Supplemental Methods & Data:

Histopathology & Immunofluorescence

For histopathology, samples were fixed, paraffin embedded and 5μm samples cut and stained with Hematoxylin and Eosin for blinded semi-quantitative GVHD scoring by an anatomical pathologist. Paneth cell numbers and Ki-67 staining were quantitated after immunohistochemical staining with anti-lysozyme or Ki-67 antibodies and goblet cells quantitated after staining with periodic acid Schiff stain. Enumeration was performed electronically using either the cytoplasmic or nuclear staining algorithms within Aperio ImageScope software (Version 12.3.2.8013). Three distinct areas devoid of processing artefact were chosen per slide and the average % of positive cells per slide assessed.

For immunofluorescence, tissues were fixed with 4% paraformaldehyde, then placed in 30% sucrose prior to being frozen. 7μm sections were cut and treated with Background Sniper (Biocare Medical) and 2% BSA for 30 min then anti-GFP and Ki-67 for 120 minutes at RT. Sections were counterstained with DAPI for 5 min and images acquired using a Zeiss 780-NLO Confocal Microscope (Zen software).

For multiplexed immunohistochemistry tissues were fixed with 10% formalin then placed in 70% ethanol prior to being paraffin-embedded. Sections (4 μm thickness on positively-charged slides) were baked for 1 hour at 60°C. The slides were then dewaxed and stained on a Leica BOND Rx stainer (Leica, Buffalo Grove, IL) using Leica Bond reagents for dewaxing (Dewax Solution), antigen retrieval/antibody stripping (Epitope Retrieval Solution 2), and rinsing after each step (Bond Wash Solution). Antigen retrieval and antibody stripping steps were performed at 100°C with all other steps at ambient temperature. Endogenous peroxidase was blocked with 3% H2O2 for 5 minutes followed by protein blocking with 10% normal mouse serum in TCT buffer (0.05M Tris, 0.15M NaCl, 0.25% Casein, 0.1% Tween 20, 0.05% ProClin300 pH 7.6) for 10 minutes. The first primary antibody (position 1) was applied for 60 minutes followed by the secondary antibody application for 10 minutes and the application of the tertiary TSA-amplification reagent (PerkinElmer OPAL fluor) for 10 minutes. A high stringency wash was performed after the secondary and tertiary applications using high-salt TBST solution (0.05M Tris, 0.3M NaCl, and 0.1% Tween-20, pH 7.2-7.6). Species specific Polymer HRP was used for all secondary applications, either Leica’s PowerVision (PV) Poly-HRP anti-Rabbit Detection or Vector Impress Rat polymer. The primary and secondary antibodies were stripped with retrieval solution for 20 minutes before repeating the process with the second primary antibody (position 2) starting with a new application of 3% H2O2. The process was repeated until five positions were completed. The stripping step was not performed after the final position. Slides were stained with DAPI for 5 minutes, rinsed for 5 minutes, and coverslipped with Prolong Gold Antifade reagent (Invitrogen/Life Technologies, Grand Island, NY).

Slides were cured overnight at room temperature, then whole slide images were acquired on the Vectra Polaris Quantitative Pathology Imaging System (Akoya Biosciences, Marlborough, MA). The
entire tissue was selected for processing using Phenochart and the images were spectrally unmixed using inForm software and exported as multi-image TIF files, which were analyzed with HALO image analysis software (Indica Labs, Coales, NM). Cellular analysis of the images was performed by first identifying cells based on nuclear recognition (DAPI stain), then measuring fluorescence intensity of the estimated cytoplasmic areas of each cell. A mean intensity threshold above background was used to determine positivity for each fluorochrome within the cytoplasm, thereby, defining cells as either positive or negative for each marker. The positive cell data was then used to define colocalized populations. Intestinal stem cells (ISC) and Paneth cells were defined as Lgr5-GFP+ EpCAM+ DAPI+ and Lysozyme+ EpCAM+ DAPI+, respectively. The average distance between every ISC to the nearest Paneth cell was calculated by Nearest Neighbor Analysis.

**Whole animal and organ Imaging**

Expansion of luciferase expressing T cells was quantitated through measurement of luciferin-luciferase signal intensity using the Xenogen imaging system (Xenogen IVIS 100; Caliper Life Sciences, CA, USA). Fur on the ventral surfaces was shaved and mice were injected with 500μg of luciferin subcutaneously and imaged 5 minutes later under continuous isoflurane-based anesthesia. After total body imaging, mice were again injected with luciferin and then euthanized and single organs were isolated and imaged. For assessment of donor T cell expansion after transplant, BALB/cLuc T cells were given at time of BMT. For assessment of APC function, expansion of TeteLuc T cells in response to selected antigen presenting cells was performed at day 15 post-transplantation, with 1-2x10⁶ flow cytometrically sorted T cells given on day 12 post-transplant.

**FITC Dextran**

Seven days post-transplantation, mice were fasted of food and water for 4 hours prior to oral gavage with 8mg of FITC labelled Dextran (MW 4kDa, Sigma-Aldrich) in 200μL of PBS. Peripheral blood was collected 4 hours later and serum separated. FITC-Dextran concentration in serum was determined using a Synergy H4 Fluorometer (Biotek) at excitation 485nm and emission 535nm.

**Cytokine analysis**

Serum IL-6, IL-17A, TNF and IFNγ were measured via murine Flex Array™ sets (BD Biosciences Pharmingen, San Diego, CA, USA) according to the manufacturer’s instructions. Samples were acquired on a BD LSR Fortessa and analyzed using FCAP Array™ Software (BD Biosciences).
Serum, SI and colon IL-28A/B were measured using the R&D Systems Mouse IL-28A/B (IFN-lambda 2/3) DuoSet ELISA on samples obtained from either serum or from mucosal intestinal homogenate as per a protocol provided by Invitrogen. The mucosa was scraped from the underlying muscle layer with a glass slide. The cells were lysed with Tris EDTA (10 mM Tris-HCl, and 1 mM EDTA, pH 7.4) containing 0.05% sodium azide, 1% Tween-80, 2 mM Phenylmethylsulfonyl fluoride (PMSF), and 1 microgram per milliliter of each of the following protease inhibitors: aprotinin, leupeptin, and pepstatin A prior to homogenization. The homogenate was then centrifuged (11,000 x g, 10 minutes at 4°C) and supernatant collected and filtered (4.5 micron filter).

qPCR

RT-qPCR was performed on RNA isolated from tissues obtained from naïve and post-transplant mice. Tissues were frozen in 500μL Trizol and then mechanically homogenized. RNA was then extracted using the QIAGEN RNeasy micro kit, converted to cDNA, and PCR performed using Taqman reagents. For *Ifnlr1* the Taqman Gene Expression Assay SM Mm00558035_m1 was used, and for the housekeeping gene SM Mm03024075_m1 (*Hprt*) was used. For *Reg3b, Reg3g, LysP* PCR GAPDH was used as the housekeeping gene and primers were used with Sybr Green Supermix using standard PCR conditions on an ABI ViiA7 PCR machine.

Mixed Lymphocyte reaction

BALB/c T cells were isolated from spleen and purified by magnetic bead selection. WT or *Ifnlr1*−/− DC were isolated from spleen via density gradient and further purified by magnetic bead selection. DC were irradiated with 2100cGy. Serial dilutions (20,000, 10,000, 5,000 and 0) of stimulator DC were plated with either CD4+ or CD8+ T cells at 200,000 T cells per well, in triplicate. After 4 days of culture, 100μL per well of 1:1000 ³H-thymidine was added. 18 hours later proportionate inclusion of ³H-thymidine was measured scintigrapically.

NK functional analysis

Congenically marked recipient B6 (CD45.2+) mice were conditioned with 1000cGy radiation on day -1 and then on day 0 co-injected with allogeneic 12x10⁶ Balb/c (CD45.1+) and syngeneic 12x10⁶ PTPxC57 (CD45.1+CD45.2+) bone marrow. 48 hours after transplantation mice were culled, spleens harvested, mashed in 2% FCS containing RPMI, and then filtered for single-cell suspensions. For in vivo cytotoxicity assays proportions of remaining syngeneic, allogeneic and recipient derived
haematopoietic cells were enumerated by flow cytometry. The index of cytotoxicity was calculated as the ratio of CD45.1+2 syngeneic derived cells to CD45.1 allogeneic derived cells. For recipient NK transcriptional profiling, splenocytes were isolated 24 hours after BMT (48 hours after irradiation) and stained with 7-AAD, CD45.1, CD45.2, CD3, NK1.1 and NKp46 for subsequent cell sorting.

16S ribosomal microbial sequencing

DNA was extracted from 50-100 mg of fecal material using an initial bead beating step followed by extraction using the Maxwell 16 Research Instrument (Promega, USA), according to the manufacturer’s protocol, with the Maxwell 16 Tissue DNA Kit (Promega, USA). DNA concentration was measured using a Qubit assay (Life Technologies, USA) and was adjusted to a concentration of 5 ng/µl. The 16S RNA gene encompassing the V6 to V8 regions was targeted using the 803F and 1392R primers modified to contain Illumina specific adapter sequence. Preparation of the 16S library was performed as described, using the workflow outlined by Illumina (#15044223 Rev.B). In the first stage, PCR products of ~466 bp were amplified according to the specified workflow, with an alteration in polymerase used to substitute Q5 Hot Start High-Fidelity 2X Master Mix (New England Biolabs, USA) in standard PCR conditions. Resulting PCR amplicons were purified using Agencourt AMPure XP beads (Beckman Coulter, USA). Purified DNA was indexed with unique 8 bp barcodes using the Illumina Nextera XT 384 sample Index Kit A-D (#FC-131-1002, Illumina, USA), in standard PCR conditions, with Q5 Hot Start High-Fidelity 2X Master Mix. Indexed amplicons were pooled together in equimolar concentrations and sequenced on the MiSeq Sequencing System (Illumina, USA), using paired end sequencing with V3 300 bp chemistry, at the Australian Centre for Ecogenomics according to manufacturer’s protocol. Heat map includes OTUs identified as significantly different \(p<0.001\) between separately housed WT and \(Ifnrl1^{-/-}\) at week 4, where OTU relative abundance exceeds 2% in at least one sample. Each column includes scaled read counts for one mouse. Read counts normalized using metagenomeSeq.

For analysis of 16S microbial sequencing, reads were cleaned of adapter sequences using Cutadapt and trimmed using Trimmomatic employing a sliding window of 4 bases with an average base quality above 15, followed by hard-trimming to 250 bases with exclusion of reads less than this length. Remaining forward reads were processed following the QIIME2 workflow using DADA2 to de-noise sequences. Taxonomy assignment was performed on amplicon sequence variants using BLAST against the SILVA reference database version 132. Differential abundance analysis was performed on raw read counts using DESeq2. Counts were normalized prior to principal component analysis (PCA) and heat map visualization using cumulative sum scaling implemented within metagenomeSeq. PCA was performed using the rda function within the vegan R package. Heat maps were generated using pheatmap.
RNAseq

For intestinal epithelial analyses single cells were isolated using the Lamina Propria dissociation kit (Miltenyi Biotec), stained with 7AAD, CD45.2 and EpCAM and sorted based on GFP expression into Lgr5+ and Lgr5− populations (see Supplemental Figure 5). For NK transcriptome analyses recipient NK cells were gated as 7-AAD−CD45.2+CD3−NK1.1+NKp46+. All subsequent cell sorting was performed on a BD FACSARia III cytometer. Sorted Lgr5+/− cells were treated with TRIzol and cryopreserved at -80°C. RNA was subsequently extracted after a second chloroform extraction step using the QIAGEN RNeasy Micro Kit. Sorted NK cells were stored at -80°C in Arcturus® PicoPure® RNA Isolation Buffer and RNA isolated as per manufacturer’s instructions. RNA libraries were prepared using the NEBnext Ultra RNA Library Prep Kit for Illumina (New England Biolabs), assessed for size, and quantified using the 2100 Bioanalyzer (Agilent Technologies) and Qubit fluorometer (Thermofischer Scientific). Libraries were sequenced using high output single-end 75 cycle sequencing kits (version 2) on the Illumina Nextseq 550 platform. Sequence reads in each fastq file were trimmed for adapter sequences using Cutadapt 32 (version 1.11) and aligned using STAR 33 (version 2.5.2a) to the mm19 assembly with the gene, transcript, and exon features of Ensembl (release 70) gene model. Expression was estimated using RSEM 34 (version 1.2.30) and was used as input to assess differential gene expression between groups.

Differential gene expression was determined using the edgeR package45 within R v3.3.4 and significance defined as $p < 0.05$ after Benjamini-Hochberg false discovery rate correction. Pathway analysis was performed by single sample gene set variation analysis via the GSVA package46 using KEGG, BioCarta, Reactome and Gene Ontology (GO) pathway databases and only gene sets between 25-500 genes considered. Heat maps were generated using heatmap.2 function in gplots v3.0.1 R package. Canonical Pathway enrichment analysis for differentially expressed genes (log2 Fold-change $>|0.58|$ and adj. p-value $<0.05$) across PEG-rIL-29-treated Lgr5+ and Lgr5− samples relative to genotype-matched PBS-treated samples was done using Ingenuity Pathway Analysis (IPA)49. IPA function enrichment was calculated using a right-tailed Fisher exact test with a threshold of significance set at P value of 0.05. Inferences in the significant activation state (z-score $>|2|$) of canonical pathways, upstream regulatory transcriptional regulators, cytokines and kinases were done using IPA. Positive z-scores reflect a predicted activation state, while negative z-scores reflect the inhibition of upstream regulatory activity.
### Experimental Models

| Model Description | Source/Identifier |
|-------------------|------------------|
| C57Bl/6J (B6)     | Animal Resources Centre, RRID:IMSR_ARC:B6 |
| BALB/c            | Animal Resources Centre, RRID:IMSR_ARC:BC |
| B6.SJL-Ptcra Pepcb/BoyJ (PTP) | Animal Resources Centre, RRID:IMSR_ARC:PTP |
| BALB/c CD45.1     | QIMRB, N/A         |
| C57Bl/6J.DBA/2 (B6D2F1) | Animal Resources Centre, N/A   |
| B6.SJL-Ptcra Pepcb/BoyJ.C57Bl/6J (PTPxC57) | QIMRB, N/A         |
| B6.lfnr1-/-       | Bristol Myers Squibb (under MTA), N/A |
| B6.129P2-Lgr5<sup>CreERT2</sup>/Ifnlr1<sup>fl/fl</sup> | The Jackson Laboratory, JAX:008875 |
| B6.Cg-Tg(Tcra,Tcrb)3Ayr/J (TEa) | Negrin Laboratory, Stanford Medicine, N/A |
| B6.FVB-Tg(CAG-luc,-GFP)L2G85Chco/JThy1<sup>fl</sup>.Thy1<sup>fl</sup> (B6<sup>Luc</sup>) | Negrin Laboratory, Stanford Medicine, N/A |
| B6.Cg-Tg(Tcra,Tcrb)3Ayr/J (TEa<sup>Luc</sup>) | The Alexander Rudensky Laboratory, MSKCC, N/A |
| B6.Cg-Tg(Tcra,Tcrb)3Ayr/J (TEa<sup>Luc</sup>) | QIMRB, N/A         |
| lfnr1<sup>fl/fl</sup> | The Kotenko Laboratory, New Jersey Medical School, N/A |
| Lgr5-EGFP-ires-creERT2.lfnr1<sup>fl/fl</sup> (Lgr5<sup>Cre</sup>,lfnr1<sup>fl/fl</sup>) | QIMRB, N/A         |
| B6(Cg)-Ncr1tm1.1(icre)Viv/Orl (NKp46<sup>Cre</sup>) | The Vivier Laboratory, Centre d’Immunologie de Marseille-Luminy, N/A |
| lfnr1<sup>fl/fl</sup>.B6(Cg)-Ncr1tm1.1(icre)Viv/Orl (NKp46<sup>Cre</sup>,lfnr1<sup>fl/fl</sup>) | QIMRB, N/A         |
| BCR-ABL1up98HoxA9 B6D2F1 primary murine leukemia | QIMRB, N/A         |
| MLL-AF9 Balb/c and B6D2F1 primary murine leukemias (Brueidigam et al., 2014) | QIMRB, N/A         |
| L-WRN (ATCC® CRL-3276™) | ATCC, Cat#CRL-3276; RRID:CVCL_DA06 |
| PK136 (ATCC® HB-191™) | ATCC, Cat#HB-191; RRID:CVCL_7695 |

**Supplemental Table 1.** Mice, tumours and cell lines used in this study.
## Antibodies

| Antibodies                                      | Source                        | Identifier          |
|------------------------------------------------|------------------------------|---------------------|
| BioMag Goat Anti-Rat IgG                       | QIAGEN                        | Cat#310107          |
| CD16/CD32 (2.4G2)                              | in-house, QIMRB               | N/A                 |
| Rabbit polyclonal anti-Lysozyme (EC 3.2.1.17)  | Agilent                       | Code#A0099          |
| Rabbit anti-Ki67 (SP6)                         | Abcam                        | Cat#16667           |
| Mouse anti-CD45.1 FITC (A20)                   | BioLegend                     | Cat#109076          |
| Mouse polyclonal anti-IL-22 PE (Poly5164)      | BioLegend                     | Cat#516404          |
| Mouse anti-CD45.2 APC (104)                    | BioLegend                     | Cat#109814          |
| Mouse anti-IL17A PE/Cy7 (TC11-18H10.1)         | BioLegend                     | Cat#506922          |
| Mouse anti-CD8a APC/Cy7 (53-6.7)               | BioLegend                     | Cat#1200714         |
| Mouse anti-IFN-γ Brilliant Violet 605 (53-2.1) | BioLegend                     | Cat#140318          |
| Mouse anti-CD4 BV786 (GK1.5)                   | BD Biosciences               | Cat#563331          |
| Mouse anti-H-2D^d FITC (KH95)                  | BioLegend                     | Cat#111506          |
| Mouse anti-CD4 PerCP-Cy5.5 (RM4-5)             | BD Biosciences               | Cat#550954          |
| Mouse anti-H-2D^d PE (34-2-12)                 | BioLegend                     | Cat#110608          |
| Mouse anti-CD19 PE-CF594 (1D3)                 | BD Biosciences               | Cat#562291          |
| Mouse anti-IFN-γ PE-Cy7 (XMG1.2)               | BioLegend                     | Cat#505826          |
| Mouse anti-IL-17A Alexa Fluor 700 (TC11-18H10.1)| BioLegend                    | Cat#506914          |
| Mouse anti-CD8a V500 (53-6.7)                  | BD Biosciences               | Cat#560776          |
| Mouse anti-CD8a PerCP-Cy5.5 (53-6.7)           | BD Biosciences               | Cat#551162          |
| Mouse anti-IFN-γ PE (XMG1.2)                   | BioLegend                     | Cat#505808          |
| Mouse anti-CD4 PE-Cy7 (RM4-5)                  | BD Biosciences               | Cat#552775          |
| Mouse anti-CD45.1 APC/Cy7 (A20)                | BioLegend                     | Cat#110716          |
| Mouse anti-CD122 PerCP-eFluor 710 (TM-b1)     | Thermo Fisher                | Cat#46-1222-82      |
| Mouse anti-TCR γ/δ PE (UC7-13D5)               | BioLegend                     | Cat#107502          |
| Mouse anti-CD17 PE-CF594 (1D3)                 | BD Biosciences               | Cat#532291          |
| Mouse anti-CD35 (NKp46) PE/Cy7 (29A1.4)       | BioLegend                     | Cat#137618          |
| Mouse anti-CD3c APC (145-2C11)                 | BioLegend                     | Cat#100312          |
| Mouse anti-CD62L Alexa Fluor 700 (MEL-14)      | BioLegend                     | Cat#104426          |
| Mouse anti-CD4 APC/Cy7 (GK1.5)                 | BioLegend                     | Cat#100414          |
| Mouse anti-CD44 Brilliant Violet 421 (IM7)     | BioLegend                     | Cat#103040          |
| Rabbit polyclonal anti-GFP (ab290)             | Abcam                        | Cat#ab290           |
| Mouse polyclonal anti-Ki-67 (MIB-5)            | Dako                          | Cat#MT248           |
| Mouse anti-NK1.1 PE (PK136)                    | Biologics                     | Cat#108708          |
| Mouse anti-NK1.1 (PK136)                       | ATCC                          | #HB-191             |
| Rabbit Polyclonal anti-GFP Rbt PV 570          | Invitrogen                    | A11122              |
| Rabbit anti-CD8α Rbt PV 620 (D4W22)            | Cell Signalling              | 98941               |
| Rabbit polyclonal anti-Lysozyme Rbt PV 480    | DAKO                          | A0099               |
| Rabbit anti-EpCAM Rbt PV 690 (EPR20533)        | Abcam                         | Ab23785             |
| Rat anti-CD4 Rat Impress HRP 520 (4SM95)       | ebioscience                   | 14-9766-32          |

**Supplemental Table 2.** Antibodies used in this study.
| REAGENT                                      | SOURCE                      | IDENTIFIER               |
|----------------------------------------------|-----------------------------|--------------------------|
| PEG-riL-29                                   | ZymoGenetics                | Lot#ZO0702/A3023F        |
| D-Luciferin                                  | Gold Biotechnology          | Cat#LUCK-1G; CAS: 115144-35-9 |
| 7-Aminoactinomycin D                         | Sigma Aldrich               | Cat#A9400-5MG            |
| Thymidine, [Methyl-3H]-                      | Perkin Elmer                | Cat#NET027/E005MC        |
| Phorbole 12-myristate 13-acetate (PMA)        | Sigma Aldrich               | Cat#P1585-1MG            |
| Iononycin                                    | Sigma Aldrich               | Cat#I0634-1MG            |
| Brefeldin A Solution (1000X)                 | BioLegend Inc               | Cat#420601               |
| Carboxyfluorescein succinimidyl ester (CFSE) | Sigma Aldrich               | Cat#21888-25MG-F         |
| Cultrex PathClear Reduced Growth Factor BME  | R&D Systems                 | Cat#RDS353301002         |
| SB431542                                     | Selleck Chemicals           | Cat#S1067-50mg           |
| Y-27632 dihydrochloride                      | Tocris Bioscience          | Cat#RDS125410            |
| G 418 disulfate salt solution                | Sigma Aldrich               | Cat#G8168-10ML           |
| Hygromycin B Gold                            | Invivogen                   | Cat#ant-hg-1             |
| Ethylenediaminetetraacetic Acid Disodium salt dihydrate AR | Chem-supply | CAS#6381-92-6            |
| Trypsin 2.5%                                 | Thermo Fisher               | Cat#15090-046            |
| TRIzol reagent                               | Thermo Fisher               | Cat#15596-026            |
| Arcturus® PicoPure® RNA Isolation Kit        | Thermo Fisher               | Cat#K10204               |
| Bayer Baytril 50 Injection 50ml (enrofloxacin 50mg/mL) | Provet | Cat#BAYT 1               |
| Fluorescein isothiocyanate–dextran 500mg     | Sigma Aldrich               | Cat# 46944-500MG-F       |
| Agencourt AMPure XP                          | Beckman Coulter             | Cat# A63881              |
| Q5® Hot Start High-Fidelity 2X Master Mix    | New England Biolabs         | Cat#M0494L               |

**Supplemental Table 3.** Chemicals, peptides, and recombinant proteins used in this study.
| Symbol | Gene Description                                                                 | logFC  | P Value      | FDR          |
|--------|----------------------------------------------------------------------------------|--------|--------------|--------------|
| Ncr1   | natural cytotoxicity triggering receptor 1                                        | -0.491| 3.06E-46     | 2.91E-42     |
| Syne2  | synaptic nuclear envelope 2                                                       | -0.398| 6.44E-10     | 3.06E-06     |
| Cdkn1a | cyclin-dependent kinase inhibitor 1A (P21)                                        | 0.301 | 5.32E-09     | 1.48E-05     |
| Phlda3 | pleckstrin homology-like domain, family A, member 3                              | 0.272 | 6.24E-09     | 1.48E-05     |
| Cma1   | chymase 1, mast cell                                                             | 0.264 | 1.20E-08     | 2.29E-05     |
| Fam46a | family with sequence similarity 46, member A                                     | -0.189| 2.34E-07     | 0.000291     |
| Ephx1  | epoxide hydrolase 1, microsomal                                                  | 0.347 | 2.45E-07     | 0.000291     |
| Wscd2  | WSC domain containing 2                                                          | 0.300 | 2.27E-07     | 0.000291     |
| Abca1  | ATP-binding cassette, sub-family A (ABC1), member 1                               | -0.236| 3.06E-07     | 0.000323     |
| Slco3a1| solute carrier organic anion transporter family, member 3a1                       | -0.416| 6.89E-07     | 0.000596     |
| Jun    | Jun oncogene                                                                     | 0.327 | 6.48E-07     | 0.000596     |
| Zfp26  | zinc finger protein 26                                                           | -0.279| 1.19E-06     | 0.000943     |
| Gdf11  | growth differentiation factor 11                                                | -0.403| 2.95E-06     | 0.00216      |
| Plk2   | polo-like kinase 2                                                               | 0.281 | 8.87E-06     | 0.00527      |
| S100a11| S100 calcium binding protein A11 (calgizzarin)                                   | 0.143 | 8.80E-06     | 0.00527      |
| Psrc1  | proline/serine-rich coiled-coil 1                                                | 0.238 | 8.41E-06     | 0.00527      |
| Aldh2  | aldehyde dehydrogenase 2, mitochondrial                                          | 0.267 | 1.14E-05     | 0.00542      |
| St8sia4| ST8 alpha-N-acetyl-neuraminide alpha-2,8-sialyltransferase 4                     | -0.171| 1.10E-05     | 0.00542      |
| Gm10116| predicted pseudogene 10116                                                       | 0.318 | 1.09E-05     | 0.00542      |
| Gm1966 | predicted gene 1966                                                              | -0.308| 9.86E-06     | 0.00542      |
| 2010016i18Rik | RIKEN cDNA 2010016i18 gene                                                        | -0.282| 1.41E-05     | 0.00638      |
| Hist1h1c| histone cluster 1, H1c                                                            | 0.446 | 1.72E-05     | 0.00742      |
| Pygl   | liver glycogen phosphorylase                                                      | 0.387 | 3.03E-05     | 0.0101       |
| Herc2  | hect (homologous to the E6-AP (UBE3A) carboxyl terminus) domain and RCC1 (CHC1)-like domain (RLD) 2 | -0.182 | 3.09E-05 | 0.0101 |
| Mga    | MAX gene associated                                                              | -0.183| 3.01E-05     | 0.0101       |

**Supplemental Table 4.** Top 25 differentially expressed genes in NKp46\(^{Cre}\).\(^{Ifnlr1^{fl.fl}}\) versus NKp46\(^{Cre-}\).\(^{Ifnlr1^{fl}}\) cells. RNA sequencing from sort purified single cells from 48hrs post-transplant (n = 6 mice per group). LogFC = the log2 transformed fold change obtained from edgeR analyses. FDR (false discovery rate) = the Benjamini-Hochberg (FDR) adjusted P value obtained from edgeR analyses.
| Symbol | Gene Description                                      | logFC | P Value  | FDR    |
|--------|-------------------------------------------------------|-------|----------|--------|
| Lgals9 | lectin, galactose binding, soluble 9                  | 3.73  | 2.70E-15 | 9.73E-12|
| Ddx58  | DEAD (Asp-Glu-Ala-Asp) box polypeptide 58             | 3.10  | 2.38E-15 | 9.73E-12|
| Xaf1   | XIAP associated factor 1                              | 4.82  | 1.36E-15 | 9.73E-12|
| Adar   | adenosine deaminase, RNA-specific                     | 2.19  | 6.45E-15 | 1.43E-11|
| Rnf213 | ring finger protein 213                               | 3.98  | 6.63E-15 | 1.43E-11|
| Stat1  | signal transducer and activator of transcription 1    | 3.51  | 8.27E-15 | 1.49E-11|
| Ifi44  | interferon-induced protein 44                         | 7.55  | 1.09E-14 | 1.69E-11|
| Irf9   | interferon regulatory factor 9                        | 2.52  | 1.97E-14 | 2.66E-11|
| Dhx58  | DEXH (Asp-Glu-X-His) box polypeptide 58               | 4.70  | 2.87E-14 | 3.11E-11|
| Ogfr   | opioid growth factor receptor                         | 2.01  | 2.84E-14 | 3.11E-11|
| Parp9  | poly (ADP-ribose) polymerase family, member 9        | 2.80  | 3.47E-14 | 3.41E-11|
| Parp14 | poly (ADP-ribose) polymerase family, member 14       | 3.10  | 4.28E-14 | 3.85E-11|
| Trim30a| tripartite motif-containing 30A                       | 4.96  | 5.52E-14 | 4.26E-11|
| Lgals3bp| lectin, galactoside-binding, soluble, 3 binding protein | 3.11 | 5.32E-14 | 4.26E-11|
| Rtp4   | receptor transporter protein 4                        | 5.11  | 6.51E-14 | 4.69E-11|
| Gbp6   | guanylate binding protein 6                           | 4.28  | 8.13E-14 | 5.50E-11|
| Mitd1  | MIT, microtubule interacting and transport, domain containing 1 | 2.06  | 1.09E-13 | 6.94E-11|
| Tap1   | transporter 1, ATP-binding cassette, sub-family B (MDR/TAP) | 3.57  | 1.41E-13 | 8.48E-11|
| Ifi35  | interferon-induced protein 35                         | 2.76  | 1.63E-13 | 9.25E-11|
| Ly6e   | lymphocyte antigen 6 complex, locus E                 | 2.79  | 2.41E-13 | 1.24E-10|
| Stat2  | signal transducer and activator of transcription 2    | 3.65  | 2.31E-13 | 1.24E-10|
| Hsh2d  | haematopoietic SH2 domain containing                   | 2.93  | 2.89E-13 | 1.42E-10|
| Gbp3   | guanylate binding protein 3                           | 5.99  | 3.49E-13 | 1.64E-10|
| H2-Q10 | histocompatibility 2, Q region locus 10               | 2.63  | 4.96E-13 | 2.23E-10|
| Irgm1  | immunity-related GTPase family M member 1             | 3.56  | 5.70E-13 | 2.47E-10|

**Supplemental Table 5.** Top 25 differentially expressed genes in ISC treated *in vivo* with PEG-rIL-29. RNA sequencing from sort purified single colonic stem cells (LGR5+) derived from either rIL-29 or PBS treated mice (n = 5 mice per group). LogFC = the log2 transformed fold change obtained from edgeR analyses. FDR (false discovery rate) = the Benjamini-Hochberg (FDR) adjusted P value obtained from edgeR analyses.
### Supplemental Table 6.

Top 25 differentially expressed genes in intestinal epithelial cells treated *in vivo* with PEG-rIL-29. RNA sequencing from sort purified single colonic epithelial cells (LGR5−) derived from either rIL-29 or PBS treated mice (n = 5 mice per group). LogFC = the log2 transformed fold change obtained from edgeR analyses. FDR (false discovery rate) = the Benjamini-Hochberg (FDR) adjusted P value obtained from edgeR analyses.

| Symbol | Gene Description                                           | logFC | P Value    | FDR       |
|--------|------------------------------------------------------------|-------|------------|-----------|
| Xaf1   | XIAP associated factor 1                                   | 4.61  | 3.26E-15   | 3.52E-11  |
| Ifi44  | interferon-induced protein 44                              | 7.55  | 1.37E-14   | 7.40E-11  |
| Irf9   | interferon regulatory factor 9                             | 2.45  | 3.21E-14   | 9.90E-11  |
| Dhx58  | DEXH (Asp-Glu-X-His) box polypeptide 58                    | 4.60  | 4.46E-14   | 9.90E-11  |
| Trim30a| tripartite motif-containing 30A                             | 5.02  | 5.49E-14   | 9.90E-11  |
| Rtp4   | receptor transporter protein 4                             | 5.25  | 5.46E-14   | 9.90E-11  |
| Ddx58  | DEAD (Asp-Glu-Ala-Asp) box polypeptide 58                  | 2.49  | 8.50E-14   | 1.31E-10  |
| Lgals9 | lectin, galactose binding, soluble 9                       | 2.92  | 1.25E-13   | 1.32E-10  |
| Parp9  | poly (ADP-ribose) polymerase family, member 9             | 2.58  | 1.35E-13   | 1.32E-10  |
| Stat1  | signal transducer and activator of transcription 1         | 2.90  | 1.59E-13   | 1.32E-10  |
| Adar   | adenosine deaminase, RNA-specific                           | 1.82  | 1.47E-13   | 1.32E-10  |
| Parp14 | poly (ADP-ribose) polymerase family, member 14            | 2.88  | 1.43E-13   | 1.32E-10  |
| Rnf213 | ring finger protein 213                                    | 3.29  | 1.22E-13   | 1.32E-10  |
| Gbp3   | guanylate binding protein 3                                | 5.95  | 5.21E-13   | 4.02E-10  |
| Gbp6   | guanylate binding protein 6                                | 3.74  | 6.00E-13   | 4.32E-10  |
| Sp100  | nuclear antigen Sp100                                     | 4.50  | 9.37E-13   | 6.02E-10  |
| Trim34a| tripartite motif-containing 34A                            | 2.92  | 9.46E-13   | 6.02E-10  |
| Lgals3bp| lectin, galactoside-binding, soluble, 3 binding protein    | 2.58  | 1.01E-12   | 6.07E-10  |
| Mx2    | myxovirus (influenza virus) resistance 2                   | 6.15  | 1.55E-12   | 7.96E-10  |
| Mitd1  | MIT, microtubule interacting and transport, domain containing 1 | 1.77  | 1.47E-12   | 7.96E-10  |
| Usp18  | ubiquitin specific peptidase 18                            | 6.99  | 1.54E-12   | 7.96E-10  |
| Qgfr   | opioid growth factor receptor                               | 1.58  | 1.75E-12   | 8.22E-10  |
| Apol9a | apolipoprotein L 9a                                        | 5.99  | 3.49E-13   | 1.64E-10  |
| Stat2  | signal transducer and activator of transcription 2         | 2.63  | 4.96E-13   | 2.23E-10  |
| Ifi2712b| interferon, alpha-inducible protein 27 like 2B            | 3.56  | 5.70E-13   | 2.47E-10  |
Supplemental Figure 1. Histological examination of naïve WT and Ifnlr1−/− GI tissues. A) Representative H&E stained images of colon and SI from naïve WT and Ifnlr1−/− mice. B) Pre-transplant weight of WT and Ifnlr1−/− mice; WT and Ifnlr1−/− were not aged matched (WT n = 14, Ifnlr1−/− n = 20, combined from 2 experiments). C) Representative period acid-schiff stained images of colon and SI from naïve WT and Ifnlr1−/− mice demonstrating goblet cells. D) Enumeration of goblet cells in WT and Ifnlr1−/− mice (n = 3). E) Representative images of colon and SI from naïve WT and Ifnlr1−/− mice stained by immunohistochemistry for lysozyme showing Paneth cells. F) Enumeration of Paneth cells in WT and Ifnlr1−/− mice (n = 3). Data are presented as mean ± SEM. P values calculated using two tailed Mann-Whitney T test.
Supplemental Figure 2. GVL capacity of WT and Ifnrlr1−/− donor grafts against leukemic cell lines. A) Ifnrlr1−/− BM + T BMT grafts retain GVL capacity. Balb/c recipients were transplanted with BM ± T cells from WT or Ifnrlr1−/− donors, together with recipient type MLL-AF9 leukemia expressing GFP. The absolute number of GFP+ leukemia cells in peripheral blood was determined thereafter (n = 18, combined from 3 replicate experiments). B) B6D2F1 recipients were transplanted with BM ± T cells from WT or Ifnrlr1−/− donors, together with recipient type BCR-ABL nup98hoxA9 leukemia expressing GFP. The absolute number of GFP+ leukemia cells in peripheral blood was determined thereafter (n = 12, combined from 2 experiments). Data are presented as mean ± SEM.
Supplemental Figure 3. The effect of conditional deletion of Ifnlr1 in NK cells following allogeneic BMT. A-B) RNAseq from sort purified NKP46\textsuperscript{Cre+}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} and NKP46\textsuperscript{Cre−}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} splenic NK cells isolated 24 hours after allogeneic BMT (48hrs after lethal irradiation). A) Heat map showing the pattern of differential gene expression between NKP46\textsuperscript{Cre+}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} and NKP46\textsuperscript{Cre−}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} splenic NK. B) Heat map showing canonical gene sets associated with functional pathways identified in NKP46\textsuperscript{Cre+}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} versus NKP46\textsuperscript{Cre−}.\textsuperscript{Ifnlr1}\textsuperscript{fl/fl} (n = 6 per group) by GSVA analysis.
Supplemental Figure 4. Paneth cell evaluation in WT and Ifnlr1−/− GI tissues following allogeneic BMT. A) Representative images and B) Enumeration of Paneth cells from SI at day 7 post-transplant (WT, n = 8, Ifnlr1−/−, n = 7, TCD, n = 6 combined from 2 experiments). C) Day 7 post-transplant qPCR enumeration of Cryptidins (pan-cryptidin), Lysozyme P (Lysp) and Regenerating islet-derived protein III gamma (Reg3g) defensin expression from WT and Ifnlr1−/− recipient mice lethally irradiated (1000cGy), and transplanted with BALB/c derived BM and T-cells (n = 4). Data are presented as mean ± SEM. P values calculated using two tailed Mann-Whitney T test, **p < .01.
Supplemental Figure 5. Sort strategy for isolation of Lgr5+ cells from GI tissues. A) Representative images from FACS sorting strategy for isolation of GFP+ Lgr5+ cells from colonic tissue samples. B) Expression of intestinal stem cell markers from GFP+ and GFP− sorted fractions by RNA sequencing (n = 5, combined from 4 experiments). Data are presented as mean ± SEM. p values calculated using the Benjamini-Hochberg procedure corrected for multiple comparisons. ****p < .0001.
Supplemental Figure 6. Canonical Pathway enrichment analysis for differentially expressed genes across sorted Lgr5⁺ and Lgr5⁻ epithelial cells following PEG-rIL-29-treatment relative to genotype-matched PBS-treated samples. Representation of all canonical pathways found to be enriched in the dataset. Bubbles represent significant pathway enrichment, as determined by Fisher exact test. Bubble diameter represents the -log10 P value as determined by Fisher's exact test. Crosses signify a lack of significant pathway enrichment. Color indicates predicted pathway activation (red) or predicted inhibition (blue). White bubbles represent significant functional enrichment of pathways with no available prediction patterns. Data are presented as log2 Fold-change >|0.58| and adj. p-value <0.05).
Supplemental Figure 7. Interferon Lambda treatment effects on hematopoietic engraftment following allogeneic BMT. (A) Frequencies and (B) absolute numbers of donor derived WBC, neutrophils, monocytes, B cells, eosinophils, dendritic cells, T cells and NK cells in peripheral blood 7 and 14 days after Balb/c → B6 BMT in the presence and absence of peri-transplant PEG-rIL29 treatment. Data are presented as mean ± SEM. P values calculated using two tailed Mann-Whitney T test, **p < .01.
Supplemental Figure 8. Interferon Lambda treatment effects on GI tissues following allogeneic BMT. A) Representative images and B) Enumeration of Lysozyme+ Paneth cells at day 7 post-BMT from ileum from PBS or PEG-rIL-29 B6 recipients of BALB/c BM + T-cells following NK cell depletion (n = 9, combined from 2 experiments). C) Quantification of distance of GFP+ Lgr5+
intestinal cells from Paneth cells as for A) and B). D) Enumeration of CD4^+ and CD8 T cells from ileum and spleen as for A) and B). E) Numbers of GFP^+ Lgr5^+ cells isolated from small intestine and ileum from digested gut preparations at day 7 post-BMT (BALB/C → B6) from PBS or PEG-rIL-29 treated recipients without recipient NK depletion (n = 6, combined from 2 experiments). L) Representative immunoflourescent images as for E). G) Exemplar dual immunoflourescent images as for K, with secondary staining for GFP and Ki-67. Data are presented as mean ± SEM. p values calculated using two tailed Mann-Whitney T test. *p < .05, **p < .01, ***p < .001, ****p < .0001.