

Supporting information

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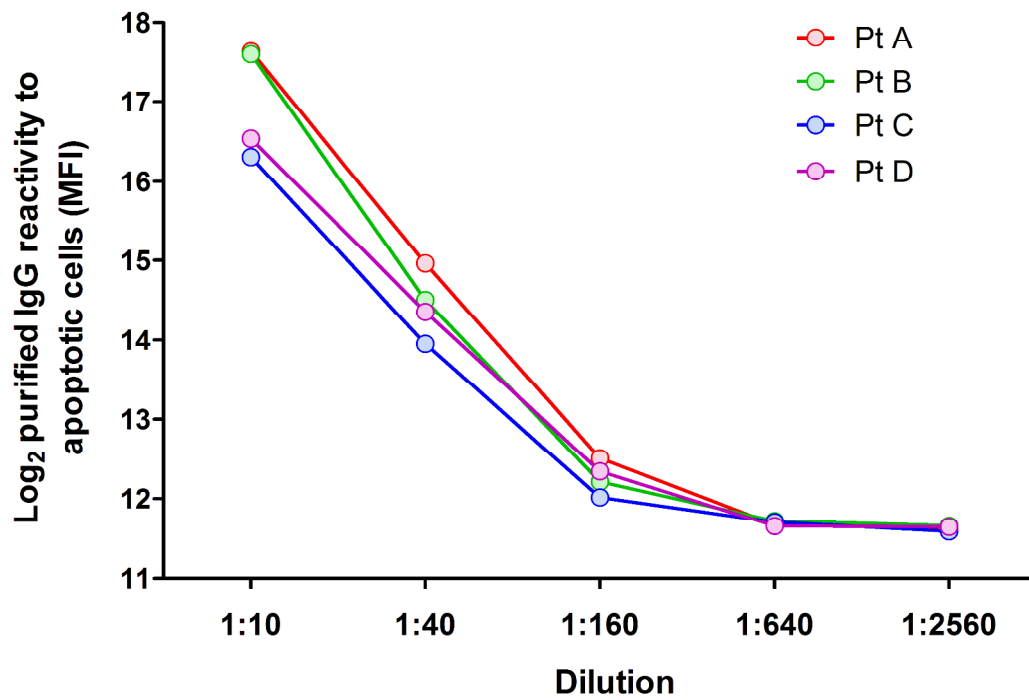


Figure S1. Titration of purified serum IgG reactivity to apoptotic Jurkat cells.

Serum samples from 4 patients were assessed by flow cytometry for their reactivity to apoptotic cells after serial dilution. Results are reported as \log_2 values of the MFI.

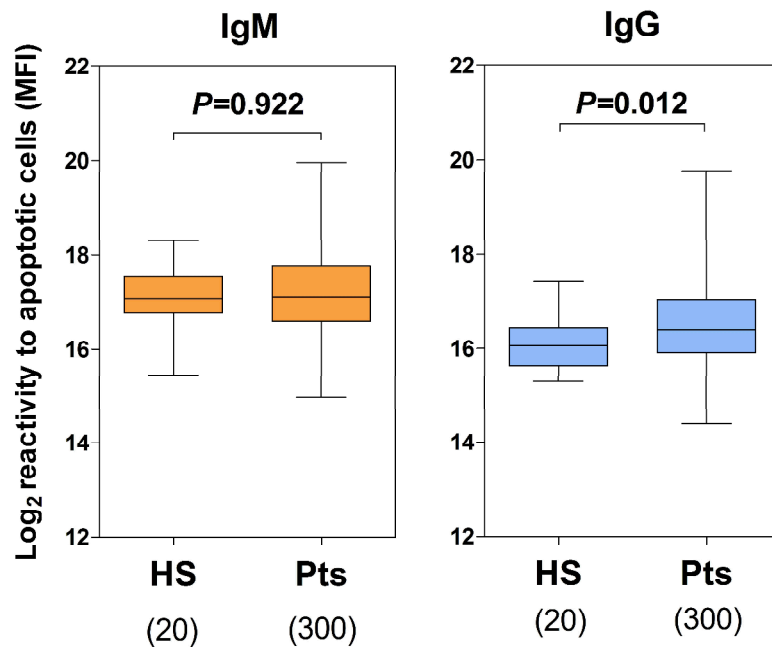


Figure S2. Serum IgM and IgG reactivity to apoptotic cells. The serum IgM and IgG reactivity to apoptotic Jurkat cells was measured by flow cytometry in samples collected pre-transplant from kidney transplant recipients as well as healthy subjects. Log₂ values of MFI are reported (y-axis). The numbers of samples tested in each group are shown below the box plot. The horizontal bar represents the median value; the bottom and top of each box represent the 25th and 75th percentiles; the lower and upper bars of each box represent the minimum and maximum values.

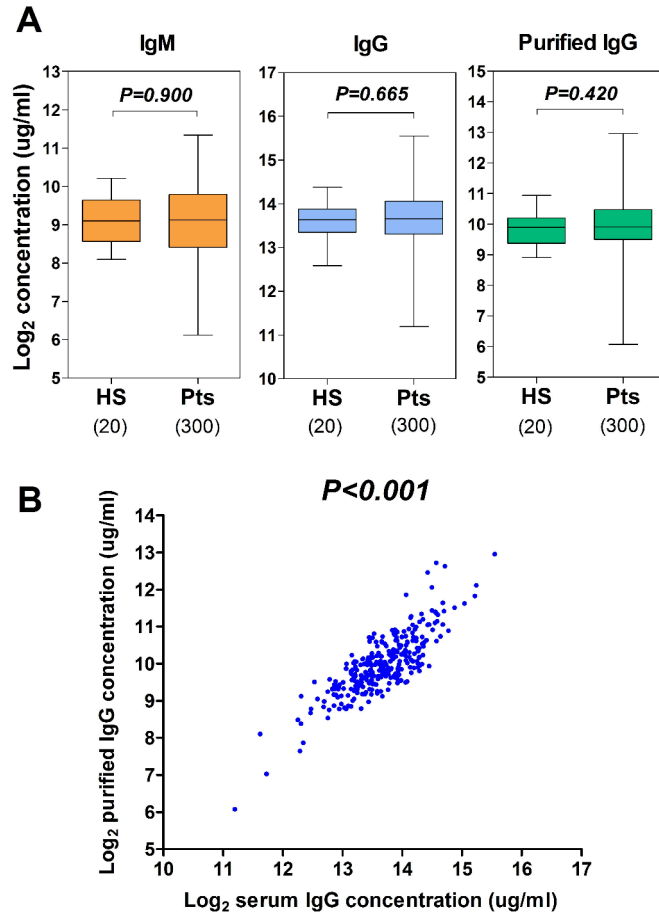


Figure S3. Serum IgM, IgG and purified IgG concentration. (A) Serum IgM, IgG and purified IgG concentrations in pre-transplant serum samples and healthy subjects. The numbers of samples analyzed in each group are shown below the box plot. The horizontal bar represents the median value; the bottom and top of each box represent the 25th and 75th percentiles; the lower and upper bars of each box represent the minimum and maximum values. **(B)** Correspondence between serum IgG concentration before and after purification. Serum IgG concentrations (x-axis) for all patients (N = 300) are plotted with purified IgG concentrations (y-axis) for the same patients. Statistical analysis is based on a two-tailed non parametric spearman's test.

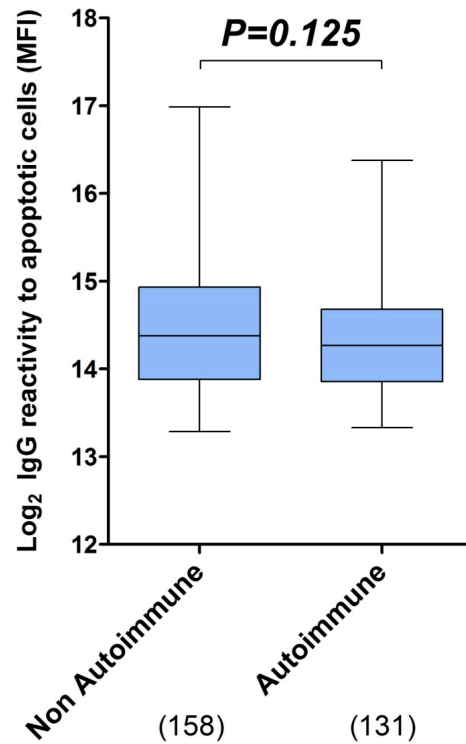


Figure S4. Comparison of purified serum IgG reactivity to apoptotic cells between patients with autoimmune disease and patients with non-autoimmune disease. Purified IgG reactivity to apoptotic cells (log₂ MFI; y axis) is shown for patients grouped by autoimmune (primary FSGS, IgA nephropathy, Type I diabetes, SLE, Immune complex diseases) or non-autoimmune original diseases. The numbers of samples analyzed in each group are shown below the box plot. The original cause of end-stage renal disease was unknown for eleven patients. The horizontal bar represents the median value; the bottom and top of each box represent the 25th and 75th percentiles; the lower and upper bars of each box represent the minimum and maximum values.

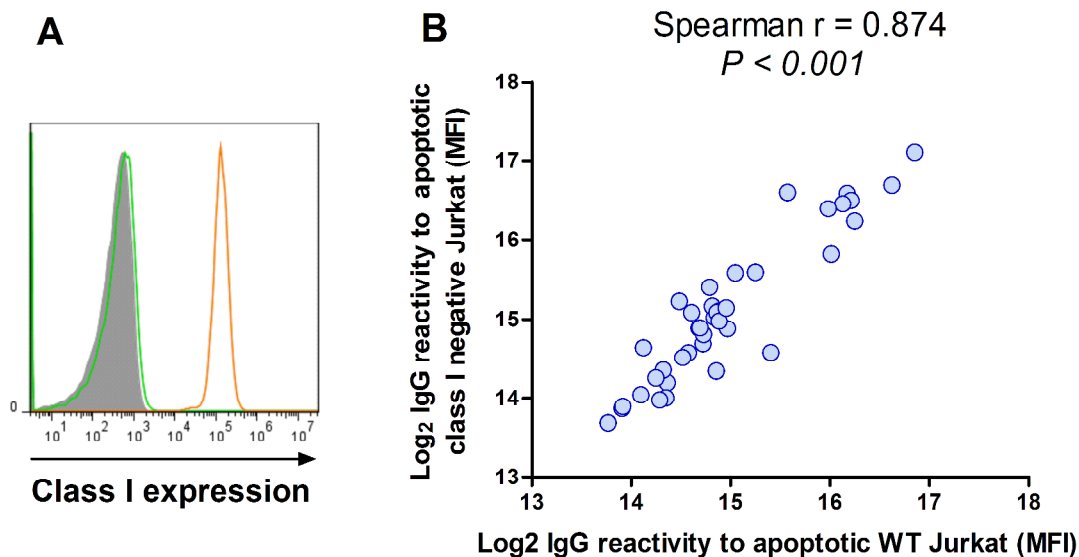


Figure S5. Purified IgG reactivity to apoptotic wild type and class I negative Jurkat cells. (A) HLA class I expression on wild type and β -2 microglobulin knocked down Jurkat cells. Filled gray histograms show the signal generated by staining with isotype control antibody alone. HLA class I expression on wild type and β -2 microglobulin knocked down Jurkat are depicted as orange and green solid line histograms, respectively. (B) Correspondence between purified IgG reactivity to apoptotic wild type and HLA class I negative Jurkat cells. Purified IgG reactivity to apoptotic wild type and HLA class I negative Jurkat cells were detected in 39 selected patients with high reactivity to HLA class I (MFI > 1000). The reactivity to apoptotic wild type Jurkat cells (x - axis) are plotted with reactivity to apoptotic HLA class I negative Jurkat cells (x - axis) for the same patients. Statistical analysis is based on a two-tailed non parametric spearman's test.

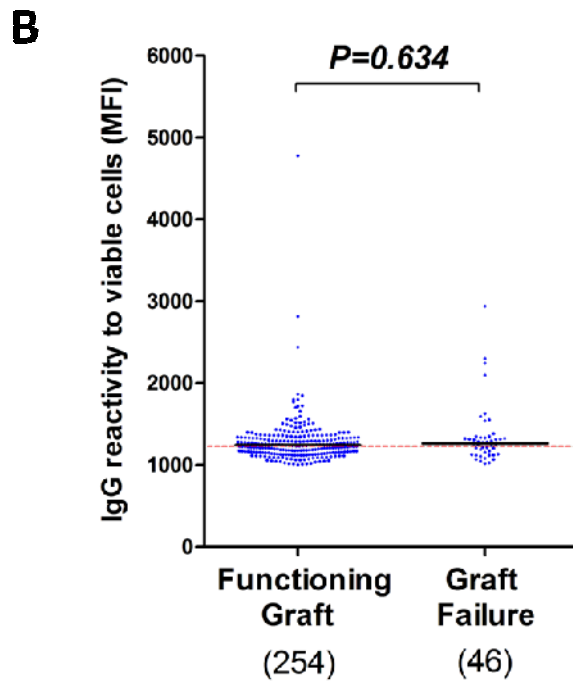
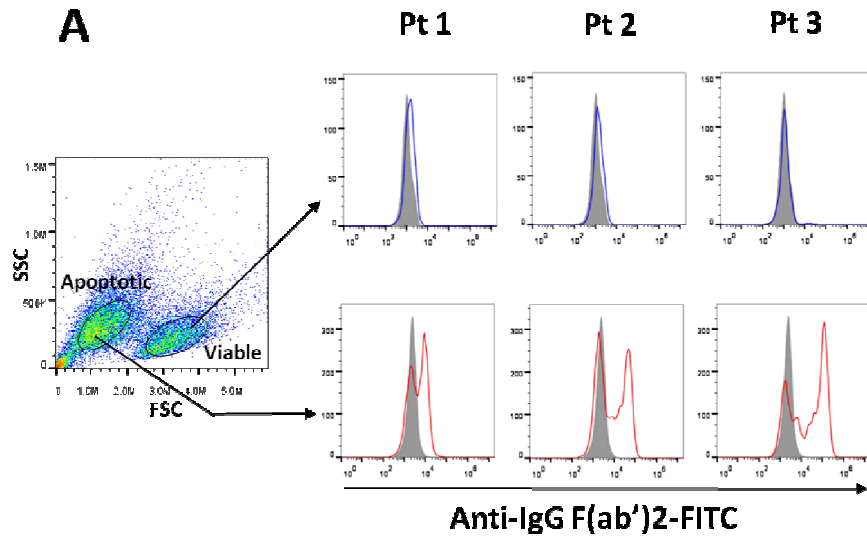


Figure S6. Purified IgG reactivity to viable and apoptotic Jurkat cells. (A) Purified IgG reactivity to viable and apoptotic Jurkat cells are shown for 3 representative pre-transplant patient samples. Results are reported after gating on viable cells (upper panel, blue solid line) or apoptotic cells (lower panel, red solid line). Filled gray histograms show the signal generated by staining with the secondary antibody alone. **(B)** Pre-

transplant purified IgG reactivity to viable Jurkat cells is reported for patients with functioning grafts and patients who experienced graft loss. The numbers of samples analyzed in each group are shown below the scatter plots. Each dot represents a patient. The black bars give the median value for each group. Red dot line represents value (MFI) generated by staining with the secondary antibody alone.

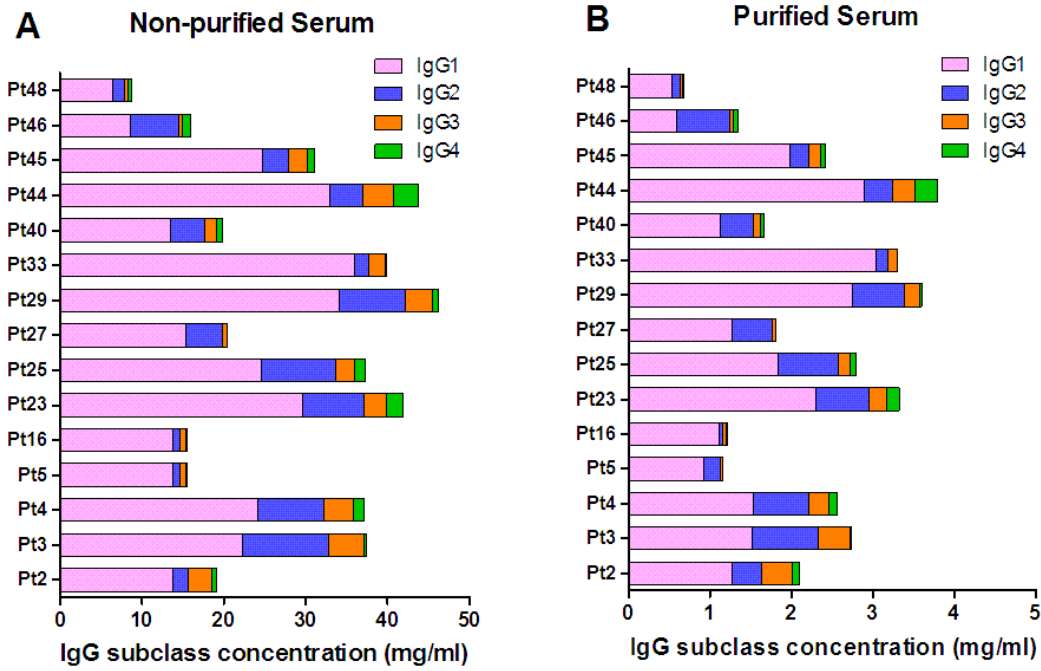


Figure S7. Concentration of IgG subclasses before and after purification. The concentration of the 4 different IgG subclasses (IgG1 ~ IgG4) was assessed in 15 randomly picked pre-transplant serum samples patients before (A) and after (B) IgG gel purification. Serum samples were diluted 1:10 during IgG purification. The distribution of all subclasses concentration is depicted with color-coded stacked bars.

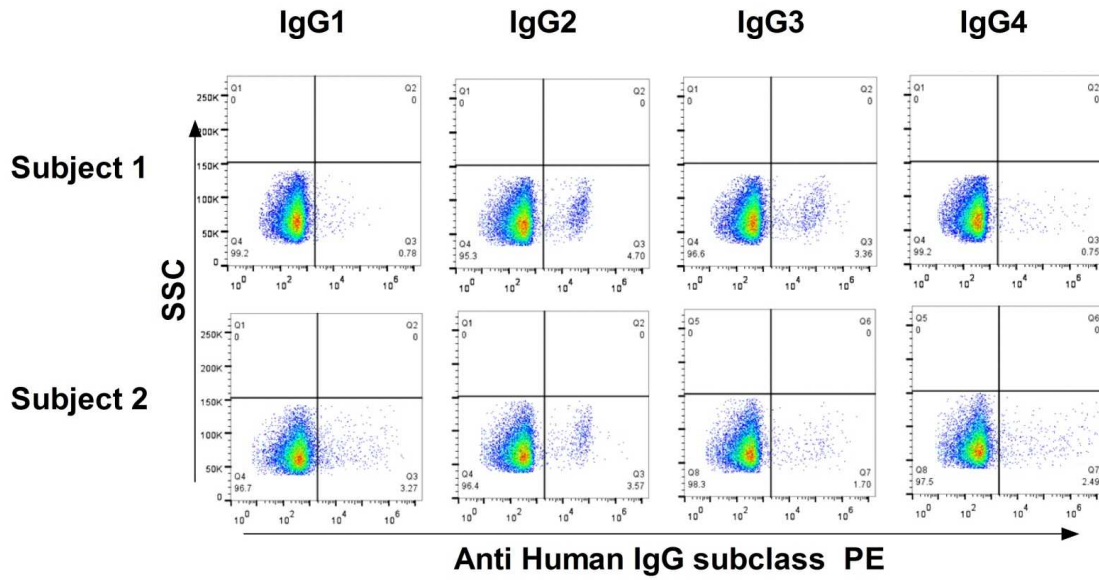


Figure S8. Expression of IgG subclasses on CD19+ B cells from healthy subjects. Peripheral Blood Mononuclear Cells (PBMC) from two healthy subjects were stained using anti-CD19 APC (BD Bioscience) and anti-human IgG1, IgG2, IgG3 and IgG4-PE (Southern Biotech), respectively. Expression of IgG subclasses was assessed after gating on CD19+ cells by flow cytometry.

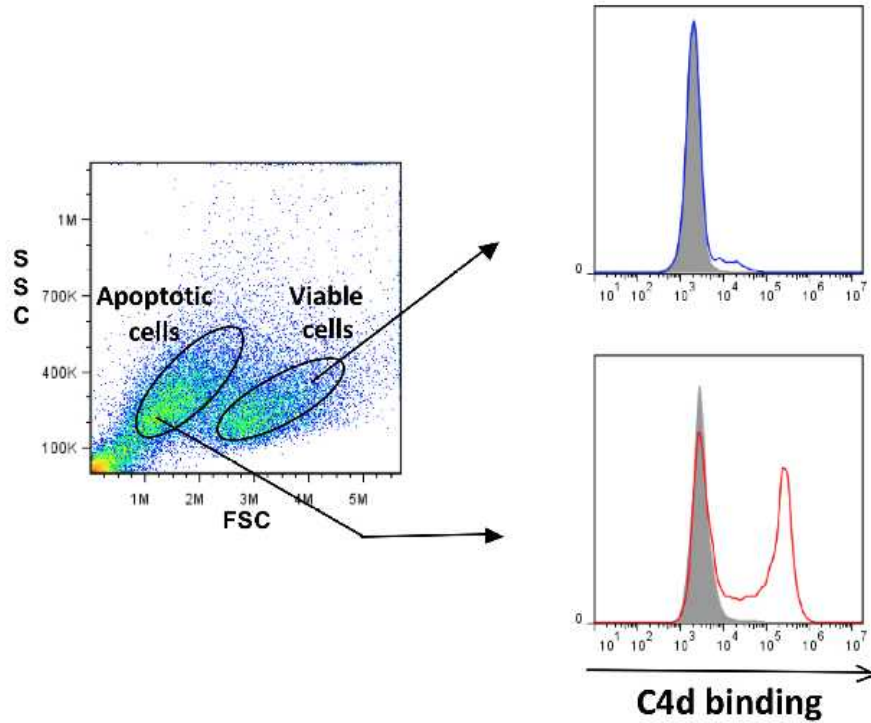


Figure S9. Complement activation and C4d deposition. C4d deposition on apoptotic and viable Jurkat cells was assessed by flow cytometry after complement activation by IgG purified from a representative pre-transplant patient serum sample. Results are reported after gating on viable cells (upper panel, blue solid line) or apoptotic cells (lower panel, red solid line). Filled gray histograms show the signal generated by staining with the secondary antibody alone.