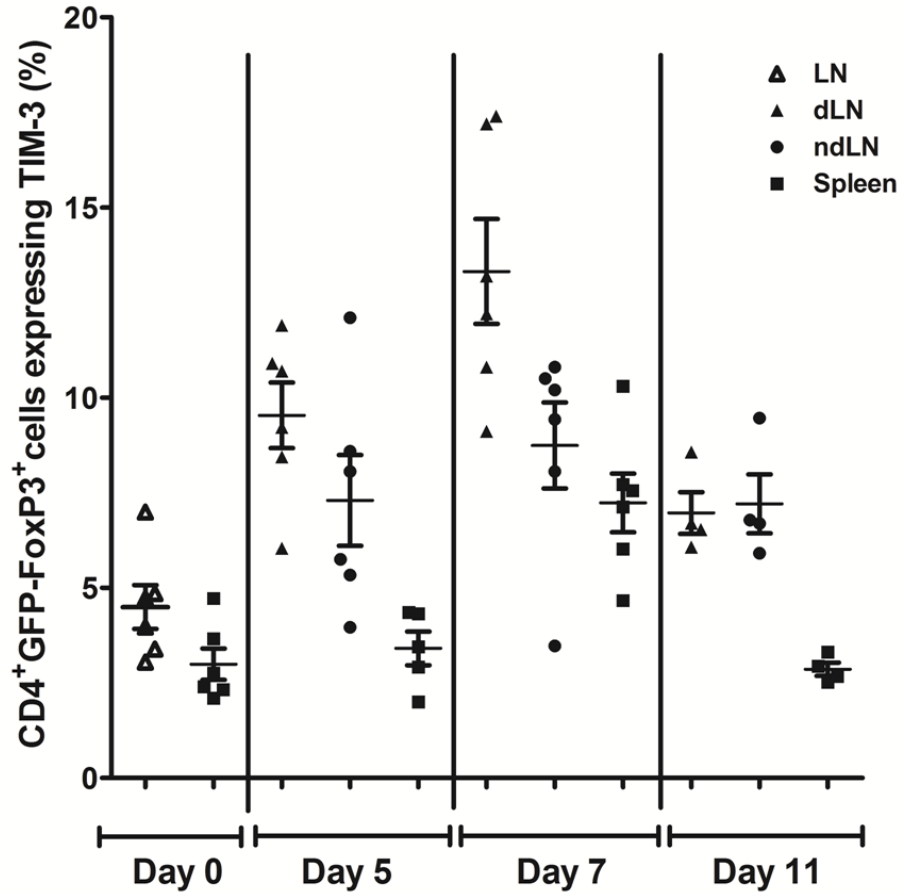


1

## Supplemental Figure 1



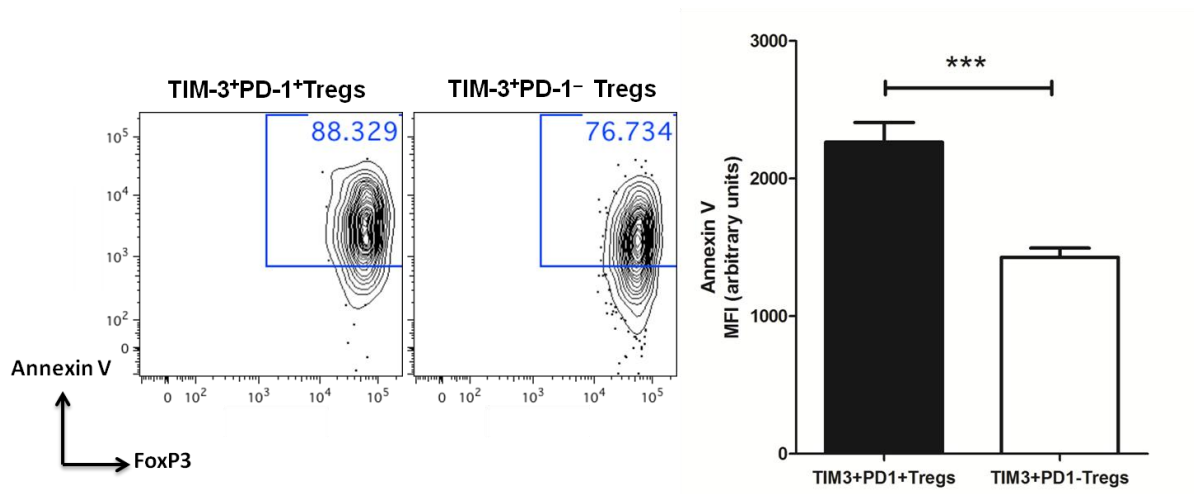
2

3 Supplemental Figure 1. *Proportion of FoxP3<sup>+</sup>TIM-3<sup>+</sup> T-cells is highest in dLN at the time*  
4 *of rejection.* C57BL/6-K1 mice grafted with BALB/c skin were analyzed for the proportion  
5 of CD4<sup>+</sup>GFP<sup>+</sup>FoxP3<sup>+</sup> cells expressing TIM-3 in the dLN, ndLN and spleen during the  
6 course of rejection as depicted. The TIM-3<sup>+</sup>FoxP3<sup>+</sup> cells are present in the highest  
7 proportions in the dLN at time of rejection on day 7. Data are presented for n<sub>≥</sub> 4  
8 animals as mean<sub>±</sub>SEM.

9

10

## Supplemental Figure 2



11

12 Supplemental Figure 2: *TIM-3<sup>+</sup> Tregs co expressing PD-1 are more likely to perish.*

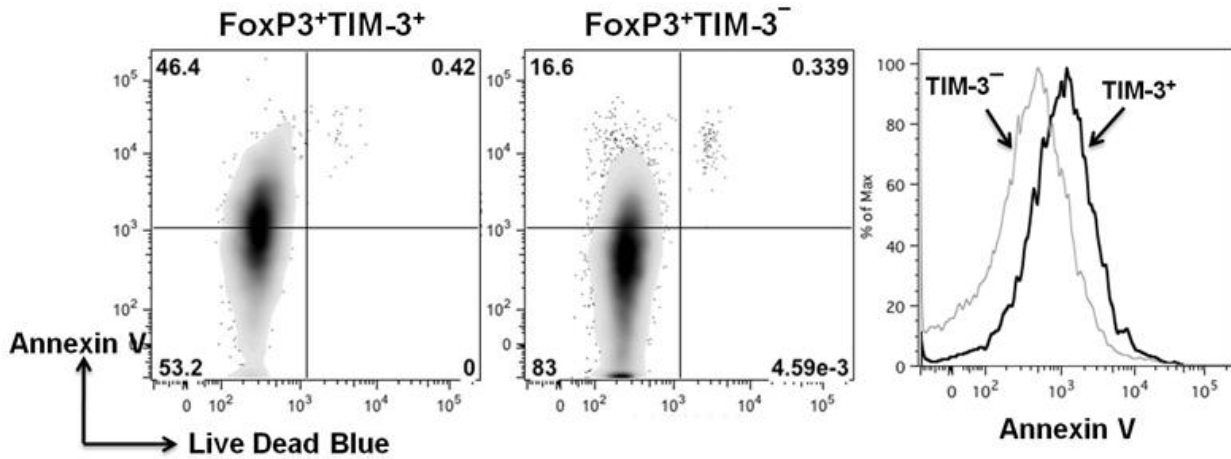
13 Draining lymph nodes of C57BL/6-KI mice grafted with BALB/c skin were analyzed for

14 the proportion of TIM-3<sup>+</sup>PD-1<sup>+</sup>/PD-1<sup>-</sup> Tregs staining with cell surface phosphatidyl serine

15 binding annexin V as depicted and a comparison of geometric mean fluorescence

16 intensity for annexin V staining was determined (n=5 mice, \*\*\*p=0.0008).

## Supplemental Figure 3



18

19 Supplemental Figure 3. *TIM-3<sup>+</sup> Tregs obtained from DBA/2 DST injected BALB/c-KI*  
 20 *mice are annexin V<sup>+</sup>*. Spleens were harvested from BALB/c-KI mice injected i.v. 5 days  
 21 prior with DBA/2 DST. *TIM-3<sup>+</sup> Tregs* were analyzed in comparison with *TIM-3<sup>-</sup> Tregs*  
 22 from spleen for their ability to stain cell surface phosphatidyl serine with annexin V and  
 23 exclude the LIVE/DEAD blue viability dye. Plots are representative of n=5 mice.