Supplementary Materials: IQGAP1 in Podosomes/Invadosomes is Involved in the Progression of Glioblastoma Multiforme Depending on the Tumor Status

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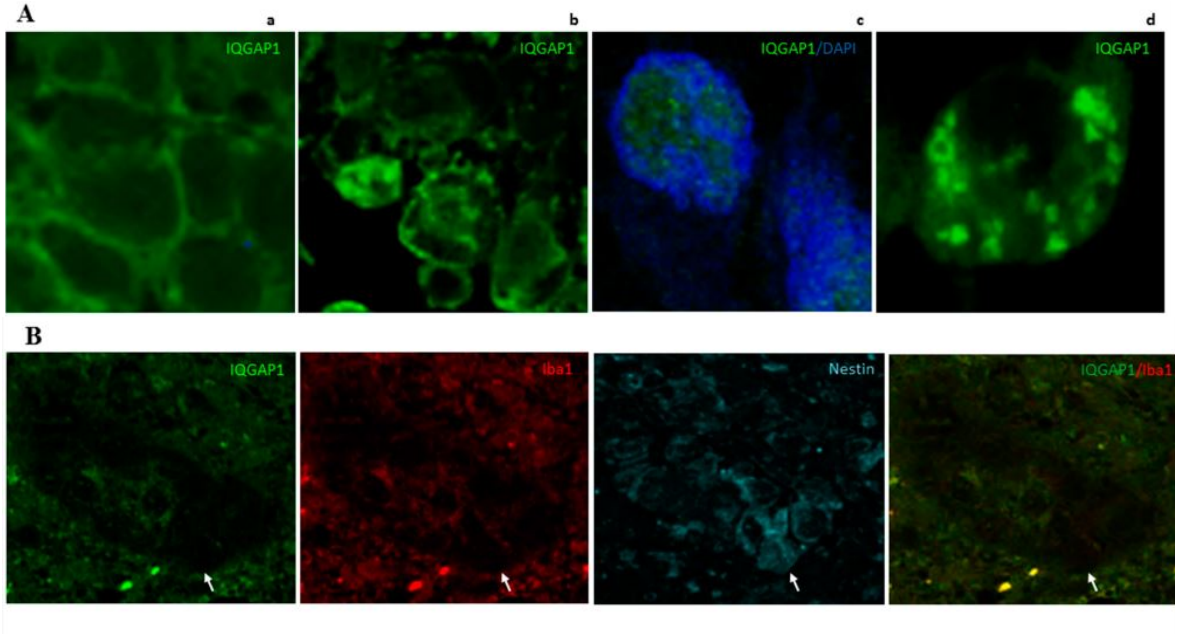

Supplemental Figure S1.

A: Higher magnification of panels A-D of Figure 1. IQGAP1 protein in GBM detected in (a) plasma membrane; (b) cell protrusions, plasma membrane and cytosol; (c) nucleus. (d) cell with podosome-like structures highly positive for IQGAP1.

B: Higher magnification of panels K-O of Figure 2. Triple immunolocalization of IQGAP1 protein (green), Iba1(red) and nestin (cyan). Arrow points to a nestin^{+}/IQGAP1^{-}/Iba1^{-} cell flanked by nestin^{+}/IQGAP1^{+}/Iba1^{+} cells.

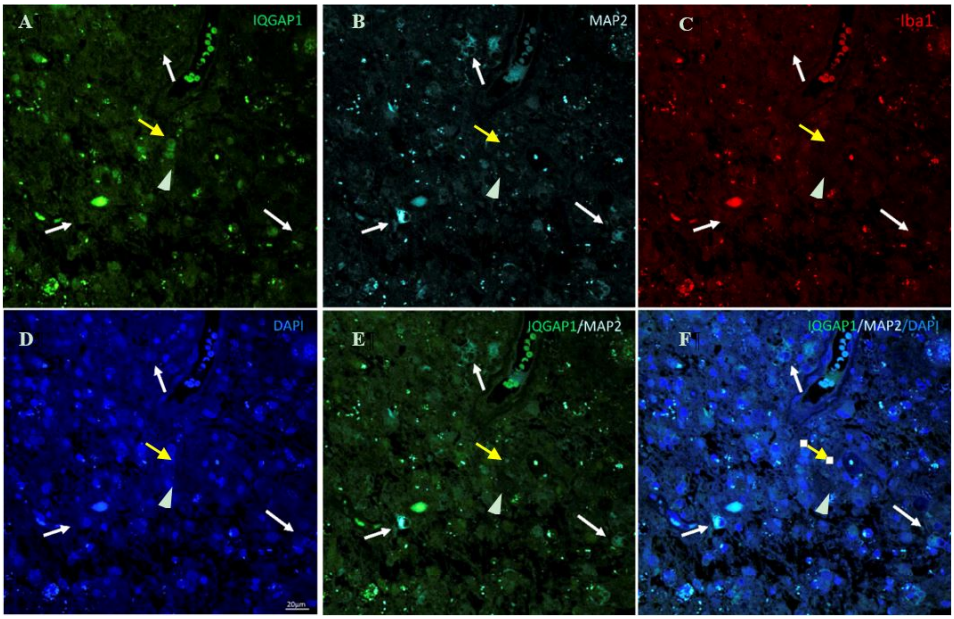

Figure S2. Triple immunolocalization of IQGAP1 protein (green), the microglia/macrophage marker Iba1 (red) and the Microtubule-associated protein 2 (MAP2, cyan) in GBM tissue sections. White arrows point to Map2^{+} neurons IQGAP1^{-}/Iba1^{-}. Yellow arrows points to a cell IQGAP1^{+}/Iba1^{+}. Arrowhead points a cell IQGAP1^{+}/nestin^{+}. Bar: 20µm.

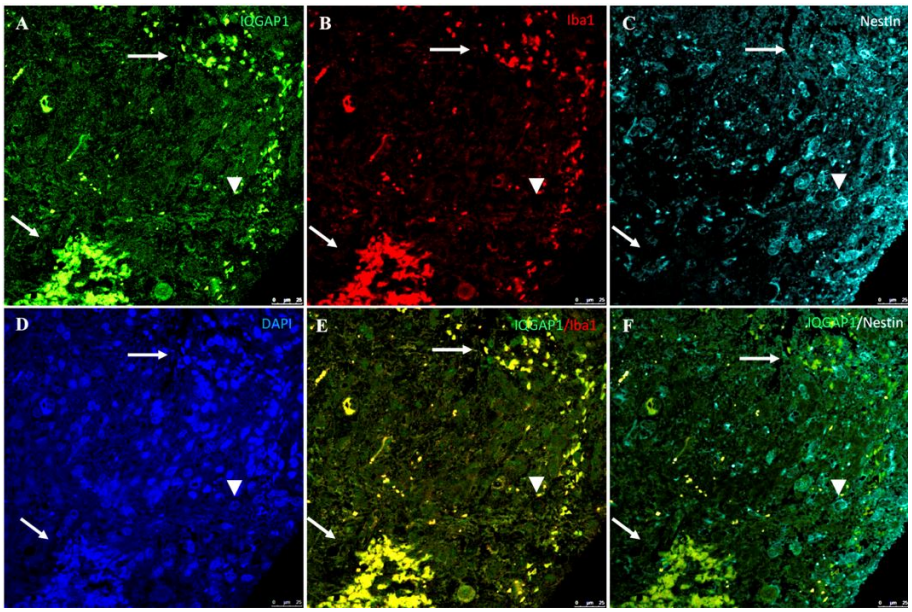

Figure S3. Triple immunolocalization of IQGAP1 protein (green), the microglia/macrophage marker Iba1 (red) and the CSC marker nestin (cyan). In this highly vascularized area of a GBM tissue section many nestin^{+} cells are observed (arrowhead). Note the massive presence of macrophages (Iba1^{+}/IQGAP1^{+}/nestin^{-}) in the periphery (arrows). Scale bar: 25µm.

Table S1. IQGAP1/PCNA and IQGAP1/GFAP colocalization. Mean and standard deviation of values obtained with the ImageJ plug-in "Manders' coefficients".

	n	R	Ch1/Ch2	M1	M2	Ch1 Thresh, Ch2 Thresh
Ch1 = GFAP Ch2 = IQGAP1	27	0.59 ± 0.05	0.995 ± 0.02	0.969 ± 0.02	0.971 ± 0.01	1;255
Ch1 = PCNA Ch2 = IQGAP1	20	0.677 ± 0.13	0.750 ± 0.29	0.851 ± 0.14	0.739 ± 0.20	1;255

Ch1 = Iba1 **Ch2 = IQGAP1**	14	0.84 ± 0.04	0.801 ± 0.15	0.954 ± 0.04	0.835 ± 0.09	1;255

n = samples analized. **R** = Manders Overlap coefficient. Value Range: 0–1, with 0 = low colocalization; 1 = high colocalization; **Ch1/Ch2** = red/ green pixel ratio. Ratio ≈ 1 allows the use of Mander's Overlap coefficient (R); **M1**, **M2** = Manders Colocalization coefficients for channel 1 (M1) and channel 2 (M2). Value range: 0–1, with 0 = no colocalization 1 = all pixels colocalize; **Ch1 thresh, Ch2 thresh** = ch1 and ch2 threshold.