Supplementary Figure 1. The R200W mutation renders pVHL less stable. **A.** Wt and R/R ES cells were treated with cycloheximide (CHX) for the indicated lengths of time and pVHL levels were determined by Western blot (arrow indicates pVHL). R/R pVHL has a shorter half-life than the wt protein (approximately 2 hours for R/R compared to greater than 12 hours for wt). Cyclin D1 was used as a positive control for the effectiveness of CHX treatment. **B.** Western blot showing that, in contrast to an antibody directed against the C-terminus, an antibody against full-length pVHL recognizes wt and R200W pVHL equivalently. **C.** Autoradiograph of wt and R200W pVHL in vitro transcribed and translated in the presence of ^{35}S-Met revealed no difference in the translation of R/R protein compared to wt. **D.** Wt and R/R ES cells were treated with DMSO vehicle (D), or one of three proteasome inhibitors: ALLN (A), lactacystin (L), or MG132 (M). Treatment with any of these compounds increased the stability of R/R pVHL above the level seen in control DMSO-treated cells. Wt pVHL is very stable and not affected by any of these compounds. Arrow indicates pVHL. p53 was used as a positive control for the effectiveness of inhibitor treatment.

Supplementary Figure 2. VEGF and Epo levels increase with age in *Vhl*$^{R/R}$ mice. Serum levels of VEGF (**A.**) and Epo (**B.**) were measured by ELISA in mice of various ages. (* $p < 0.04$; # approaching significance, $p < 0.06$; $p < 0.08$ for R/R Epo levels at 21-28 weeks).

Supplementary Figure 3. Enhanced angiogenesis in the skin of *Vhl*$^{R/R}$ mice. **A.** Representative sections of skin from wt and R/R mice stained with H&E (top, 10x magnification, scale bar represents 100μm) or with an antibody to CD31 to mark vessels (bottom, 20x magnification, scale bar represents 50μm). **B.** Quantification of the number of CD31^{+} blood vessels revealed a slight increase in the vascularity of R/R skin compared to wt controls, which may contribute to the redness observed in R/R mice.

Supplementary Figure 4. Additional phenotypes in *Vhl*$^{R/R}$ mice. **A.** Representative sections of wt and R/R livers stained with H&E (top, 10x magnification, scale bar represents 100μm) or oil red O (bottom, 40x magnification, scale bar represents 20μm) revealed no changes in fat deposition in

R/R livers. However, the vascularity of R/R livers was significantly increased compared to wt controls (**B.**). (* $p < 0.05$) **C.** Representative H&E sections of wt and two independent R/R lungs revealed that the vessels in R/R mice were congested with blood (top, 5x magnification, scale bar represents 200μm). Staining of lung tissue with α-SMA did not differ significantly between wt and R/R animals (bottom, 20x magnification, scale bar represents 50μm).

Supplementary Figure 1

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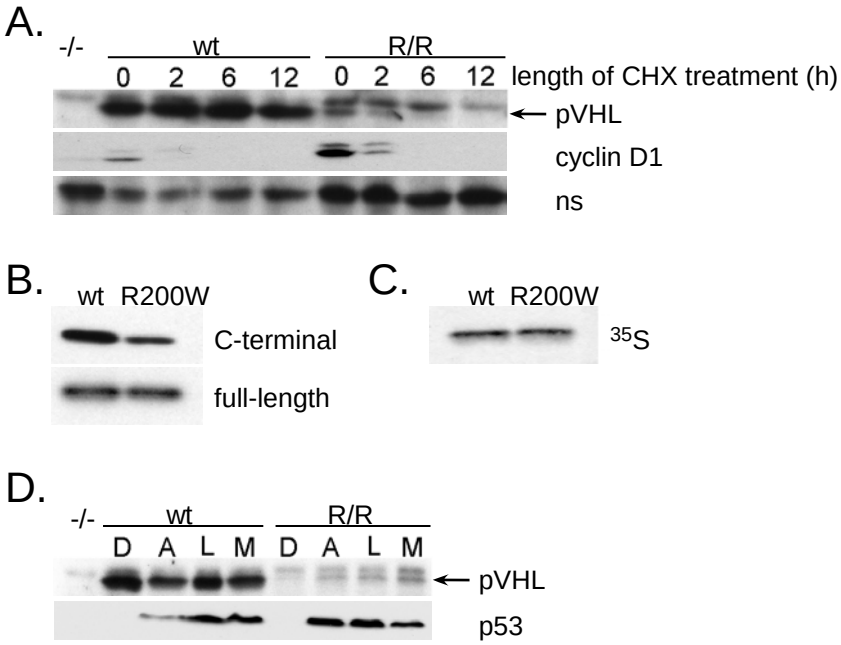
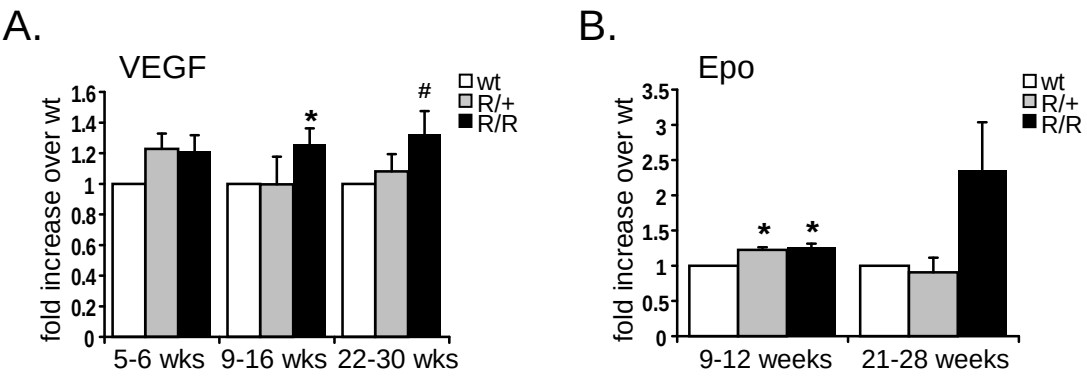
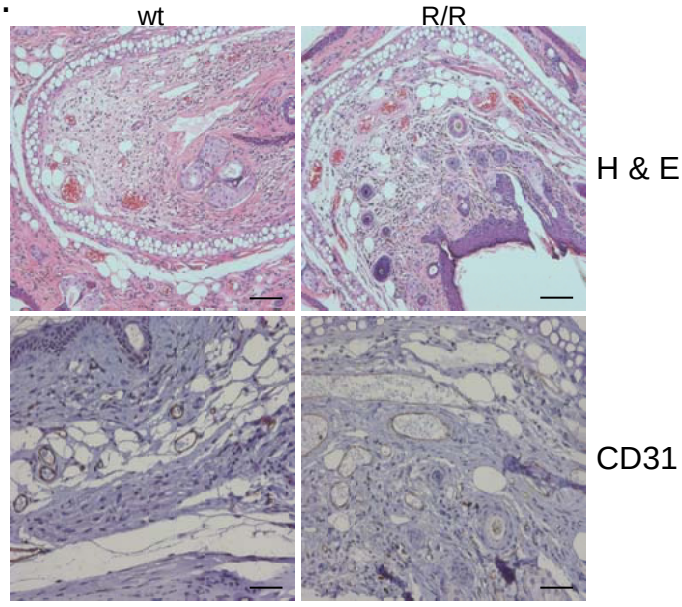
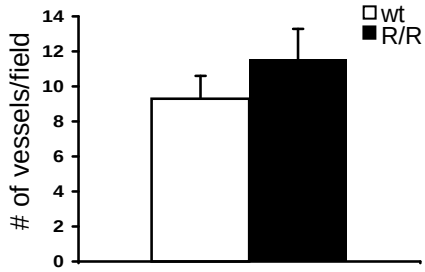


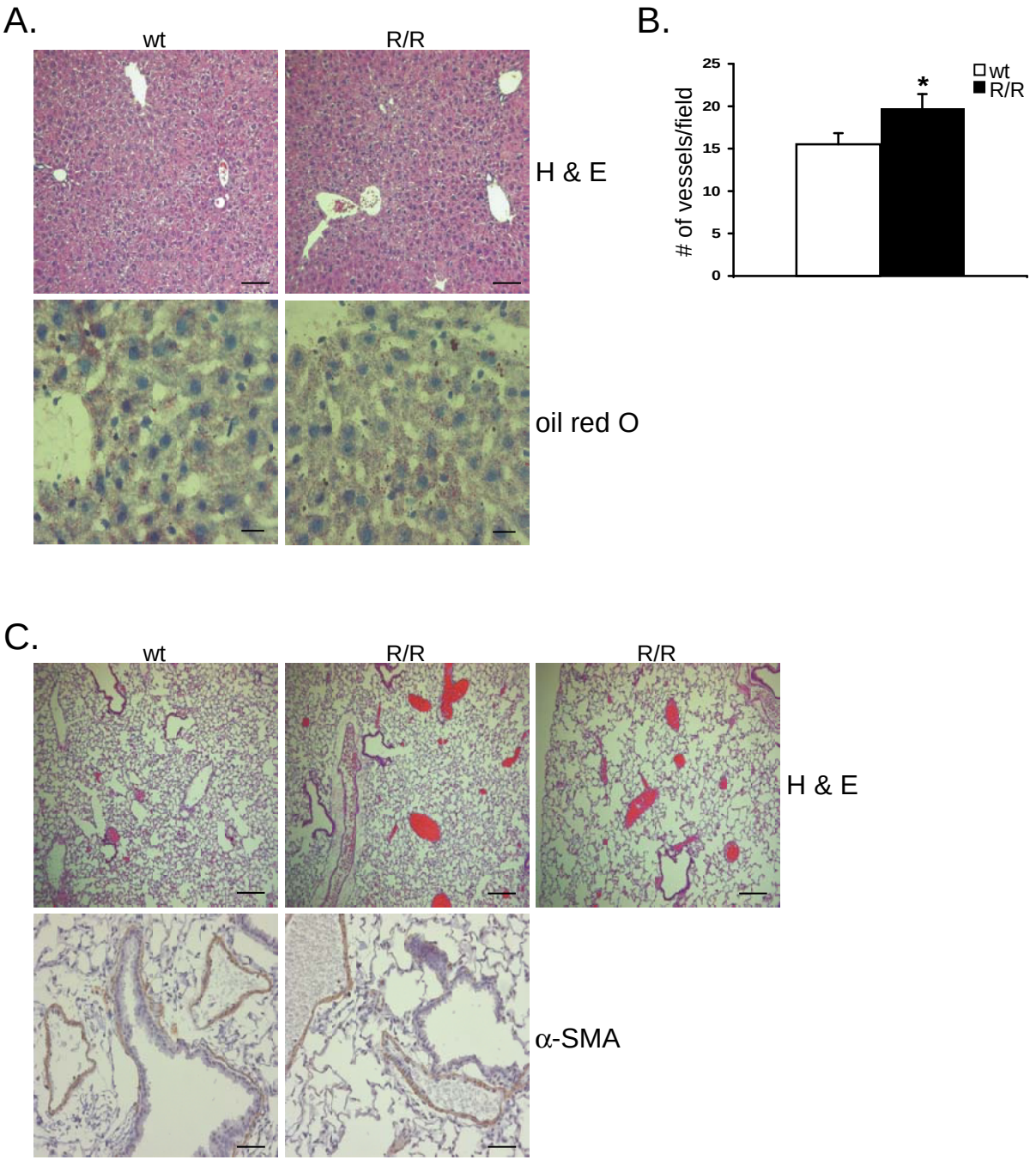

A.
VEGF
fold increase over wt
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
*
#
wt
R/+
R/R
5-6 wks
9-16 wks
22-30 wks
B.
Epo
fold increase over wt
3.5
3
2.5
2
1.5
1
0.5
0
*
*
wt
R/+
R/R
9-12 weeks
21-28 weeks

A.



B.




A.
wt
R/R
H & E
oil red O
B.
# of vessels/field
25
20
15
10
5
0
wt
R/R
*
C.
wt
R/R
R/R
H & E
α-SMA