Supplemental Figure Legends

Supplemental Figure 1. Trans-vivo DTH response to col(V) in lung transplant recipients is specific for the α1(V) chain

Col(V) is normally composed of genetically distinct α1(V) and α2(V) chains. To analyze subunit specificity of the col(V) TV-DTH response, recombinant α1(V) and α2(V) chains were prepared from gene-transferrent fibroblast cell lines (panel A). Patient PBMC responded strongly to col(V) in the TV-DTH assay over a wide dose range. The response to recombinant α1(V) was lower, but equivalent to col(V) at the highest dose tested; α2(V) did not induce a swelling response (panel B).

A) SDS-PAGE of Mr markers (lane 1), recombinant α2(V) (lane 2) and α1(V) (lane 3) preparations purified from cultures of gene-transfected mouse embryo fibroblasts. The α1(V) subunit preparation contained both a partially processed form (Mr 250 kDa) and a higher Mr pro-form containing intact N-terminal globular sequences; the α2(V) recombinant protein migrated as a single band of Mr 150 kDa. B) Dose-response in TV-DTH assay of PBMC from pts L52 and L84 to purified human col(V), or to recombinant α2(V) and α1(V). Assayed points were averaged from 2 determinations per patient. Recombinant human α1(V) homotrimerers were prepared as described previously (1). For biosythesis and purification of recombinant α2(V), we used a previously described α2(V) expression construct in pcDNA4/TO/myc-His (Invitrogen) and described methods (2).

Supplemental Figure 2. Time course of pulmonary function in patients developing BOS.
Fourteen patients who developed BOS are grouped as BOS-1 only (panel A) and BOS-2/3 (panel B). FEV-1 (forced expiratory volume in 1 sec) values are displayed on the Y-axis. Individual patients are indicated by line type. The mean time to BOS-1 for all patients who developed BOS was 2.9 ± 1.6 years, while the mean time to BOS-2 was 2.5 ± 1.0 years.

Supplemental Figure 3. Post-transplant average TV-DTH responses to col(V) are elevated compared to col(II)-specific response across all 1° lung disease categories

All post-transplant TV-DTH determinations obtained pre-BOS from each patient were averaged to give a single value per patient. Means of averaged TV-DTH values were compared between patients with different lung diseases. With the exception of AAD pts, the mean response to col(V) in each subgroup fell within the low/negative response range (grey area) and were not different from each other. The mean response to col(V) was significantly higher than responses to col(II), both overall (n=54), and within each disease group (p<.001) with the exception of IPF (p=.08).

References
Supplemental Figure 1

A

B

Net footpad Swelling (×10^{-4} In)

Amount of Protein Injected (ng)
Supplemental Figure 3

Net Footpad swelling ($10^{-4}$ in.)

CF  AAD  Other  COPD  IPF