

Rare DRB1*01 with DRB5 Haplotype in a Renal Transplant Candidate

Mary Beth Wagner BS, Ashley Hawkins BS, Mary Carmelle Philogene PhD
Histocompatibility and Immunogenetics Laboratory, VCU Surgery



Introduction

Allele associations within a haplotype are routinely verified to confirm HLA typing accuracy. In the presented case, HLA typing accuracy was questioned in the case of a DRB5 allele detected in the presence of a DRB1*01 allele for a 54yo African American female.

The initial low-resolution HLA typing rendered DR1, DR7 and DR51, DR53. While DR53 and DR7 are known to be associated, the presence of a DR51 in the absence of a DR15 or DR16 raised questions regarding the validity of the result. To rule out the possibility of chimerism, a review of the patient's medical file confirmed no previous bone marrow transplantation.

Methods

DNA was extracted from the patient's blood sample using Qiagen QIAamp DNA Mini Blood Kit. DNA concentration and purity were calculated using the NanoDrop 1000 spectrophotometer.

Low-resolution HLA typing was performed using One Lambda LinkSeq ABCDRDQDP+ RT-PCR assay. High-resolution typing was performed using CareDx AlloSeq Tx 17 Next Generation Sequencing kit.

HLA typing data from donors and patients tested between 2022-2023 were extracted from the laboratory information system to determine the frequency of DR1/DR51 in this population.

Results

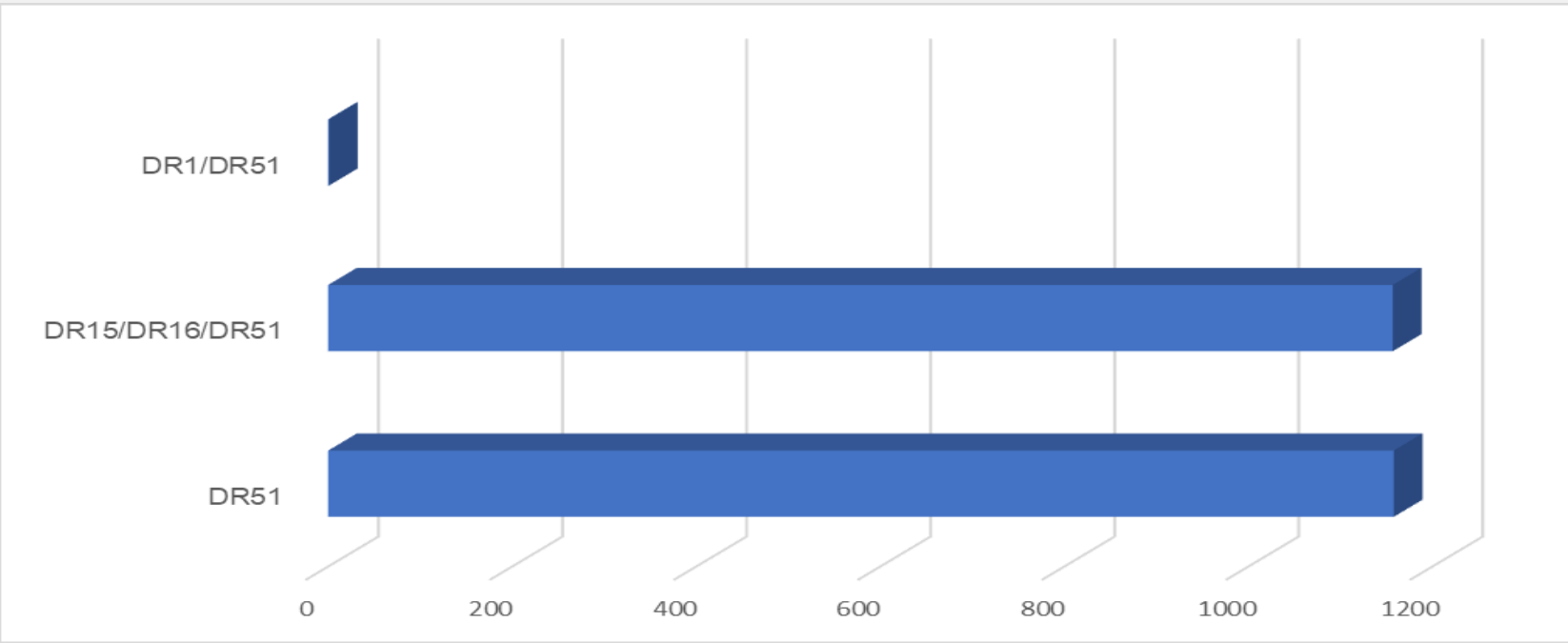


Figure 1: 4,195 HLA typing results from our database were reviewed. 1,158 samples typed as DR51 (27.60%). 1157 samples (27.58%) typed DR15 or DR16 or DR15/DR16 and DR51. The sample in question was the only sample (0.024%) in our repository that was typed as DR1, DR51.

Results

Figure 2: A DRB1*01 with DRB5 haplotype was confirmed in this patient using low-resolution and high-resolution typing methods.

HLA-A*	HLA-B*	HLA-C*	HLA-DRB1*	HLA-DQB1*	HLA-DRB3,4,5*
02:01	44:02	05:01	01:01	05:01	DRB5*01:01
30:02	51:01	16:01	07:01	02:02	DRB4*01:01

Figure 3: SureTyper detected multiple positive DRB5 wells for this patient using the LinkSeq ABCDRDQDP+ typing kits. Shown below is a result from one assay showing the strong melt curve that is also visible on multiple assays that include primers for DRB5.

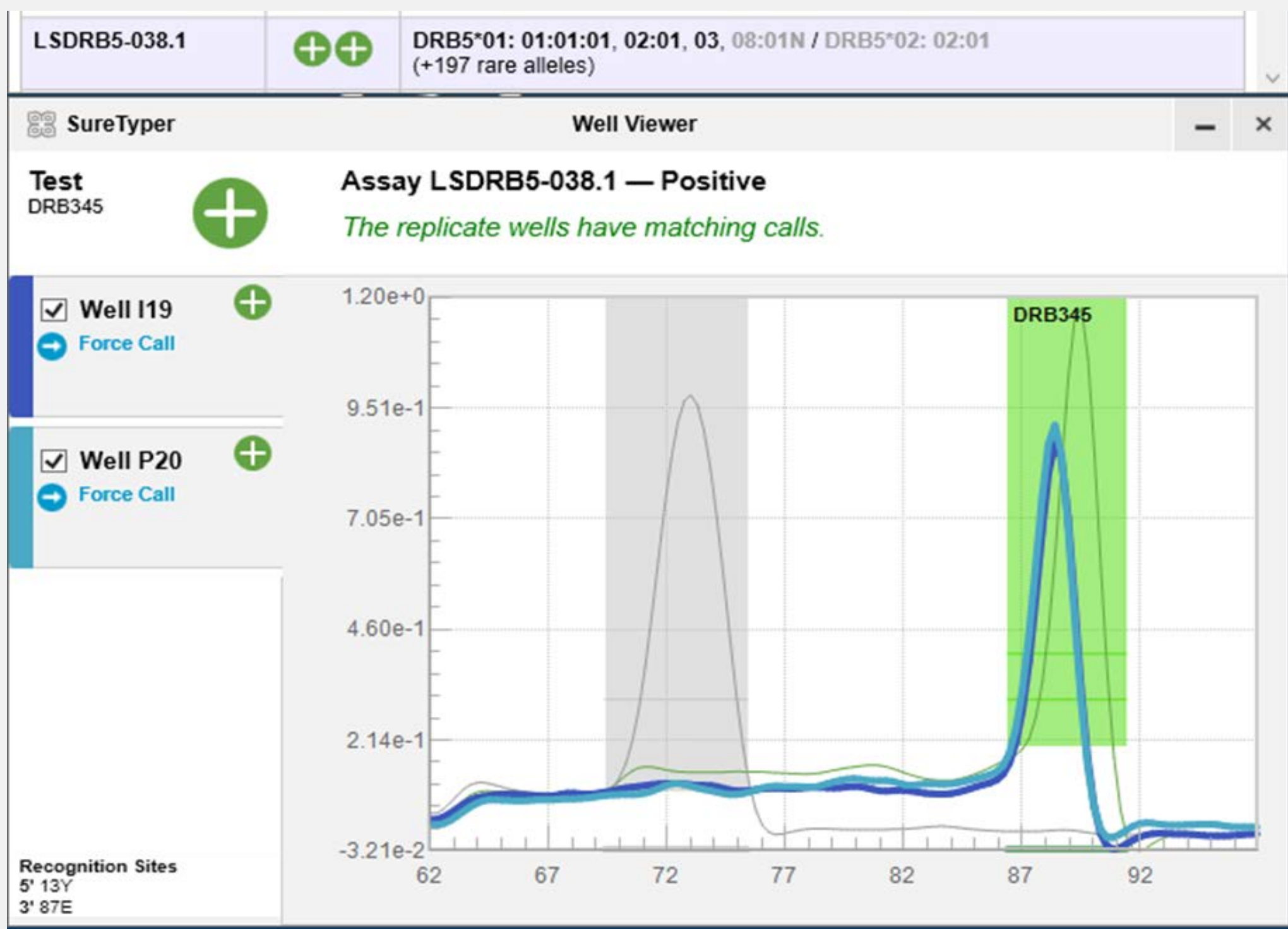
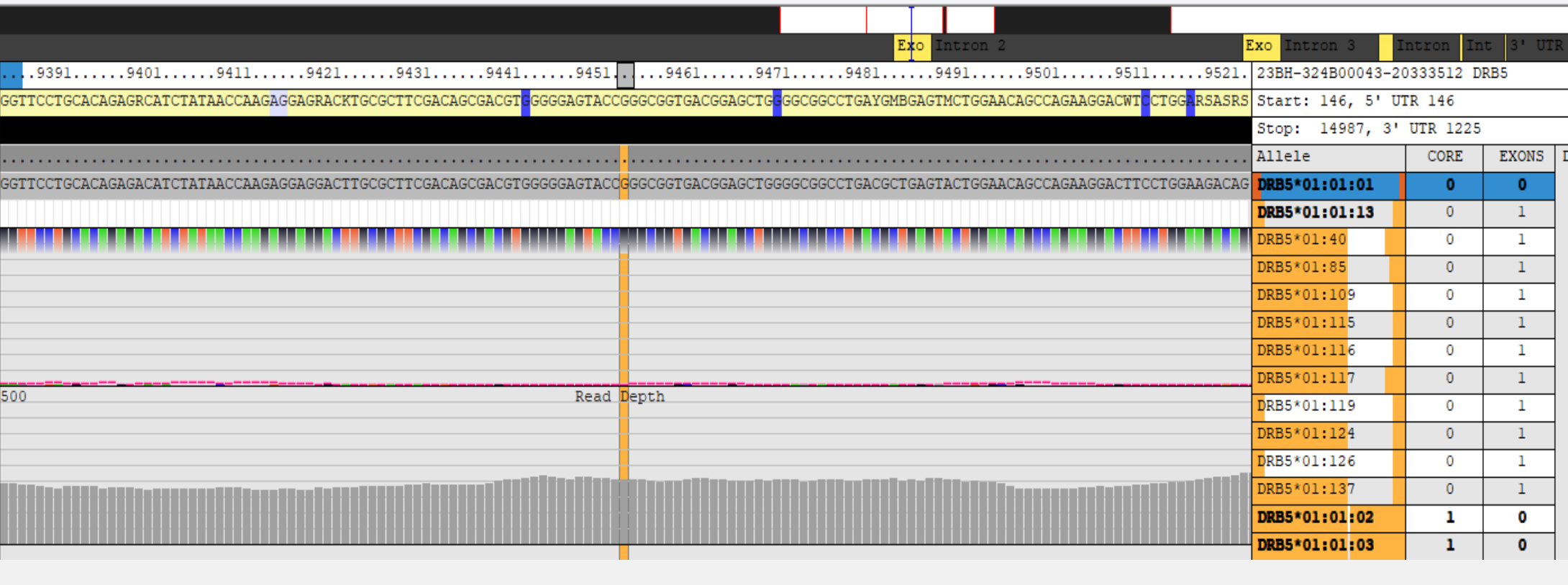


Figure 4: A portion of the exon 2 sequence for DRB5 (DR51) is shown. The AlloSeq Assign software reports a robust Mean Read Depth of 195 for the DRB5 gene (acceptable Mean Read Depth is 50 for hemizygous loci).



Discussion

A DR1, DR51 association was confirmed in one patient. Review of HLA typing records in the last 2 years corroborated that this association is rare.

The DRB subregion of the human major histocompatibility complex includes several expressed DRB genes (DRB1, DRB3, DRB4, and DRB5). While some haplotypes may express only one of these genes (e.g., DRB1*01, DRB1*08 or DRB1*10 do not express a second gene), other haplotypes express 2 of the genes (e.g., DRB1*15 also expresses DRB5). These haplotypes are thought to result from gene duplication and divergence, whereby at some point during evolution, an ancestral DRB1*15 acquired a DRB5 gene.

Based on studies from Satta et al. (Satta Y et al. 1996), the ancestral DRB1*01 gene did not undergo duplication. More recent studies are showing that, in fact, the DRB region is expanding with discovery of new haplotypes (DRB1*15 with a DRB5 null).

A search of the haplotype frequency database (haplostats.org) revealed that this haplotype is most frequent in African American populations at 5.8X10⁻⁸ and significantly less frequent in other populations. This DR1, DR51 association should be discussed among the HLA community.

Contact

Ashley Hawkins, Ashley.Tiedemann@vcuhealth.org

Beth Wagner, Mary.Wagner@vcuhealth.org

References

Satta Y, Mayer WE, Klein J. HLA-DRB intron 1 sequences: implications for the evolution of HLA-DRB genes and haplotypes. Hum Immunol. 1996 Nov;51(1):1-12. doi: 10.1016/s0198-8859(96)00155-3. PMID: 8911992.

Haplostats.org