

## Supplemental Figure 1. Generation of *Klrk1<sup>-/-</sup>* mice

We flanked exon 4 of the *Klrk1* gene with *loxP* sites and transfected this altered gene into Bruce4 embryonic stem cells derived from C57BL/6 (B6) mice. Chimeric mice were generated and germline transmission of the floxed *Klrk1* gene in the offspring of these founder mice was achieved. The targeting vector used to create the floxed *Klkr1* gene contained a neomycin-resistance element that was used for selection of ES cells with homologous recombination. In our targeting vector, the neomycin-resistance element is flanked by sites that allow for excision by the *FlpE* enzyme. Therefore, heterozygous mice with a floxed *Klrk1* gene were bred to B6 transgenic mice with a *FlpE* transgene. The offspring were genotyped to select mice with a floxed *Klkr1* allele, but lacking the neomycin-resistant element, and these mice were crossed with wild-type B6 mice to generate mice that have a floxed *Klrk1* gene, but lack the *FlpE* transgene. A subsequent cross of these mice will result in mice homozygous for the floxed *Klrk1* gene (*Klrk1*flox/flox mice). *Klrk1*-<sup>*i*</sup> mice were obtained from the breeding of *Klrk1*flox/flox mice and *Actb*-Cre deleter mice. Homozygous mice with a floxed *Klrk1* gene and an *Actb*-Cre transgene (*Klrk1*flox/flox Actb-Cre mice) were screened and mice in which NK cells do not express NKG2D were selected for the subsequent breeding. These mice were bred with *Klrk1*flox/flox mice and the offspring that do not have the *Actb*-Cre transgene in their germlines were further maintained as *Klrk1*-<sup>*i*</sup> mice.