

HLA A10 SPURIOUS REACTIVITY WITH SINGLE ANTIGEN BEADS INVESTIGATED

Maureen M. Miller, Maha Taha-Furst, John Raposa, Laurine M. Bow, PhD; Yale University School of Medicine, New Haven, CT



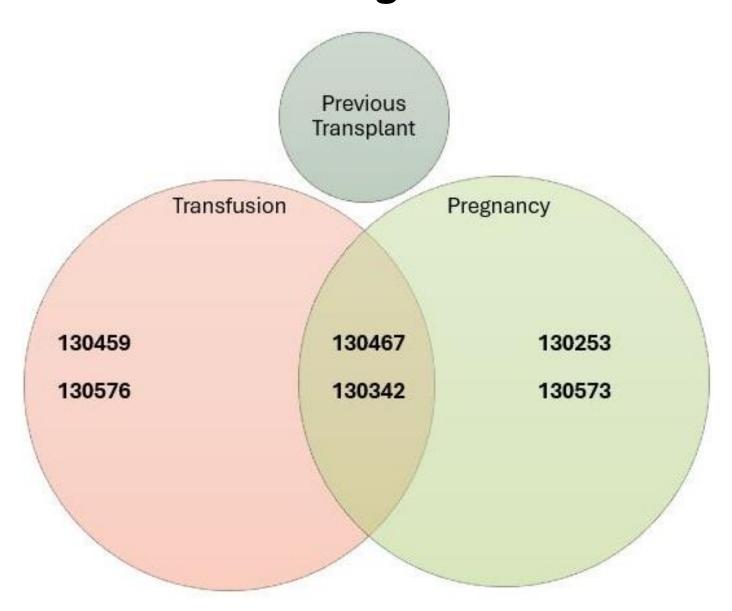
AIM

To review pre-transplant kidney and heart patients with spurious reactivity to HLA A10 (A25,26,34,66,43) on the single antigen bead assay and determine if the reactivity should be listed as unacceptable antigens in UNET. Accurately defined unacceptable antigens are critical to our workflow since most of our transplants are performed with virtual crossmatches. cPRA for A10 is 13%.

METHODS

Ten pre-transplant patients with antibody reactivity to some or all the A10 antigens were selected. The antibody testing results were examined and sensitizing events including transfusions, pregnancies, and previous transplants were reviewed. MFI of the antibody results ranged from 11,000 to 1,100. Antibody testing was performed using LABScreen™ Single Antigen, LABScreen™ PRA and LIFECODES® Single Antigen and ID kits. Crossmatches were performed by flow cytometry (FCXM) on pronase treated, T and B cells using a BD FACSLyric.

Sensitizing Events



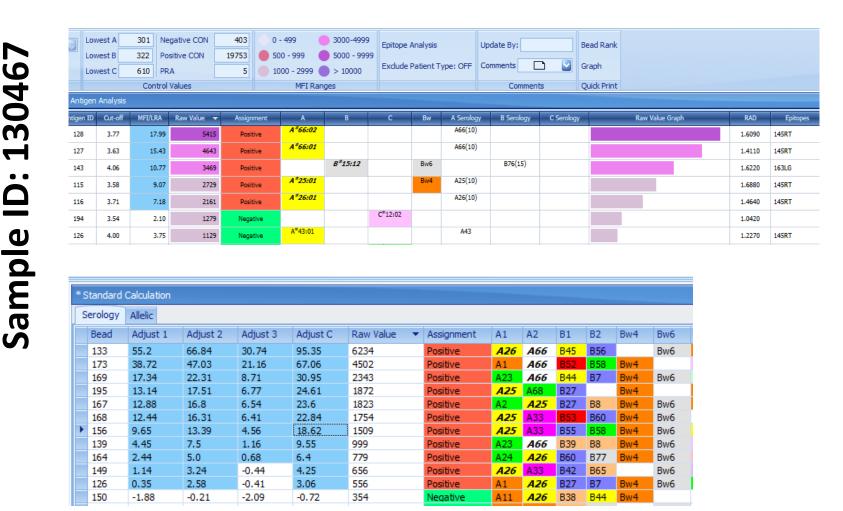
| Sensitization and Antibody Results Summary | | | | | | | | | |
|--|-----------|-----|-------|------|-----|------|------|-------|---|
| | Sample ID | Тхр | Txf | Preg | SA | ID | LSA | LC ID | Comments (SA MFI if available) |
| 1 | 130467 | NO | YES | 2 | POS | POS | POS | POS | A66 (10,000), 34:01 (10,000), 25 (7,600), A26 (5500), A43 (3900) |
| 2 | 130459 | NO | 19 | NA | POS | NEG | POS | POS | A25 (10,500), A26 (4,000), A43 (5,600), A66:01 (8,400) |
| 3 | 130290 | NO | NO | 0 | POS | NEG | NEG | NEG | A26(2400) |
| 4 | 129927 | NO | NO | NA | POS | POS | POS* | NEG | A34:01(4300), A66:01(4200), A25(1200); LSA only A*66:01 (1201) |
| 5 | 130253 | NO | NO | 1 | POS | NEG* | POS | POS | A25 (9400) |
| | | | | | | | | | |
| 6 | 130344 | NO | NO | NA | POS | NT | POS | POS | A43(11000), A26(11000), A25(8300), A66(8000), A34:01(5500) |
| 7 | 129166 | NO | NO | NA | POS | POS | NT | POS | A43(6400), A26(5500), A25(8300), A66(4000), A34:01(3400), A25(2400) |
| 8 | 130573 | NO | NO | 5 | POS | NEG | POS | POS | A66:02 (5500) with B7C,163W epitope on ID too. |
| | | | | | | | | | A34(1800), A26(1100), A66:01(1100), A25(1000); other reactivity includes A33, |
| 9 | 130342 | NO | MULTI | 2 | POS | POS | POS | POS | 68,69; and 44RMA (B13,62,75,76) |
| 10 | 130576 | NO | 2 | NΔ | POS | NEG | NO | POS | Δ26(1100), Δ66(600), Δ25(570) |

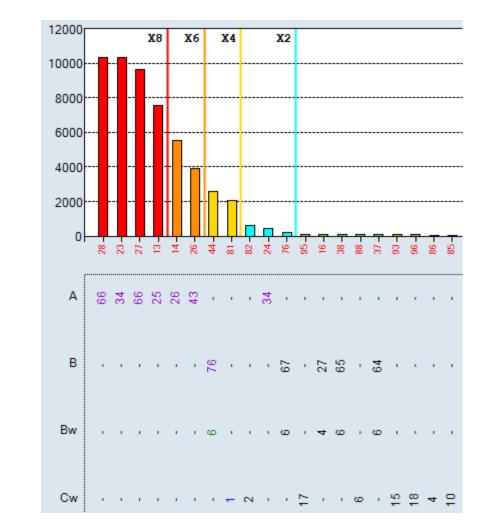
Results

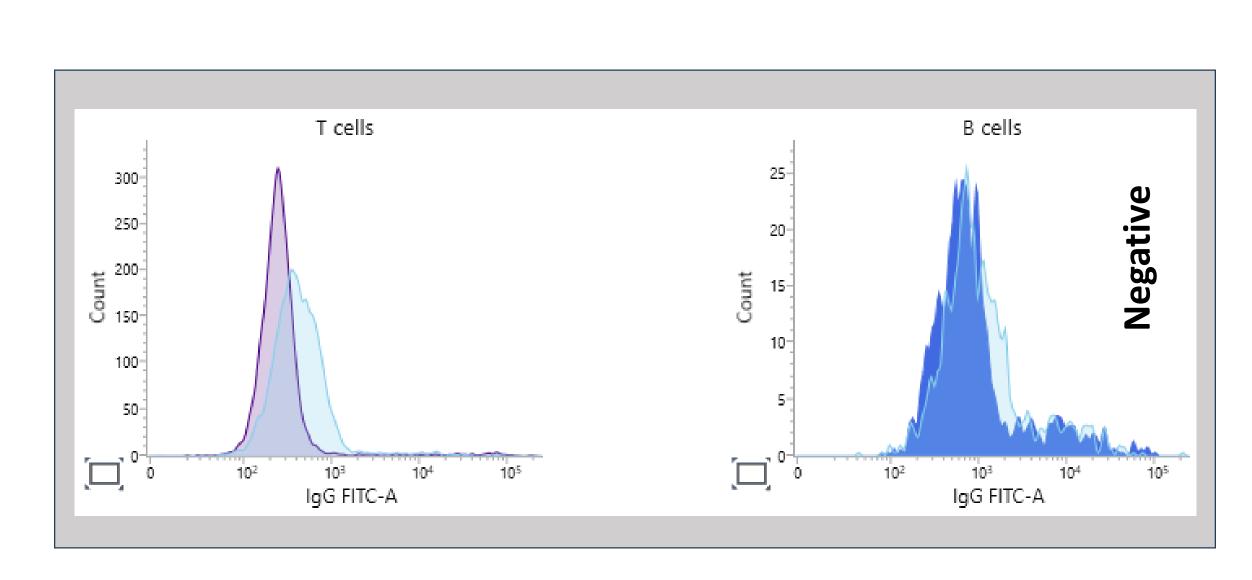
Six out of the ten patients had sensitizing events, none had previous transplants, 2 received transfusions, 2 had pregnancies, and 2 had both transfusions and pregnancies. The summary of the sensitizing events and antibody testing is attached in table 1. Six cells were tested against 10 samples for a total of 60 flow crossmatches. Fifty-six (93%) crossmatches were negative. Four crossmatches were positive, both recipients with the positive crossmatches had sensitizing events. The positive crossmatches were most likely due to a broad epitope antibody that contained some of the A10 antigens. The two epitopes that were identified on the antibody testing were 163EW and 62RR. The MFI detected on the antibody testing did not predict the positive flow crossmatch result. Eighty percent (8/10) of the patients with antibody reactivity to A10 had negative flow crossmatches.

Donor HLA type: A*01:01, 34:02; B*08:01, 51:01; C*07:01, 16:01; DRB1*03:01P, 09:01P; DRB3*01:01P; DRB4*01:01P; DQA1*03:03, 05:01; DQB1*02:01, 02:02; DPA1*01:03, 02:02; DPB1*01:01P, 04:01P

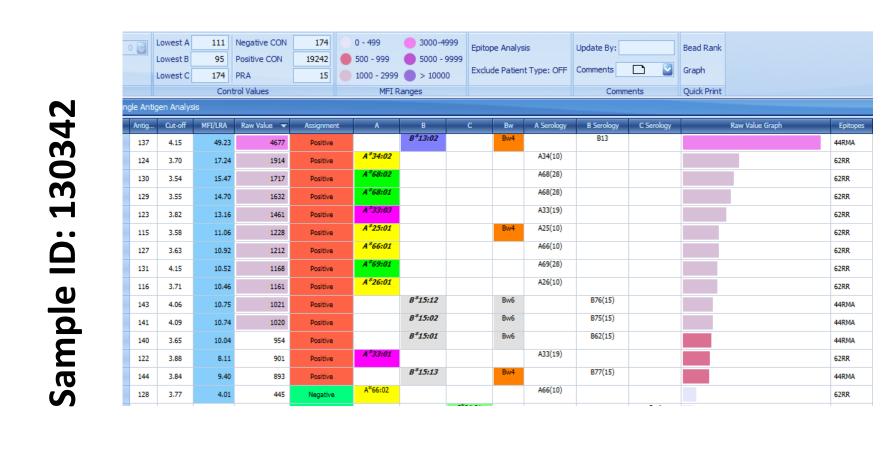
Antibody Data

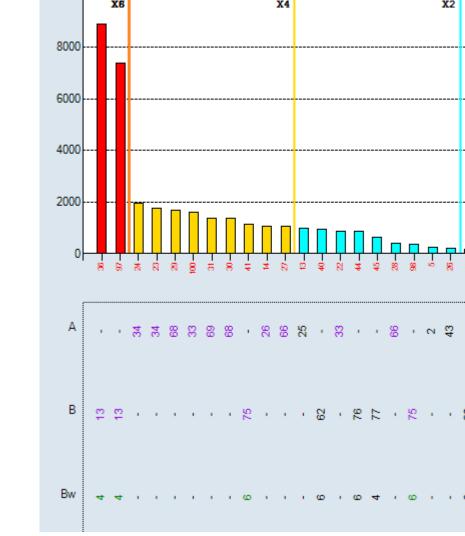


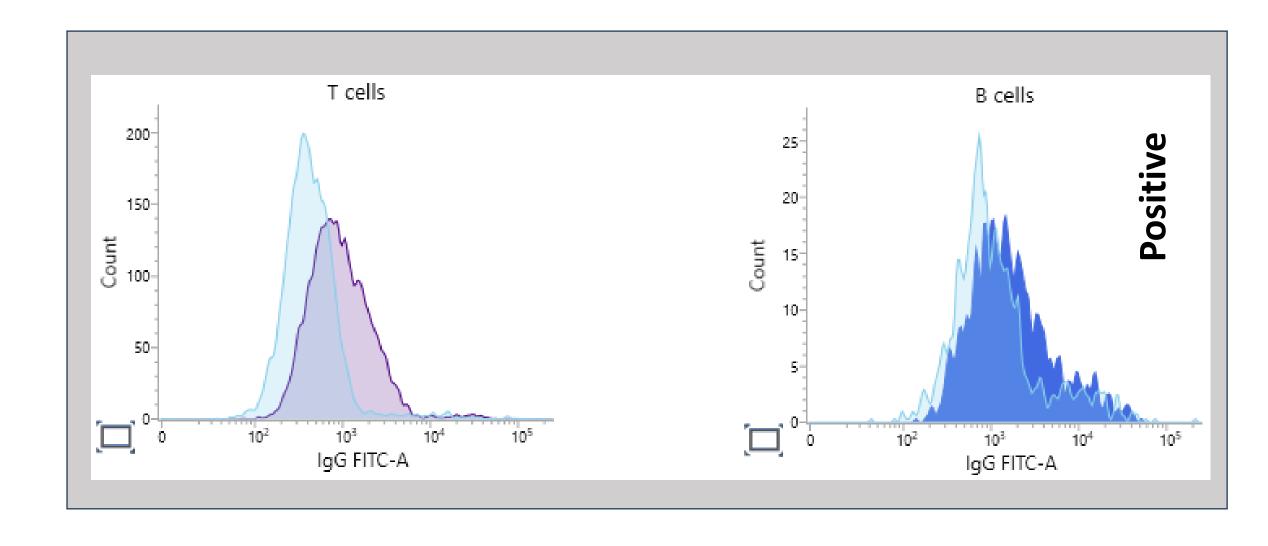




FCXM







Conclusion

Spurious reactivity on the single antigen antibody tests have been reported for years. We investigated the A10 reactivity in 10 pre-transplant patients using flow crossmatches and determined that 8 out of 10 recipients had multiple negative flow crossmatches. The unacceptable antigens were removed from UNET for these patients.