*Neurology Asia 2013; 18 (Supplement 1) : 27 – 29*

# Pharmacogenomics screening of HLA-B*1502 in epilepsy patients: How we do it in the UKM Medical Centre, Malaysia

Sue-Mian Then, Zam Zureena Mohd Rani, *Azman Ali Raymond, Rahman Jamal

*UKM Medical Molecular Biology Institute, Universiti Kebangsaan Malaysia; *Neurology Department, UKM Medical Center, Universiti Kebangsaan Malaysia (UKM), Kuala Lumpur Malaysia*

### *Abstract*

Previous studies have shown that carbamazepine-induced Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN) patients is associated with the HLA-B*1502 allele. Screening for HLA-B*1502 before using carbamazepine can prevent SJS/TEN particularly in populations with high frequency of the allele. The objective of this paper was to describe how the UKM Medical Centre, Malaysia was able to set up a cost effective screening of HLA-B*1502 for patients taking carbamazepine. The cost of in-house HLA-B⁄1502 screening was less than those commercially available, and was sensitive and specific.

## INTRODUCTION

The drug treatment of epilepsy is characterized by unpredictability of efficacy, adverse drug reactions and variable optimal doses. Pharmacogenomics is the use of the genetic makeup of an individual to predict drug response, efficacy and potential adverse drug events. Carbamazepine (CBZ) is an antiepileptic drug (AED) which is most often taken as monotherapy for long term treatment of epilepsy. CBZ is among the commonest AED that causes cutaneous adverse reactions (cADRs)[1] which includes the life-threatening Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN). Data from the WHO Uppsala Monitoring (WHO-UMC) and Novartis CBZ-SJS/TEN reports of 2000-2006 showed that the incidence of CBZ-induced cADR was highest among South East Asian countries such as Malaysia and Thailand.[2,3] Various studies have shown that CBZ therapy can cause SJS/TEN in Han Chinese, Thai and Malay patients with HLA-B*1502 polymorphism.[4-6] No other genotypes were associated with CBZ-SJS/TEN except for some haplotypes which have closely linked loci such as CW*0801, A*1101 and DRB*1202.[7] A recent report on AED usage in a Malaysian hospital showed that CBZ is the commonest AED prescribed as monotherapy for simple and complex partial seizures.[8] CBZ is also the second commonest AED prescribed after valproic acid in some Asian countries such as India, Taiwan, and Oman.[8] Therefore, according to the Malaysian Consensus Guidelines on the Management of Epilepsy 2010, it is recommended that patients of Chinese and Malay descent should be screened for the HLA-B*1502 allele prior to CBZ prescription.[9] A recent retrospective study from Thailand has shown that it is more cost-effective to screen for the HLA-B*1502 allele using the PCR-based HLA-B genotyping kit prior to prescribing CBZ, than treating SJS/TEN.[7] Our study describes how a public teaching hospital (UKM Medical Centre) in Malaysia developed a cost effective pharmacogenetics screening of HLA-B*1502 allele for epilepsy patients.

## THE WORK FLOW OF HLA-B*1502 SCREENING

Before starting CBZ in epilepsy patients, the clinicians in the Neurology Clinic of UKM Medical Centre will contact our laboratory for the HLA-B*1502 screening. Five mls of blood are taken intravenously and subjected to multiplex PCR assay and confirmed with DNA sequencing assay to rule out false positive results within one week. In some instances where the clinicians urgently need the screening results to administer the appropriate drugs, the samples are prioritized for both multiplex PCR assay and DNA sequencing. Results are given within 3 days verbally before a written report is sent to the clinicians. We earlier did a preliminary study on 27 epilepsy patients (19 Malays, 8 Chinese) who were treated with CBZ to look for HLA-B*1502. Six patients had

*Address correspondence to:* Dr Sue-Mian Then, School of Biomedical Science, Faculty of Science, The University of Nottingham Malaysia Campus, Jalan Broga, 43500 Semenyih, Selangor, Malaysia. E-mail: then.sue-mian@nottingham.edu.my

**Table 1: Cost estimation of various test method**

| Types of HLA-B*1502 testing | Estimate cost per patient (RM) | Estimate cost per patient (USD) |
|---|---|---|
| Commercial HLA-genotyping kit | 227.40 | 74.54 |
| Commercial HLA-B*1502 test (Real-time PCR) | 317.00 | 103.62 |
| Commercial HLA-B*1502 (PCR) | 263.10 | 86.00 |
| In-house Multiplex PCR protocol | 82.85 | 25.15 |
| In-house DNA sequencing protocol | 78.45 | 25.70 |
| In-house Real-time PCR protocol | <50.00 | <16.38 |

CBZ-induced SJS, 11 had CBZ induced rash and 8 were controls. Our study showed that 10 (6 Malay, 4 Chinese) patients were positive for HLA-B*1502. Of the 10 patients, 6 patients had CBZ induced SJS (p=0.0006), while 4 patients had CBZ induced skin rash. However, there were 6 Malay patients and 1 Chinese patient who had CBZ induced skin rash who were negative for the HLA-B*1502 allele. This indicates that there may be allele other than HLA-B*1502 associated with CBZ-induced hypersensitivity.[10]

## COST ESTIMATION OF HLA-B*1502 SCREENING

There are commercially available HLA-B*1502 tests for both real-time PCR and conventional PCR, but the cost of running the test is prohibitive particularly some hospital laboratories in the developing countries. We therefore developed an in-house test methods to reduce the cost of running the test. Our in-house methods cost less than RM 100 (USD 32.76). To make the HLA-B*1502 screening accessible to all our patients, we have developed a new real-time PCR method with the lowest cost thus far of <RM 50 (USD 16.38). The cost comparison of the various test methods are shown in Table 1. The new real-time PCR method with internal positive control also cuts down the running time to less than 3 hours, a 40% reduction from our multiplex PCR method, and 96% reduction when the run time for DNA sequencing was added (Figure 1). Table 2 shows the validation of our newly developed tests. The preliminary results shows that our test is 100% sensitivity and 100% specificity when compared to DNA sequencing in the 9 samples tested.

## CONCLUSION

Our pharmacogenetics screening program of HLA-B*1502 at the UKM Medical Centre serves as a viable model. The success appears to be due to a clear workflow from the clinic or ward to the

**Table 2: Comparison of our newly developed HLA-B*1502 test against the DNA sequencing gold standard.**

| | | DNA Sequencing (Gold Standard) | | |
|---|---|---|---|---|
| New Real Time -PCR Protocol | | HLA-B*1502 positive | HLA-B*1502 negative | TOTAL |
| | Tested HLA-B*1502 positive | 3 | 0 | 3 |
| | Tested HLA-B*1502 negative | 0 | 6 | 6 |
| | TOTAL | 3 | 6 | 9 |

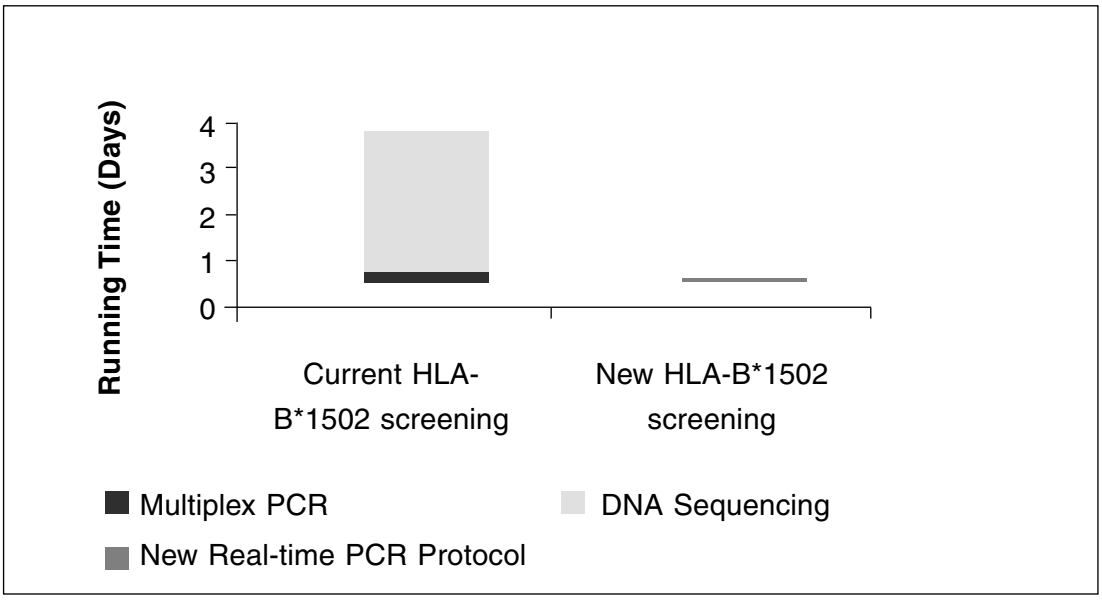


Figure 1. A comparison of the running time of the newly developed HLA-B*1502 screening method against the multiplex PCR method and DNA sequencing method.

testing laboratory and back to the clinic / ward; reliable and validated test methods; dedicated staff; and most importantly, funding source to cover the cost of the test. By reducing the cost of the test, we are able to ensure that the screening model is feasible in our hospital, and accessible for the benefit of the patients.

## ACKNOWLEDGEMENTS

This study was supported by the internal funding of UKM Medical Centre's Genomics Unit 50-6601-001-27405. The authors would like to acknowledge the contributions of Prof. Dr. Ong Lai Choo, Associate Prof. Dr. Tan Hui Jan and Assoc. Prof. Dr Wong Sau Wei in referring the study patients.

## REFERENCES

1. Arif H, Buchsbaum R, Weintraub D, *et al*. Comparison and predictors of rash associated with 15 antiepileptic drugs. *Neurology* 2007; 68:1701-9.
2. Lim KS, Kwan P, Tan CT. Association of HLA-B*1502 allele and carbamazepine-induced severe adverse cutaneous drug reaction among Asians, a review. *Neurol Asia* 2008; 13:15-21.
3. Farkas R: Carbamazepine (marketed as CABATROL, EQUETRO, TEGRETOL and genericics): Stevens-Johnson syndrome, toxic epidermal necrolysis, and HLALA-B*1502. *FDA Drug Safety Newsletter* 2008; 1(3):35-7.
4. Chung WH, Hung SI, Hong HS, *et al*. Medical genetics: a marker for Steven-Johnson syndrome. *Nature* 2004; 428:486.
5. Hung SI, Chung WH, Jee SH, *et al*: Genetic susceptibility to carbamazepine-induced cutaneous adverse drug reactions. *Pharmacogen & Genom* 2006; 16:297-306.
6. Locharernkul C, Loplumlert J, Limotai C, *et al*. Carbamazepine and phenytoin induced Steven-Johnson syndrome is associated with HLA-B*1502 allele in Thai population. *Epilepsia* 2008, 49(12):2087-91.
7. Locharernkul C, Shotelersuk V, Nattiya H. Pharmacogenomic screening of carbamazepine-induced severe cutaneous allergic reactions. *J Clin Neurosci* 2011; 18:1289-94.
8. Hasan SS, Bahari MB, Babar ZU, Ganesan V. Antiepileptic drug utilisation and seizure outcome among paedetric patients in a Malaysian public hospital. *Singapore Med J* 2010; 51:21-7.
9. Malaysian Society of Neurosciences. Consensus guidelines on the management of epilepsy 2010. In: Clinical Practice Guidelines [online]. Available at: http://www.neuro.org.my/pdf/ConsensusGuidelinesontheManagementofEpilepsy2010.pdf
10. Then SM, Rani ZZM, Raymond AA, Ratnaningrum S, Jamal R. Frequency of the HLA-B*1502 allele contributing to carbamazepine-induced hypersensitivity reactions in a cohort of Malaysian epilepsy patients. *Asian Pacific Journal of Allergy and Immunology* 2011; 29(3):290-3.