# Supplementary Appendix

This appendix has been provided by the authors to give readers additional information about their work.

Supplement to: Clatworthy MR, Watson CJE, Plotnek G, et al. B-cell–depleting induction therapy and acute cellular rejection. N Engl J Med 2009;360:2683-5.

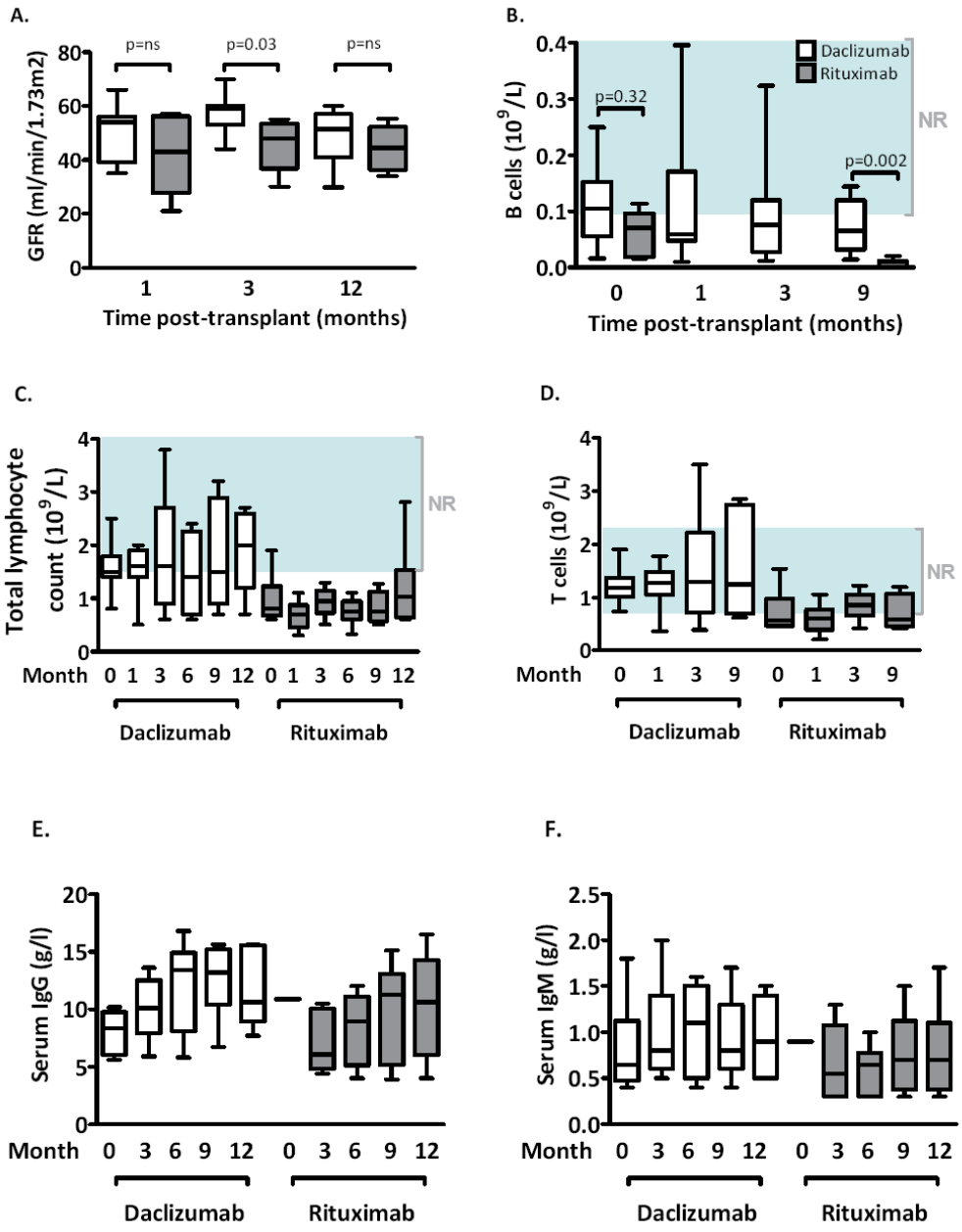
**SUPPLEMENTARY TABLE 1**: **Baseline patient characteristics and adverse events.**

| | **Daclizumab group** | **Rituximab group** |
|---|---|---|
| **Total patients** | 7 | 6 |
| **Male** (%) | 57 | 67 |
| **Age at transplant** (median years+range) | 42.1(27-58) | 44.7(23-64) |
| **Cause of end stage renal failure** | | |
| *Autosomal dominant polycystic kidney disease* | 0 | 4 |
| *IgA nephropathy* | 2 | 0 |
| *Henoch-Schönlein purpura* | 2 | 0 |
| *Sjögren's syndrome* | 1 | 0 |
| *Unknown* | 2 | 2 |
| **Dialysis mode** | | |
| *Pre-dialysis* | 1 | 0 |
| *PD* | 4 | 3 |
| *HD* | 2 | 3 |
| **Time on dialysis (months)** Median(range) | 2.3 (0-76) | 2.7 (11-108) |
| **Type of transplant** | | |
| *Living* | 4 | 0 |
| *Donation after brainstem death* | 1 | 6 |
| *Donation after cardiac death* | 2 | 0 |
| **CMV mismatch** | 3/7 | 1/6 |
| **Cold ischaemic time** (median hours,mins+ range) | 3hr 48m (1hr56m-11hr49m) | 10hr 17m (6hr6m-20h14m) |
| **Infections at 12 months*:** | | |
| ***Minor*** | 14 | 16 |
| ***Major*** *(requiring hospitalisation)* | 3 | 6 |
| ***Life-threatening*** | 0 | 0 |
| **Other adverse events at 12 months** | | |
| ***Malignancy*** | 0 | 0 |
| ***Cardiovascular*** | 3 | 2 |
| ***Neutropaenia*** | 2 | 3 |
| ***Post-transplant diabetes*** | 2 | 2 |
| ***Urethral stricture*** | 1 | 2 |

* Patients received a three month course of valganciclovir as prophylaxis against cytomegalovirus if the donor was sero-positive and the recipient sero-negative. All recipients received cotrimoxazole for 6 months and oral amphotericin for 4 weeks following transplantation.

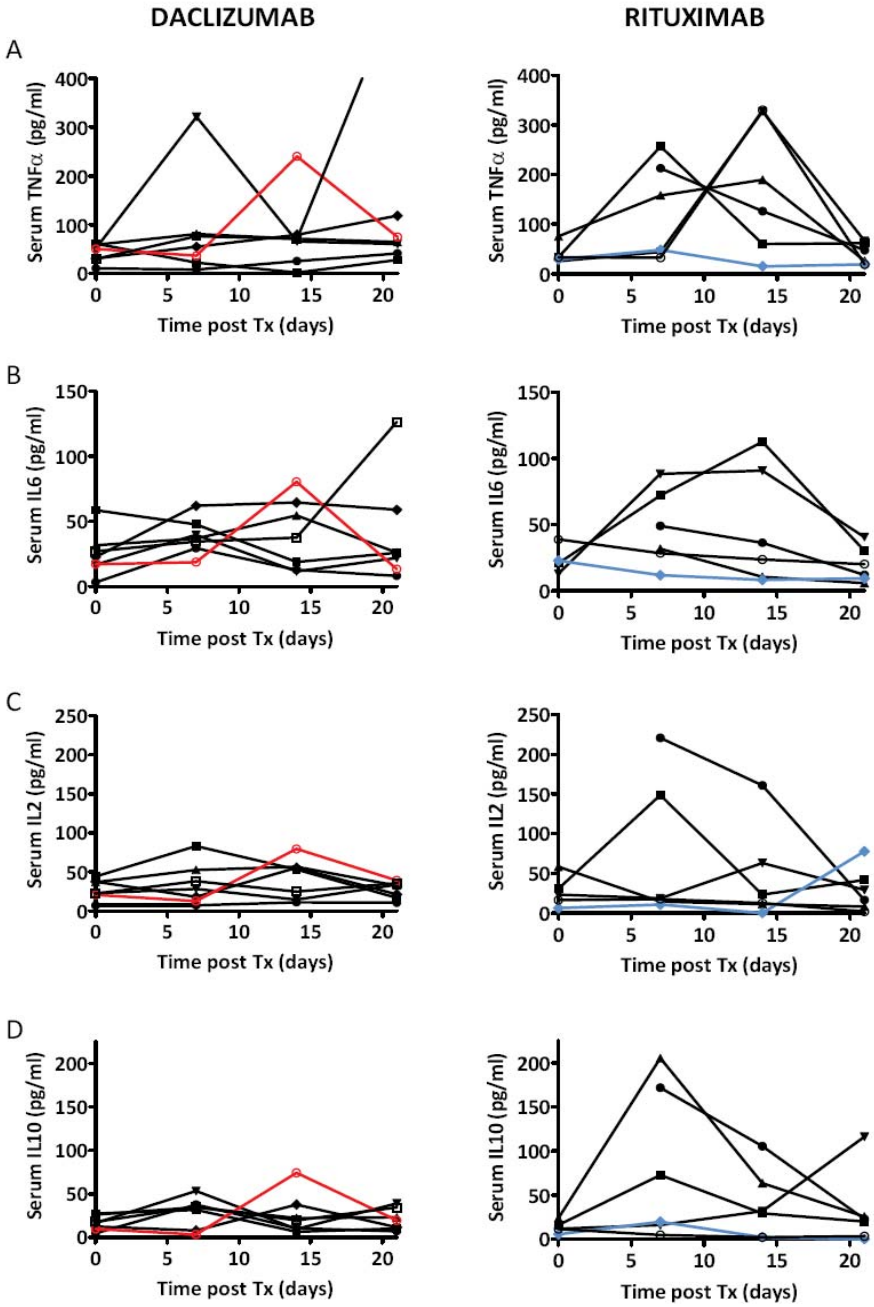
## SUPPLEMENTARY FIGURES

### SUPPLEMENTARY FIGURE 1



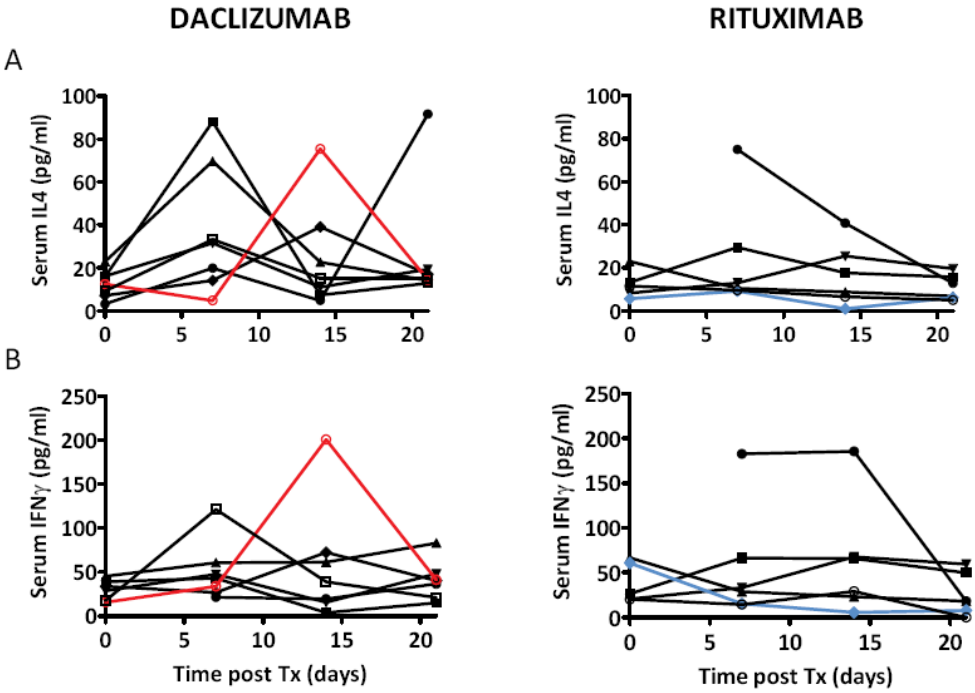

**A.** Glomerular filtration rate (GFR) normalised for body surface area, **B.** B cell counts (Normal range (NR) shown by shaded area) **C.** Total lymphocyte count, **D.** T cell counts, **E.** IgM, and **F.** IgG, in daclizumab (open boxes) versus rituximab-treated (grey boxes) patients. Whiskers show minimum and maximum values, and lines show medians. Values in the daclizumab and rituximab groups are compared using a Mann Whitney test. All patients were mildly lymphopaenic prior to transplantation (**C**) a well-recognised phenomenon in patients with end-stage renal disease (Yoon JW *et al*. Kidney Int 2006;70:371-6). There was no significant fall in mean total lymphocyte count or CD3 count in either of the study groups following transplantation (**D**).

**SUPPLEMENTARY FIGURE 2**



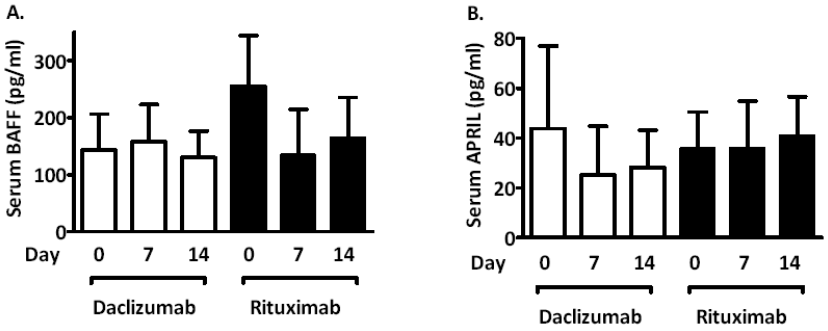

Serum levels of **A.** TNFα, **B.** IL-6, **C.** IL-10, **D.** IL-2 at days 0, 7, 14 and 21 in daclizumab (left hand panel) versus rituximab-treated (right hand panel) patients. The daclizumab- treated patient with rejection is shown in red, whilst the rituximab-treated patient who did not develop rejection is shown in blue. The rise in IL-10 is of interest, since this cytokine is classically considered to play an immunomodulatory role (Ishida H, et al. J Exp Med 1994;179:305-10). However, it is a cytokine with both pro- and anti-inflammatory effects (Moore KW, et al. Annu Rev Immunol 1993;11:165-90). For example, IL-10 can enhance T cell proliferation (MacNeil IA, *et al.* J Immunol 1990;145:4167-73) and IL-10 production by T cells is associated with increased titres of anti-dsDNA antibodies and with increased proteinuria in NZB/W mice (Enghard P, *et al.* Scand J Rheumatol 2006;35:209-16).

**SUPPLEMENTARY FIGURE 3**




Serum levels of **A.** IL-4, and **B.** IFN-γ at days 0, 7,14, 21 in daclizumab (left hand panel) versus rituximab-treated (right hand panel) patients. The daclizumab-treated patient with rejection is shown in red, whilst the rituximab-treated patient who did not develop rejection is shown in blue. Overall, serum IL-4 levels were increased at day 7 and 14 in the daclizumab group compared with baseline(**A**). There was no change in IFN-γ levels post-transplantation in either group (**B**).

**SUPPLEMENTARY FIGURE 4**




Mean serum levels of **A.** BAFF, and **B.** APRIL at days 0, 7 and 14 in daclizumab (open boxes) versus rituximab-treated (black boxes) patients. Error bars show standard error of mean. Serum BAFF and APRIL levels have been shown to increase following B cell depletion (Pers JO, et al. Arthritis Rheum 2007;56:1464-77) and can drive T cell activation (Mackay F and Leung H. Immunol 2006;18:284-9). However, there was no significant change in BAFF and APRIL levels in either rituximab or daclizumab-treated patients in the post-transplant period.