Figure Legend and Method:

Supplemental Figure 1. mTRAIL does not have a cytotoxic effect on normal spleen, kidney, heart and brain cells from mice.

Supplemental method: Cr-release assay in normal primary cells from mice: Spleen, liver, kidney, cardiac muscle, brain were isolated from 8-weeks old BABL/c female mice as described below.

Splenocytes were isolated by pressing spleen on a stainless steel mesh (mesh size 100 μm) in a Petri dish containing Hank's solution. Red blood cells were lysed, remaining cells were washed and seeded in RPMI-1640 medium containing 10% FBS in cell culture dishes in a cell incubator at 37 °C (5% CO_2).

For liver cells, livers were perfused with Hank's solution and cut into tiny pieces, digested with 0.1% collagenase IV (Sigma Chemical Co., St. Louis, MO), for 45 min, and filtered through a stainless steel mesh (mesh size 100 μm). Cells were washed with Hank's solution. Cells were seeded in RPMI-1640 medium in collagen-coated 60-mm polystyrene dishes in a cell incubator at 37 °C (5% CO_2). After 2 hours medium was changed with RPMI-1640 medium containing 10% FBS.

To isolate kidney cells, the kidneys were minced into tiny pieces and digested in 1 mg/ml collagenase I (Sigma) for 30 min. The tissue was gently pressed through a stainless steel mesh (100-μm mesh size), and washed with Hank's solution. The cells were then cultured in RPMI-1640 medium containing 10% FBS.

Cardiac muscle cells were isolated by immersing the heart tissue strips 24 hours in 50% glycerine buffer solution containing 0.15 M NaCl, 20 mM sodium phosphate, 10 mM EGTA, 0.1 mM phenylmethylsulfonyl fluoride (PMSF). The tissues were dissociated by homogenization in Hank's solution. The cells were maintained in RPMI-1640 medium containing 10% FBS.

Single cells were prepared from brain tissue by slicing and digested with collagenase I (Sigma) at1.5 mg collagenase/ml buffer for 1.5 hours at 36 °C, followed by washing. Cells were cultured in medium 199 (Invitrogen Life Technologies, Inc., Grand Island, NY) containing 10% charcoal-treated sheep serum and antibiotics/antimycotics.

After two or three days of culture, the above cells were harvested by trypsinization and used as target cells in Cr-release assay, with 2PK-3 or mTRAIL/2PK-3 cells as effector cells. 1×10^4 ^{51}Cr-labeled target cells and effector cells were mixed in U-bottom wells of a 96-well microtiter plate at a 100:1 effector/target ratio. After 8 – 16 hours incubation, cell-free supernatants were collected, and radioactivity was measured in a gamma counter. The percentage of specific ^{51}Cr release was calculated as described previously (26).