Supplementary Data to the manuscript:

Silk based scaffolds with immunomodulatory capacity: anti-inflammatory effects of nicotinic acid

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Supplementary Fig. S1. Krypton gas adsorption–desorption isotherm of cross-linked silk scaffold (three times measurement per sample).



Supplementary Fig. S2. Metabolic activity (%) of MG63 cells in medium supplemented with different concentrations of nicotinic acid was measured using a PrestoBlue assay. (A) After 1 day of seeding. (B) After 3 days of seeding (±SD, n=3).



Supplementary Fig. S3. Confocal images of MG63 cells seeded on the scaffolds after 7 days of culture stained for actin filaments (green) and cell nuclei (blue). Scale bars = $100 \mu m$.



Supplementary Fig. S4. Relative gene expression of pro-inflammatory markers TNF- α , CXCL10 and CD197 in response to medium supplemented with different concentrations of nicotinic acid. Expression levels ±SD were normalized to M ϕ macrophages seeded in drug-free medium (TCP (M ϕ)). RPL37a was used as a housekeeping gene. n=3 (****p< 0.0001, ***p< 0.001, **p< 0.01).



Supplementary Fig. S5. Total DNA content after 1 day of seeding M1-like macrophages on the scaffolds was measured using a Hoechst assay. $(\pm SD, n = 3)$



Supplementary Fig. S6. Relative gene expression of anti-inflammatory marker IL-10 in response to different concentrations of nicotinic acid in the scaffolds (SNP) and in the medium (NA). Expression levels \pm SD were normalized to M ϕ macrophages. RPL37a was used as a housekeeping gene (n=3).