Figure S1. Representative histograms demonstrating the phenotype characteristics of M-MDSCs (A) and PMN-MDSCs (B) on the 3rd day after IL-10 silencing. M-MDSC, monocytic myeloid-derived suppressor cells; PMN-MDSC, polymorphonuclear myeloid-derived suppressor cells.

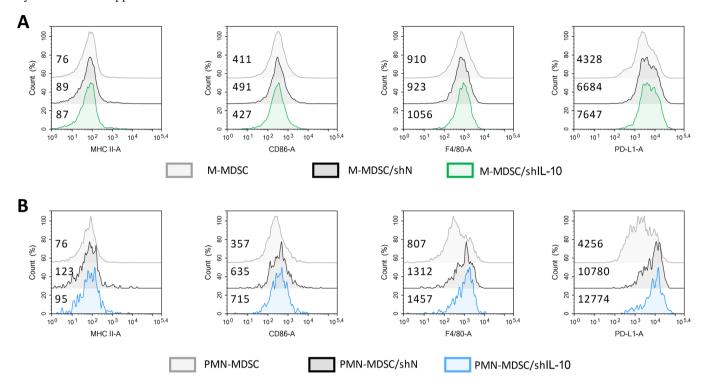


Figure S2. Characteristics of CD11c<sup>+</sup> with the silenced expression of IL-10 identified in the *in vitro* differentiating MDSC culture. (A) Representative flow cytometric data and bar plots showing the changes in the proportion of CD11c<sup>+</sup> cells in the culture and the percentage of EGFP<sup>+</sup> cells (as a transduction efficacy control) among identified cell subpopulation on the 3rd day after IL-10 silencing. (B) Bar plots and representative histograms demonstrating the phenotype characteristics of CD11c<sup>+</sup> cells on the 3rd day after IL-10 silencing. The results are given as the mean ± SD calculated for three independent experiments measured in triplicates. Statistical significance was calculated using Welch's ANOVA followed by post hoc Dunnett's T3 multiple comparison test. Differences with a P<0.05 were regarded as significant (\*P<0.05, \*\*P<0.01, \*\*\*\*P<0.001, \*\*\*\*P<0.0001). ctrl, non-transduced cells; shN, cells transduced with LVs encoding shIL-10 sequence; MFI, mean fluorescence intensity.

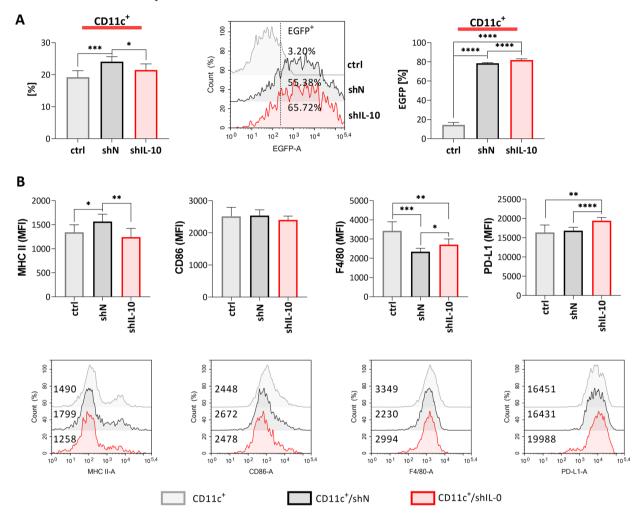


Figure S3. Scheme of flow cytometry analysis of CFSE-based proliferation assay.

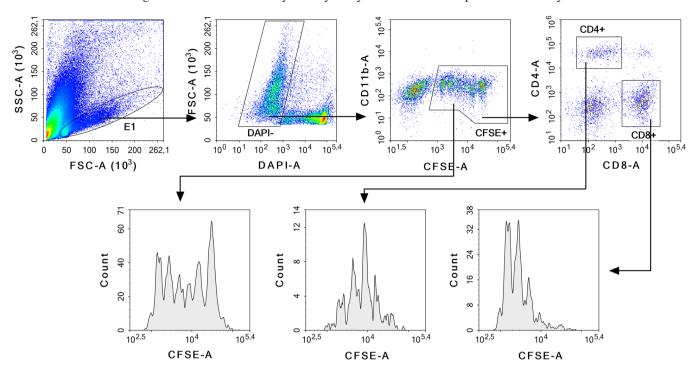


Figure S4. Influence of IL-10 silencing on the suppressive activity of (A and C) M-MDSCs and (B and D) PMN-MDSCs. CFSE-labeled splenocytes obtained from healthy mice were cultured with M-MDSCs and PMN-MDSCs (isolated from the *in vitro* culture of MDSCs using the FACS Aria sorter) at the final ratio of 4:1. (A and B) Proliferation of CD4<sup>+</sup> and CD8<sup>+</sup> spleen-derived T lymphocytes was measured by CFSE dilution and presented as mean intensity of fluorescence (MFI). Splenocytes cultured with ConA and IL-2 (splc ctrl) were used as the positive control of proliferation. The results are presented as the mean inverse fold change of CFSE MFI calculated by reference to the splc ctrl group  $\pm$  SD. (C and D) Concentrations of IFN- $\gamma$  and IL-10 in supernatants collected after 72 h co-culture of M-MDSCs or PMN-MDSCs with splenocytes measured using ELISA. The results are expressed as the mean  $\pm$  SD. Data obtained for two independent experiments measured in 1-3 repeats. Statistical significance was calculated using Welch's ANOVA followed by post hoc Dunnett's T3 multiple comparison test. Differences with a P-value <0.05 were regarded as significant (\*P<0.05). M-MDSC, monocytic myeloid-derived suppressor cells; MFI, mean fluorescence intensity.

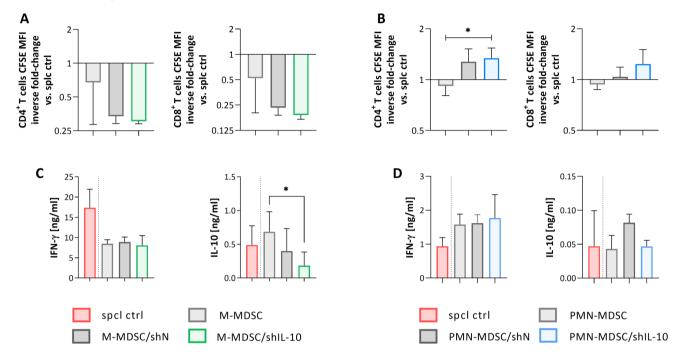


Figure S5. Expression of EGFP in MC38 tumors after triple injection of LVs. Tumors were dissected and analyzed using the flow cytometry method on the (A) 6th or (B) 10th day after triple injection of LVs or on the (C) 8th or (D) 12th day after intraperitoneal administration of CY followed by triple injection of LVs. Fold change of EGFP MFI was calculated in relation to untreated control [100% control value-(A) 128, 192, 203, (B) 146, 129, 223, (C) 340, 239, 170, (D) 215, 54, 225 for CD45<sup>neg</sup> cells, lymphoid cells and myeloid cells, respectively]. The number of mice per group was: (A and B) 7-10; (C) 3; and (D) 9-10. shN LVs, lentiviral vectors encoding shN sequence; shIL-10 LVs, lentiviral vectors encoding shIL-10 sequence; CY, cyclophosphamide; EGFP, enhanced green fluorescent protein; MFI, mean fluorescence intensity.

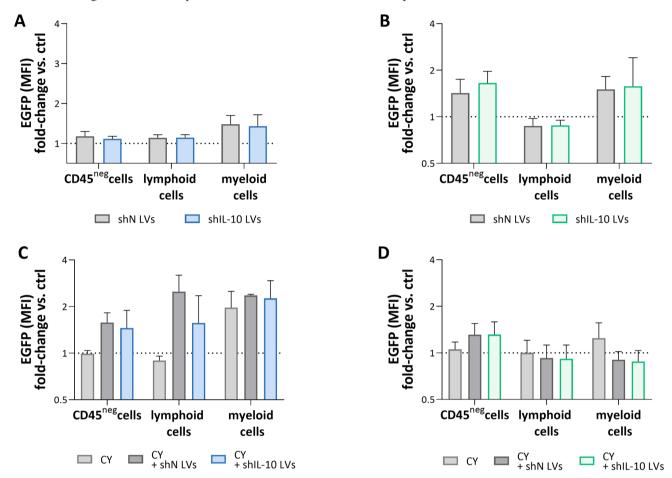


Figure S6. Scheme of multicolor flow cytometry analysis of myeloid cells infiltrating MC38 tumors. TAM, tumor-associated macrophages; DC, dendritic cells; M-MDSC, monocytic myeloid-derived suppressor cells; PMN-MDSC, polymorphonuclear myeloid-derived suppressor cells; Mf, macrophages.

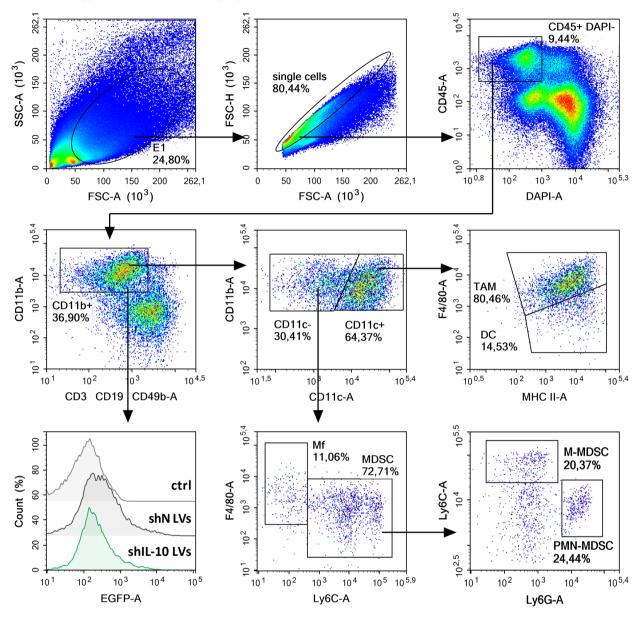


Figure S7. Scheme of multicolor flow cytometry analysis of lymphoid cells infiltrating MC38 tumors.

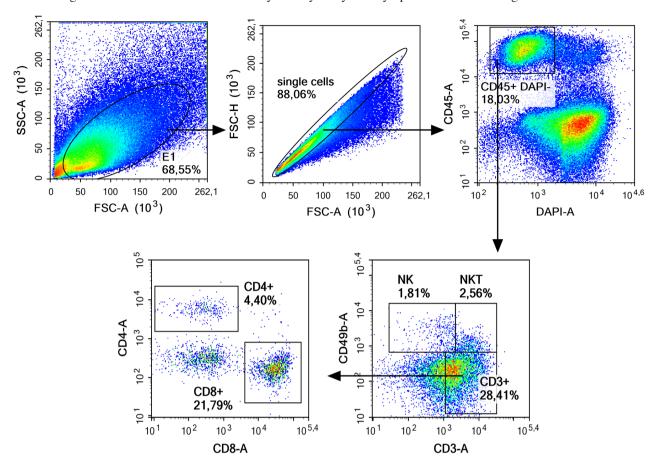


Figure S8. Scheme of multicolor flow cytometry analysis of CTLs in tLNs.

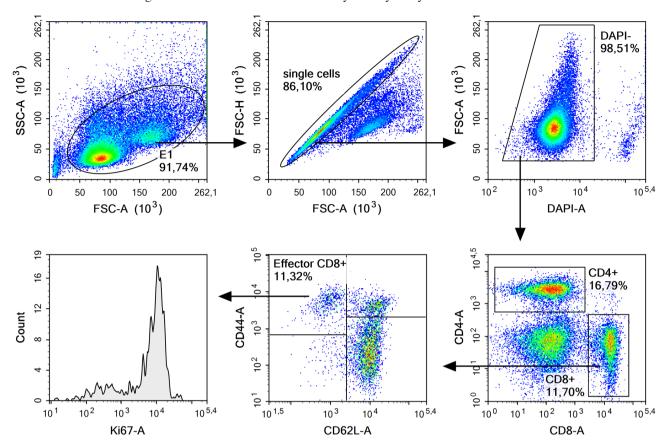


Figure S9. MHC II and CD86 expression on the surface of tumor-infiltrating myeloid cell subpopulations determined on the (A and C) 8th or (B and D) 12th day after intraperitoneal administration of CY followed by triple injections of LVs. The results are expressed as the mean ± SD. The number of mice per group in the experiment presented on A and C was 3 and in the experiment presented on B and D was 9-10. Statistical significance was calculated using (A, C and D) Welch's ANOVA followed by post hoc Dunnett's T3 multiple comparison test or (B) the non-parametric Kruskal-Wallis test followed by post hoc Dunn's multiple comparison test. Differences with a P<0.05 were regarded as significant (\*P<0.05, \*\*P<0.01, \*\*\*P<0.001, \*\*\*\*P<0.0001, \*\*\*\*P<0.0001, \*\*\*\*P<0.001, \*\*\*\*P<0.0001, \*\*\*

