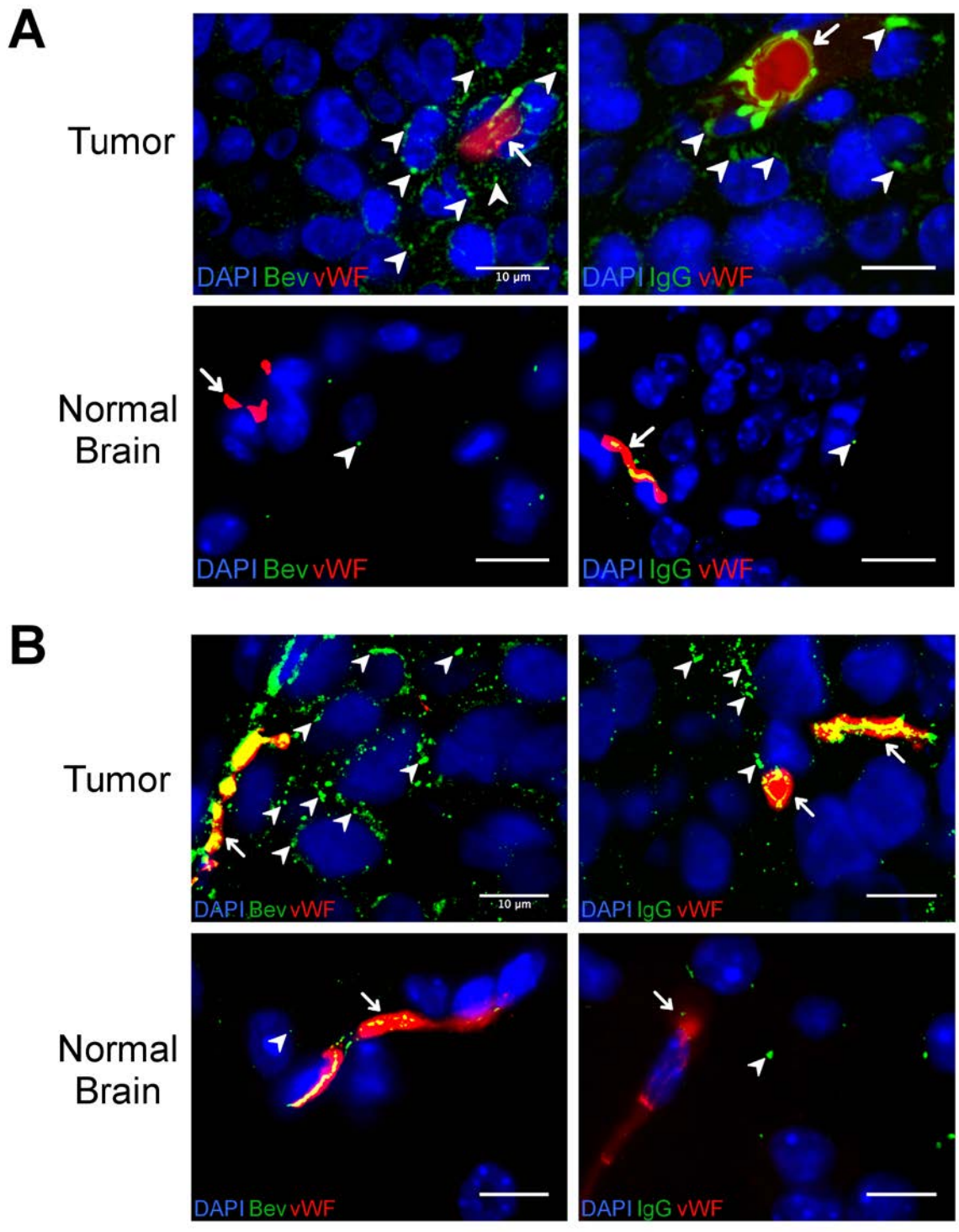
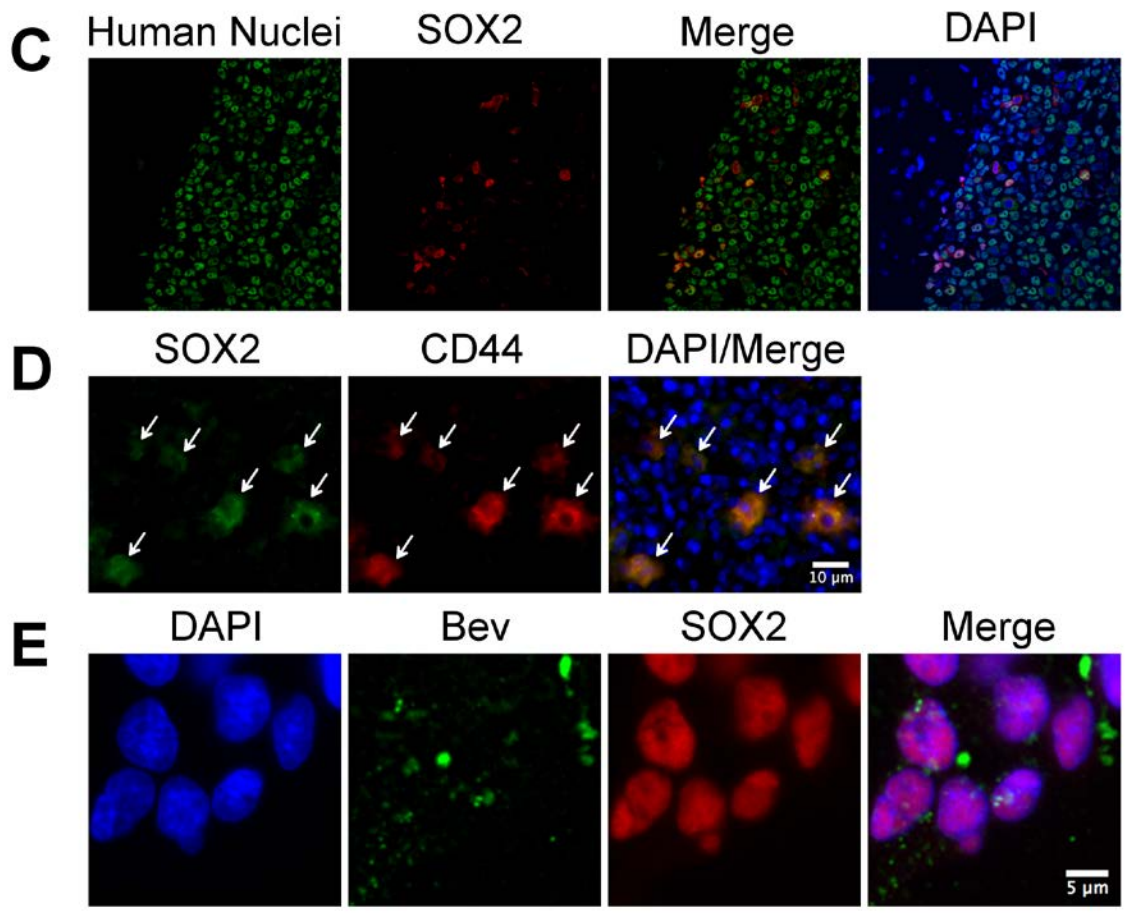
Suppl Figure 1





SFigure 1. Bevacizumab gains access to the perivascular tumor space and is internalized by tumor cells when administered to an orthotopic xenograft model of GBM and confirmation of cancer stem-like cell markers in vivo. **A&B**, Luciferase-labeled CSCs (15,000) were injected intracerebrally into the nude mouse brain, at 14 days bioluminescent imaging was initiated and once the tumor reached a predetermined size bevacizumab or IgG administration was begun and continued for 5 days. The mice were then euthanized, the brains harvested, fixed in buffered formalin and embedded in paraffin (A), or fixed in 4% paraformaldehyde, immersed in 20% sucrose and frozen (B). Sections were reacted with Alexa-488-anti-human IgG (green), and anti-vWF IgG,

followed by an Alexa-594-secondary antibody (red). Area of tumor from mouse administered bevacizumab or IgG is shown along with adjacent normal mouse brain. Arrows denote ECs (blood vessels) and arrowheads denote bevacizumab. Scale bars denote 10-µm. **C**, Perivascular human tumor cells detected with rabbit anti-human nuclei and mAb anti-SOX2 in the xenograft model of GBM from A&B. Sections were double-labeled with mAb anti-Sox2 (2 µg/ml) and rabbit anti-human nuclei (5 µg/ml, Millipore cat# MAB1281A4), followed by Alexa-488 and Alexa-594 secondary antibodies, respectively, and DAPI nuclear stain. **D**, Perivascular CD44+ cells also mark with mAb anti-Sox2 in the immune competent PDGF-B-induced mouse model of GBM. Tumors were established as described in Figure 1E. Sections were reacted with sheep anti-mouse CD44 (5 ug/ml; R&D Systems cat#AF6127) and with mAb anti-Sox2 (2 µg/ml), followed by Alexa-488 and Alexa-594 secondary antibodies, and DAPI nuclear stain. **E**, $CD133^+$ GBM tumor cells are $Sox2^+$. $CD133^+$ cells were plated and treated with bevacizumab as described in Figure 3A&B. Cells were reacted with Alexa-488 anti-human IgG (5 µg/ml) (green) and with mAb anti-Sox2 (2.5 µg/ml), followed by an Alexa-594-secondary antibody (red), DAPI nuclear stain, cover slipping and microscopy.