Supplemental Methods

PET and CT Imaging. Mice were fasted for 4-6 h before [¹⁸F]FDG injection and placed on a heating pad (30°C) starting 30 min before [¹⁸F]FDG injection. For tracer injection and imaging, mice were anesthetized with 2% isoflurane. Mice were imaged in a chamber that minimizes positioning errors between PET and CT to less than 1 mm. Imaging was started 60 min after an i.p. injection of 7.4MBg (80µCi) [¹⁸F]FDG or [¹⁸F]HBG via tail vein. Image acquisition time was 10 min. Images were reconstructed using a combination of OSEM3D (3D-Ordered Subset-Expectation Maximization) and MAP (Maximus A Priori). The number of OSEM subsets used was 16. The number of MAP iterations used was 18. Image counts per pixel per second were calibrated to activity concentrations (Bq/ml) by measuring a 3-cm cylinder phantom filled with a known concentration of [¹⁸F]FDG. Immediately after the PET scan, the mice underwent a 8-min microCT scan using routine image acquisition variables (70kVP and 500 µA). For display, activity concentrations were expressed as percent of the decay-corrected injected activity per gram of tissue by using Osirix software. Spherical regions of interest (2-mm diameter) were placed in the area of the tumor with the highest [¹⁸F]FDG or ¹⁸F]HBG uptake. All image analysis was done using the OsiriX software.

Supplemental Figure Legends

Figure S1. MART-1 specific CTLs limit growth of MART-1/HLA-A*0201 (M202) melanoma tumors. Tumor metabolic activity was measured by levels of FDG uptake using PET imaging. The squares (**a**) represent the growth of the M207 (HLA-A*0201 MART⁺) tumor while the circles (**•**) the M202 (HLA-A*0201⁺MART⁺). The M207 tumor was used as a negative control. The tumors were measured at the endpoint, week 6. Control mice are mice that received non-transduced hematopoietic stem cells. The unit %ID/g stands for injected dose per gram of body weight. Statistical significance for all the experiments (unless otherwise indicated) was determined by a paired Student's t-test with significant values set at p<0.05. There as no statistically significant differences in tumor metabolic activity between the two tumors in control mice as well as the M207 in the transgenic mice versus the tumors in their control counterparts.

Figures S2-6. Surface expression of the MART-1 specific TCR and other endogenous V β chains. In addition to the V β 5.1, we assessed by flow cytometric analysis whether the MART-1 specific TCR was co-expressed with other endogenous chains on the transgenic CD8 T cell surface. We assayed for V β 13.1 (S3), V β 9 (S4), V β 5.3 (S5), V β 23 (S6), V β 20 (S7).

Figures S7-11. Intracellular expression of endogenous V β chains in transgenic CD8 T cells. In addition to the V β 5.1, we assessed by intracellular flow cytometric analysis whether the other endogenous chains failed to assemble on the cell surface of transgenic CD8 T cells. We assays for V β 20 (S8), V β 23 (S9), V β 5.3 (S10), V β 13.1 (S11), V β 9 (S12).











































m32

















10³ Vb20

10²

0

0.873

10⁵ –

MART-1 Tetramer

10³

10²

















Control