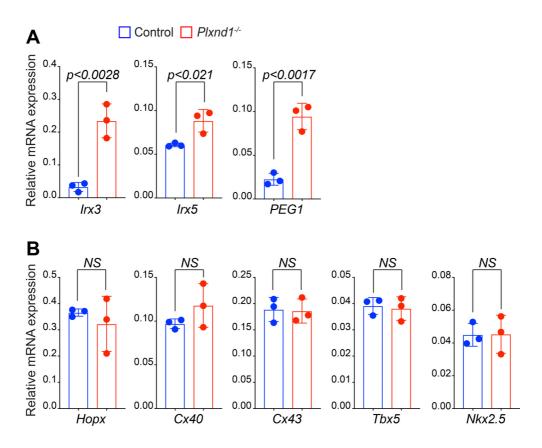


**Figure S1. PlexinD1 expression in the developing heart.** Immunohistochemistry for PlexinD1 was performed on E12.5, E13.5 and E17.5 wildtype heart sections. Scale bars are 200µm respectively.



**Figure S2. Expression of myocardial trabeculation and cardiac chamber genes in** *Plxnd1*<sup>-/-</sup> **hearts**. Real-time qPCR analysis of cardiac chamber genes using RNA isolated from E14.5 control and *Plxnd1*<sup>-/-</sup> hearts (A-B). *n*=3 for each genotype.

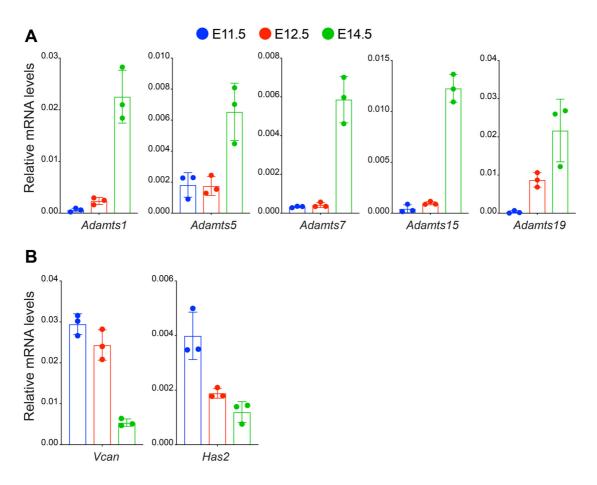
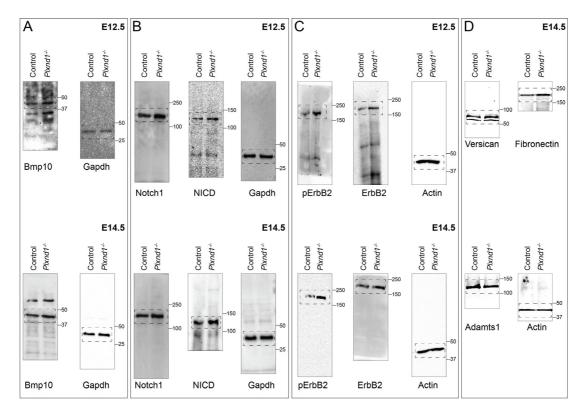


Figure S3. Expression of ECM proteolytic and synthesis genes in wildtype hearts at different embryonic stages. Real-time qPCR analysis of ECM proteolytic genes using RNA isolated from E11.5, E12.5, and E14.5 wildtype hearts (A). Real-time qPCR analysis of ECM synthesis genes using RNA isolated from E11.5, E12.5, and E14.5 wildtype hearts (B). n=3 for each genotype.



**Figure S4.** Uncropped images of western blot images presented in Figure 3B (A). Uncropped images of western blot images presented in Figure 5C and E (B-C). Uncropped images of western blot images presented in Figure 4E (D). Dotted box indicates areas that were cropped for use in figures.