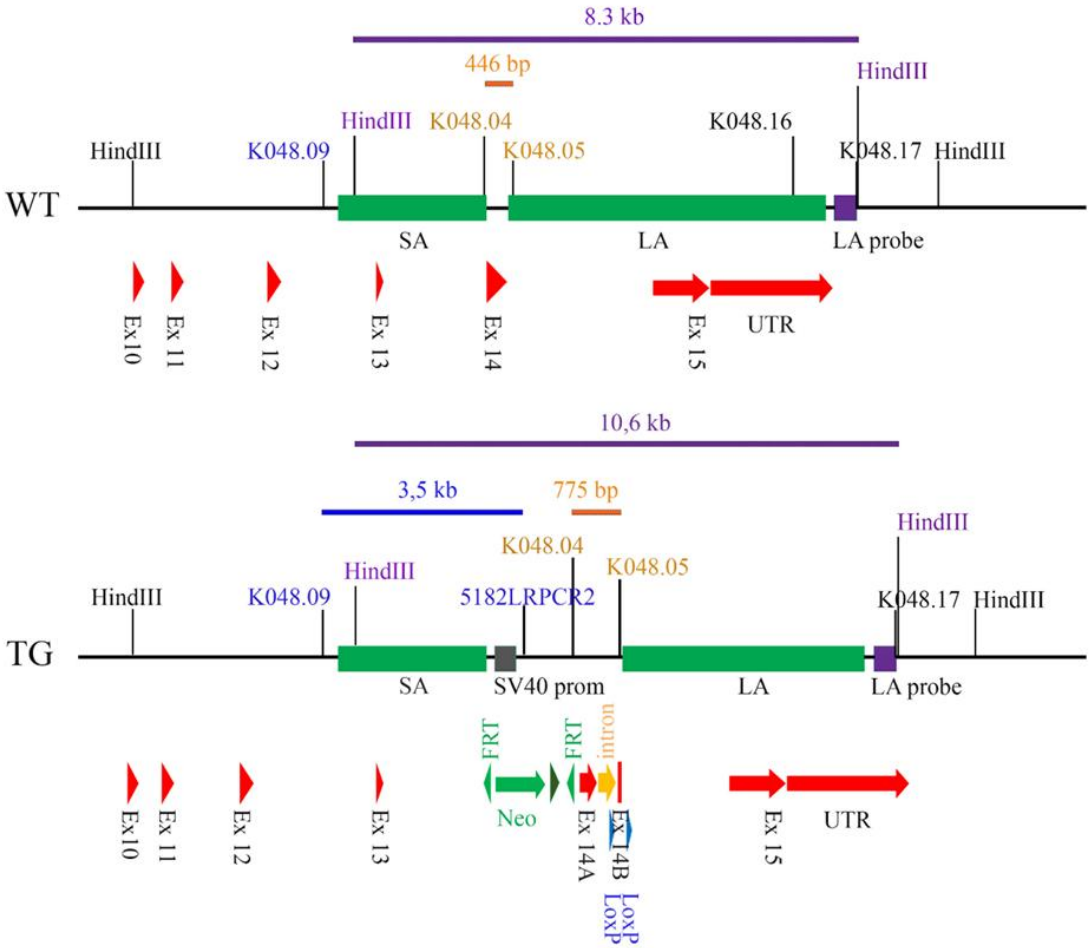
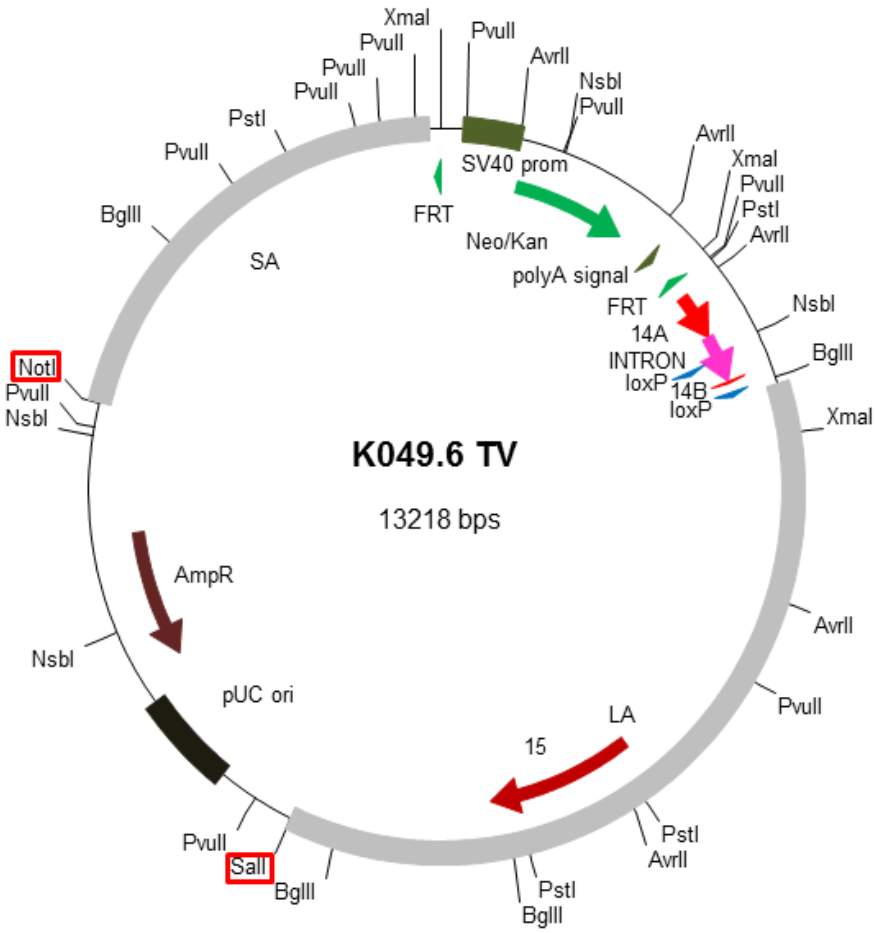
Supplemental Figure 1



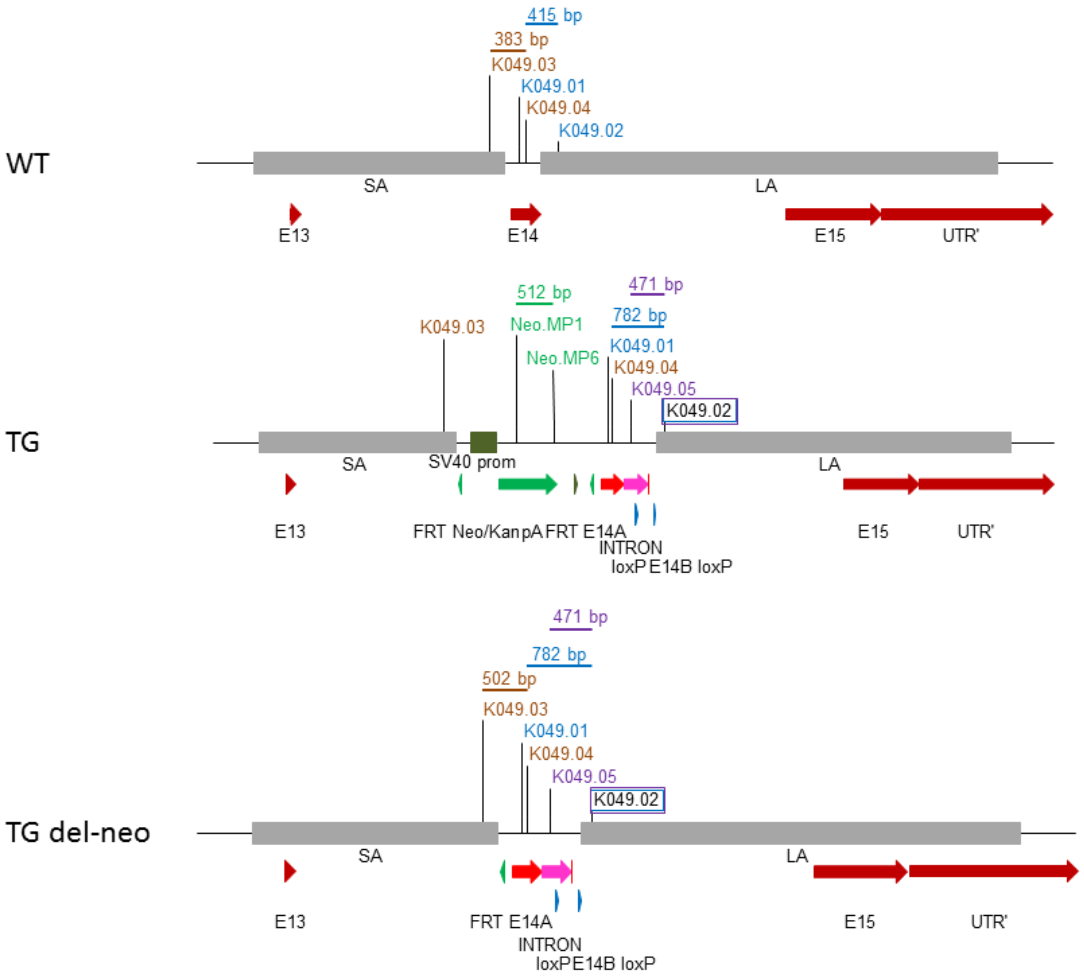
Supplemental Figure 1: Schematic drawing of the different *Dsg2* alleles carried by heterozygous ES cells. The upper panel shows the WT allele, the lower panel the TG allele. Homologous regions (SA: short arm of homology; LA: long arm of homology) used for recombination is depicted as grey box. Dsg2 exon parts Ex 14A and Ex 14B are marked in red, separated by the artificial intron (orange), which contains the LoxP site (blue). For selection, an FRT-flanked neomycin resistance cassette (green) is inserted. The primers K048.9 and 5182LRPCR2 were used for PCR screening of recombined ES cells. The corresponding fragment size is indicated in blue. Primers used for confirmation of the presence of the intron including the LoxP sites are indicated in brown. The restriction enzyme HindIII, used for Southern blot analysis, and the corresponding external probe (LA-probe) and fragments are indicated in purple.

Supplemental Figure 2



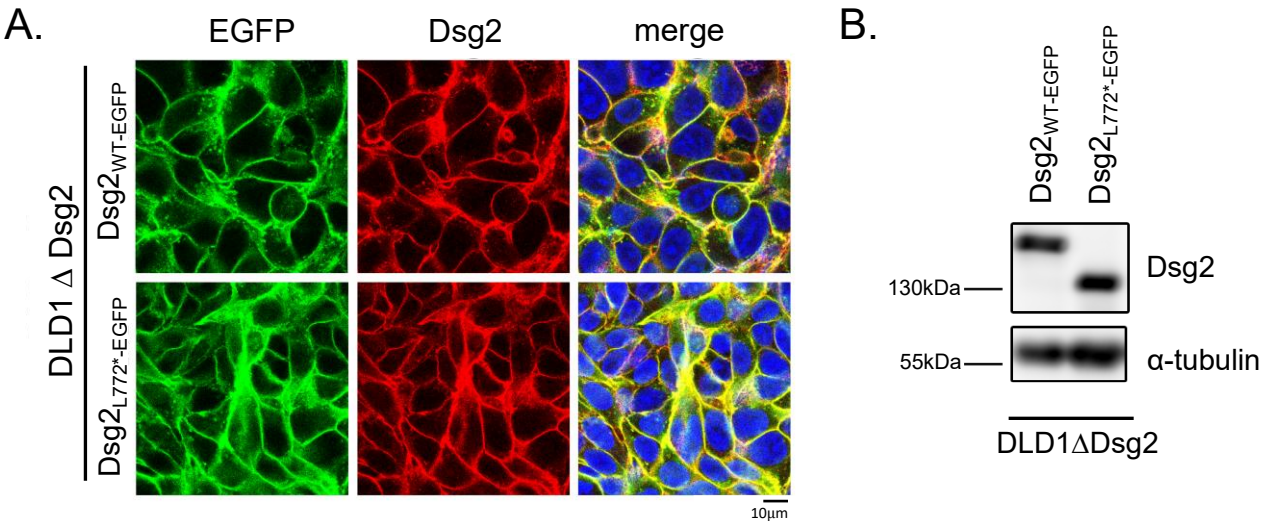
Supplemental Figure 2: Targeting vector K046.6 TV
Dsg2 exon parts 14A and 14B are marked in light red, separated by the artificial intron (pink), which contains the LoxP site (blue). For selection, an FRT-flanked Neo resistance cassette (green) is inserted. The LA has a length of 4.87 kb whereas the SA of homology extends for 2.72 kb (LA and SA; grey boxes). Restriction enzymes used for vector verification are indicated.

Supplemental Figure 3



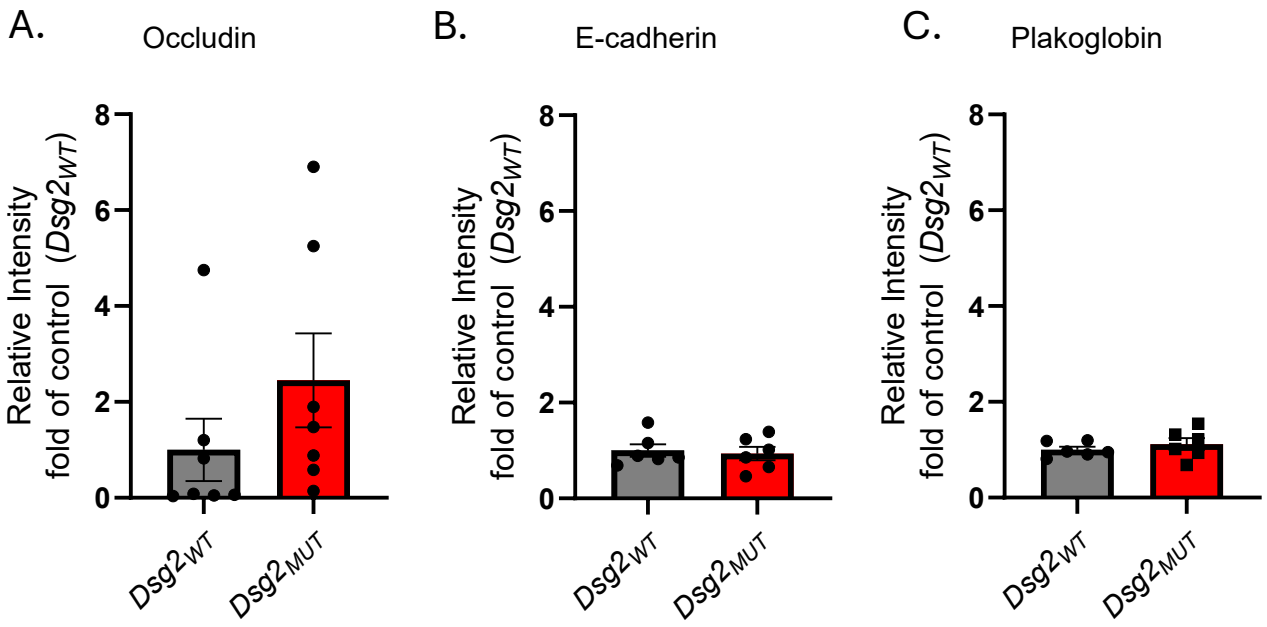
Supplemental Figure 3: **Schematic drawing of the WT locus and the targeted loci of *Dsg2*, with (TG) and without (TG del-neo) the presence of Neo resistance cassette**. *Dsg2* exon parts Ex 14A and Ex 14B are marked in light red, separated by the artificial intron (pink), which contains the LoxP site (blue). The primers used for genotyping and the corresponding amplicon sizes (brown: del-neo PCR, green: neomycin PCR, blue and purple: LoxP PCRs) are mapped on the different loci.

Supplemental Figure 4



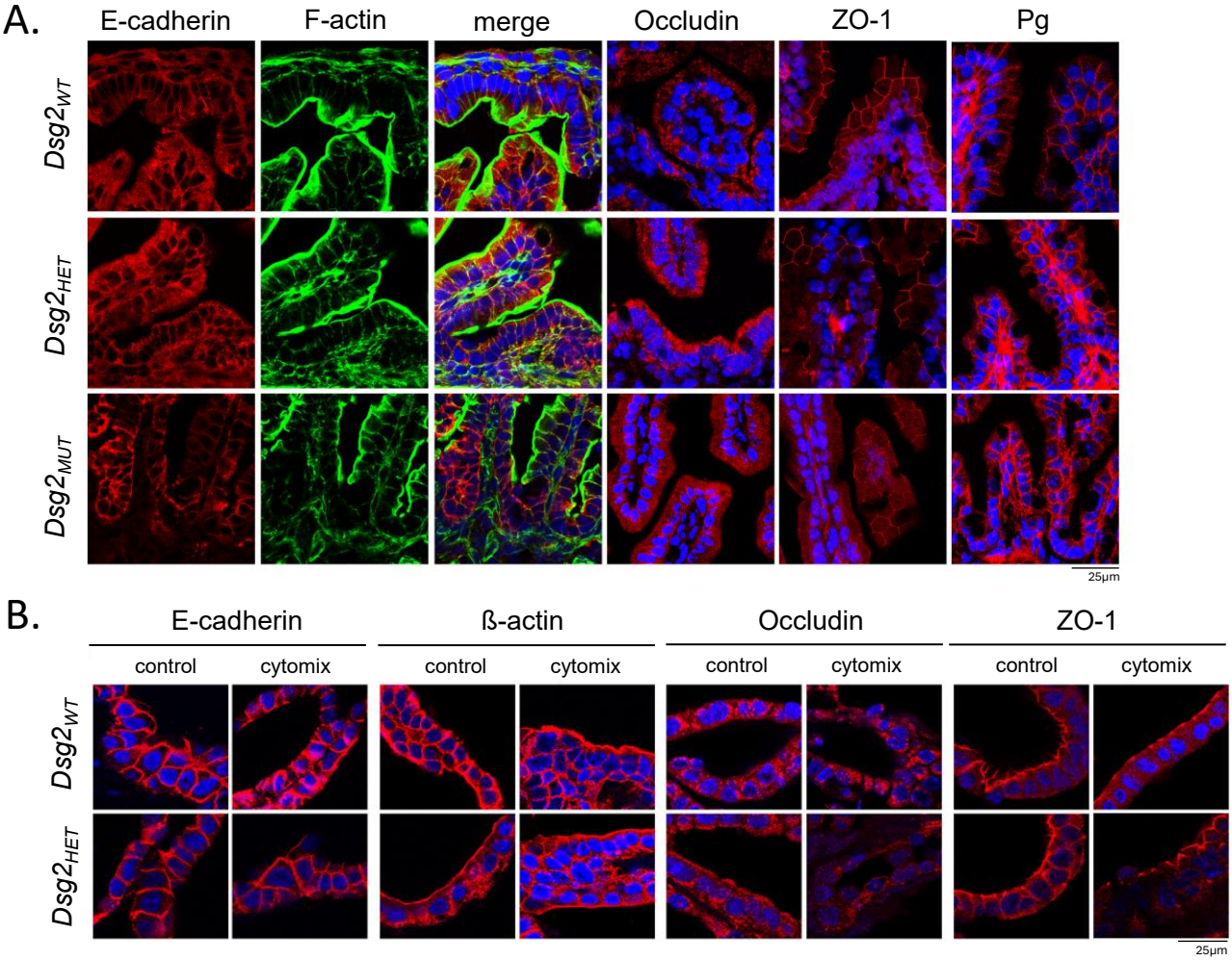
Supplemental Figure 4: Validation of lentiviral transduction. (A) Immunofluorescence staining demonstrating successful lentiviral transduction. Co-staining with the respective autoantibody against the extracellular domain of Dsg2 shows overlapping signals, confirming expression of the transduced cells. (n=3, Scale bar: 10µm). (B) Western Blotting confirming the expression of Dsg2. The full-length EGFP-tagged Dsg2 protein is detected at the expected molecular weight, while the truncated form appears at the corresponding lower molecular weight (n=3).

Supplemental Figure 5



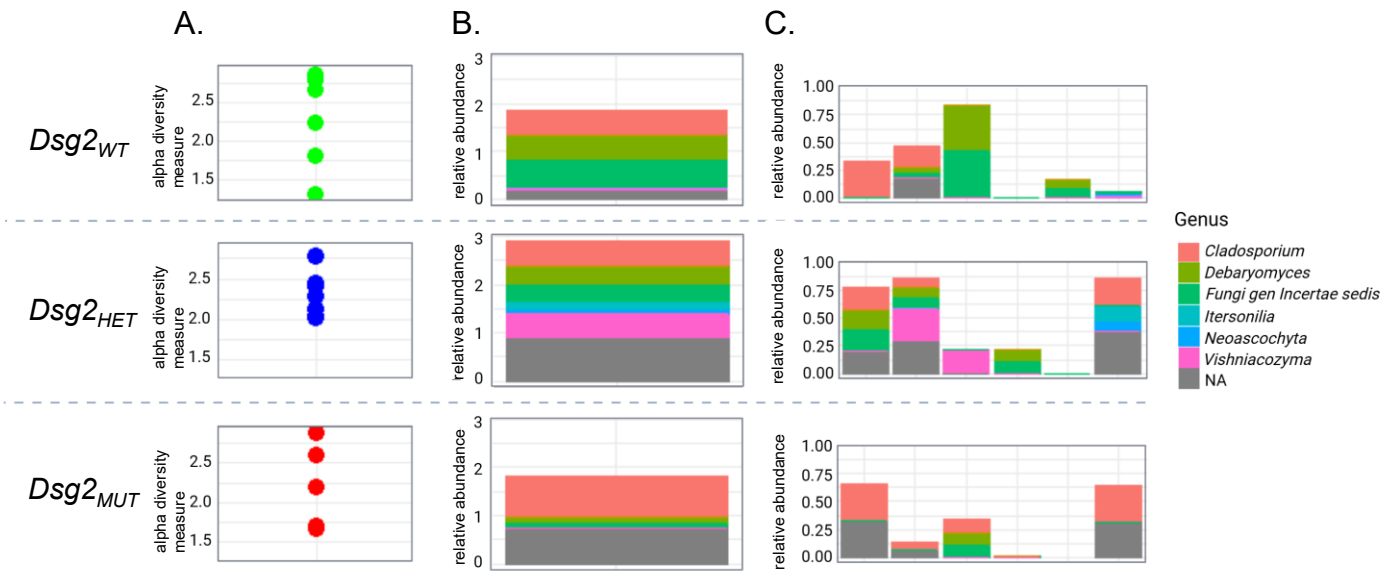
Supplemental Figure 5: Quantification of Western blots. (A) Occludin protein levels showed a trend toward increased levels in $Dsg2_{MUT}$ mice. This difference reached statistical significance only when $Dsg2_{WT}$ and $Dsg2_{MUT}$ littermates were directly compared using the paired Wilcoxon matched-pairs signed-rank test (p=0.0156) (n=7), whereas no significant difference was detected using other statistical tests (t-test; Mann-Whitney U test). (B) E-cadherin (n=6) and (C) plakoglobin (n=6) levels did not differ significantly between $Dsg2_{WT}$ and $Dsg2_{MUT}$ mice.

Supplemental Figure 6



Supplemental Figure 6: (A) Immunostaining of mouse intestinal tissue from $Dsg2_{MUT}$, $Dsg2_{HET}$ and $Dsg2_{WT}$ mice reveals reduced and disorganized localization of the tight junction proteins occludin and ZO-1 along cell borders in $Dsg2_{MUT}$ tissue compared with $Dsg2_{HET}$ and $Dsg2_{WT}$ mice. No differences in E-cadherin, F-actin, or plakoglobin (Pg) staining are observed between wildtype, mutant and heterozygous mice under basal conditions (n≥3). (B) Immunostaining of intestinal organoids derived from $Dsg2_{WT}$ and $Dsg2_{HET}$ mice treated with or without cytomix. Following cytomix incubation, $Dsg2_{HET}$ organoids showed a marked reduction of occludin and ZO-1 at cell–cell junctions, whereas E-cadherin and ß-actin remains unchanged (n≥3).

Supplemental Figure 7



Supplemental Figure 7: *Dsg2* genotype-dependent changes of the intestinal mycobiome. Microbial DNA was extracted from fecal samples of Dsg_{WT} (n=6), Dsg_{HET} (n=6) and Dsg_{MUT} (n=6) mice and used for ITS1 analysis. (A) Alpha diversity of the fecal mycobiome. (B) Relative abundance of the most frequent fungal ASVs at genus level discovered in all samples of the corresponding *Dsg2* genotype. (C) Relative abundance of the most frequent fungal ASVs at genus level per sample according to *Dsg2* genotype.

Supplementary Table 1

Negative regulation interleukin-17	***pValue***	***FDR***
V-set immunoregulatory receptor (Vsir)	0.030356939	0.236585343
arginase type II (Arg2)	0.415993	0.781048
interferon gamma (Ifng)	0.083232	-
Interleukin-12b (Il12b)	0.28306	0.677772
tumor suppressor 2, mitochondrial calcium regulator (Tusc2)	0.369351	0.749816
Positive regulation interleukin-17		
Interleukin-12b (Il12b)	0.28306	0.677772
Interleukin-18 (Il18)	0.011149	0.133856
myeloid differentiation primary response gene 88 (Myd88)	0.664958	0.903328
oncostatin M (Osm)	0.233009	0.630194
solute carrier family 7 (cationic amino acid transporter, y+ system), member 5 (Slc7a5)	0.032111	0.244523
tyrosine kinase 2 (Tyk2)	0.456559	0.803112
Cellular response to interleukin-17		
chemokine (C-X-C motif) ligand 1 (Cxcl1)	0.031767	0.242957
chemokine (C-X-C motif) ligand 10 (Cxcl10)	0.573762	0.863656
decapping mRNA 1B (Dcp1b)	0.07894	0.393192
Interleukin-17 receptor A (Il17ra)	0.001651	0.038491
TRAF3 interacting protein 2 (Traf3ip2)	0.213769	0.610486
signal transducer and activator of transcription 3 (Stat3)	0.020511	0.190591
suppressor of cytokine signaling 3 (Socs3)	0.001973	0.043087
IL-17 Signaling pathway		
CCAAT/enhancer binding protein (C/EBP), beta (Cebpb)	0.006188	0.091293
S100 calcium binding protein A8 (calgranulin A) (S100a8)	9.26E-17	5.95E-13
S100 calcium binding protein A9 (calgranulin B) (S100a9)	1.87E-20	1.99E-16
TNF receptor-associated factor 5 (Traf5)	0.290761	0.683929
caspase 8 (Casp8)	0.634428	0.890484
inhibitor of kappaB kinase epsilon (Ikbke)	0.219683	0.617172
Interleukin-1b (Il1b)	0.001556	0.036893
Interleukin-17F (Il17f)	0.779071	-
lipocalin 2 (Lcn2)	2.04E-06	0.000334
matrix metallopeptidase 9 (Mmp9)	0.754325	0.933922
mucin 5, subtypes A and C, tracheobronchial/gastric (Muc5ac)	0.521459	-

Supplemental Table 1: Genes contained within each of the GO enrichment categories presented in Fig. 5D and their corresponding pValue and FDR.

Supplementary Table 2

UPREGULATED				DOWNREGULATED
ADM2	GIP	MIRLET7D	SNORD17	DSC3
ALKAL1	GPR141	MUC5AC	SULT1E1	FAM43B
ARRDC5	GPRC5A	NT5C1A	SYT8	HSD17B6
ATP6V1B1	HEMGN	PHLDA2	TACSTD2	LY6G5C
CCL8	IFNG	PLA2G2A	TEX22	MCOLN3
CHAC1	IL1B	PRSS2	TEX43	SLC23A1
CHIT1	ITGAD	PTPRQ	TIGIT	TAF7L
CIB3	KLRC3	RASAL1	TMEM249	
CNFN	KMO	REG3G	TNFRSF17	
CPLX3	L1TD1	S100A3	TNFRSF8	
CTLA4	LCN12	S100A8	TPSAB1	
CXCL3	LCN2	S100A9	TRIM29	
CXCL5	LRG1	SIM2		
CYTL1	LYPD5	SLPI		
DPEP3	MIR200A	SNORA30		
FOS	MIR27A	SNORA64		

Supplemental Table 2: List of “common” genes in Venn´s diagram differentially regulated in both human and mouse from Fig. 5E.

Supplemental Table 3

Purpose/Aim	Primer name	Sequence 5'-> 3'
Generation of external probe for Southern Blot	K048.16	TTGGCTGGTCATGAGTTG
Generation of external probe for Southern Blot	K048.17	TGACCTCTGCCCACATAG
Screening PCR for successful homologous recombination in ES cells	K048.9	ACTCGTGCACTCAAGCACATAC
Screening PCR for successful homologous recombination in ES cells	5182LRPCR2	GTTGTGCCCAGTCATAGCCGAATAG
Screening for the presence of artificial intron and LoxP sites in ES cell clones	K048.04	GATCCCTGCAGGCACATCTTCCTGCCAAGTACAC
Screening for the presence of artificial intron and LoxP sites in ES cell clones	K048.05	TCCCTCATCACCATCCATAG
Screening for presence of the Neo resistance cassette in F1 generation	Neo.MP1	GCTGTGCTCCACGTTGTCAC
Screening for presence of the Neo resistance cassette in F1 generation	Neo.MP5	GGAAAGCTGGGCTTGCATCTC
Screening for presence of the Neo resistance cassette in F1 generation	Neo.MP6	GGAGCGGCGATACCGTAAAG
Screening for deletion of Neo resistance cassette in F1 generation	K049.03	GTGTCCTCCTCTGGTTGTAG
Screening for deletion of Neo resistance cassette in F1 generation	K049.04	TCTTCCCATCTTCCGTCAAC
Screening for the presence of artificial intron and LoxP sites in F1 generation	K049.01	CAGCTCAGCTTCCGTTACC
Screening for the presence of artificial intron and LoxP sites in F1 generation	K049.02	ACTCAGCCACTGGCTATTTC
Screening for the presence of artificial intron and LoxP sites in F1 generation	K049.05	CCTCATCCGTGGTTAAGG
Screening for Flp recombinase allele in F1 generation	SD24	CTAATGTTGTGGGAAATTGGAGC
Screening for Flp recombinase allele in F1 generation	SD25	CTCGAGGATAACTTGTTTATTGC
Screening for correct genomic DNA sequence	mDsg2-gDNA-752bp-FW	CATGAGCTGTCTGAGGTTGAC
Screening for correct genomic DNA sequence	mDsg2-gDNA-752bp-REV	CTCAGCCACTGGCTATTTCAGTG
Screening for correct splicing using cDNA as a template	mDsg2-cDNA-608bp-FW	CTGGTGGCCGATCATGCAG
Screening for correct splicing using cDNA as a template	mDsg2-cDNA-608bp-REV	GGTTACCGCTTGCTCATAGTGC

Supplemental Table 3: Generation of the *Dsg2*$_{TRUNC}$ mouse model: Primer combinations.