

SUPPLEMENTAL MATERIAL

Webster et al., <http://www.jem.org/cgi/content/full/jem.20082824/DC1>

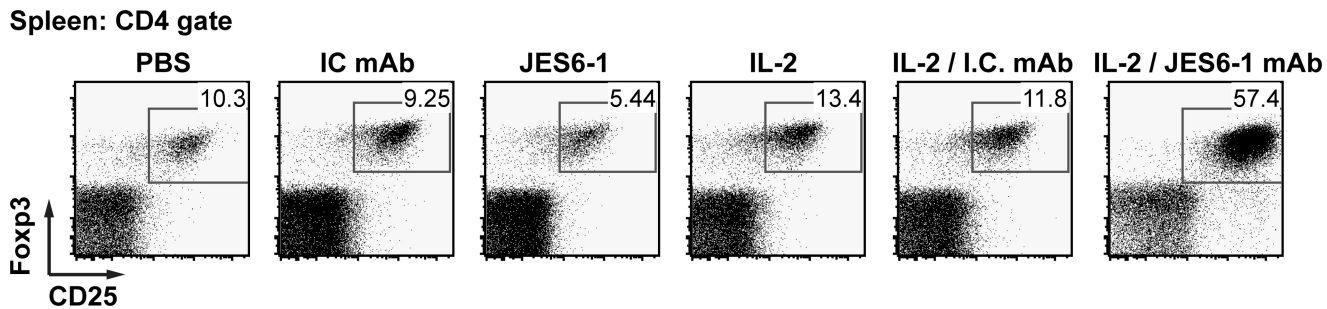


Figure S1. Proportion of CD25⁺Foxp3⁺ T reg cells in the splenic CD4⁺ population after treatment with combinations of control antibody, IL-2, or IL-2 mAb. Mice were treated with PBS, isotype control antibody (5 μ g rat IgG2a), 5 μ g JES6-1, 1 μ g IL-2, 1 μ g/5 μ g IL-2–isotype control antibody, or 1 μ g/5 μ g IL-2–JES6-1 mAb on days 0, 1, and 2. On day 5, mice were sacrificed and the splenic T reg cell population was analyzed by flow cytometry. Data are shown as percentages of the CD4⁺ T cell population. Data are representative of two independent experiments. Flow cytometry plots depict log₁₀ fluorescence.

Mouse	BGL (mmol/liter)	
	Before nephrectomy	After nephrectomy
1	5.9	>32
2	6.7	23
3	5.2	31
4	7.4	25
5	6.8	>32

Figure S2. BGLs before and after nephrectomy of graft-bearing kidneys. Nephrectomy of the graft-bearing kidney was performed on C57BL/6 mice with long-term BALB/c islet grafts. Blood glucose was monitored before and after nephrectomy to determine if the islet graft was responsible for the euglycemia. A return to hyperglycemia (>16 mmol/liter) after nephrectomy was indicative of graft-dependent euglycemia. The surgery on five mice was performed in three independent experiments.

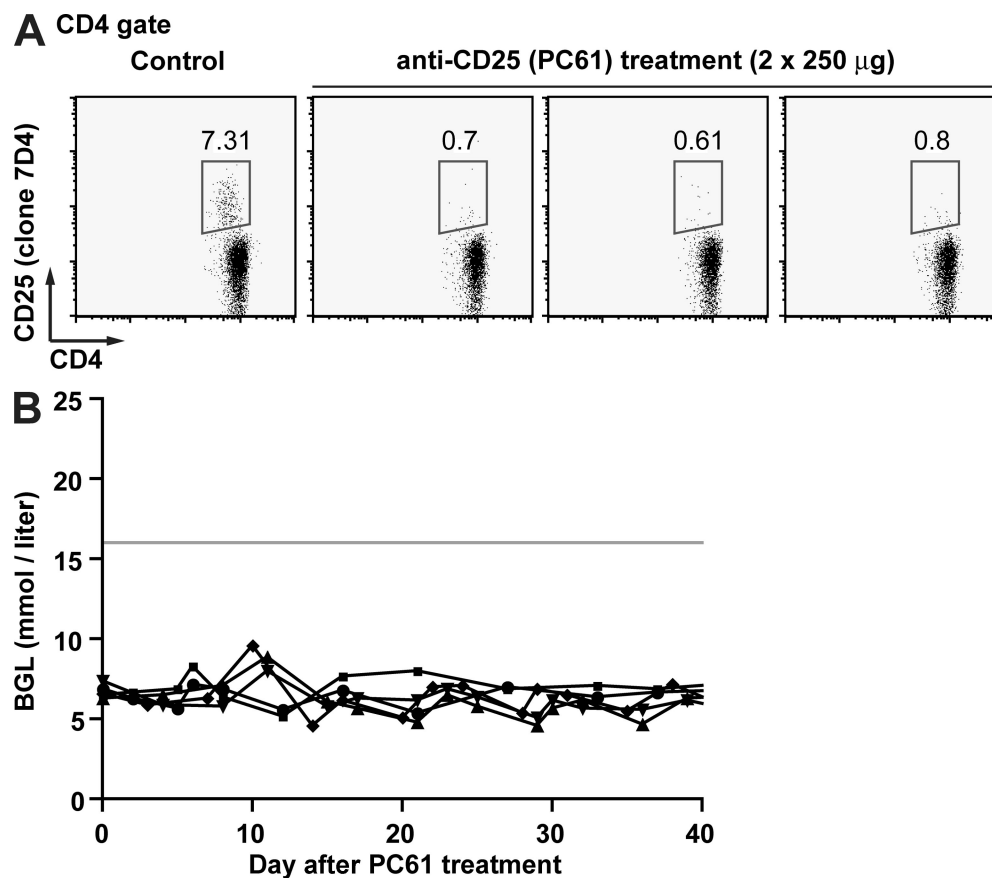


Figure S3. Treatment of BALB/c allograft-tolerant mice with anti-CD25 mAb (PC61). Mice were injected i.p. twice 2 d apart with 250 µg PC61. Five mice were treated: three 100 d before transplant (starting on day 32) and two >100 d after transplant (days 118 and 196). Efficiency of depletion was monitored by flow cytometry using CD25 mAb (clones 7D4 and PC61), CD4 mAb, and Foxp3 mAb. (A) Expression of CD25 (clone 7D4) on CD4⁺ T cells 7 d after PC61 treatment compared with untreated control. This disappearance of CD25 was not paralleled by a complete depletion of Foxp3⁺ cells, with only a 50% reduction observed in CD4⁺Foxp3⁺CD25⁻ cells (not depicted), indicating that the reduction in the percentage of CD25⁺ cells was due in part to CD25 modulation rather than depletion of cells. (B) BGLs of BALB/c islet allograft-tolerant mice after challenge with 2 × 250 µg anti-CD25 mAb (*n* = 5). The mice were treated and analyzed in two independent experiments. Flow cytometry plots depict log₁₀ fluorescence. The horizontal gray line indicates a BGL of 16 mmol/liter, the threshold for rejection.