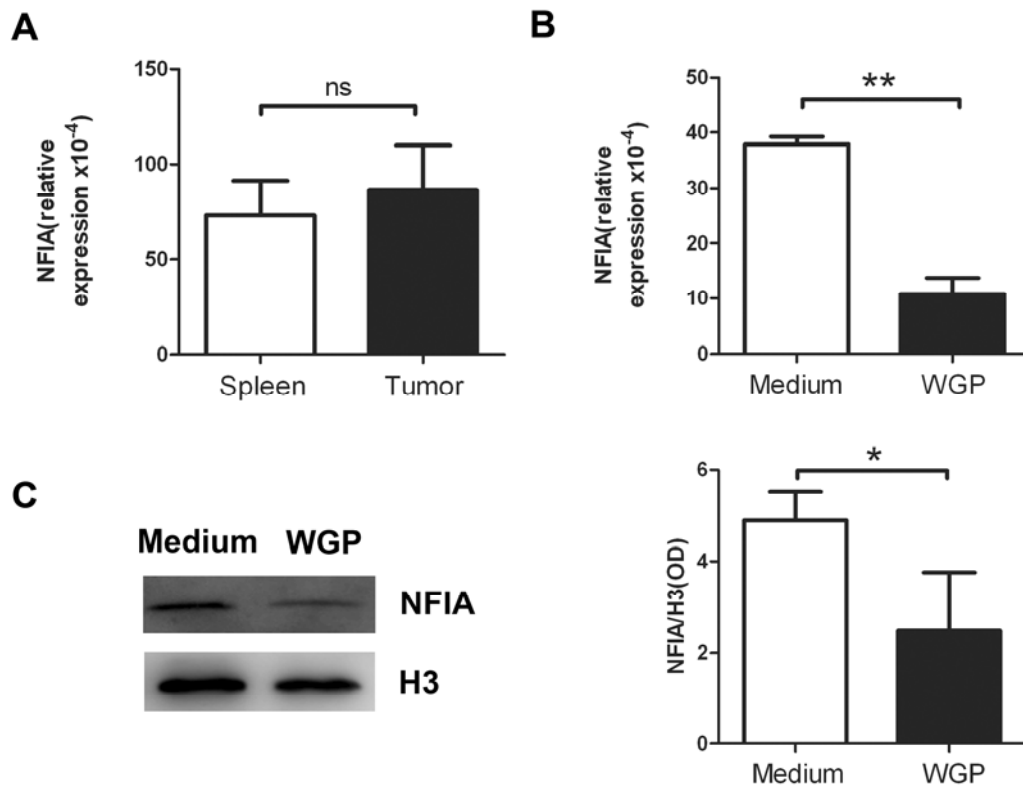


Particulate β -glucan regulates the immunosuppression of granulocytic myeloid-derived suppressor cells by inhibiting NFIA expression

Supplement Figure 1.



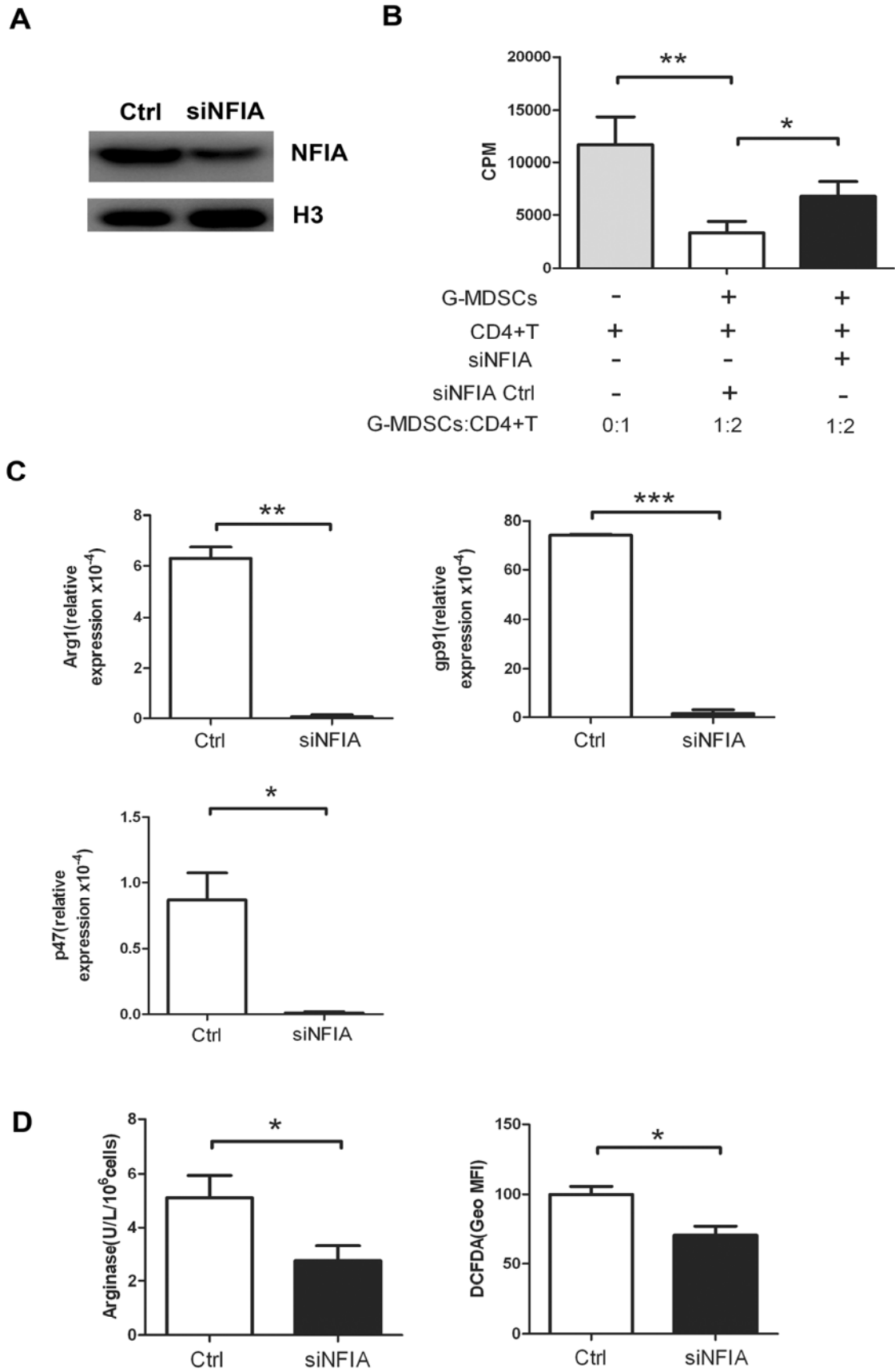
Supplement Figure 1. WGP inhibits the expression of NFIA in G-MDSCs isolated from tumor tissue of tumor-bearing mice.

A total of 3×10^6 Lewis lung carcinoma cells (LLCs) were injected s.c. into C57BL/6 mice. After 2 weeks, tumor tissue collected was prepared to the single-cell suspension and G-MDSCs were sorted. (A) Total RNA isolated from G-MDSCs derived from spleen and tumor tissue was subjected to qRT-PCR to measure NFIA expression. (B-C) Sorted G-MDSCs were cultured in the presence or absence of WGP (100 μ g/mL) for 24 h/48 h. (B) NFIA mRNA level in G-MDSCs was measured by qRT-PCR. (C)

Western-Blot analysis was developed with anti-NFIA antibody. Histone 3 served as a loading control. The amount of NFIA protein was calculated by gray scanning.

**p<0.01, *p<0.05, ns: no significance.

Supplement Figure 2.



Supplement Figure 2. NFIA knockdown decreases the suppressive capacity of G-MDSCs isolated from tumor tissue.

G-MDSCs sorted from tumor tissue of tumor-bearing mice were transfected with NFIA siRNA (100 nM). (A) Western blot analysis confirmed NFIA knockdown in G-MDSCs transfected with siNFIA. (B) G-MDSCs sorted from tumor tissue of tumor-bearing mice were transfected with NFIA siRNA (100 nM), then cells were harvested and cocultured with CD4⁺ T cells at different ratios in the presence of anti-CD3 mAb and anti-CD28 mAb for 72 h. ³H-thymidine incorporation was used to detect T-cell proliferation. (C) NADPH oxidase complex which consists of phox p47 and gp91 appears to be a major source of ROS. Here, p47 and gp91 were chosen to represent the ROS production. Arg1 and gp91/p47 mRNA levels in G-MDSCs transfected with siNFIA were detected by qRT-PCR. (D) Arg1 activity of G-MDSCs transfected with siNFIA was measured. Production of ROS in G-MDSCs was analyzed by flow cytometry. ***p<0.001, **p<0.01, *p<0.05, Geo MFI: geometric mean fluorescent intensity.