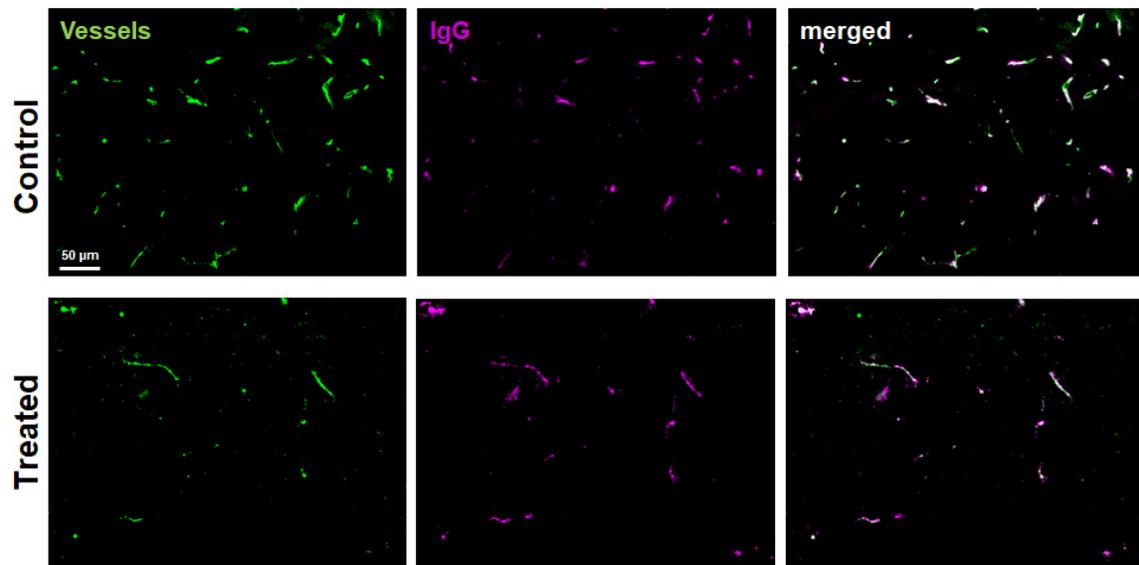


Multimodal and multiscale optical imaging of nanomedicine delivery across the blood-brain barrier upon sonopermeation

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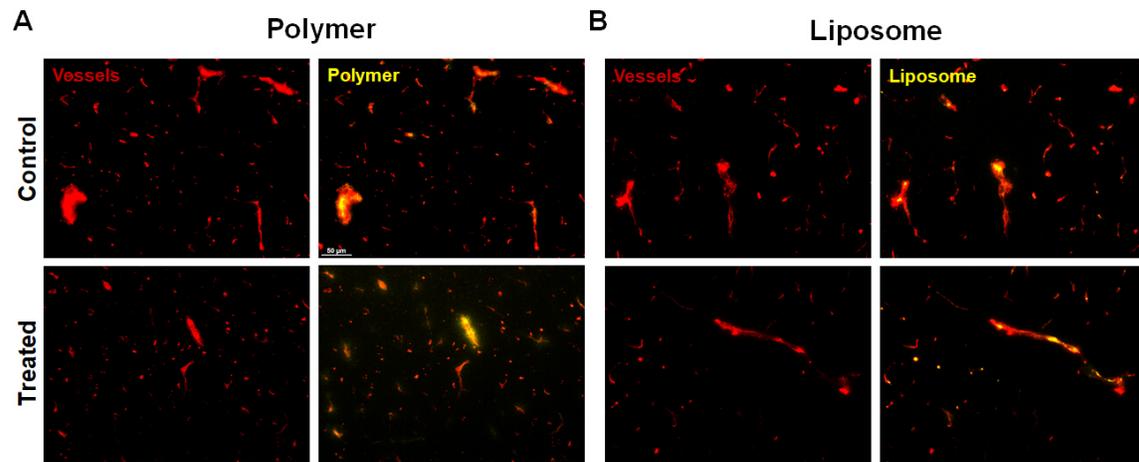
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Supplementary Figure 1:



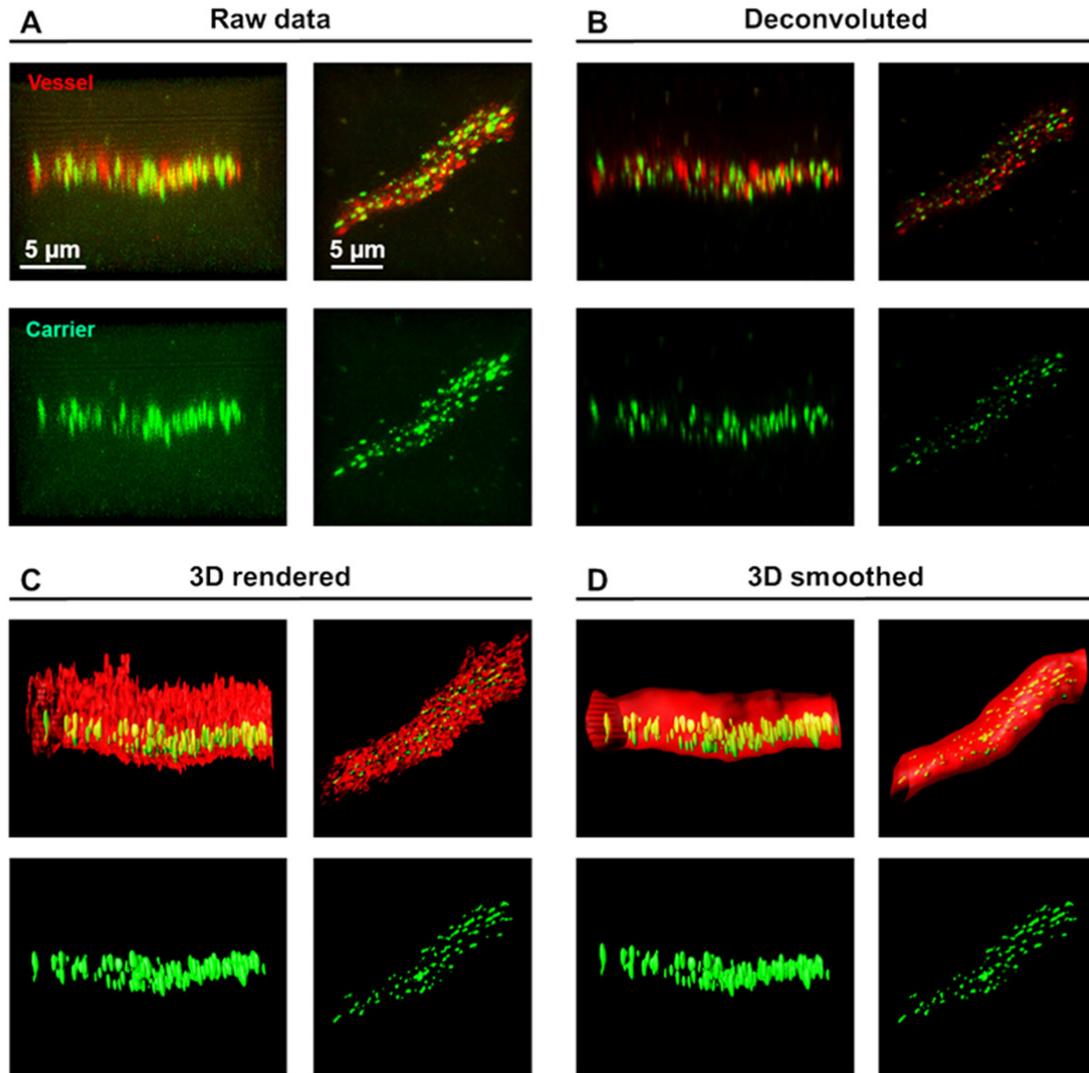
Supplementary Figure 1: Fluorescence microscopy of IgG extravasation. Immunofluorescence staining of endogenous IgG (in purple) and blood vessels (CD31 staining; in green) in the brain showed extravasated IgG only in case of sonopermeation treatment. Images at higher resolution are provided in Figure 3.

Supplementary Figure 2:



Supplementary Figure 2: Fluorescence microscopy of polymer and liposome extravasation. Analysis of nanocarrier (in yellow; fluorophore-labeled) extravasation and penetration out of brain blood vessels (in red; rhodamine-lectin staining) upon sonopermeation treatment. Higher resolution images are provided in Figure 4.

Supplementary Figure 3:



Supplementary Figure 3: STED image processing and 3D rendering. STED images of a vessels in the brain (stained using rhodamine-lectin; in red) and nanomedicine formulations (in green), at different stages of image processing. A: Raw data with background and shifted signal. B: Deconvoluted images with background reduction and correction for potential signal shifts, automatically performed by the Huygens Professional software (Scientific Volume Imaging). C: 3D-rendered images based on the gained signals. D: 3D-smoothed final images, with realistic vessel shapes and surfaces.