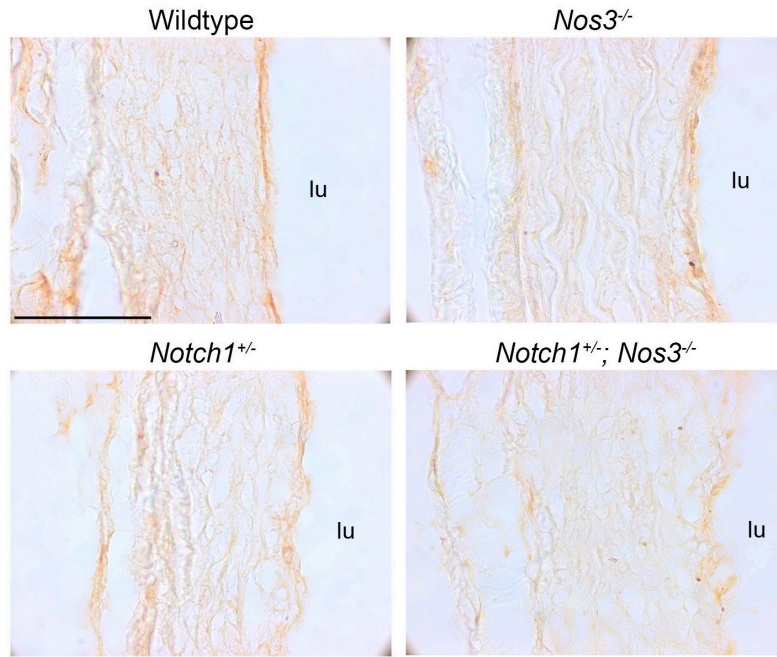
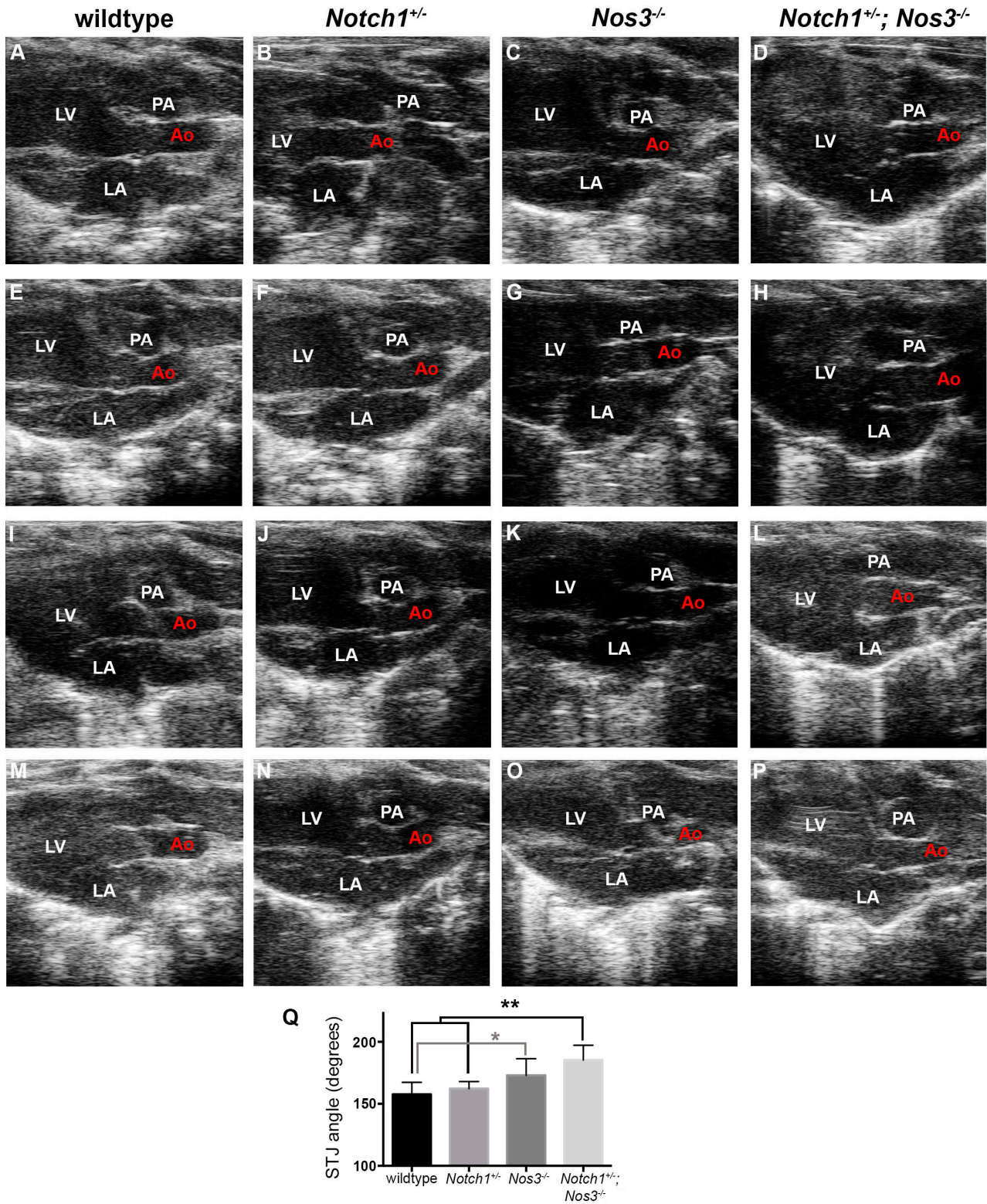


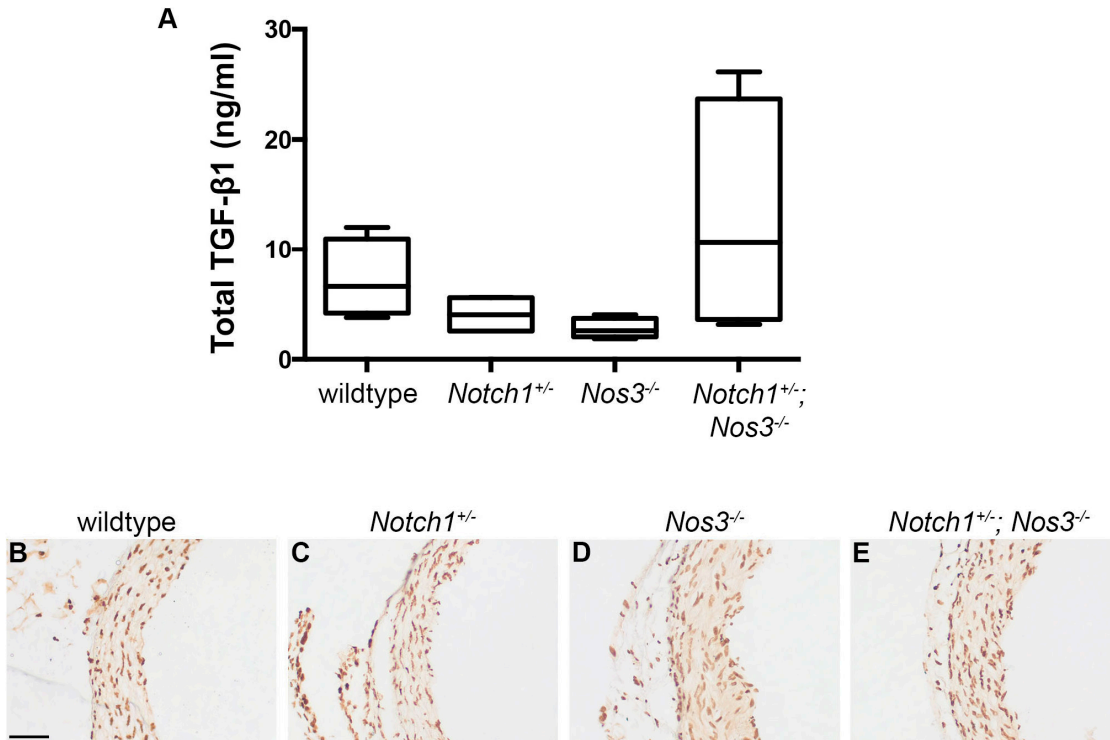
## Supplementary Materials



**Figure S1.** Expression of Notch1 intracellular domain (activated Notch1) by immunohistochemistry is decreased in endothelial cells lining the lumen (lu) of *Notch1*<sup>+/-</sup> and *Notch1*<sup>+/-</sup>; *Nos3*<sup>-/-</sup> mice as compared to wildtype and *Nos3*<sup>-/-</sup>. Scale bar = 50 microns.



**Figure S2.** Representative echocardiographic images of ascending aorta from 6 month old mice of each genotype (A–P). Each image represents a different animal (4 mice per genotype are shown). LV, left ventricle; Ao, aorta. (Q) Quantified aortic sinus-ascending aorta angle in 6 month old mice (\* *p* value < 0.05, \*\* *p* value < 0.01).



**Figure S3.** TGF-β signaling is not consistently elevated in *Notch1*<sup>+/-</sup>; *Nos3*<sup>-/-</sup> mice. **(A)** Serum levels of total circulating TGF-β1 in serum from 8 month old mice of all four genotypes demonstrate no statistically significant differences. **(B–E)** Expression of phosphorylated SMAD2 by immunohistochemistry in the aortic wall is unchanged between genotypes. Aortas of 8 month old mice were examined at  $n = 3$  per genotype.