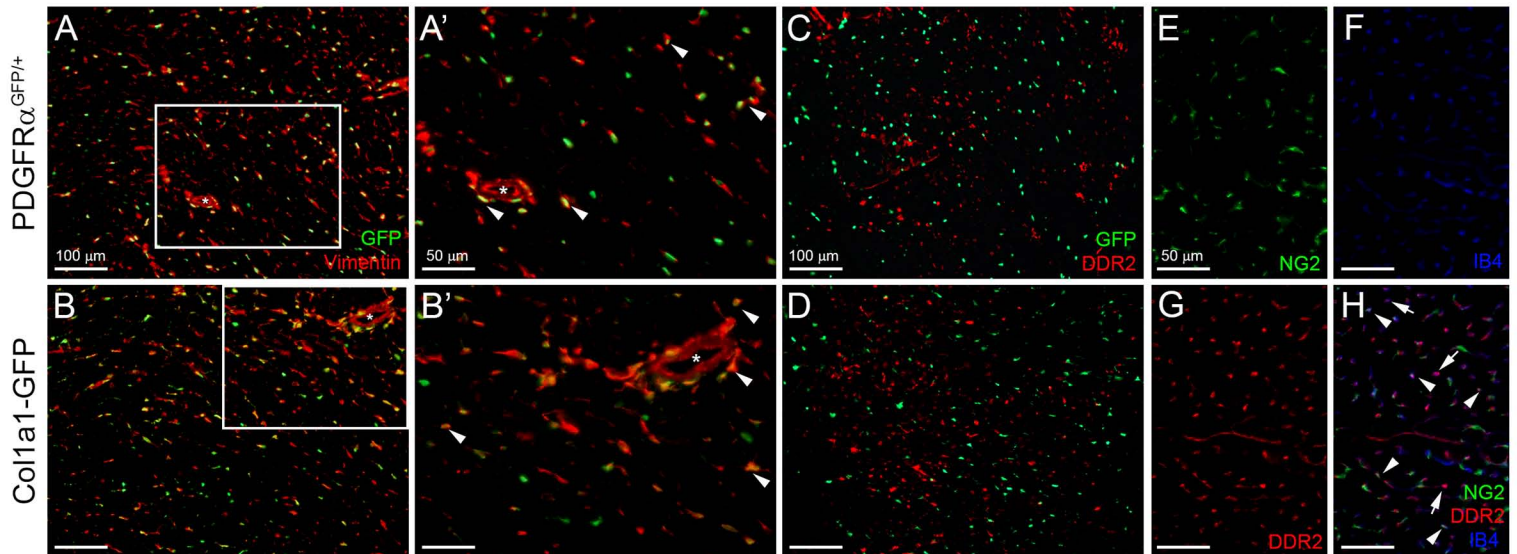
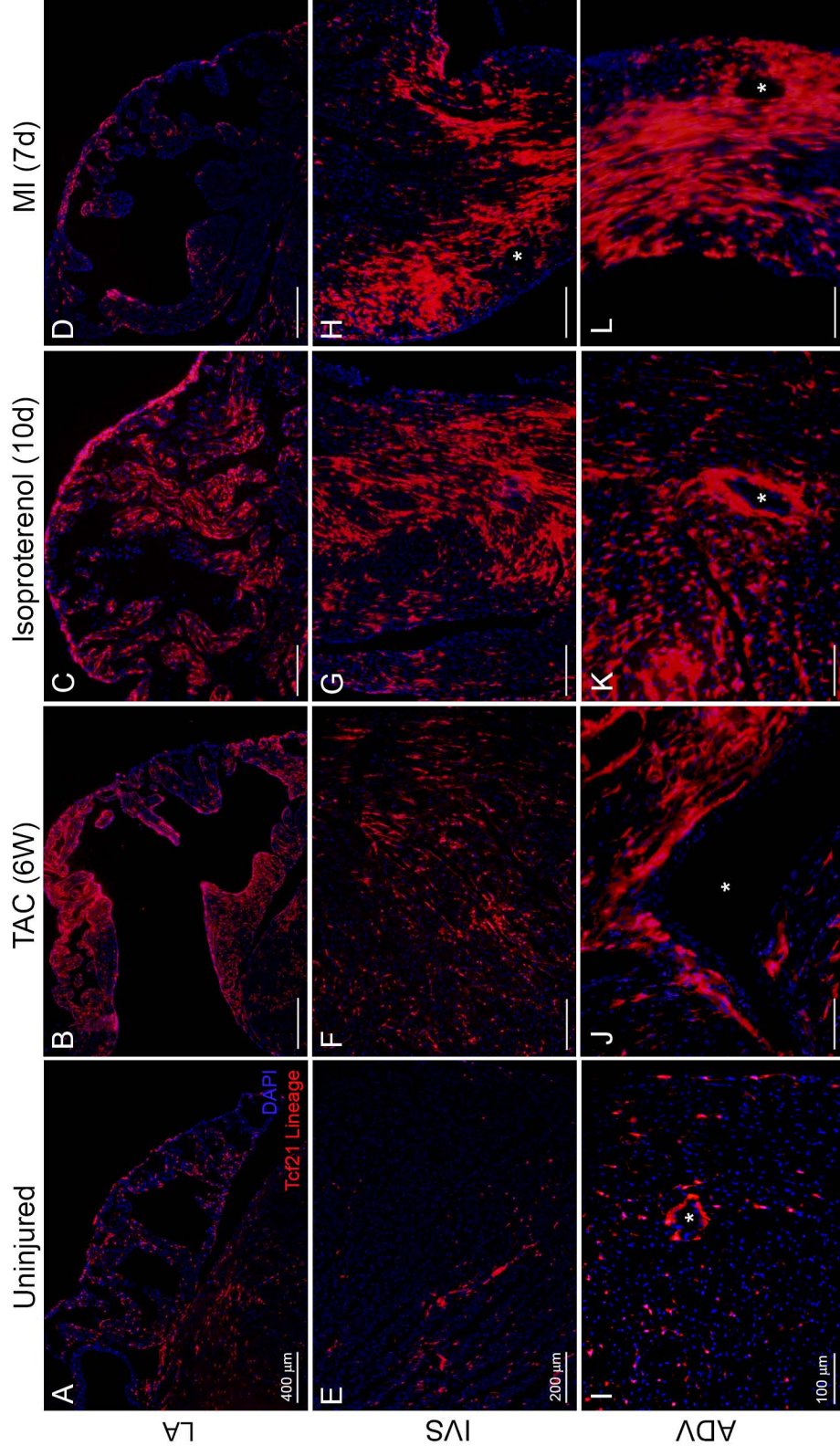


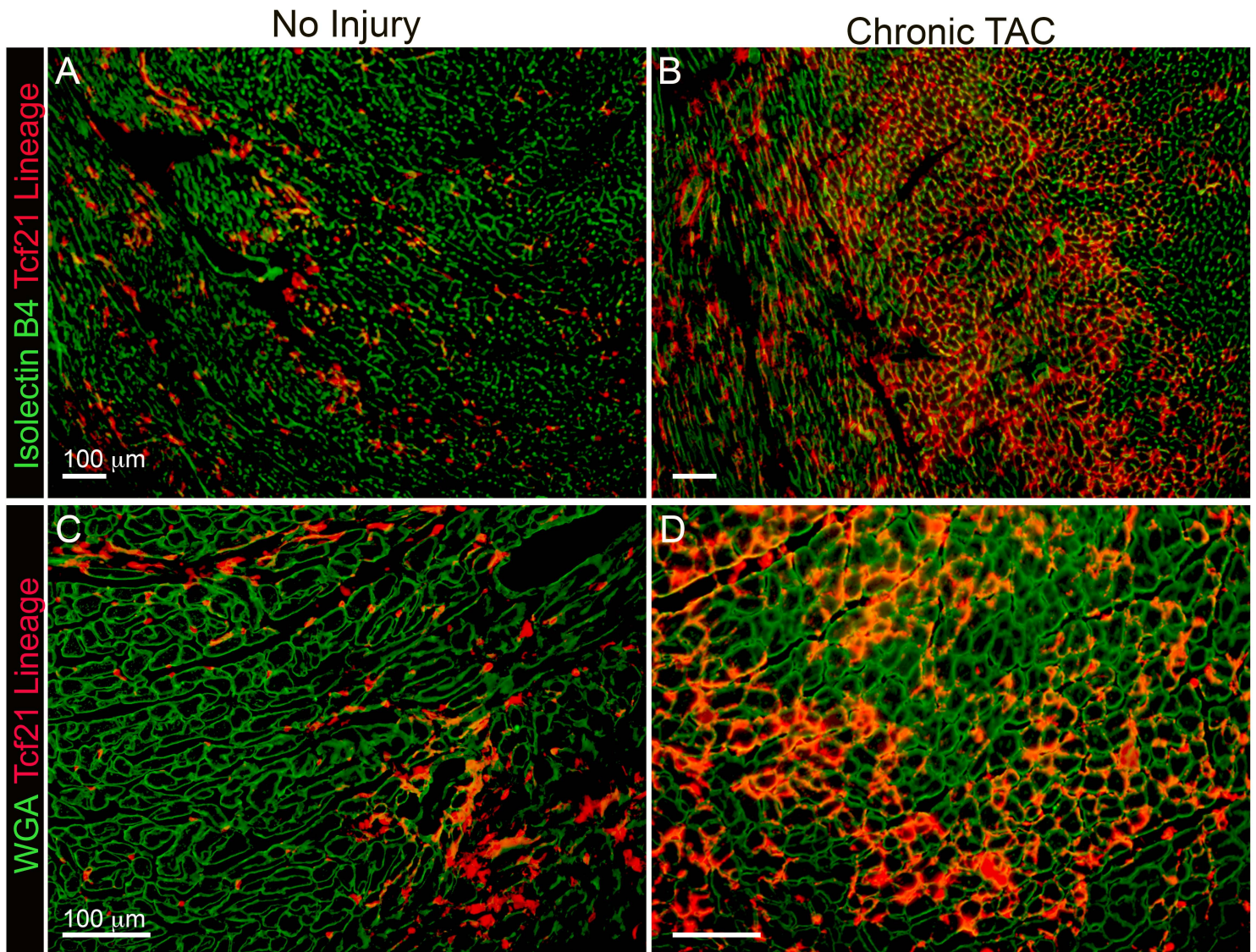
Supplemental Figure 1. Comparison of GFP fibroblast reporter lines. (A) Images of GFP expression from the left ventricle of $PDGFR\alpha^{GFP/+}; Col1a1-GFP$ double transgenic hearts. Arrows indicate GFP^+ nuclei ($PDGFR\alpha^{GFP}$) that are embedded within cells expressing cytoplasmic GFP ($Col1a1-GFP$). E: epicardium. Scale bars: $100\mu m$. (B) Number GFP^+ cells determined by flow cytometry from either $PDGFR\alpha^{GFP/+}$ or $Col1a1-GFP$ hearts. $n=6$ per genotype. Results are mean \pm SD.



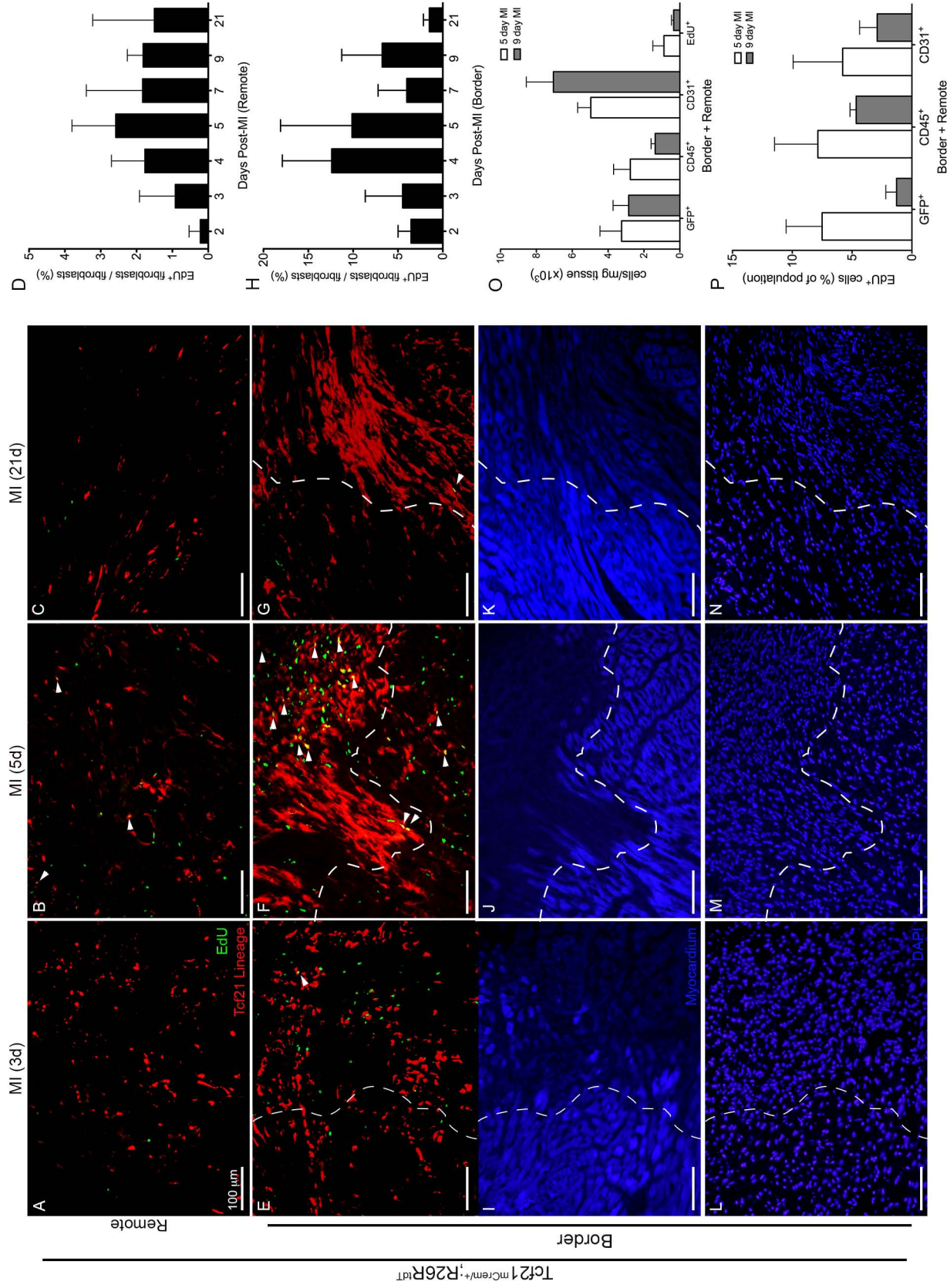
Supplemental Figure 2. Alternative cardiac fibroblast markers. (A-B) Representative images of vimentin (red) staining. **(A'-B')** Higher power view of images shown in A-B. Arrowheads indicate GFP⁺ / vimentin⁺ cells. **(C-D)** Representative images of DDR2 (red) staining. **(E-H)** Representative images of cardiac tissue stained with NG2 (green), IB4 (blue), and DDR2 (red). Asterisks indicate blood vessels. Arrowheads indicate DDR2⁺/NG2⁺ cells. Arrows indicate DDR2⁺/IB4⁺ cells. Scale bars: (A-D) 100 μ m, (A'-B') 50 μ m, and (E-H) 50 μ m.



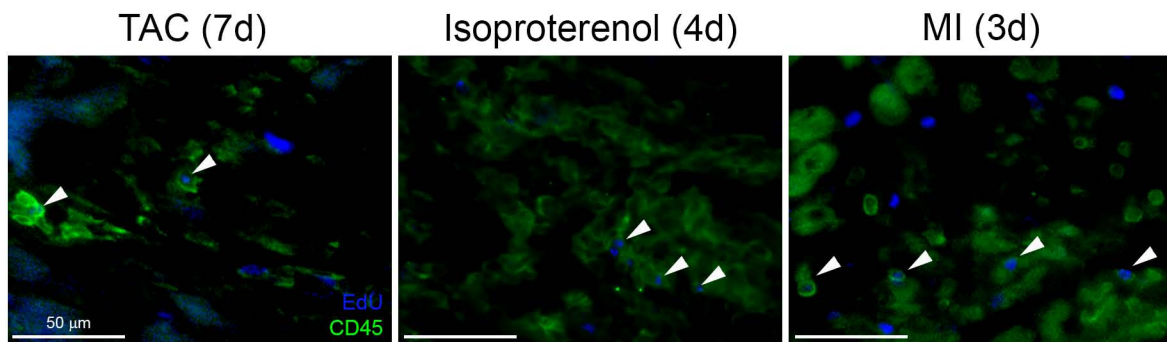
Supplemental Figure 3. Atrial, septal, and adventitial fibroblast expansion in three injury models. (A-D) Atrial, (E-H) septal, and (I-L) adventitial fibroblasts after indicated treatments. Mode of tamoxifen induction: uninjured (gavage), TAC (chow), MI (gavage or postnatal intragastric injection), isoproterenol (gavage). Sections from TAC hearts were stained with DsRed antibody to detect tdTomato protein. Asterisk indicates blood vessel. LA: left atria; IVS: interventricular septum; ADV: adventitia. Scale bars: (A-D) 400μm, (E-H) 200μm, and (I-L) 100μm.



Supplemental Figure 4. Fibroblast distribution in aged and chronic pressure overload heart. Tcf21 lineage fibroblast accumulation in (A, C) One year old uninjured heart and (B, D) 1 year old heart after 7 months of mild pressure overload. (A-B) Isolectin B4 (green) and (C-D) WGA (green) staining. Fibroblasts accumulate in a perivascular and perimysial pattern predominantly after chronic pressure overload. Fibroblasts: tdTomato (red). WGA: wheat germ agglutinin. Scale bars: 100μm.



Supplemental Figure 5. Border and remote area proliferation after MI. (A-C) Remote and (E-G) border zone Tcf21 lineage fibroblasts after permanent LAD ligation. Fibroblasts: tdTomato (red); proliferating cell: EdU (green); myocardium: autofluorescence (blue, I-K), nuclei: DAPI (blue, L-N). (D, H) Proliferation analyzed by immunohistochemistry. (O) Number of cells per mg of tissue and the (P) percent of proliferating cells of the fibroblast (Col1a1-GFP⁺), immune cell (CD45⁺) and endothelial cell (CD31⁺) populations in the border and remote regions determined by flow cytometry. Results are mean ± SD. n=2-5 mice per group. Scale bars: 100µm.



Supplemental Figure 6. Immune cell proliferation after injury. Representative images of CD45 (green) and EdU (blue) staining at indicated time points. Arrowheads identify CD45⁺/EdU⁺ costained cells. Scale bars: 50μm.