# MALDI-TOF MS ANALYSIS OF 36 BLOOD GROUP ALLELES AMONG 398 THAI SAMPLES REVEALS REGION-SPECIFIC VARIANTS

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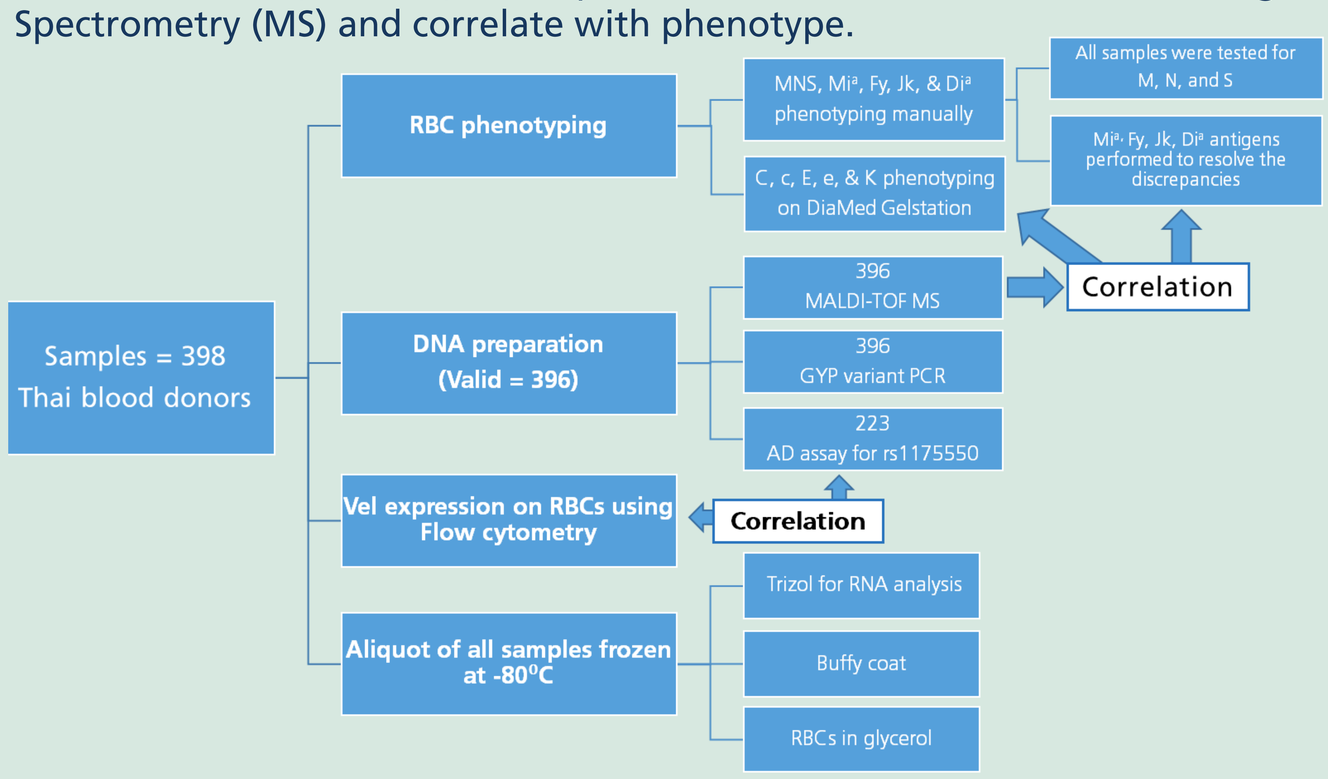
## Introduction

Blood group phenotype variation in different populations has been attributed to potential pathogen resistance. We wanted to investigate the blood group antigen distribution in two groups of blood donors from different regions of Thailand, one north (Lampang) and one central (Saraburi), to map variation. We characterised the blood group allele profile of Thai blood donors by MALDI-TOF Mass Spectrometry (MS) and correlate with phenotype.



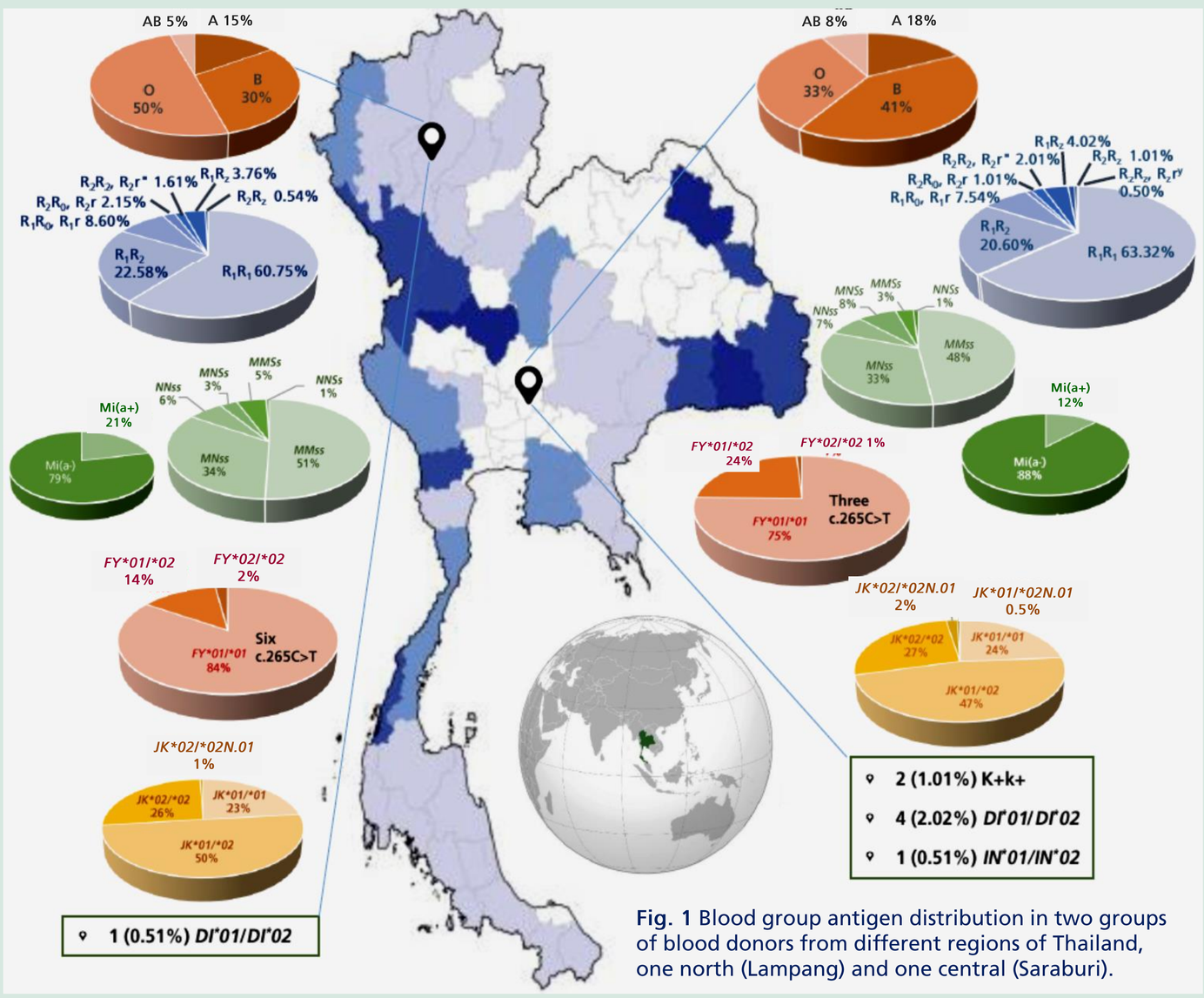




**Fig. 1** Blood group antigen distribution in two groups of blood donors from different regions of Thailand, one north (Lampang) and one central (Saraburi).

## Results

- Genotyping/phenotyping for K, and S/s showed 100% concordance. Genotyping predicted the correct RhD and RhCE phenotypes in 100% and 99.2% of all samples tested, respectively.
- Serological investigation of 3 outliers with a panel of monoclonal anti-e revealed an e-variant antigen (Table 1). Sequence analysis identified heterozygosity for the *RHCE*02.22* allele in these samples. This allele had been shown previously in Caucasians associated with a weak C antigen expression (Fig. 4).
- Discrepancies with MN typing in 44 samples revealed glycophorin variants of which 41/44 were Mi(a+) (Fig. 2).
- Six samples (1.5%) were heterozygous for the *JK*02N.01* allele, of which five genotyped as *JK*02/JK*02N.01* and one as *JK*01/JK*02N.01.* The latter typed Jk(a+b-). The null alleles were more common in the central region (5/6 samples).
- All samples were homozygous for wild type *SMIM1.* AD of rs1175550 revealed homozygosity for the AA allele in 216/223 samples tested (97%), the remaining 7 samples genotyped as AG (Fig. 3).

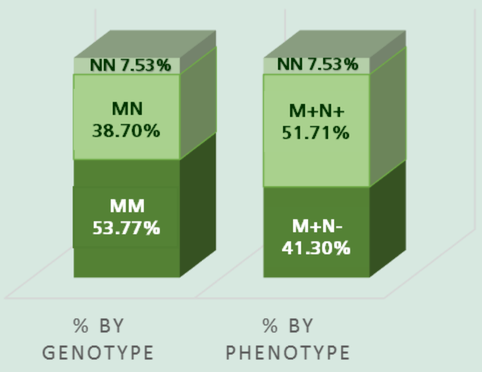



**Fig. 2** MALDI-TOF MS identified 44 *MM* samples that showed serological typing as M+N+. The discrepancies revealed glycophorin variants of which 41/44 were Mi(a+).

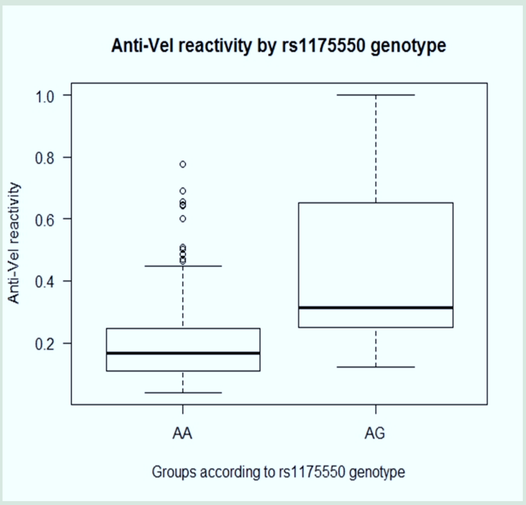



**Fig. 3** Homozygosity for rs1175550A showed lower MFI for Vel antigen expression than heterozygous samples.

**Table 1. Reactivity of the outliers' RBCs with a panel of monoclonal anti-e**

| Reagent | | Anti-e | | | | | |
|---|---|---|---|---|---|---|---|
| Source | Rh | Bio-Rad | Millipore | | | | |
| Clone / ID | phenotype | MS16/MS21/MS63 | MS16 | MS21 | MS62 | MS63 | MS69 |
| Positive control | | 4+ | 4+ | 4+ | 4+ | 4+ | 3+ |
| Negative control | | 0 | 0 | 0 | 0 | 0 | 0 |
| 16-050 | $R_1R_z$ | 4+ | 4+ | w+ | 0 | 0 | 1+w |
| 16-220 | $R_1R_z$ | 4+ | 4+ | 0 | 0 | 0 | 0 |
| 16-276 | $R_1R_2$ | 4+ | 4+ | 1+w | 0 | 0 | 2+ |

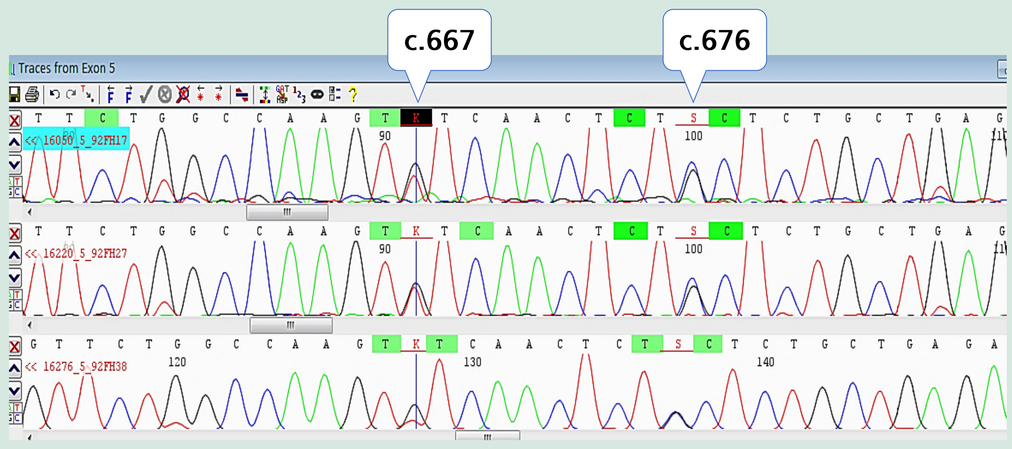



Fig. 4 Sequence analysis of RHCE exon 5 of the Rh outliers. All samples showed heterozygosity for c.676(E/e) and c.667 which defined *RHCE*02.22*

## Conclusions

MALDI-TOF MS is an efficient method for rapid genotyping and correlated well with the phenotype. We observed the expected high prevalence of the Mi(a+) phenotype in donors from both regions. However, other variants were identified indicating further diversity at this locus. Interestingly, nine samples (2.3%) carried the c.265C>T mutation on a *FY*01* allele. This allele is commonly associated with *FY*02* in other parts of the world but was recently reported in the Thai population. The Duffy protein is a known ligand for *Plasmodium vivax* which is endemic in this region. The prevalence of *FY*01* is 98.44% in our cohort and one may speculate that modification of this protein by c.265C>T may reduce susceptibility to invasion. This remains to be investigated.

### References

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