Supplementary Materials

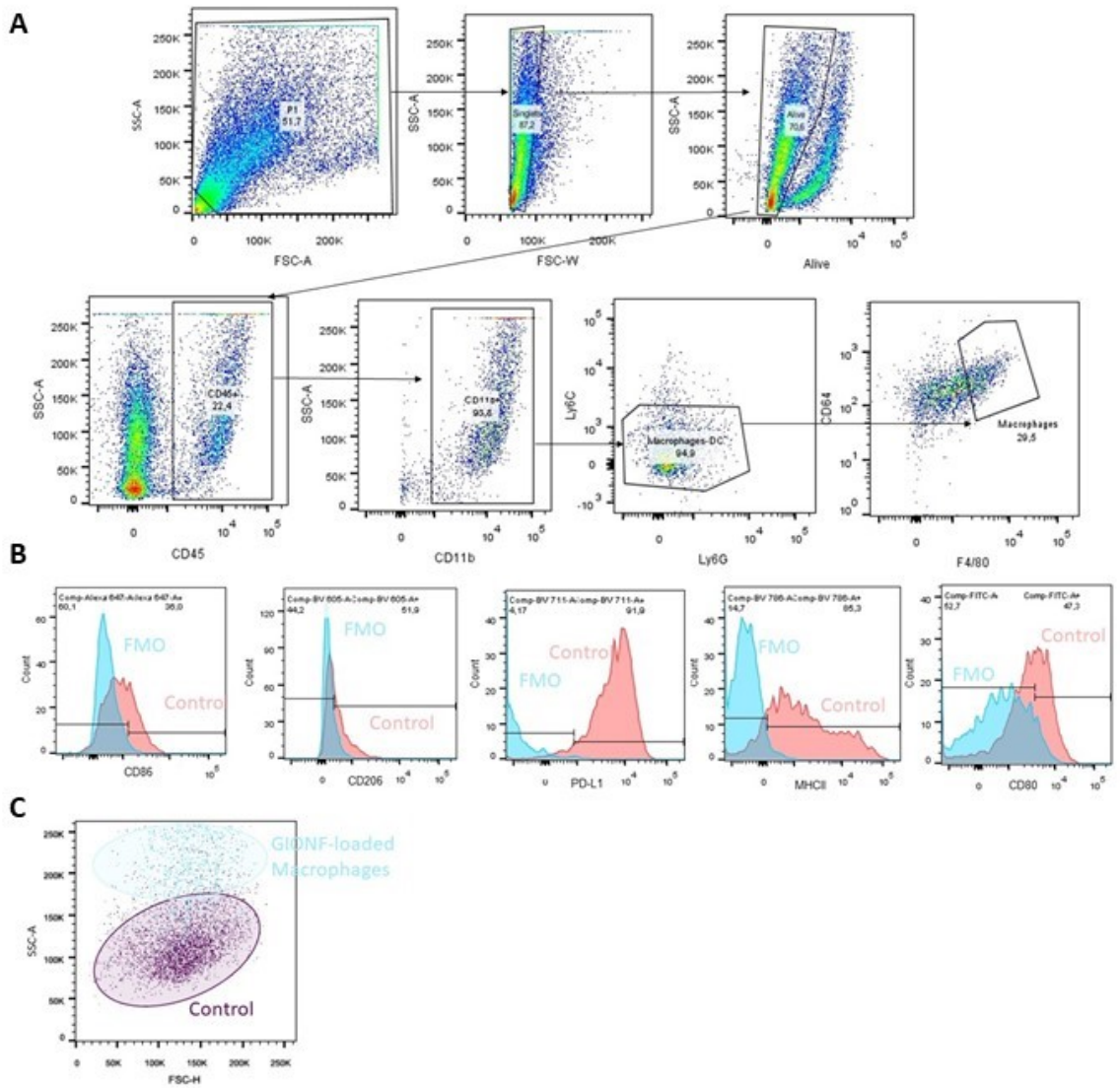

Figure S1. A) Gating strategy for macrophage profiling. Macrophages are gated as CD45+ CD11b+ Ly6C- Ly6G- F4/80+ CD64+. B) Once the macrophage was gated, the percentage of positive cells for each of the markers was determined. To establish the gating between positive and negative cells in activation markers (CD86, CD206, PD-L1, MHCII, CD80), Fluorescence Minus One (FMO) controls have been used (blue histogram vs controls in red). C) Comparing the side scatter of control macrophages and GIONF-loaded macrophages.

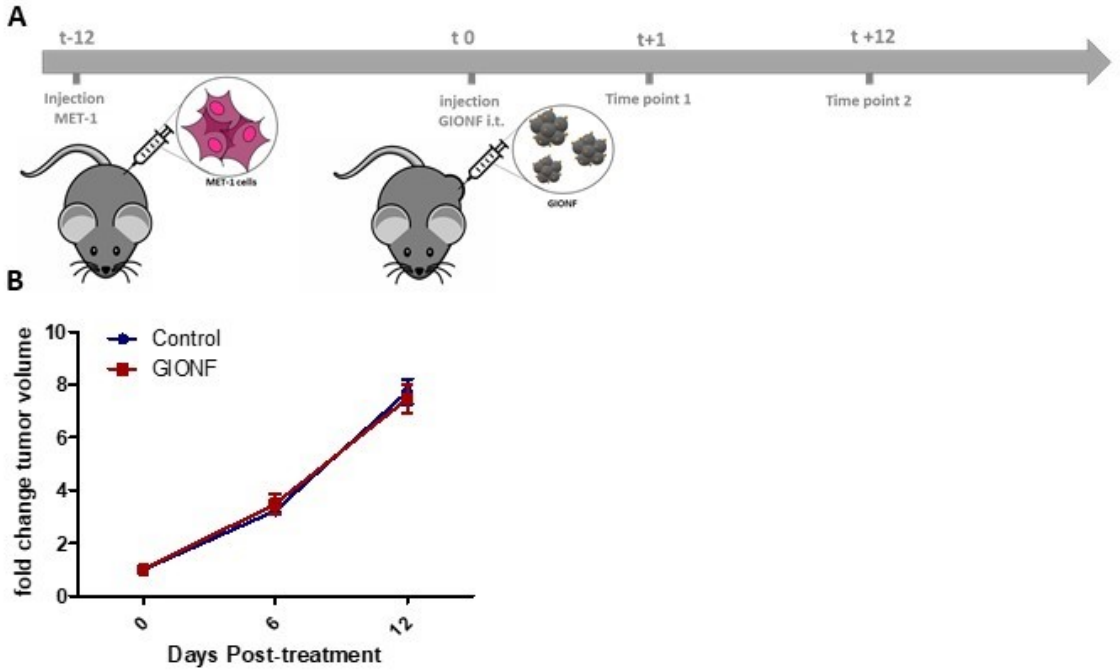

Figure S2. Effect of GIONF on the tumor immune infiltrate. A) Scheme of the experimental procedure. The tumor immune infiltrate was evaluated 1 day and 12 days after GIONF injection. B) MET-1 tumor volume fold change of PBS (Control) and GIONF-injected tumors (n=6/group).

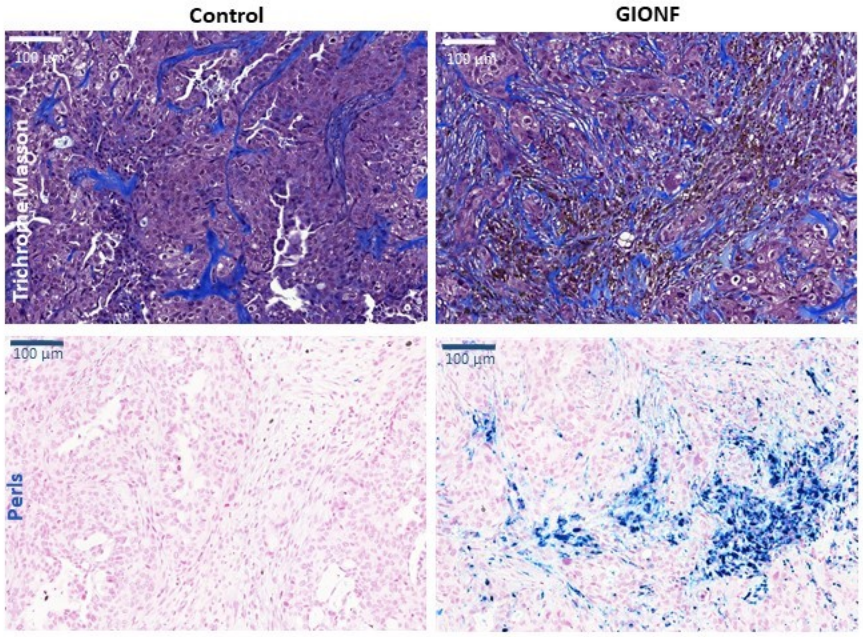

Figure S3. Histological analysis of MET-1 tumor 24 hours after GIONF intratumoral injection. Trichrome Masson and Perls staining of MET-1 tumors (consecutive slices). Perls staining labels iron in blue, allowing the detection of GIONF location. Scale bar = 100 µm.

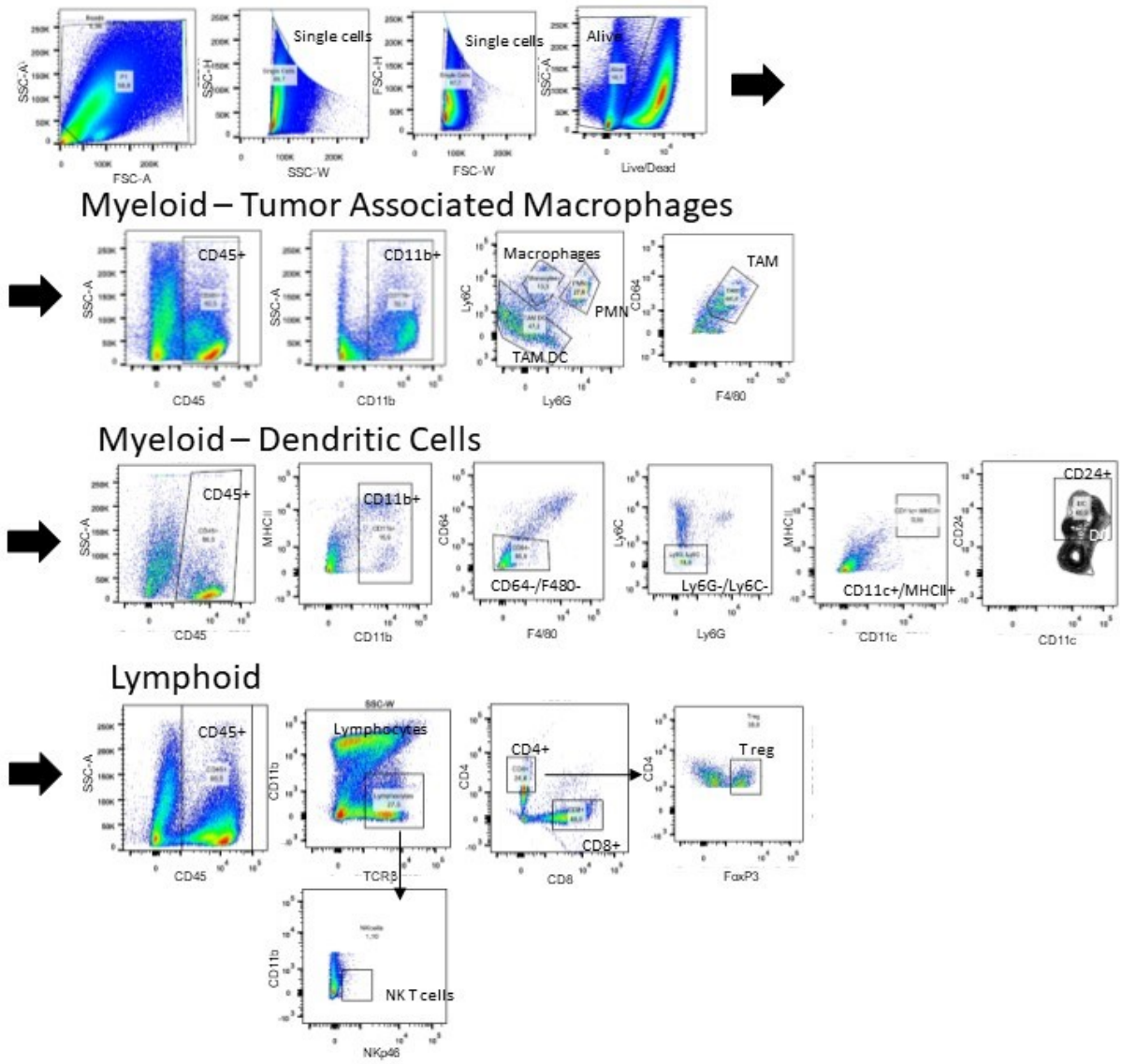

Figure S4. Gating strategy for tumor immune infiltrate profiling. Three different strategies are used: myeloid cell gating strategy to identify tumor-associated macrophages (CD45+ CD11b+ Ly6C- Ly6G- F4/80+ CD64+), myeloid cell gating strategy to identify tumor dendritic cells (CD45+ CD11b+ F4/80- CD64- Ly6C- Ly6G- CD11c+ MHCII+CD24+),and lymphoid cells gating strategy to identify tumor infiltrating lymphocytes.

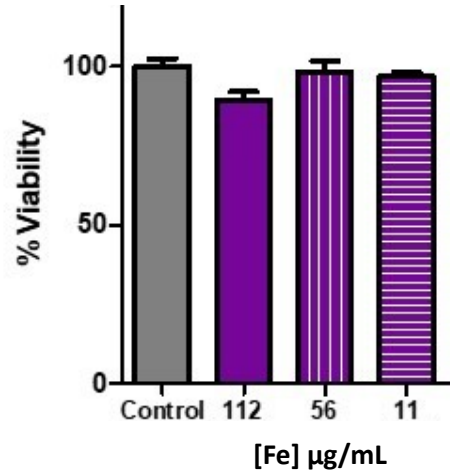

Figure S5. GIONF are not cytotoxic to MET-1 cells. Cell viability was measured using Alamar Blue after 24 hours in contact with GIONF at different concentrations.

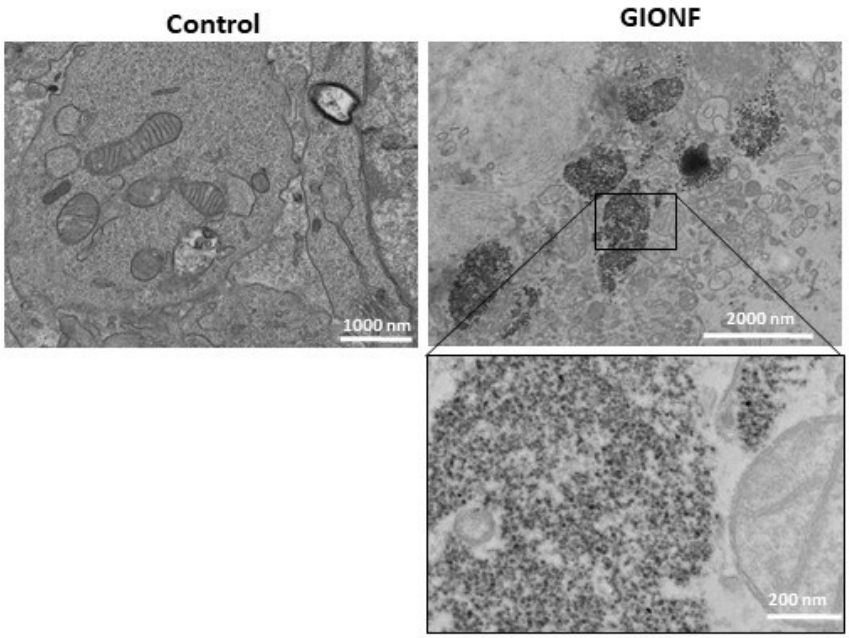

Figure S6. TEM images of MET-1 tumors treated with GIONF-mediated PTT. GIONF are located as large clusters in cells showing destructuration and apoptosis features.