***T- Cell Cultures***

T cell potential of human cells harvested from transplanted mice was assessed by co-culturing in bulk ($3x10^3$ cells/well) in 96 well plates on Delta-like-1-expressing OP9 (OP-DL1) stromal cell line (generous gift of Dr Juan C. Zunniga-Pflucker, University of Toronto, Toronto, Canada) into a 1:1 mixture of T cell medium and human thymic stroma conditioned medium +/- CID. T cell medium consisted of RPMI 1640, 5% FCS, 50 mM 2-ME, Penicillin (50 U/mL)/ Streptomycin (50 µg/mL), L- glutamine (200 mM), FL (5 ng/mL), IL-7 (5 ng/mL; R&D) and (Tpo; 10 ng/ml). Human thymic stroma conditioned medium consisted of Isocove's Modified Dulbecco's Medium [IMDM: BioWhittaker], 15% horse serum(Omega Scientific, Tarzana, CA), 50 mM 2-ME, Penicillin (50 U/mL)/ Streptomycin (50 µg/mL) and L- glutamine (200 mM). After 3-4 weeks in culture cells were harvested and assessed by FACS for expression of human CD3, CD4 and CD8.

**PCR analysis of Mpl expression**

RNA was extracted from FACS isolated cells according to manufacturer's guidelines using the RNA STAT-60 kit (Tel-Test, Friendswood, TX) and reverse-transcribed using the Omniscript RT kit (Qiagen, Valencia, CA) to generate complementary DNA (cDNA). One-half of each sample was subjected to RT PCR (1RT) for cDNA production, and the other half was used as a negative control (2RT). The 1RT and 2RT products were then each divided in half for PCR detection of the *c-MPL* gene

and *ß2-microglobulin ($ß_2M$)* (used as a positive control for loading of cDNA). Primers for detection of c-MPL expression in non-transduced progenitor populations were designed based on the published cDNA and genomic sequences to span at least one intron, as follows: 5' TCCCAAGAACTCCACTGGTC, and 3' GGAGTCCTGAGATGAGGCAG. This primer pair amplified a 215bp sequence.

PCR conditions for detection of c-Mpl were as follows: 95°C (15 min for one cycle), 94°C (1 min), 55°C (1 min), 72°C (1 min) for 35 cycles, then 72°C (10 min for one cycle) using HotStar Taq master mix (Qiagen). Gels were imaged using the Strata gene Eagle Eye system (La Jolla, CA).

**PCR detection of Human Alu sequences**

DNA was extracted from cells according to manufacturer's guidelines using the DNeasy Tissue Kit (Qiagen). Primers for detection of human Alu in hematopoietic cells were designed based on the published cDNA and genomic sequences, as follows: 5' TCACCCAACAGCCTAAATGA and 3'TGTGCAATAACACCTTACAGTCAA. This primer pair amplified a 160bp sequence under the following conditions: 95°C (15 min for one cycle), 94°C (1 min), 53°C (1 min), 72°C (1 min) for 35 cycles, then 72°C (10 min for one cycle) using HotStar Taq master mix.