Assessing the utility of HISTO SPOT® HLA AB for interpreting HLA antibody profiles compared with Luminex-based platforms

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INTRODUCTION

Luminex-based platforms have revolutionised the detection of anti-HLA antibodies. However, single antigen beads (SAB) reactions can sometimes give rise to false positive results that can potentially interfere the assignment of specific antibodies. Recently, a new platform has been established that utilises a single antigen chip array to detect HLA antibodies, called HISTO SPOT® HLA AB. The HISTO SPOT HLA AB test is a miniaturised and multiplexed solid phase enzyme linked immunosorbent assay. Each well of a microtiter plate is printed with recombinant HLA antigens in individual spots (see figure 1). The test is performed using an automated microarray processor, MR SPOT® 2.0 and sample analysis performed with HISTO MATCH HLA AB software. The purpose of this study was to assess the performance of HISTO SPOT® HLA AB when testing a panel of known questionable antibody profiles generated using LABScreen platform and serum samples from the 2023 APHIA Quality Assurance program (QAP).

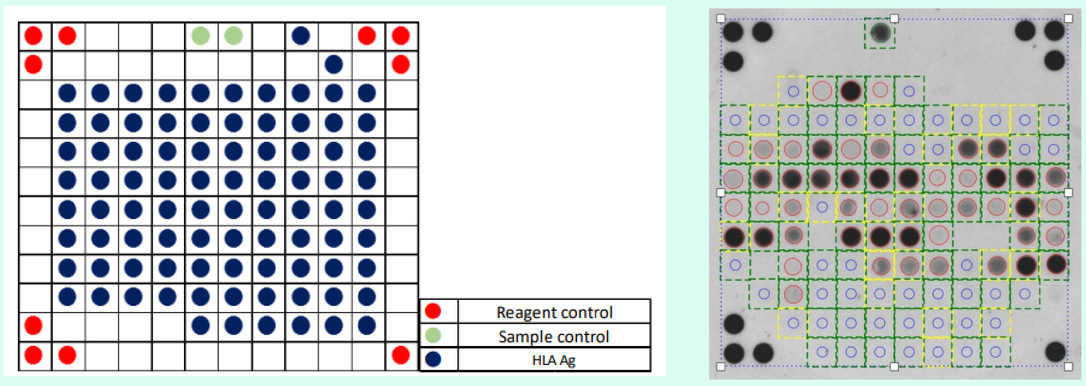

Figure 1: Schematic representation of the HISTO SPOT® HLA AB ID Class I (Left) and example of a sample with Class I Antibodies (Right).

METHOD

Part 1: Serum from the 2023 APHIA Antibody QAP March to August sendouts (n=9) were tested using HISTO SPOT® HLA AB Class I and Class II tests and results compared with the reported consensus specificities.

Part 2: Patient serum (n=14) with known questionable antibody profiles using LABScreen were tested using HISTO SPOT® HLA AB Class I or Class II tests and results compared with Halifaster Flow XM and/or Adsorption Crossmatch Cells and Elution (AXE) results.

RESULTS

Part 1: HISTO SPOT® HLA AB demonstrated good concordance with the reported 262 Class I and 52 Class II APHIA consensus specificities across 9 serum tested. A total of 250 Class I and 46 Class II specificities were detected using HISTO SPOT® HLA AB (see Figure 2). All tests achieved a concordance of ≥80% with consensus, which would satisfy the programs guidelines.

RESULTS

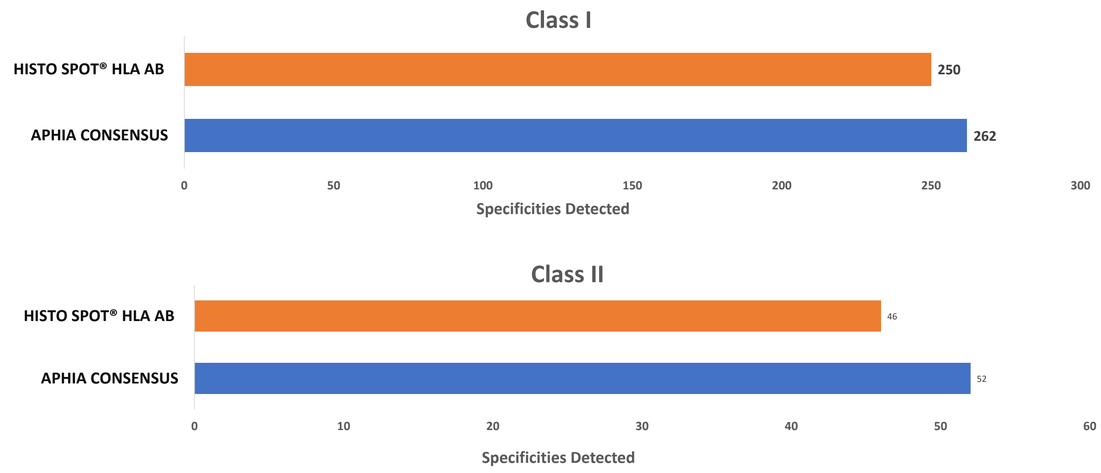

Figure 2: Performance with APHIA QAP serum

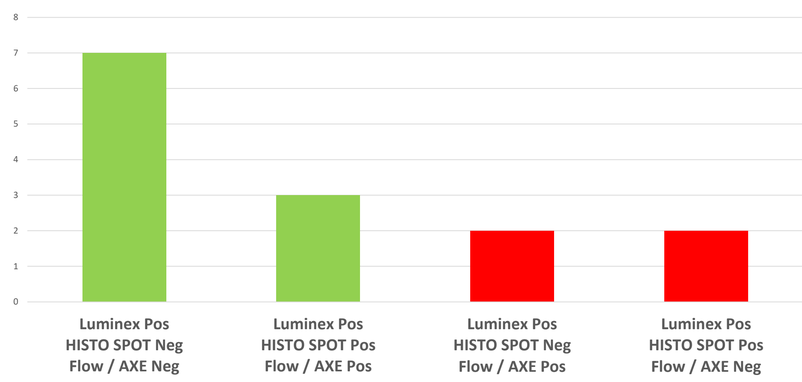

Figure 3: Summary Of Results

Part 2: 10/14 serum with known questionable antibody profiles tested with HISTO SPOT® HLA AB were concordant with Halifaster Flow XM and/or AXE results. Of these 10 concordant results, 7 confirmed the LABScreen result as spurious (Negative HISTO SPOT and Negative Flow XM/AXE Results) and 3 confirmed the LABScreen results are real (Positive HISTO SPOT® HLA AB and Flow XM/AXE results). 4/14 serum with known questionable antibody profiles tested with HISTO SPOT® HLA AB were not concordant with Halifaster Flow XM and/or AXE results.

CONCLUSION

This study has demonstrated that the HISTO SPOT® HLA AB method gives broadly consistent results with the Luminex Based platforms. There does appear to be some utility in using HISTO SPOT® HLA AB as a complementary method when investigating questionable antibody profiles generated on Luminex based platforms. However, the method also has its own spurious positive reactions. Some variations in the antigen panels do exist and the most notable is with Class II antigens and in particular a reduction in DQB/DQA and DPB/DPA combinations. This may cause challenges with Class II antibody assignment. Further studies are recommended to determine the utility of HISTO SPOT® HLA AB as an alternative method to Luminex based systems.

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DISCLAIMER

The following relevant financial relationships existed related to this presentation: no relationships to disclose.