS/TEN) if given carbamazepine. Given the complexity of in vivo drug metabolism, it is plausible that the activity of enzyme systems unrelated to specific drug metabolism may be important. Although multiple biomarkers have been identified in unique ethnic groups, there has yet to be a study investigating the presence of the slow metabolizing allele of CYP2C19, denoted CYP2C19*2, in diverse groups and the risk of developing SJS/TEN.

Patients and methods: This study looked into the carrier status of CYP2C19*2, a poor metabolizing variant of CYP2C19, in patients diagnosed with SJS/TEN. We looked at its status in our series as a whole and when patients were divided by ethnicity. Genomic DNA was extracted from formalin-fixed paraffin-embedded tissue of patients with biopsy-proven SJS/TEN and real-time polymerase chain reaction was used to assess for the presence of CYP2C19*2.

Results: CYP2C19*2 status was determined in 47 patients. Twenty-nine of these 47 patients had a single medication implicated as causing their disease, and eight of these patients were heterozygous or homozygous for CYP2C19*2. There was insufficient evidence to conclude that the presence of CYP2C19*2 is an independent predictor of risk for developing SJS/TEN in our series as a whole. This analysis also confirmed that the frequency of the CYP2C19*2 polymorphism within the different ethnicities in our series did not vary statistically from reported ethnic rates.

Conclusion: Our study was unable to show a relationship between CYP2C19*2 status and predisposition toward SJS/TEN. We had a heterogeneous population, making it difficult to control for possible confounding factors.

Keywords: drug reactions, drug metabolism, adverse events, dermatology

Introduction

Stevens-Johnson syndrome and toxic epidermal necrolysis (SJS/TEN) are uncommon severe cutaneous adverse drug reactions in dermatology with an unclear pathogenesis; however, polymorphisms in the cytochrome p450 enzymes are thought to play a role. Multiple studies have associated altered drug metabolism with an increased risk of these events when the offending drug is a substrate of the altered metabolic pathway.[1–3] For example, one study showed a statistically significant association between the development of SJS/TEN in Thai children taking phenobarbital, a substrate of CYP2C19, and the slow metabolizing allele of CYP2C19, designated CYP2C19*2.[3] We sought to determine if the presence of CYP2C19*2 was an independent risk factor for the development of SJS/TEN in an ethnically diverse group of patients taking

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medications not principally metabolized through CYP2C19. A secondary goal was to investigate if CYP2C19*2 was increased in specific ethnicities compared to background population frequencies.

Patients and methods

Forty-seven patients with biopsy-proven SJS/TEN were identified through a 10-year retrospective review by Chung K (US Army Institute of Surgical Research [USAISR], Fort Sam Houston, TX, USAISR Study No. H-12–004, institutional review board (IRB) Log No. M-10225, unpublished data). Patient ethnicities were classified as White, Black, Hispanic, or other if no ethnicity was listed in the medical record. Genomic DNA was extracted from formalin-fixed paraffin-embedded (FFPE) tissue using the GeneRead DNA FFPE kit (Qiagen, Hilden, Germany). Real-time polymerase chain reaction was performed on the Rotor-Gene Q thermocycler (Qiagen) using Taqman Genotyping Master Mix and Taqman SNP Genotyping Assay primers and probes specific for the CYP2C19*2 mutation (Applied Biosystems/Thermo-Fisher Scientific, Waltham, MA, USA). Selected samples were verified by Sanger Sequencing on the 3130XL Genetic Analyzer (Applied Biosystems).

This database was a preexisting one. The IRB (Wilford Hall Ambulatory and Surgical Center, JBSA Lackland, TX, USA) deemed no patient consent was needed. This IRB protocol was titled: "Retrospective genetic analysis of formalin fixed paraffin embedded tissue in patients with severe cutaneous adverse reactions to drugs". IRBNet#: 409483-1, FWH: 20150039E.

Results

We determined CYP2C19*2 status in a series of 47 patients. When observing the data from the standpoint of causative medication, 29 out of 47 patients had a single causative medication listed, and 8 of these 29 patients were found to be heterozygous (7) or homozygous (1) for this allele (Table 1). In seven of eight cases, the offending drug was not a primary substrate of CYP2C19. The remaining patient was taking voriconazole, which is a substrate of CYP2C19. When observing the data from the standpoint of ethnicity, only 9 patients out of the 47 total patients had both CYP2C19*2 and a listed ethnicity, with the remaining CYP2C19*2-positive patient having an unlisted ethnicity (Table 2).

A logistic regression on incidence of SJS/TEN was fit to patient ethnicity and CYP2C19*2 status using population data from the Exome Aggregation Consortium and Single Nucleotide Polymorphism Database.[4,5] This analysis did not

Table 1 Twenty-nine patients with a single causative medication, CYP2C19 status, and ethnicity

Causative agent	CYP2C19*2 status	Ethnicity
Allopurinol	Negative	Hispanic
Allopurinol	Negative	Other
Allopurinol	Negative	White
Amikacin	Negative	White
Amoxicillin	Heterozygous	Black
Azithromycin	Heterozygous	Unlisted
Carbamazepine	Negative	White
Ceftriaxone	Heterozygous	Hispanic
Ceftriaxone	Negative	Unlisted
Ciprofloxacin	Negative	Other
Colchicine	Homozygous	Black
Diclofenac	Negative	White
Phenytoin	Negative	White
Donnatal	Negative	White
Ibuprofen	Negative	White
Lamotrigine	Heterozygous	White
Lamotrigine	Heterozygous	White
Levofloxacin	Negative	White
TMP/SMX	Heterozygous	White
TMP/SMX	Negative	Black
TMP/SMX	Negative	Black
TMP/SMX	Negative	Hispanic
TMP/SMX	Negative	Other
TMP/SMX	Negative	Other
TMP/SMX	Negative	Other
TMP/SMX	Negative	White
TMP/SMX	Negative	White
TMP/SMX	Negative	White
Voriconazole	Heterozygous	White

Abbreviations: SMX, sulfamethoxazole; TMP, trimethoprim.

Table 2 Ten patients with the presence of CYP2C19*2 and their ethnicities

Causative drug	CYP2C19*2 status	Ethnicity
Amoxicillin	Heterozygous	Black
Azithromycin	Heterozygous	Unlisted
Ceftriaxone	Heterozygous	Hispanic
Colchicine	Homozygous	Black
Lamotrigine	Heterozygous	White
Lamotrigine	Heterozygous	White
TMP/SMX	Heterozygous	White
Voriconazole	Heterozygous	White
Unknown	Heterozygous	White
Unknown	Heterozygous	White

Abbreviations: SMX, sulfamethoxazole; TMP, trimethoprim.

show an increase in CYP2C19*2 allele frequency within our population as a whole independent of ethnicity. We also confirmed that the frequency of the CYP2C19*2 polymorphism within the different ethnicities in our series did not vary statistically from reported ethnic rates (Figure 1).

Additional analysis revealed that the most commonly identified medication in the 29 out of 47 patients who had a medication identified as being causative was trimethoprim/
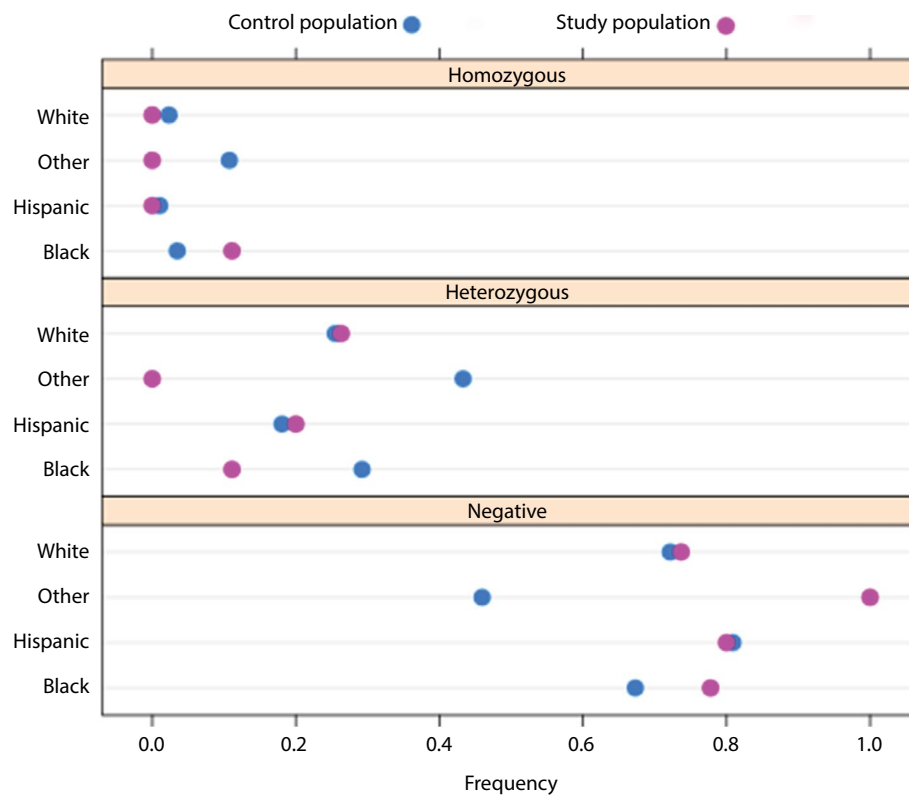
Figure 1 Dot plot comparing the frequency of the CYP2C19*2 genotype (homozygous, heterozygous, and negative) among different ethnic groups in our study population (pink dots) to a control population obtained from dbSNP and ExAC (blue dots). Units for frequency are the fractions of the population.

sulfamethoxazole followed by allopurinol, then ceftriaxone, and lamotrigine (Table 1).

Discussion

CYP2C19*2 was not found to be an independent risk factor for the development of SJS/TEN, both in our series as a whole and when different ethnic populations were compared to the published rates of the CYP2C19*2 frequency. Many of the patients were taking medications that were not necessarily metabolized through CYP2C19, which may or may not have led to the drug reaction. It is known that one medication can be metabolized through multiple enzymes and that its intermediates can interact with multiple targets.[6] This polygenic model makes finding one causative genetic polymorphism difficult. Also, the effect of a cytochrome p450 mutation on metabolism can be substrate dependent, with some drugs being barely affected and others rising to toxic levels.[7] Regardless, the presence of CYP2C19*2 may play more of a role when a patient is taking a medication that is processed by the CYP2C19 pathway, as was the case for the patient in our cohort taking voriconazole. While this number was too small to draw any conclusions about the direct relationship between culprit medications and the

CYP2C19*2 polymorphism, we did not observe any patients taking a medication that was a known substrate of CYP2C19 who did not have a CYP2C19*2 polymorphism.

Our study had several limitations. First, we used genomic DNA extracted from FFPE specimens, which usually is fragmented and chemically modified compared to genomic DNA from peripheral blood cells. Because our study was performed retrospectively using a previously constructed database, we were limited to this type of genomic DNA. Second, the number of patients in this study may have been too small to be able to make a concise conclusion about the effect of ethnicity on CYP2C19*2 status. We only had two Black patients and one Hispanic patient who had a listed medication and were positive for CYP2C19*2, far too few to make definitive conclusions. Also, the frequency of CYP2C19*2 in Asian populations is roughly two to three times that of African and American allele frequencies, ranging from 29% to 35% in Asian populations based on region (East, Central, and South Asia).[8] In a diverse series like ours, more patients would be needed to make up for the low baseline frequency of CYP2C19*2. Finally, prior studies reporting genetic polymorphisms linked with medication toxicities, such as CYP2C9*3 with phenytoin and CYP2C19*2 with

phenobarbital, have been performed in populations of the same ethnicity with similar medical histories and medication exposure.[1,3] Our population was varied with regard to ethnicity, age, comorbidities, and medication history, so there were many confounding factors that could have altered our results.

Conclusion

This study has investigated a previously understudied link between a polymorphism in CYP2C19, CYP2C19*2, and its possible role as an independent risk factor for SJS/TEN. We were not able to prove statistical significance between this polymorphism and an increased risk of developing SJS/TEN in our series as a whole or when divided by ethnicity. While our results were similar to established frequencies of CYP2C19*2 in different ethnic populations, this research has suggested that patients taking substrates of CYP2C19 with the CYP2C19*2 polymorphism may be at increased risk of SJS/TEN development. Further research using a large cohort of patients taking medications primarily metabolized through CYP2C19 could help clarify results. Similarly, performing this study in a group of patients of the same ethnicity, regardless of causative drug, could also help to reduce confounding factors.

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Disclosure

The authors report no conflicts of interest in this work.

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