

Association of HLA-B*1502 allele and carbamazepine-induced Stevens-Johnson syndrome among Indians

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INTRODUCTION

Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN) are acute life-threatening reactions of the skin to drugs. SJS and TEN are two related mucocutaneous blistering disorders with significant mortality (especially with TEN) and prolonged morbidity. Patients develop an acute exanthema, which progresses toward a widespread blistering (TEN) or more limited purpura, vesicles (SJS) and erosion of the skin and mucous membranes, resulting from apoptosis of keratinocytes. The incidence of SJS is estimated to be one to six cases per million person-years and of TEN at

0.4–1.2 cases per million person-years. Various drugs are reported to be associated with a high risk for SJS and TEN. These include several antiepileptic agents, especially carbamazepine, phenytoin, phenobarbital and lamotrigine, antibacterials, anti-inflammatory drugs of the oxicam family and allopurinol.^[1,2]

Chung and colleagues^[3] reported a very strong association between carbamazepine-induced SJS in Han Chinese patients and the human leucocyte antigen (HLA)-B*1502 allele. They studied 44 Han Chinese patients with carbamazepine-induced SJS and two groups of controls, 101 tolerant patients

ABSTRACT

Background: Stevens–Johnson Syndrome (SJS) and toxic epidermal necrolysis are severe cutaneous reactions caused by certain drugs, including antiepileptic carbamazepine. A strong association has been reported between human leucocyte antigen (HLA)-B*1502 and carbamazepine-induced SJS in Han Chinese patients. European studies suggested that HLA-B*1502 is not a universal marker but is ethnicity-specific for Asians. **Aim:** To study the association between HLA-B*1502 and carbamazepine-induced SJS in Indian patients. **Methods:** Eight individuals who fulfilled the diagnostic criteria of SJS induced by carbamazepine were identified and HLA-B molecular typing was performed. HLA-B genotyping was carried out by polymerase chain reaction using sequence-specific primers. **Results:** Out of eight patients studied for genotype, six patients were found to have the HLA-B*1502 allele. **Conclusion:** This study suggests an association between HLA-B*1502 and carbamazepine-induced SJS in Indian patients.

Key words: Carbamazepine, HLA-B*1502, Hindu, Indian, Stevens–Johnson syndrome

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and 93 healthy controls. All 44 SJS or TEN patients had an HLA-B*1502 allele in contrast with 3% of the carbamazepine-tolerant patients and 8.6% of the unexposed controls, respectively.^[3] In Europe, HLA-B genotyping was carried out in 12 patients of carbamazepine-induced SJS. Of the 12, only four had HLA-B*1502. Remarkably, all the four had an Asian ancestry and originated from China, Vietnam, Cambodia, and the Reunion Island. It was suggested that HLA-B*1502 is not a universal marker for carbamazepine-induced SJS but is ethnicity-specific for Asians.^[4] Studies in other Asian populations from Malaysia, Thailand and Hong Kong have also reported an association between HLA-B*1502 and SJS induced by carbamazepine.^[5-7] None of these studies have been reported from India although the incidences of SJS were found to be higher among Indians compared with Europeans.^[8-11] Carbamazepine is a widely prescribed antiepileptic in India and is found to be the most commonly implicated drug responsible for SJS/TEN among Indians.^[12] In the light of the above observations, we have undertaken an HLA genotyping study of Indian patients suffering from carbamazepine-induced SJS.

METHODS

Patient recruitment and sample collection

Eight individuals who fulfilled the diagnostic criteria of SJS induced by carbamazepine were identified at Dr. Rasiklal Shah Sarvajanic Hospital and Samarpan Centre for Treatment of Diseases of Skin, STD and Leprosy, Modasa, India. All the patients were assessed by dermatologists who had received photographs, pathological slides and medical records. Patients with definite diagnosis and ready to sign written consent were included in the study. Ten unrelated normal controls from the same population were selected randomly. The study was approved by the Institutional

Review Board of Dr. Rasiklal Shah Sarvajanic Hospital, Modasa. The diagnostic criteria of SJS were based on the clinical morphology defined by Roujeau.^[13] We defined SJS as skin detachment up to 10% of the body surface area and overlap SJS/TEN as skin detachment of 10–30%. Our five patients (no. 2, 3, 4, 7, 8) received carbamazepine as an antiepileptic therapy. Two patients (no. 5, 6) received carbamazepine as a post head injury anticonvulsant prophylaxis. In the remaining one patient (no. 1), trigeminal neuralgia was the indication for carbamazepine. Peripheral blood was collected in ethylene diamine tetra acetic acid-coated sterile vials and stored in a refrigerator. Table 1 shows the clinical characteristics of eight patients with carbamazepine-induced SJS identified in our study.

DNA extraction

Genomic DNA from peripheral lymphocytes was extracted by the standard phenol–chloroform method.

HLA-B genotyping

The quality and quantity of the extracted DNA was checked on 0.8% agarose gel and a spectrophotometer. The concentration of each DNA sample was finally adjusted to 30 ng/μl, which was used for HLA-B molecular typing. HLA-B genotyping was carried out by polymerase chain reaction (PCR) using sequence-specific primers (Micro SSP HLA-B kits; One Lambda Inc., Canoga Park, CA, United States. Low-resolution HLA-B genotyping was carried out with the One Lambda HLA-B kit in a PCR with sequence-specific primers according to the protocol and recommendations of the manufacturer (PCR-SSP, One Lambda Inc.).

Statistical analysis

Allele frequency between the two groups was compared by the Chi-square method with Yates' correction by constructing 2 x 2 tables. The odds ratio

Table 1: Clinical characteristics of patients with carbamazepine-induced Stevens-Johnson syndrome

Age/sex	Phenotype	Exanthem (%)/ blister or detachment (%)	Mucous membrane erosion, number and sites	Systemic manifestations
45/F	SJS	70–80/<10	2: oral, eyes	Fever
20/F	SJS	85–95/<10	3: oral, eyes, genital	Leukocytosis
16/F	SJS	50–60/<10	2: oral, eyes	Eosinophilia
18/F	SJS	50–60/<10	2: oral, eyes	Eosinophilia
16/M	SJS/TEN	>90/20–30	3: oral, eyes, genital	Fever, leukocytosis, minor LFI and RFI
10/M	SJS	50–60/<10	3: oral, eyes, genital	Fever, leukocytosis, minor LFI and RFI
32/M	SJS/TEN	80–90/20–30	3: oral, eyes, genital	Fever, leukocytosis, minor LFI and RFI
26/M	SJS/TEN	80–90/10–20	3: oral, eyes, genital	Fever, leukocytosis, minor LFI and RFI

LFI, liver function impairment; RFI, renal function impairment. *Exanthem: widespread purpuric macules or flat atypical targets. Both exanthem and blister/detachment were expressed as % of total body surface area. †Odds ratio: 71.40 (95% CI, 3.0–1698); *P* = 0.0014.

Table 2: Genotypes in eight patients with carbamazepine-induced SJS and ten normal controls[†]

Patients with SJS		Normal controls	
B*1502	B*35	B*1521/95 (75)	B*44
B*1502	B*13	B*1521/95 (75)	B*5209
B*1508	B*1508	B*44	B*5711
B*1502	B*72/40 (B*1546/53; 4011/2)	B*77 (15)	B*5209
B*1502	B*55	B*60 (40)	B*44
B*1502	B*44	B*08	B*44
B*07	B*61 (40)	B*62 (1505)	B*44
B*1502	B*35	B*61 (40)	B*5137
-	-	B*61 (40)	B*57
-	-	B*08	B*53

was calculated with Haldane's modification, which adds 0.5 to all cells to accommodate a possible zero count.

RESULTS

We determined the HLA-B genotypes of eight carbamazepine-induced cases and 10 normal controls by PCR using sequence-specific primers (Micro SSP HLA-B kits; One Lambda Inc.). We found that six out of eight patients had the HLA-B*1502 allele (all being heterozygous). One patient was found to be homozygous with HLA-B*1508 allele. All the 10 controls were found to be negative for HLA-B*1502. The genotypes observed are shown in Table 2. The odds ratio was found to be 71.40 (95% CI, 3.0–1698) and the *P*-value was found to be 0.0014.

DISCUSSION

HLA-B*1502 as a marker for carbamazepine-induced SJS is well established in the Han Chinese by different studies. The allele frequency of HLA-B*1502 in the Han Chinese population is 8%, which is relatively higher than any other population of the world.^[14] From India, a 0–6% (average 2.5%) prevalence of HLA-B*1502 in different communities has been reported. Most of them are sub-Hindu communities, except Parsi, in which a 0% prevalence is reported.^[15–20] Our eight patients also belong to the Hindu community, which is the biggest community of India comprising of one billion Indians. Our six out of eight patients had HLA-B*1502 while none of the 10 controls were found to be positive. This clearly indicates a significant association (Odds ratio: 71.40 [95% CI, 3.0–1698]; *P* = 0.0014) between carbamazepine-induced SJS and HLA-B*1502 among Indians.

Because the association is not 100%, as observed in the Han Chinese community, other genes or combinations of genes might be playing a role. To conclude, we suggest HLA-B*1502 testing before initiating carbamazepine drug therapy in Indians. To obtain a clear Indian picture, the study should be extended to different subethnic groups from different regions.

REFERENCES

- Bastuji-Garin S, Rzany B, Stern RS, Shear NH, Naldi L, Roujeau JC. Clinical classification of cases of toxic epidermal necrolysis, Stevens-Johnson syndrome, and erythema multiforme. *Arch Dermatol* 1993;129:92-6.
- Roujeau JC, Kelly JP, Naldi L, Rzany B, Stern RS, Anderson T, *et al*. Medication use and risk of Stevens-Johnson syndrome or toxic epidermal necrolysis. *New Engl J Med* 1995;333:1600-7.
- Chung WH, Hung SI, Hong HS, Hsieh MS, Yang LC, Ho HC, *et al*. A marker for Stevens-Johnson syndrome. *Nature* 2004;428:86.
- Lonjou C, Thomas L, Borot N, Ledger N, de Toma C, LeLouet H, *et al*. A marker for Stevens-Johnson syndrome: Ethnicity matters. *Pharmacogenomics J* 2006;6:265-8.
- Chang CC, Too CL, Murad S, Hussein S. Association of HLA-B*1502 with carbamazepine-induced toxic epidermal necrolysis and Stevens-Johnson syndrome in Malaysian population. *Proceeding in 7th Asian- Oceanian Epilepsy Congress, Xiamen. 2008.*
- Locharernkul C, Loplumert J, Limotai C, Korkij W, Desudchit T, Tongkobpetch S, *et al*. Carbamazepine and phenytoin induced Stevens-Johnson syndrome is associated with HLA-B*1502 allele in Thai population. *Epilepsia* 2008;49:2087-91.
- Man CB, Kwan P, Baum L, Yu E, Lau KM, Cheng AS, *et al*. Association between HLA-B*1502 Allele and antiepileptic drug-induced cutaneous reactions in Han Chinese. *Epilepsia* 2007;48:1015-8.
- Uppal R, Jhaj R, Malhotra S. Adverse drug reactions among inpatients in a North Indian referral hospital. *Natl Med J India* 2000;13:16-8.
- Naldi L, Conforti A, Venegoni M, Troncon MG, Caputi A, Ghiotto E, *et al*. Cutaneous reactions to drugs. An analysis of spontaneous reports in four Italian regions. *Br J Clin Pharmacol* 1999;48:839-46.
- Hunziker T, Künzi UP, Braunschweig S, Zehnder D, Hoigné R. Comprehensive hospital drug monitoring (CHDM): Adverse skin reactions, a 20-year survey. *Allergy* 1997;52:388-93.
- Noel MV, Sushma M, Guido S. Cutaneous adverse drug reactions in hospitalized patients in a tertiary care center. *Indian J Pharmacol* 2004;36:292-5.
- Devi K, George S, Criton S, Suja V, Sridevi PK. Carbamazepine - The commonest cause of toxic epidermal necrolysis and Stevens-Johnson syndrome: A study of 7 years. *Indian J Dermatol Venereol Leprol* 2005;71:325-8.
- Roujeau JC. The spectrum of Stevens-Johnson syndrome and toxic epidermal necrolysis: A clinical classification. *J Invest Dermatol* 1994;102:28-30.
- Geer L, Terasaki PI, Gjertson DW. HLA frequency. In: Gjertson DW, Terasaki PI, editors. *HLA 1998*. Lenexa, KS: American Society for Histocompatibility and Immunogenetics; 1998. p. 327-63.
- Shankarkumar U. HLA-A, -B and -Cw allele frequencies in a Bhil population from Western India. *Hum Immunol* 2004;65:960-61.
- Shankarkumar U. HLA-A, -B and -Cw allele frequencies in a Maratha population from Mumbai, India. *Hum Immunol* 2004;65:954-6.
- Shankarkumar U. HLA-A, -B and -Cw allele frequencies in a Pawra population from Khandesh, India. *Hum Immunol* 2004;65:952-53.

18. Shankarkumar U. HLA-A, -B and -Cw allele frequencies in a Parsi population from Western India. *Hum Immunol* 2004;65:992-93.
19. Rajalingam R, Parham P, Mehra NK. HLA-A, -B, -Cw, -DQA1, -DQB1 and -DRB1 Alleles and KIR Alleles in a Hindu Population from Punjab, India. *Hum Immunol* 2004;65:958-60.
20. Rani R, Marcos C, Lazaro AM, Zhang Y, Stastny P. Molecular diversity of HLA-A, -B and -C alleles in a North Indian population as determined by PCR-SSOP. *Int J Immunogenet* 2007;34:201-8.

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