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Supplemental Information

Locally controlling mesenchymal stem cell morphogenesis by 3D PDGF-BB gradients towards the establishment of an in vitro perivascular niche

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Figure S1

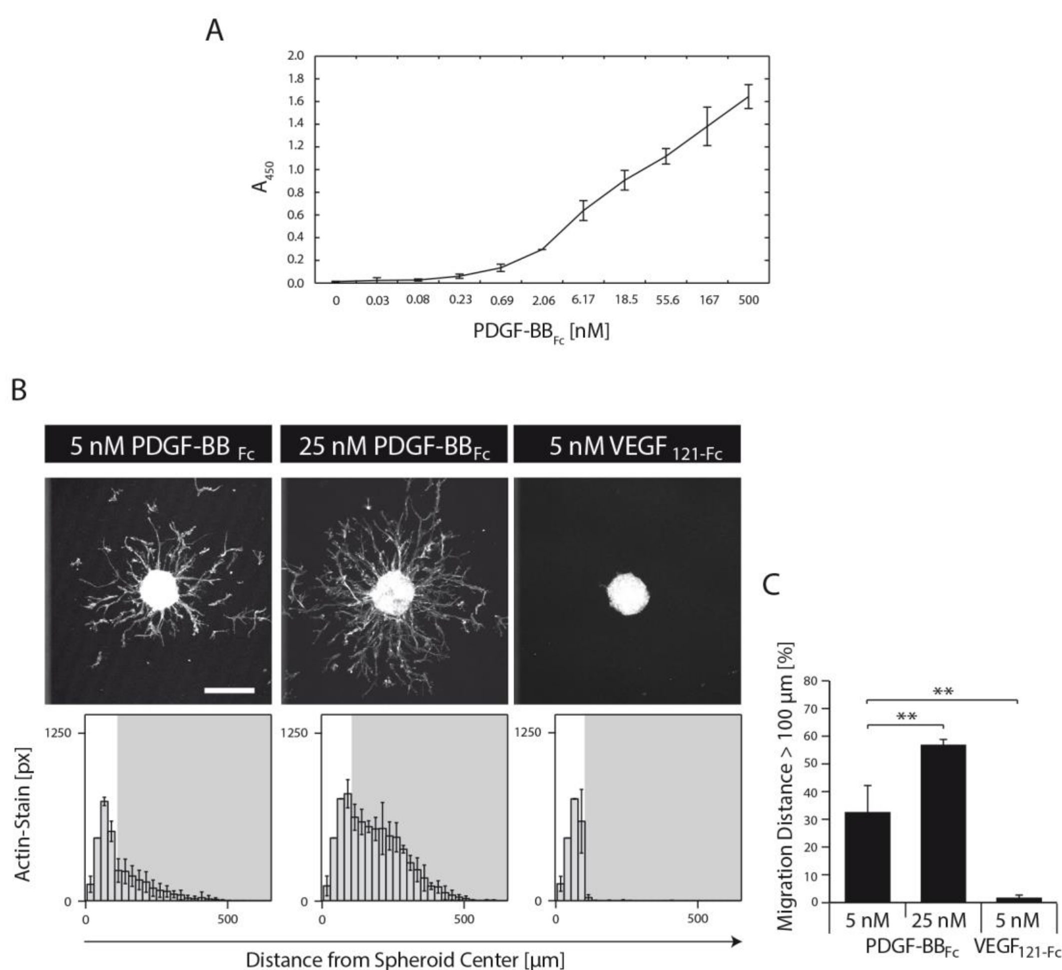

Fig. S1 Soluble PDGF-BB_{Fc} specifically binds to Gln-ZZ and is biologically active. A) Solid phase binding assay for detection of PDGF-BB_{Fc} binding to Gln-ZZ that was adsorbed to a plastic surface. B) MSC microtissues consisting of approx. 750 cells were embedded in synthetic matrices and stimulated with the indicated Fc tagged growth factor for 20 hours under serum-free conditions. Z-stack projections of F-actin stained MSC microtissues (upper panel, scale bar = 200 µm). Histograms showing the distribution of MSCs from microtissue center when stimulated with the respective growth factor (lower panel). C) Bar graph shows the percentage of actin-stained pixels being located at a distance larger than 100 µm from the centre of the microtissue. Data is depicted as mean ± SD ($n \geq 4$, **, $p < 0.01$).

Figure S2

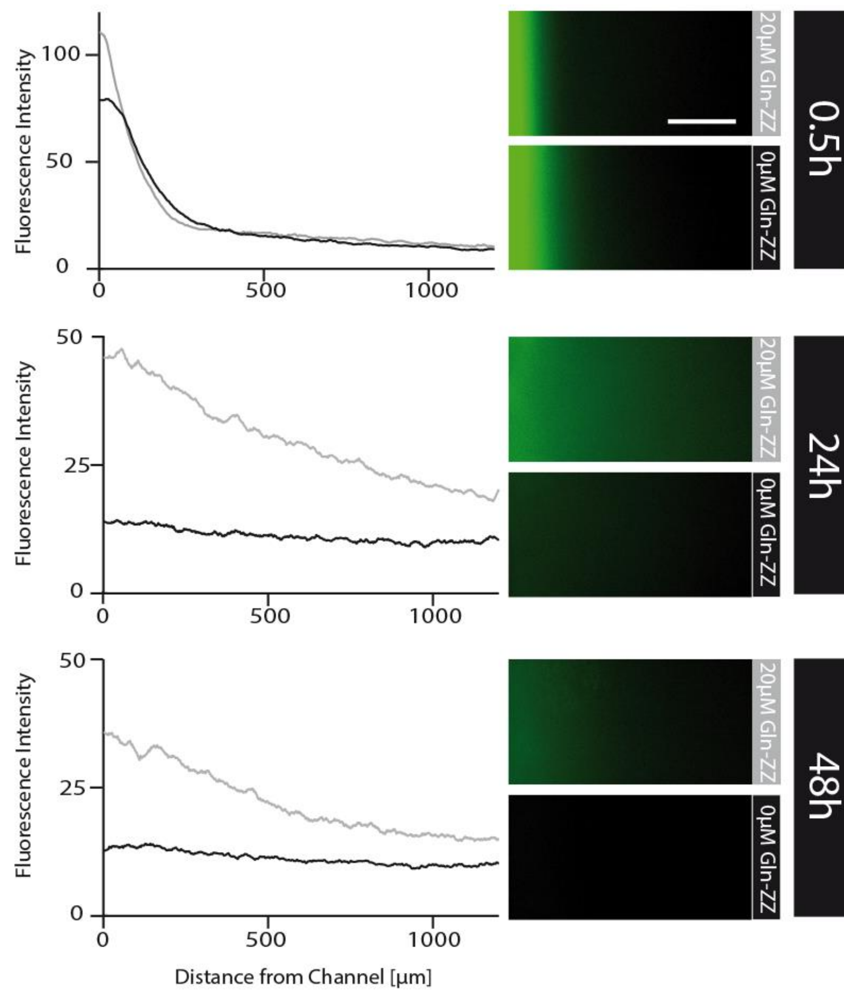

Fig. S2 Gln-ZZ containing TG-PEG gel allows to bind PDGF-BB$_{FC}$ by affinity and build a stable gradient providing local growth factor supply in presence of encapsulated MSCs (0.25 * 10^6 cells ml^{-1}). The graph shows the evolution of labeled PDGF-BB$_{FC}$ diffusing in the gel over time. 4 µM PDGF-BB$_{FC}$ were perfused in the gel and images were taken at 0.5, 24 and 48 hours after perfusion. The upper and lower images of each time point depict the gel containing 20 µM and 0 µM Gln-ZZ, respectively. The plots on the left panel show the fluorescence intensity for each condition. The grey line relates to the 20 µM Gln-ZZ condition (upper panel) and the black line to the 0 µM Gln-ZZ condition (lower panel).

Figure S3

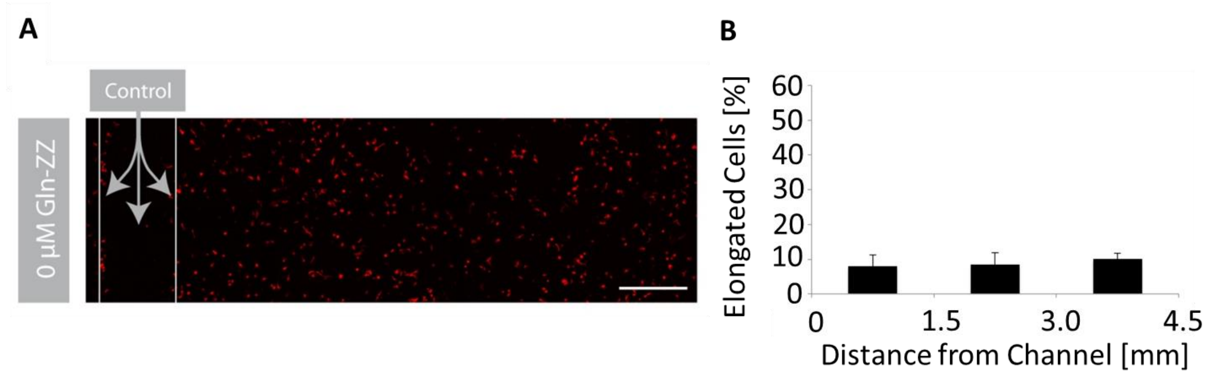

Fig. S3 Evaluation of MSCs in a perivascular niche model without PDGF-BB. MSCs were encapsulated (0.25 * 10^6 cells ml^{-1}) in TG-PEG hydrogels and channels for biomolecule perfusion were formed by electrochemical inhibition of hydrogel polymerization. A) Representative Z-stack images (total stack size 500 µm) of F-actin stained MSCs after 48 hours in culture (Scale bar = 500 µm). Channel in unaltered TG-PEG hydrogel was perfused with control medium. B) Histogram depicting percentage of cells with a circularity ratio below 0.25 relative to distance to channel. Data is depicted as mean ± SD (n ≥ 4).