# (F)Utility of Flow Crossmatch in the Presence of Weak Donor-Specific Antibodies

**Elaine Chou-Wu, Danny Youngs, Idoia Gimferrer**

Immunogenetics/HLA Laboratory, Bloodworks Northwest, Seattle, WA, USA

## Introduction

In the presence of weak donor-specific HLA antibodies (DSAs), transplant teams often rely on prospective flow cytometric crossmatch (FXM) results to guide organ acceptance decisions. This study sought to assess the association between FXM reactivity and the characteristics of pre-transplant weak DSA, and to evaluate the clinical utility of FXM in predicting early post-transplant DSA kinetics.

## Objectives

- ❑ Evaluate the association between pre-transplant weak DSA characteristics and FXM reactivity.
- ❑ Determine whether weak DSA MFI can predict FXM reactivity.
- ❑ Assess the utility of FXM in predicting early post-transplant DSA persistence.

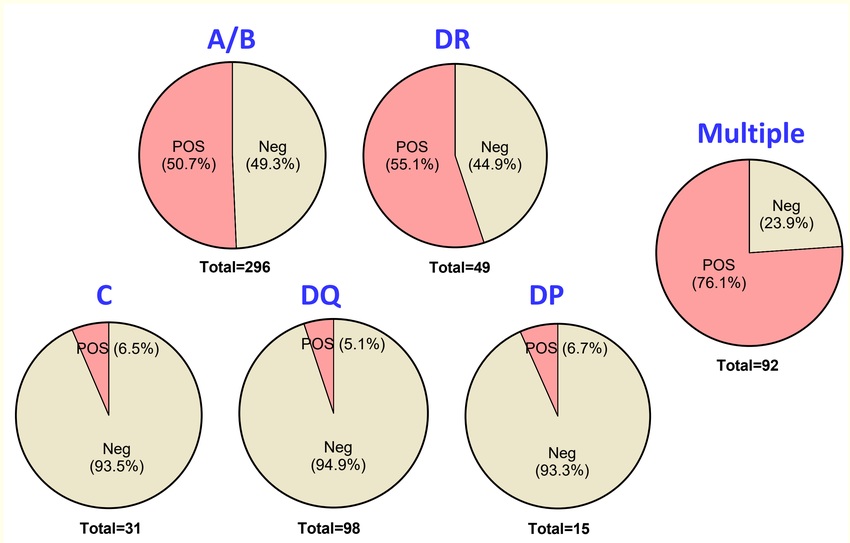
## Methods

A total of 489 prospective FXMs performed between 04/2024 and 01/2026 in the presence of single weak DSAs (defined as MFI < 2,000) were analyzed to evaluate the association between DSA characteristics and FXM positivity. Early post-transplant DSA persistence was assessed in a separate cohort of 181 patients who underwent solitary kidney transplantation between 2015 and 2025 in the presence of preformed weak DSAs. DSA persistence at 3 months and 1 year post-transplant was compared between FXM- and FXM+ transplants using the chi-square test.

## Results

### ➢ Percentage of Positive FXM by DSA Loci

As expected, single weak DSAs directed against lower-expressed HLA loci (HLA-C, -DQ, and -DP) predominantly associate with negative FXMs. Approximately 50% of FXMs were positive in the presence of single weak DSAs against HLA-A, -B, or -DRB1 antigens, highlighting the substantial variability in FXM outcomes and demonstrating that DSA MFI alone is a poor predictor of FXM reactivity. FXM positivity increased markedly when multiple DSAs were present.




### ➢ Pre-Txp FXM Reactivity and 3-Month Post-Txp DSA Persistence

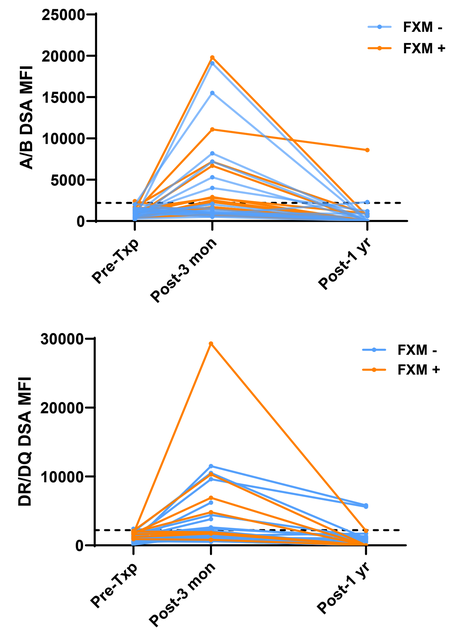
Within the first three-month post-transplant, DSA persistence was more common for antibodies targeting class II HLA-DRB1 (52%) and -DQB1 loci (47%), compared to those targeting HLA-A/B antigens (31%). Importantly, no significant difference in DSA persistence was observed between recipients with positive and negative pre-txp FXM results, suggesting that FXM reactivity was not predictive of early post-txp DSA persistence.

| | A/B DSA Persistence | | | DRB1 DSA Persistence | | | DQB1 DSA Persistence | | |
|---|---|---|---|---|---|---|---|---|---|
| | Negative | Positive | | Negative | Positive | | Negative | Positive | |
| Total (n) | 73 (69%) | 33 **(31%)** | | 12 (48%) | 13 **(52%)** | | 26 (53%) | 23 **(47%)** | |
| **FXM–** | 45 (70%) | 19 **(30%)** | p = 0.693 | 10 (53%) | 9 **(47%)** | p = 0.645 | 23 (55%) | 19 **(45%)** | p = 0.861 |
| **FXM+** | 28 (67%) | 14 **(33%)** | | 2 (33%) | 4 **(67%)** | | 3 (43%) | 4 **(57%)** | |

## Results-cont.

### ➢ Pre-Txp FXM Reactivity and 1-year Post-Txp DSA Persistence

Although some DSAs demonstrated early post-txp rebound, persistence of antibodies at levels >2,000 MFI at 1 year was uncommon, occurring in only 1/33 (3%) HLA-A/B DSAs and 2/36 (5.6%) HLA-DRB1/DQB1 DSAs. No clear correlation was observed between pre-transplant FXM reactivity and 1-year post-transplant DSA persistence.




## Conclusions

- ❑ The relationship between weak DSAs and FXM reactivity is locus-dependent, with unpredictable FXM outcomes for HLA-A, -B, and -DRB1 DSAs and predominantly negative FXM results for HLA-C, -DQ, and -DP DSAs.
- ❑ FXM reactivity does not predict DSA persistence at either 3 months or 1 year post-transplant.
- ❑ These findings question the clinical utility of prospective FXM as a determinant of organ offer acceptance in the setting of weak DSAs.

Bloodworks Northwest