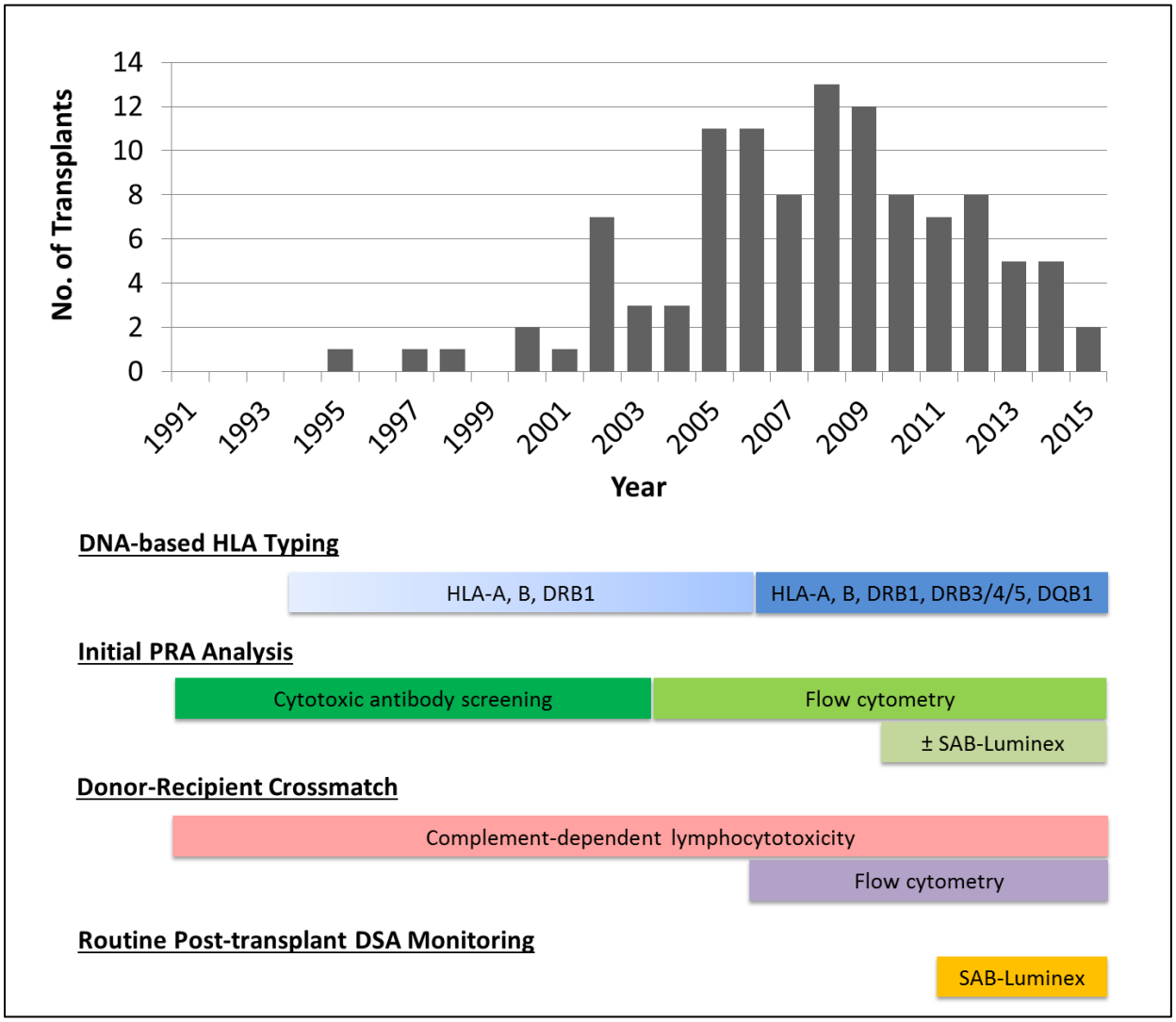
SUPPLEMENTAL DIGITAL CONTENT



This schematic illustrates the evolution of HLA typing and antibody testing methods over the analysis period. Also shown are the numbers of patients transplanted in each given year.

- ***HLA Typing*** – Our Tissue Typing Lab transitioned from serological to molecular methods of HLA typing in the early 1990s. Since the first patient in the analysis cohort was transplanted in 1995, all recipients were typed at the HLA-A, -B and –DRB1 loci using DNA-based methods. DRB3/4/5 and DQB1 typing were routinely performed for all transplants beginning in 2006.
- ***Initial PRA Screen*** – The pretransplant evaluation included determination of PRA by cytotoxic antibody screening in the early years, but is now measured by the flow cytometry. When Luminex single-antigen bead testing became available in 2010, candidates with positive PRA by flow cytometry underwent Luminex testing to determine antibody specificities and strengths.

- ***Donor-Recipient Crossmatch*** – At the time of transplantation, donor-recipient T- and B-cell CDC crossmatches (XM) are performed. This was supplemented with T- and B-cell flow cytometry XMs beginning in 2006. A mean channel fluorescence shift of >50 channels for the T-cell peak, or >150 channels for the B-cell peak, constituted a positive flow cytometry XM. A pronase-treated crossmatch is performed if the untreated crossmatch is positive in the absence of DSA.
- ***Post-transplant DSA Monitoring*** – Routine monitoring for DSA post-transplant began in late 2011, and recipients underwent testing with the Luminex single-antigen bead technique at 1, 3, 6, 12 months after ITx, and semiannually thereafter. Additional testing may be performed if clinically-indicated. High-risk recipients are also tested more frequently within the first post-transplant year according to protocol. An MFI value of 1000 or greater was used as the threshold for DSA-positivity.