Supplemental Figure 1: Assessment of right ventricular (RV) pressures. Representative RV pressure tracing from wild type and *Id1 null* mice exposed to normoxia or 3 weeks of hypoxia showing hypoxia-induced increases in RVSP.

Supplemental Figure 2: Hypoxia-induced muscularization of the peripheral pulmonary vasculature. **A-B:** Representative two color immunofluorescence staining using mouse monoclonal anti- α-SMA (green) and rabbit polyclonal anti-von Willebrand factor (vWF) antibodies (red) in lung sections from wild type and *Id1* deficient mice exposed to normoxia (A-F), 1 week (G-L) and 3 weeks hypoxia (M-R). **D, E, F, J, K, L, P, Q, R:** Inset from A, B, C, G, H, I, M, N, O respectively. **S:** Negative staining using mouse and rabbit IgG controls. Arrows denote representative non-muscularized peripheral pulmonary vessels, arrowheads muscularized peripheral pulmonary vessels. Size bar = 50 μm.

Supplemental Figure 3: Specificity of anti-ID1 antibodies. Immunoblot for ID1 and β-actin in normoxic, 1 week and 3 week hypoxic wild type, *Id1*$^{+/-}$, and *Id1 null* lung lysates as indicated. Specificity of staining was determined by the absence of signal in lung lysates from *Id1 null* mice using rabbit monoclonal anti-ID1 antibodies Clones 195-14 (**A**) and 37-2 (**B**) from Biocheck and a rabbit polyclonal anti-ID1 antibody from Santa Cruz (SC-488) (**C**). Note that ID1 is detected as a 15-17 kDa doublet which is absent in *Id1 null* mouse lungs.

Supplemental Figure 4: Specificity of rabbit monoclonal anti-ID3 Clone 17-3 antibody. **A:** Immunoblot for ID3 and β-actin in normoxic wild type and *Id3 null* mutant mice. **B-G:** Immunoperoxidase staining for ID3 in lung sections from normoxic wild type (B-D) and *Id3 null*

(E-G) mice. Specificity was determined by the absence of signal in *Id3 null* mice. * indicates vessels, § indicates muscularized airways.

Supplemental Figure 5: Localization of ID1 expression using EC and smooth muscle cell markers. **A-F:** Sequential sections of 1 week hypoxic wild type mouse lungs were double stained for ID1 using rabbit monoclonal anti-ID1 antibody Clone 37-2 (A-C, red) or the EC marker vWF (D-F, red) and the smooth muscle marker α-SMA (green). Nuclear staining with DAPI is shown in blue. Prominent airways are outlined to orient sequential sections A and D, and the same vessel is boxed. **B-C:** inset of vessel from A. **E-F:** inset of vessel from D. Dominant EC expression of ID1 is determined by overlap with vWF staining (B vs. E). Size bar = 100 μm.

Supplemental Figure 6: Localization of ID3 expression using EC and smooth muscle cell markers. **A-F:** Sequential sections of 1 week hypoxic wild type mouse lungs were double stained for ID3 using rabbit monoclonal antibody Clone 17-3 (A-C, red) or the EC marker vWF (D-F, red) and the smooth muscle marker α-SMA (green). Nuclear staining with DAPI is shown in blue. Prominent airways are outlined to orient sequential sections A and D, and the same vessel is boxed. **B-C:** inset of vessel from A. **E-F:** inset of vessel from D. Dominant EC expression of ID3 is determined by overlap with vWF staining (B vs. E). Size bar = 100 μm.

Supplemental Figure 7: Localization of ID2 expression using EC and smooth muscle cell markers. **A-F:** Sequential sections of 1 week hypoxic wild type mouse lungs were double stained for ID2 using rabbit monoclonal antibody Clone 98-2 (A-C, red) or the EC marker vWF (D-F, red) and the smooth muscle marker α-SMA (green). Nuclear staining with DAPI is shown in

blue. Prominent airways are outlined to orient sequential sections A and D, and the same vessel is boxed. **B-C:** inset of vessel from A. **E-F:** inset of vessel from D. Dominant VSMC expression of ID2 is determined by overlap with α-SMA staining (B). Bar is 100 μm.

Supplemental Figure 8: SMAD1/5/8 phosphorylation in the hypoxic mouse lung. **A:** Immunoblot for phospho-SMAD1/5/8 and total SMAD1 expression relative β-actin in normoxic, 1 week and 3 weeks hypoxic wild type whole lung lysates. **B:** Densitometry for phospho-SMAD1/5/8 relative to total SMAD1 expression from A. **C:** Densitometry for total SMAD1 relative to β-actin expression from A. A light chain specific secondary antibody was employed in order to resolve SMAD1 from heavy chain of IgG. Data are expressed as mean ± SEM. *a*: $p<0.05$ vs. normoxic control by One-Way ANOVA with Bonferroni correction.

Supplemental Figure 9: Immunohistochemical staining using different ID1 antibodies. Immunoperoxidase staining for ID1 in lung sections from 1 week hypoxic wild type and *Id1 null* mice as indicated. **A-F:** Rabbit polyclonal anti-ID1 antibody from Santa Cruz (SC-488). Arrowheads denote nuclear staining in ECs which is absent in *Id1 null* lungs. Thick arrows indicate non-specific, cytoplasmic staining seen in vessel walls and bronchial smooth muscle cells in wild type and *Id1 null* mouse lungs. **G-L:** Rabbit monoclonal anti-ID1 antibody Clone 37-2. Nuclear staining of ECs (arrowheads) and VSMCs within the vessel walls (thin arrows) is seen in wild type but not *Id1 null* mouse lungs. Sections were counterstained with hematoxylin. **I, L:** Inset from H and K, respectively. Size bar = 50 μm. * indicates vessels.

Supplemental Figure 10: EC expression of ID1 in hypoxic lungs. **A:** Immunofluorescence staining for ID1 in a larger diameter vessel cut in longitudinal section and negative staining of bronchial smooth muscle cells using rabbit monoclonal anti-ID1 antibody Clone 37-2 (red), counterstained with mouse anti-α-SMA antibodies (green) to identify smooth muscle cells. **B:** inset from A. Size bar = 100 μm. * indicates vessel, arrowheads indicate EC staining, arrows bronchial smooth muscle cells.

Supplemental Figure 11: Comparison of ID2 localization in wild type mouse lungs using two different anti-ID2 antibodies. **A-C:** Immunoperoxidase staining for ID2 in lung sections from wild type mice exposed to 1 week of hypoxia using rabbit monoclonal anti-ID2 Clone 98-2 from BioCheck. **D-F:** Immunoperoxidase staining for ID2 in lung sections from wild type mice exposed to hypoxia for 1 week using rabbit polyclonal anti-ID2 from Santa Cruz (C-20, SC-489) **G-I:** Negative staining with rabbit IgG control. Sections were counterstained with hematoxylin. Size bar = 50 μm. * indicate vessels.

Supplemental Figure 12: Lack of hypoxic regulation of ID2 expression in cultured PASMCs. **A:** Immunoblot for ID2 using the rabbit polyclonal Santa Cruz antibody (SC-489) and β-actin in serum starved mouse PASMCs maintained in normoxia, 1% oxygen or 3% oxygen ± 200 ng/ml Noggin for 24 hours, as indicated. **B:** Densitometry of ID2 from A relative to β-actin. **C:** Representative immunoblot for ID2 and β-actin in serum starved mouse PASMCs treated ± 10 ng/ml recombinant human BMP4 for 4 hours, as indicated. The experiment was repeated in triplicate. **D:** Densitometry of ID2 from C relative to β-actin. Data are expressed as mean ±

SEM. No significant between group differences were detected by one-way ANOVA (B) or two-tailed t-test (D).