Supplementary

Table S1 xeno-GVHD clinical scoring system

Clinical signs	0= no signs of xeno-GVHD	1= moderate signs of xeno-GVHD	2= severe signs of xeno-GVHD
Weight	No weight loss	Weight loss ≤15%	Weight loss >15%
Anemia	Normal blood test	Slight decrease in RBC, HB, PLT	Massive decrease in RBC, HB, PLT
Behavior	Normal mobility and posture	Decreased mobility and convex back	Lack of mobility and extremely convex back

PBMCs were collected from NSCLC patients and HDs and injected into NSG-SGM3 mice to create hu-PBL models. A scoring system was used to determine the severity of xeno-GVHD signs in hu-PBL mice. Mice were monitored biweekly for clinical signs of xeno-GVHD (weight, anemia, and behavioral changes [pain signs and response to stimulation]) and were given a score between 0-2. PBMCs, peripheral blood mononuclear cells; hu-PBL, human peripheral blood lymphocytes; NSCLC, non-small cell lung cancer; HDs, healthy donors; xeno-GVHD, xeno-graft versus host disease; RBC, red blood cell; HB, hemoglobin; PLT, platelet.

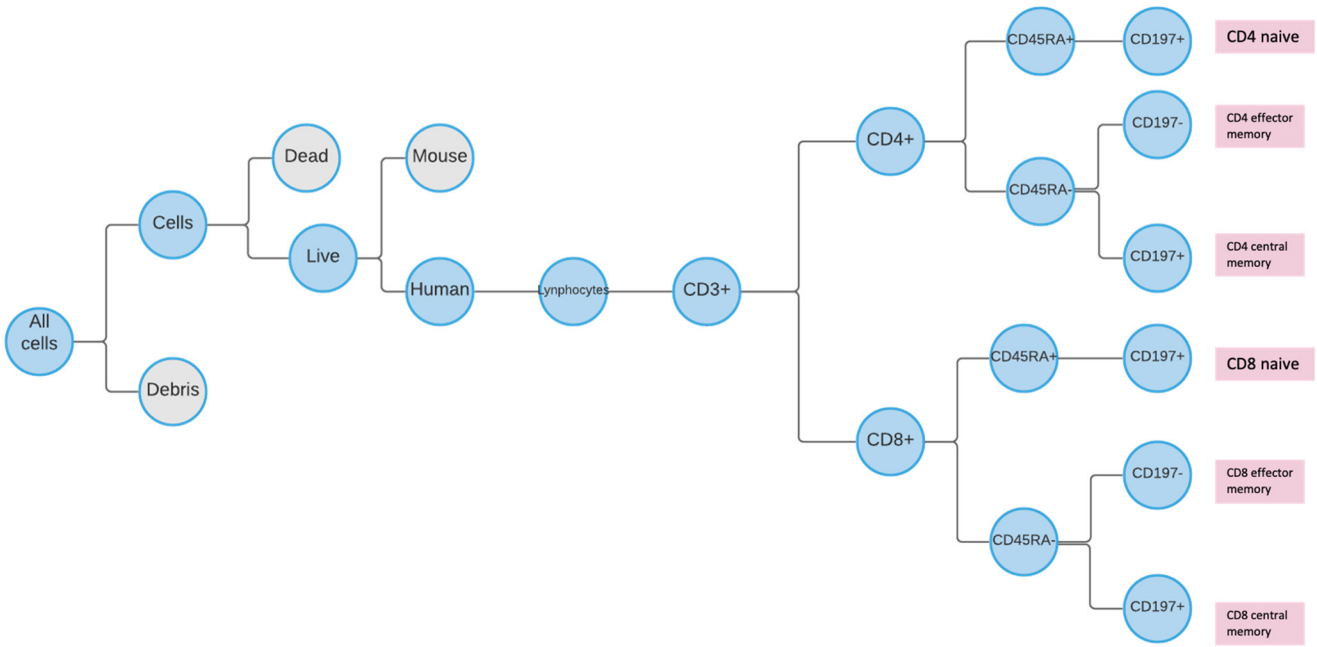

Figure S1 Flow cytometry gating strategy for T-cell compartments and subpopulations. Peripheral blood was collected from hu-PBL NSG-SGM3 mice and analyzed using flow cytometry. First, erythrocytes were removed, and PBMCs were isolated and stained with specific surface antibodies. A panel of antibodies was used to characterize the T-cell compartment and its subpopulations, allowing for the assessment of human immune cells in the hu-PBL NSG-SGM3 mice and the investigation of the xeno-GVHD process. A ghost dye was used to distinguish live and dead cells, and anti-mouse $CD45^+$ antibodies facilitated differentiating between human and mouse immune cells. The characterization of PBMCs included staining with anti-human CD45, followed by CD3, CD4, and CD8 to identify the T-cell subpopulations. The phenotypes of central memory, effector memory, and naïve T-cells were determined using anti-human CD45RA and CD197-specific antibodies. PBMCs, peripheral blood mononuclear cells; hu-PBL, human peripheral blood lymphocytes.

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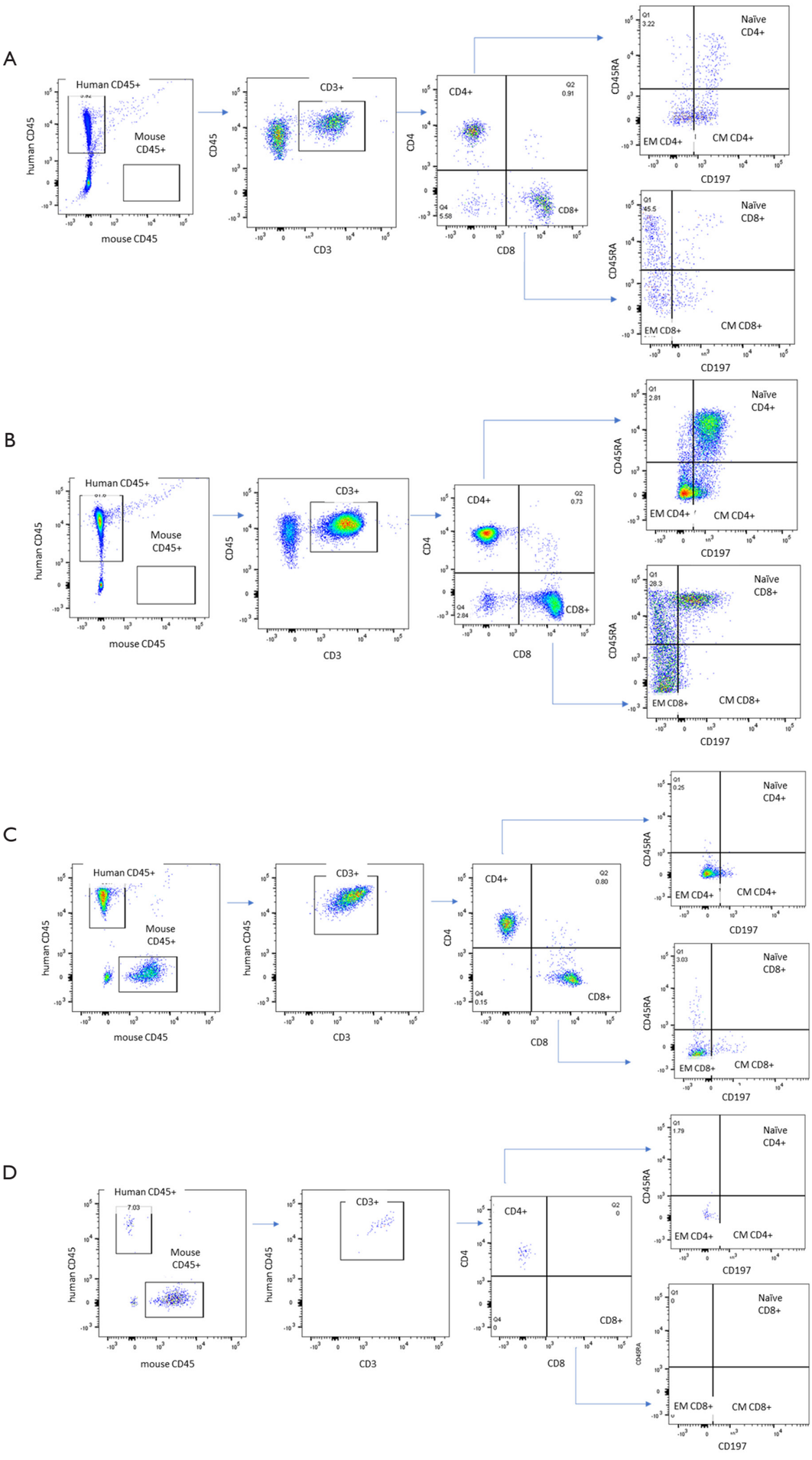

Figure S2 Flow cytometry raw data results of T-cell analysis of PBMCs injected into hu-PBL mice model. The representative results from a flow cytometric analysis of T-cell subpopulations derived from NSCLC patients and HDs hu-PBL models. Initially, human and mouse PBMCs were distinguished using specific $CD45^+$ antibody staining. $CD3^+$ cells were stained and analyzed for their $CD4^+$ and $CD8^+$ subpopulations. The flow cytometry panel presented shows $CD4^+$ T cells in the upper section and $CD8^+$ T cells in the lower section. Within this panel, the upper left quadrant represents nonspecific cells, the upper right quadrant indicates naive cells, the lower left quadrant denotes effector memory (EM) cells, and the lower right quadrant shows central memory (CM) cells. The T-cell baseline results at week "0" for patient Pt3 (A) and healthy donor HD3 (B) are shown in panels A and B, respectively. Panels C and D display the T-cell results at week "6" post-PBMC injection for HD4 (C) and Pt4 (D). PBMCs, peripheral blood mononuclear cells; hu-PBL, human peripheral blood lymphocytes; NSCLC, non-small cell lung cancer; Pts, patients; HDs, healthy donors

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