

Molecular Basis for the high affinity interaction between the non-classical MHC class I Thymic Leukemia (TL) Antigen and the CD8 $\alpha\beta$ molecule.

Antoine Attinger¹, Olga Naidenko¹, Donald Martin¹, Yiran Wang-Zhu¹, Peter Jensen², Yi Xiong³, Ellis L. Reinherz³, Mitchell Kronenberg¹ and Hilde Cheroutre¹

¹La Jolla Institute for Allergy and Immunology, Developmental Immunology, San Diego, CA 92121,

²Emory University School of Medicine, Department of Pathology, Atlanta, GA 30322, ³Laboratory of

Immunobiology, Dana-Farber Cancer Institute, Boston, MA 02115.

The thymic leukemia (TL) antigen is a non-classical and non-polymorphic MHC class I expressed predominantly in the epithelium of the small intestine. Recently we have demonstrated that TL-tetramers stain intraepithelial lymphocytes (IEL) but not the lymphocytes from spleen and lymph nodes. Moreover, we have shown that CD8 $\alpha\beta$ but not CD8 $\alpha\alpha$ molecule interacts with a relatively high affinity for TL, in a TCR independent manner.

In contrast to TL, the classical MHC class I molecule H-2K^b binds only weakly to CD8 $\alpha\beta$. Since TL and H-2K^b display the overall same structure, subtle changes in the amino acid composition are likely to be responsible for this difference in affinity. To define the region of the TL molecule that is responsible for the CD8 binding, we have generated chimeric molecules composed of the α_1 and α_2 domain of H-2K^b with the α_3 domain of TL (H-2K^b $\alpha_1\alpha_2$ TL α_3). The inverse chimera, TL $\alpha_1\alpha_2$ H-2K^b α_3 , also has been generated. The affinity of the chimeric molecules for CD8 $\alpha\beta$ has been evaluated by BIAcore and by tetramer staining on CD8 $\alpha\beta$ transfected cells. Our results demonstrate that the strong affinity of TL for CD8 $\alpha\beta$ is mediated by its α_3 domain. We are currently performing site directed mutagenesis in the α_3 domain of TL to map the exact amino acid side chains responsible for its high affinity binding to CD8 $\alpha\beta$.