



Should PAK LX HLA Class I Bead Be Used As A Stand-Alone Assay For Platelet Transfusion?

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Background:

In a previous study we have shown that PAK LX (a Luminex based assay that screens for platelet-specific glycoproteins and HLA class I antibodies) reactivity to HLA antibodies may not be an accurate indicator of HLA immunization, as it showed a 37% discrepancy with single antigen Luminex based assay (SAG) in a single center small size cohort making it risky to use as a standalone assay for HLA class I antibody screening.

Objective:

In the current study we have extended the study cohort to further investigate this phenomenon and identify the trends governing this observed discrepancy.

Methods:

A cohort of 137 individuals from two American National Red Cross Histocompatibility laboratories in both Charlotte, NC and Portland, OR, underwent screening using both PAK LX (Werfen, Inc) and SAG (Thermofisher, Inc) methods. Demographic data, antibody positivity rates, discordance rates, mean fluorescence intensity (MFI) values, and calculated percent reactive antibodies (cPRA) were obtained.

Results:

The study population comprised 51% female and 49% male individuals, with a majority being Caucasian (52%) and 34% diagnosed with leukemia. Seventy-three percent tested positive for HLA class I antibodies by SAG, while 36% showed discordant results between PAK LX and SAG. Most discordant cases were false negatives (98%) from PAK LX, with two cases false positive. The mean cPRA of discordant cases was 29% (range 0.02-96%). Class I ABC antibodies showed mean MFI values of 4500 (range 1,000-17,000), 4079 (range 1,100-18,000), 3758 (range 1,200 – 19,000), respectively. The MFI of class I antibodies among discrepant cases was mainly for specificities with MFI below 6000 with a broad range from 2000 to 19,000.

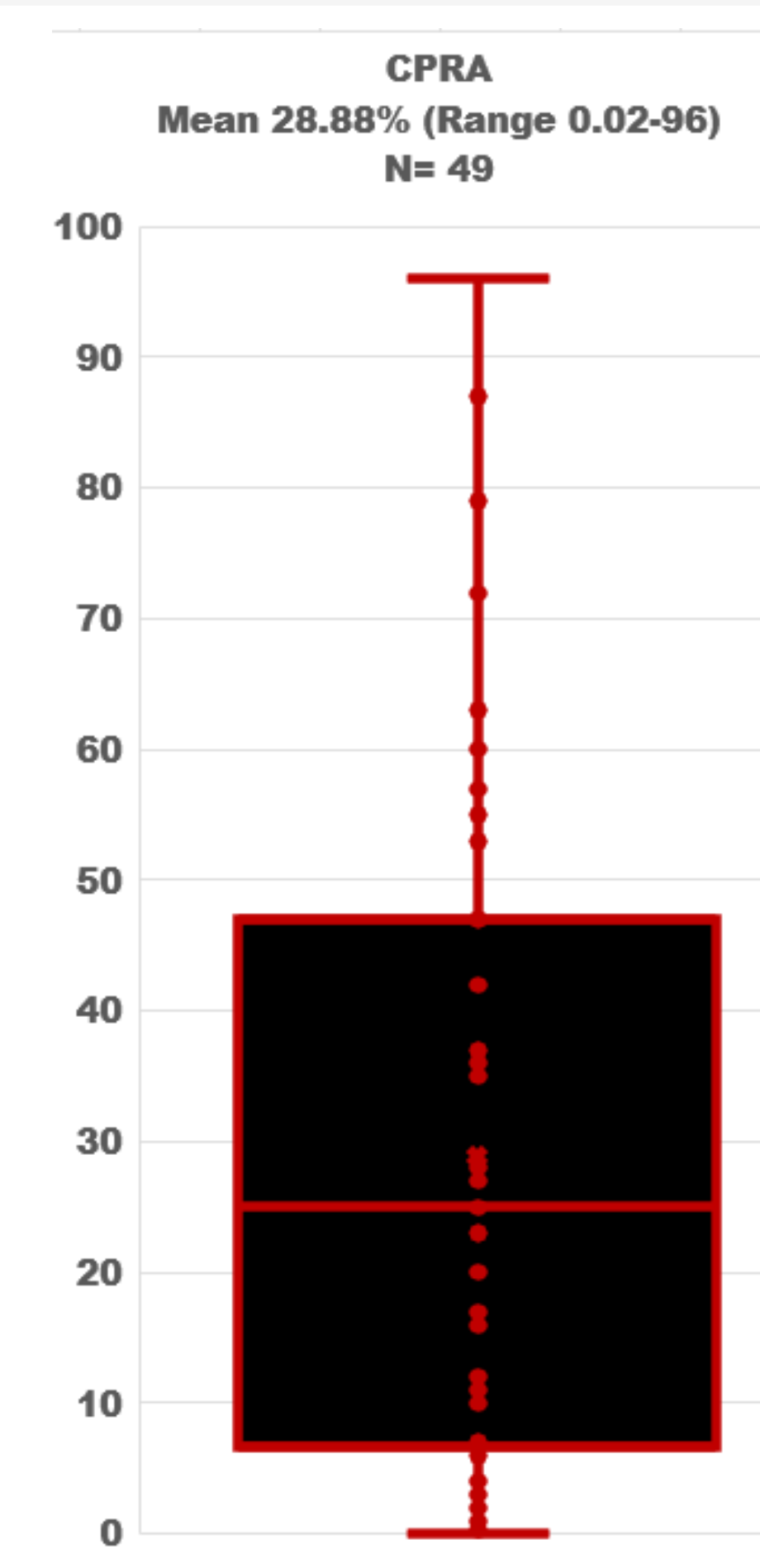


Figure 1A

Figure 1A:

A box-and-whisker plot distribution of cPRA values based on Anti-HLA Class I antibodies detected by SAB but not showing reactivity on PAK-LX.

Figure 1B:

A box-and-whisker distribution of mean fluorescence intensity (MFI) values for class I specificities detected by SAG and missed by PAK-LX assay.

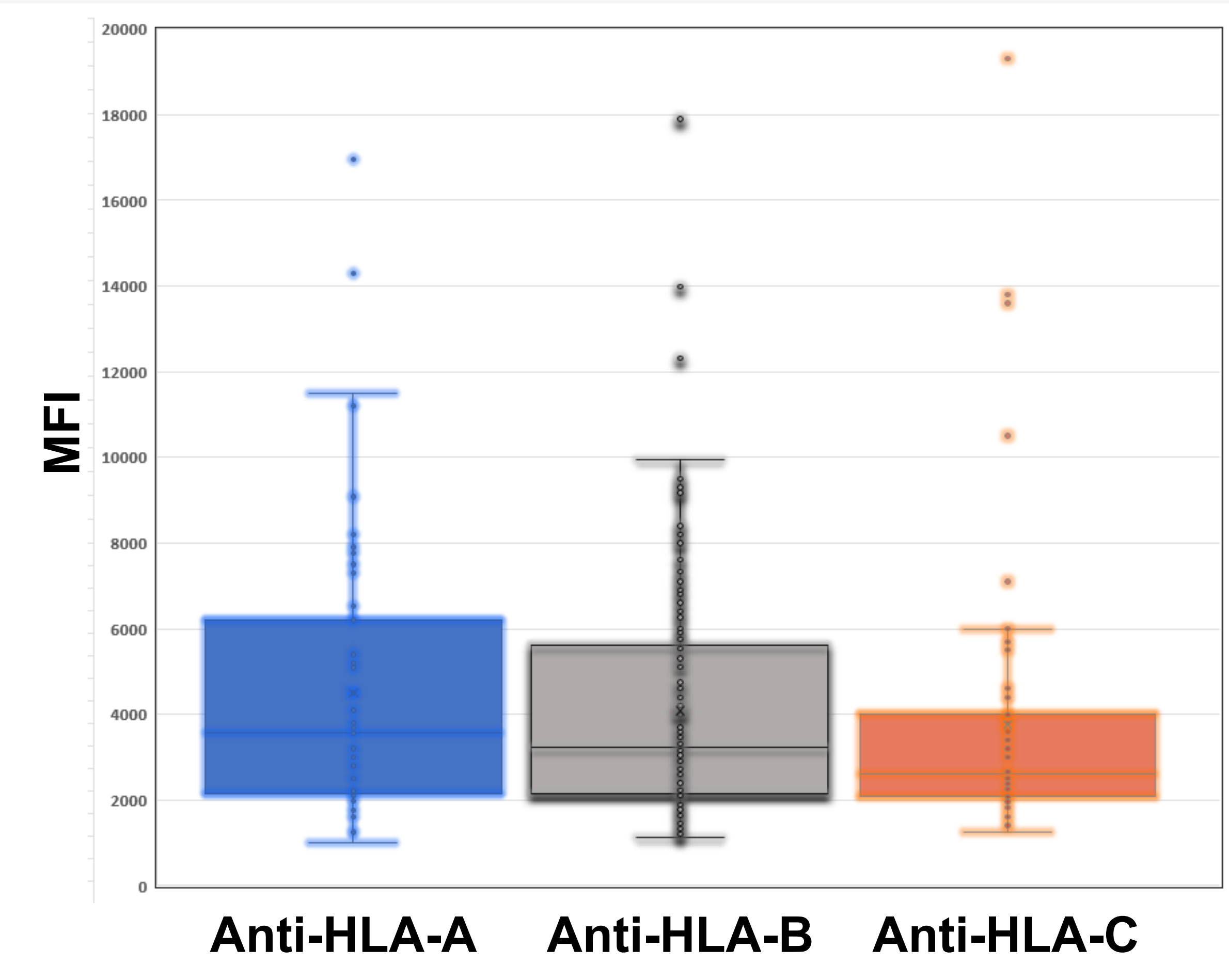


Figure 1B

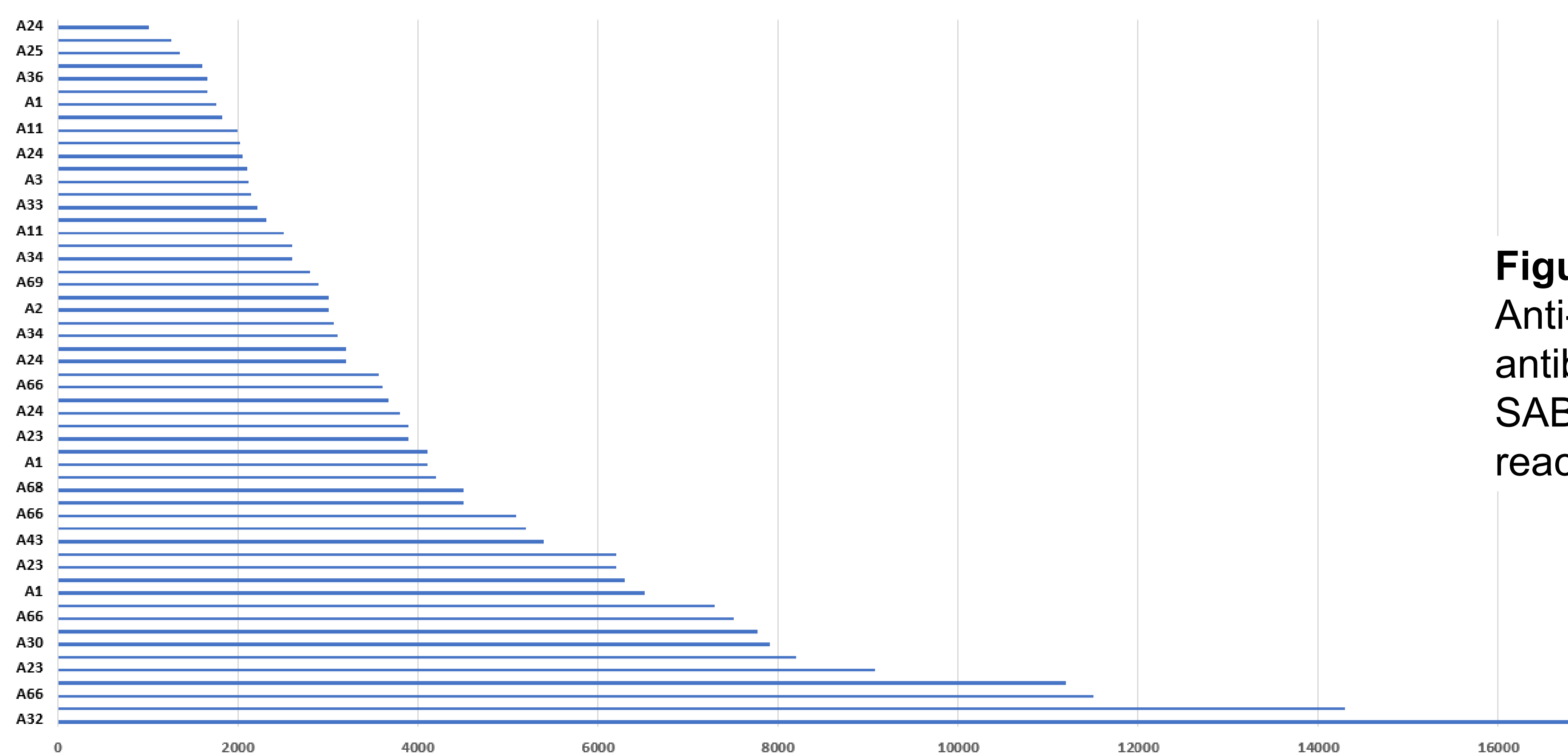


Figure 2A: Class I A specificities and MFIs

Figure 2 A&B:

Anti-HLA-A & B Class I antibodies (MFI) detected by SAB but not showing reactivity on PAK-LX assay.

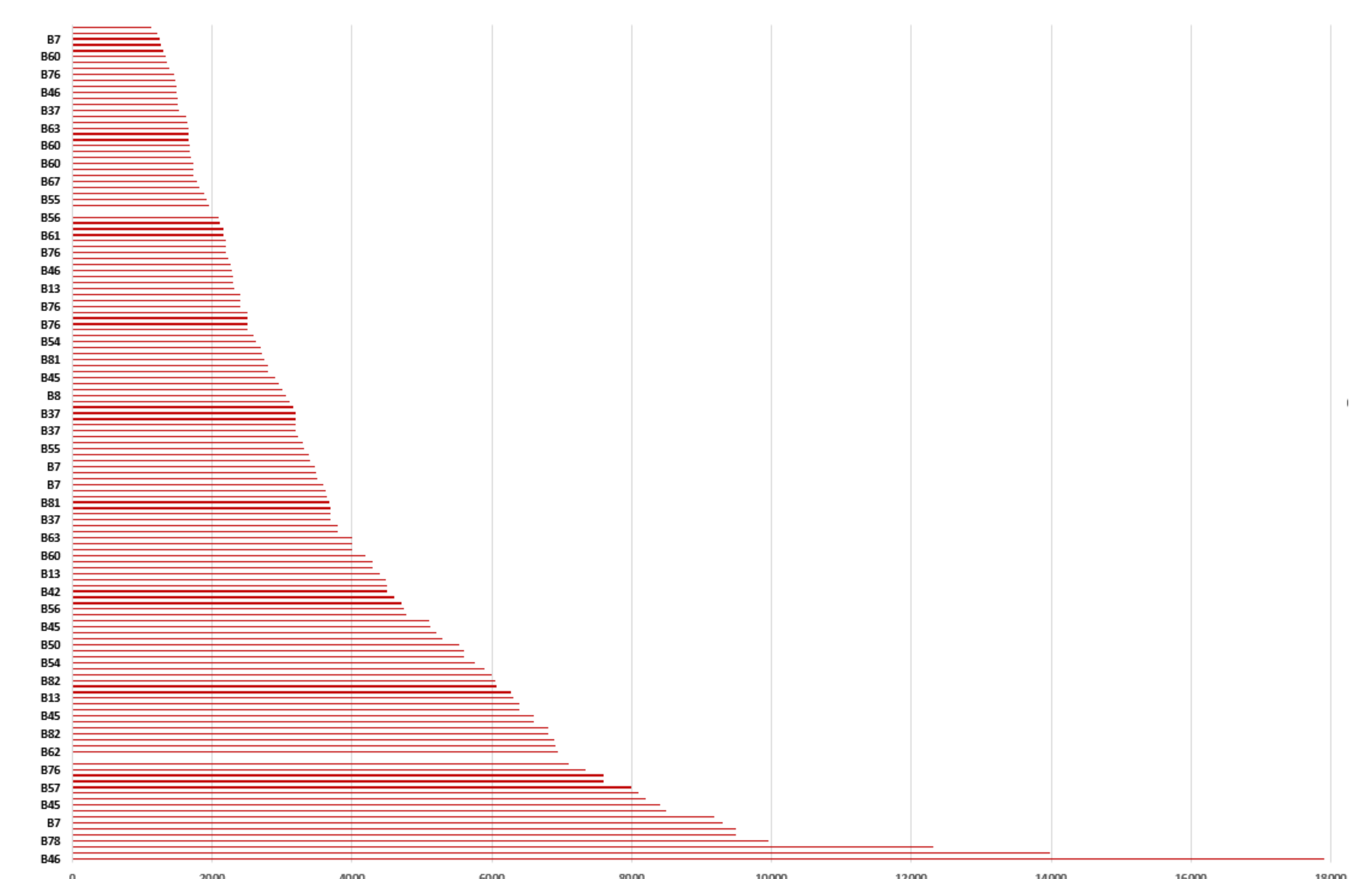


Figure 2B. Class I B specificities and MFIs

Conclusion:

Missing HLA class I antibody specificity is a significant risk factor for platelet transfusion refractoriness that may lead to ineffective treatment and increase the risk of bleeding. Our current study confirmed previous observation that PAK LX HLA class I bead should not be used as a standalone method for detecting HLA class I antibody. It also gave some insight into the nature of HLA class I specificities missed by PAK LX.

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