Supplemental Figures/Tables

Supplemental Table 1: *Rpl27a^(low/+):Mdm2^(+/-) (RP27M2)* **was not observed post-E16.5 from** *Rpl27a^(low/+)* **x** *Mdm2^(+/-)* **crosses**
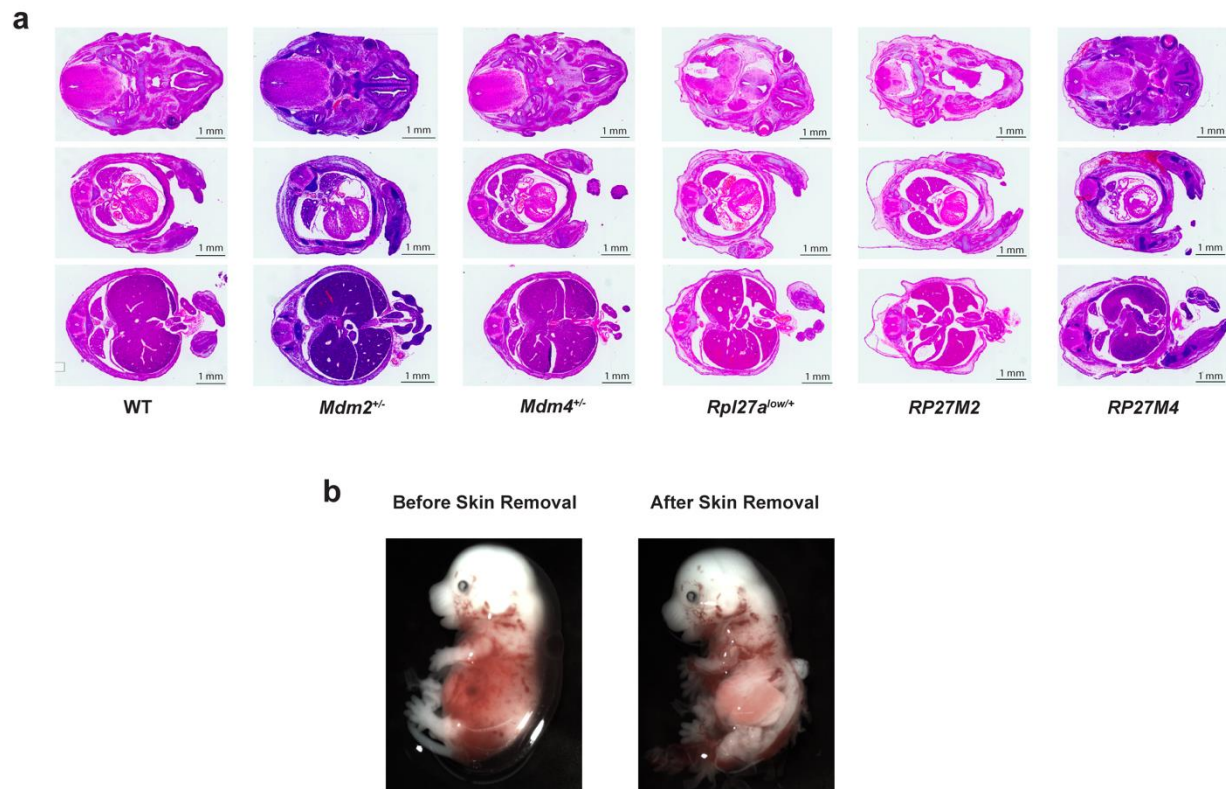
Gestational Age	N	WT (%)		*Rpl27a^(low/+)* (%)		*Mdm2^(+/-)* (%)		*Rpl27a^(low/+):Mdm2^(+/-)* (%)	
		Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected
11.5	24	7 (29)	6 (25)	8 (33)	6 (25)	6 (25)	6 (25)	3 (13)	6 (25)
12.5	31	1 (3)	7.75 (25)	12 (39)	7.75 (25)	10 (32)	7.75 (25)	8 (26)	7.75 (25)
13.5	89	34 (38)	22.25 (25)	19 (21)	22.25 (25)	14 (16)	22.25 (25)	22 (25)	22.25 (25)
14.5	147	37 (25)	36.75 (25)	39 (27)	36.75 (25)	34 (23)	36.75 (25)	37 (25)	36.75 (25)
15.5	80	18 (23)	20 (25)	24 (30)	20 (25)	17 (22)	20 (25)	21 (25)	20 (25)
16.5	54	15 (28)	13.5 (25)	15 (28)	13.5 (25)	12 (22)	13.5 (25)	12 (22)	13.5 (25)
18.5	12	1 (8)	3 (25)	5 (42)	3 (25)	6 (50)	3 (25)	0 (0)	3 (25)
P28	100	34 (34)	25 (25)	32 (32)	25 (25)	34 (34)	25 (25)	0 (0)	25 (25)

Supplemental Table 2: *Rpl27a$^{low/+}$:Mdm4$^{+/-}$ (RP27M4)* **was not observed post-E16.5 from** *Rpl27a$^{low/+}$* **x** *Mdm4$^{+/-}$* **crosses**

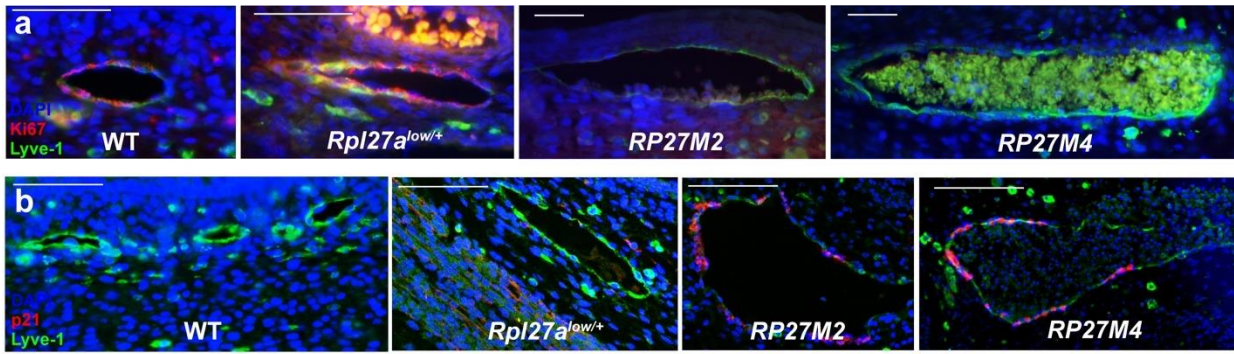| Gestational Age	N	WT (%)		Rpl27a$^{low/+}$ (%)		Mdm4$^{+/-}$ (%)		Rpl27a$^{low/+}$:Mdm4$^{+/-}$ (%)	
		Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected
11.5	18	3 (17)	4.5 (25)	6 (33)	4.5 (25)	4 (22)	4.5 (25)	5 (28)	4.5 (25)
12.5	54	18 (33)	13.5 (25)	9 (17)	13.5 (25)	14 (26)	13.5 (25)	13 (24)	13.5 (25)
13.5	143	40 (28)	35.75 (25)	32 (22)	35.75 (25)	41 (29)	35.75 (25)	30 (21)	35.75 (25)
14.5	172	48 (28)	43 (25)	36 (21)	43 (25)	46 (27)	43 (25)	42 (24)	43 (25)
15.5	83	22 (27)	20.75 (25)	24 (29)	20.75 (25)	22 (27)	20.75 (25)	15 (17)	20.75 (25)
16.5	30	7 (23)	7.5 (25)	9 (30)	7.5 (25)	9 (30)	7.5 (25)	5 (17)	7.5 (25)
18.5	9	6 (66)	2.25 (25)	3 (34)	2.25 (25)	0 (0)	2.25 (25)	0 (0)	2.25 (25)
P28	48	16 (33)	12 (25)	15 (31)	12 (25)	17 (36)	12 (25)	0 (0)	12 (25)
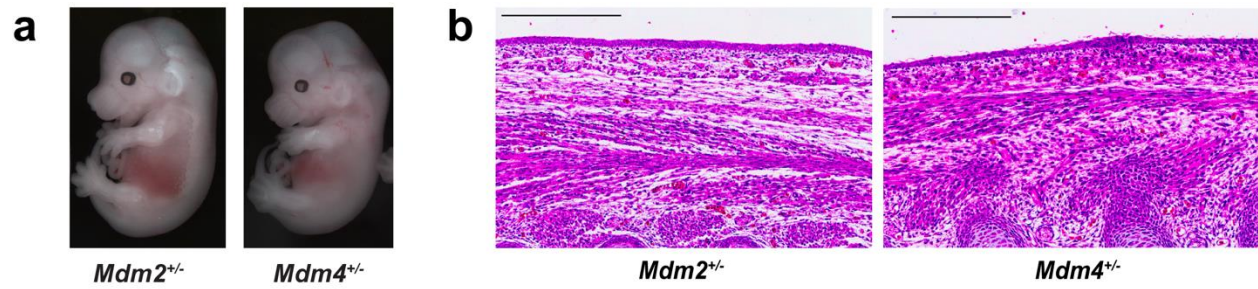
Supplemental Figure 1. E14.5 *Mdm2*$^{+/-}$ and *Mdm4*$^{+/-}$ embryos are normal. **a)** Representative image taken with Leica M165 FC stereoscope. **b)** H&E staining of dorsal skin. Scale bar represents 300 μm.

a

WT   *Mdm2*^{+/-}   *Mdm4*^{+/-}   *Rpl27a*^{low/+}   *RP27M2*   *RP27M4*

**b**   Before Skin Removal   After Skin Removal

Supplemental Figure 2. Major organs in mutant embryos are normal. **a)** H&E staining of E15.5 embryos. **b)** Picture of a mutant embryo before and after skin removal showing a clear view of the liver and no internal hemorrhaging.
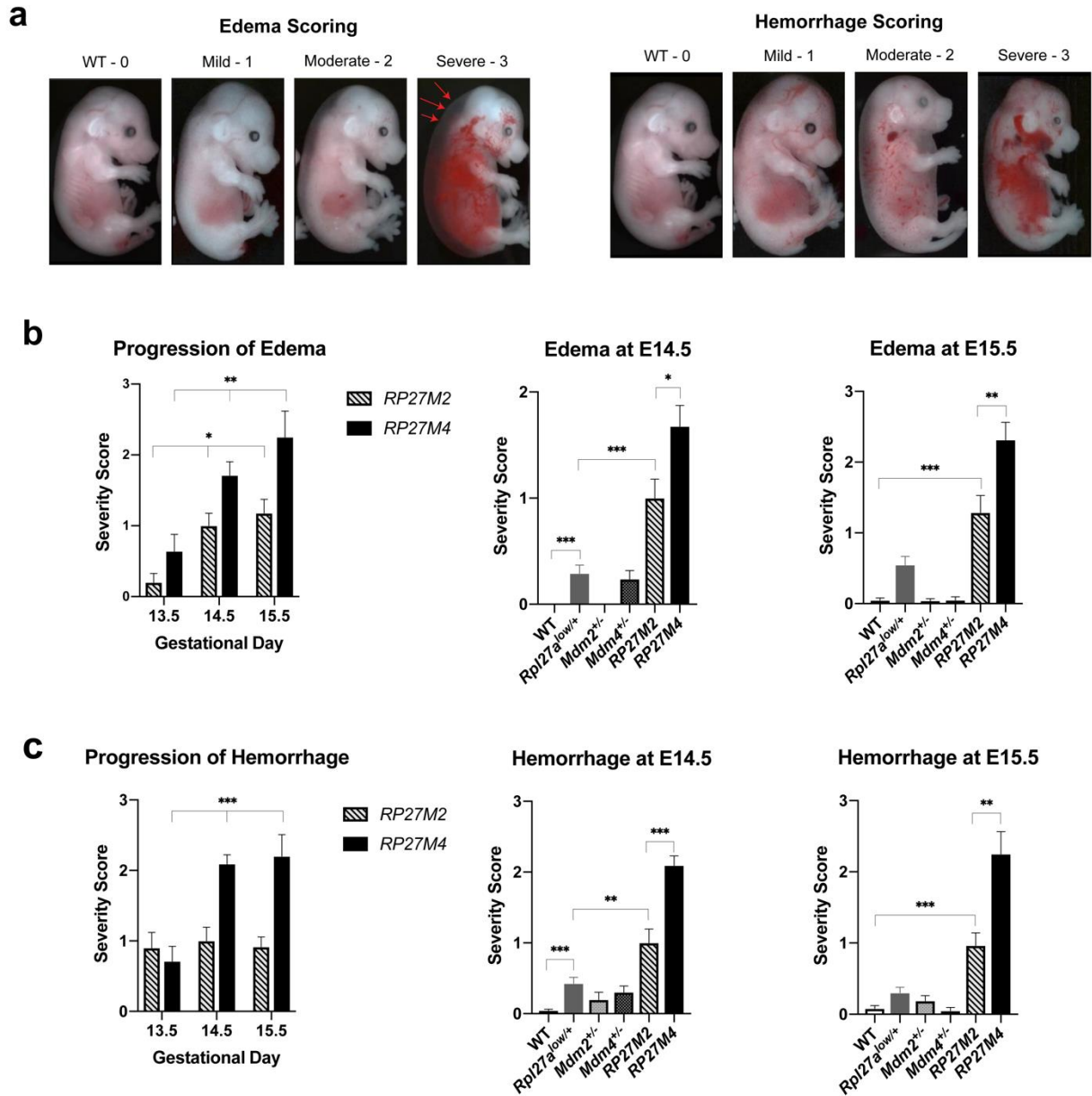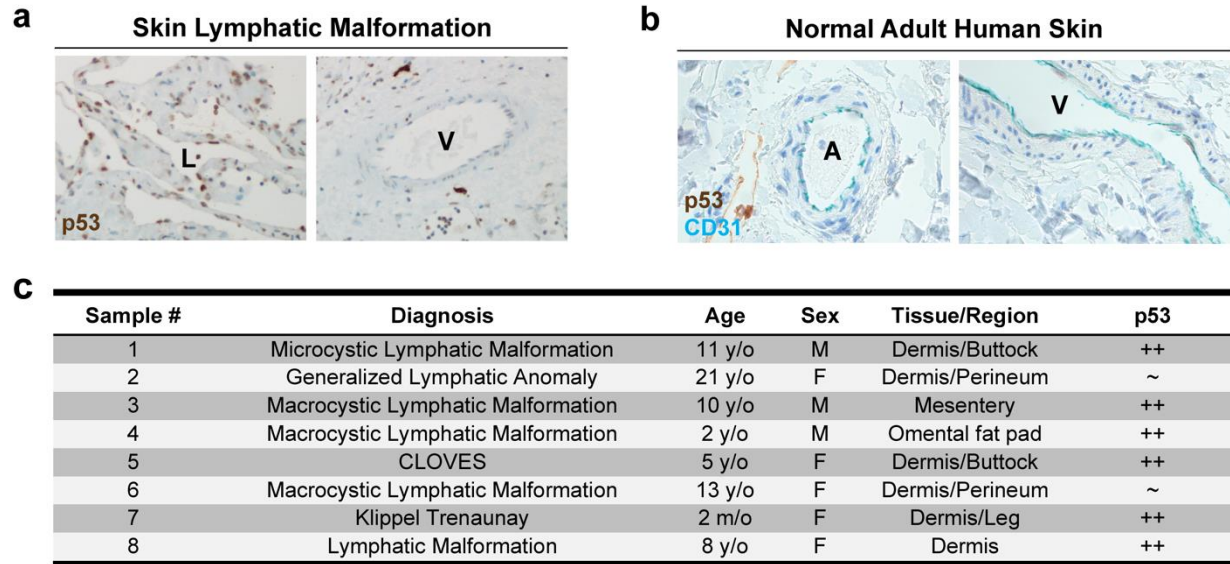
Supplemental Figure 3. Edema and cutaneous hemorrhaging are progressive and more severe in *RP27M4*. **a)** Edema (red arrows) and hemorrhaging scoring criteria used by three researchers during evaluation to avoid bias. **b)** Severity scoring of edema by gestational age. **c)** Severity of hemorrhaging by gestational age. Sample sizes for (a) and (b) are at E13.5: 10 *RP27M2* and 14 *RP27M4*; at E14.5: 38 WT, 42 *Rpl27a$^{low/+}$*, 25 *Mdm2$^{+/-}$*, 36 *Mdm4$^{+/-}$*, 32 *RP27M2*, and 38 *RP27M4*; at E15.5: 31 WT, 33 *Rpl27a$^{low/+}$*, 14 *Mdm2$^{+/-}$*, 19 *Mdm4$^{+/-}$*, 30 *RP27M2*, and 10 *RP27M4* embryos. Statistical significance determined by one-way ANOVA (for the first panels of a & b) and *t* tests. NS= not significant, * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$.

Supplemental Figure 4. IF staining of *Mdm2*$^{+/-}$ and *Mdm4*$^{+/-}$ skin shows normal size lymphatic vessels comparable to WT. **a)** Lyve-1 staining of E15.5 skin. **b)** whole-mount staining of E14.5 skin. **c)** Ki-67 and Lyve-1 double staining of E15.5 skin. **d)** p21 and Lyve-1 double staining of E15.5 skin. **e)** Ultramicroscopy imaging of E11.5 CV, pTD and superficial LECs. **f)** whole-mount staining of E14.5 mesentery and **g)** E16.5 mesentery.

Supplemental Figure 5. Immunofluorescence staining of E15.5 lymphatic vessels **a)** Dorsal skin double stained with Ki-67 and Lyve-1. Magnification 40X for WT and *Rpl27a^low/+^*, 20X for *RP27M2* and *RP27M4*. **b)** p21 expression in lymphatic endothelium. Data are representative of more than four biological samples per genotype. Scale bars are 100μm for **b** and 50μm for **a**.
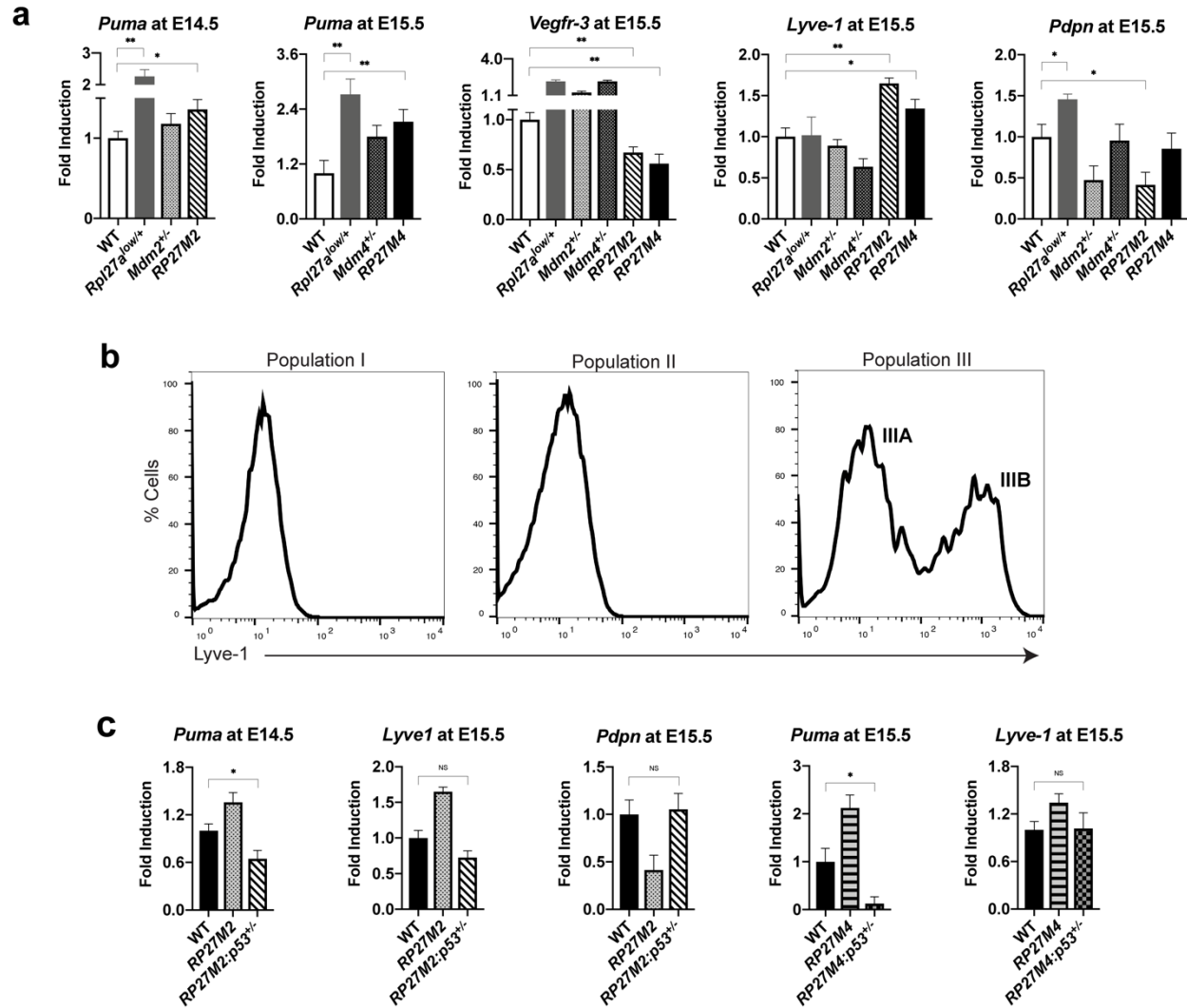
Supplemental Figure 6. Gene Expression differences and CD31$^+$ cell distributions are rescued by the loss of one functional *p53* copy. **a)** Gene expression assays by qPCR (mean $\pm$ SEM) in *RP27M2* and *RP27M4* skin. **b)** Representative lyve-1 expression plot of the four CD31$^+$ cell populations in WT mice. **c)** Gene expression assays by qPCR (mean $\pm$ SEM) in *RP27M2:p53$^{+/-}$* and *RP27M4:p53$^{+/-}$* skin show restoration to WT levels.

a Skin Lymphatic Malformation

b Normal Adult Human Skin

c

Sample #	Diagnosis	Age	Sex	Tissue/Region	p53
1	Microcystic Lymphatic Malformation	11 y/o	M	Dermis/Buttock	++
2	Generalized Lymphatic Anomaly	21 y/o	F	Dermis/Perineum	~
3	Macrocystic Lymphatic Malformation	10 y/o	M	Mesentery	++
4	Macrocystic Lymphatic Malformation	2 y/o	M	Omental fat pad	++
5	CLOVES	5 y/o	F	Dermis/Buttock	++
6	Macrocystic Lymphatic Malformation	13 y/o	F	Dermis/Perineum	~
7	Klippel Trenaunay	2 m/o	F	Dermis/Leg	++
8	Lymphatic Malformation	8 y/o	F	Dermis	++

Supplemental Figure 7. Lymphatic endothelium is positive for p53 in majority of human lymphatic diseases. **a)** p53 IHC staining of lymphatic endothelium (L), artery (A), or vein (V) in pediatric lymphedema specimen and **b)** normal human skin. **c)** 6 out of 8 human lymphatic cases are highly positive for p53.