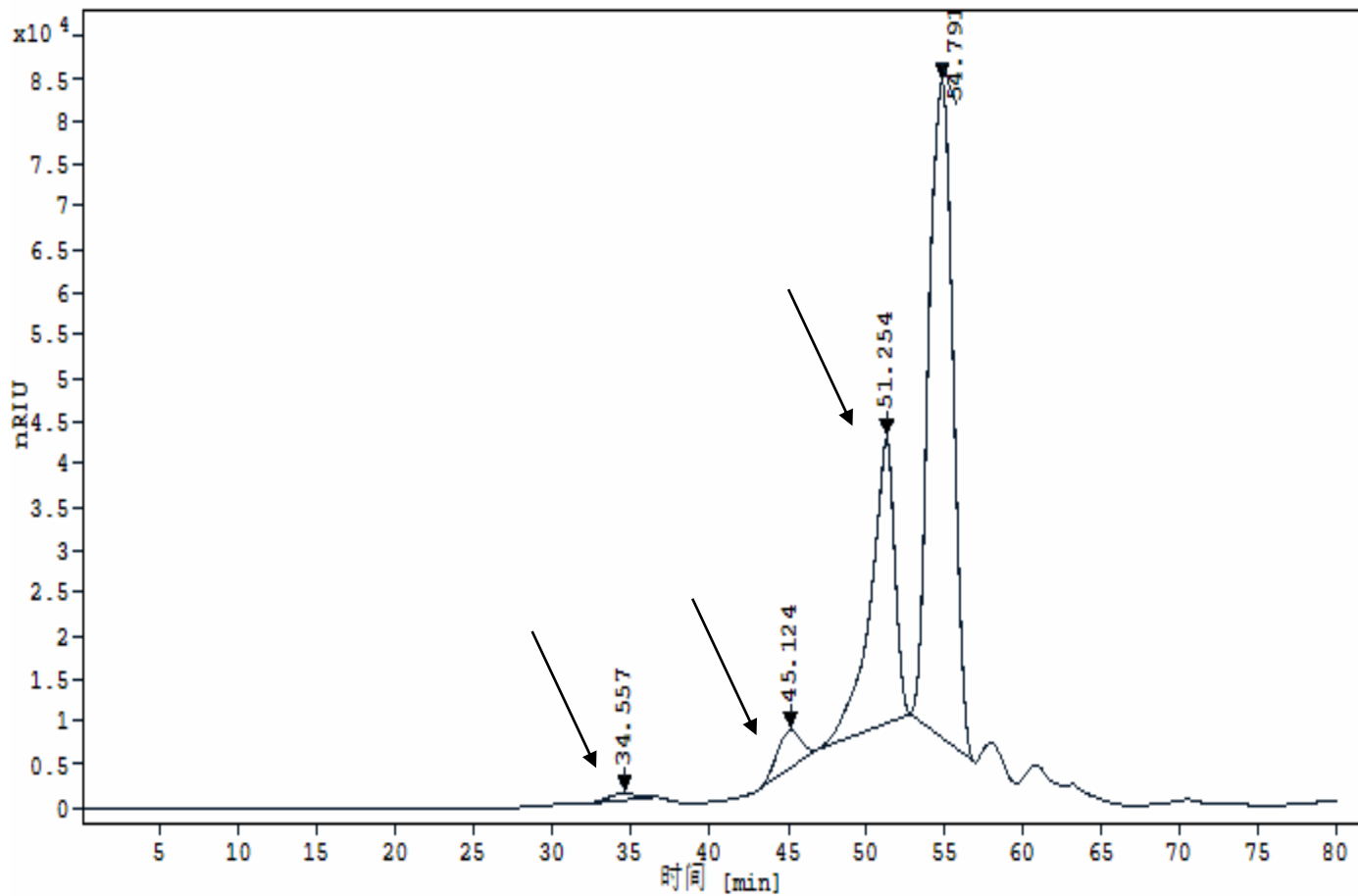


Supplementary Figure S1. Inhibition of tumor growth.

The second day after 4T1 cells implantation into mice, the mice were given intraperitoneal injections of GFI (100 mg/kg/per mouse) or 0.9% saline once every other day up to 32 days.

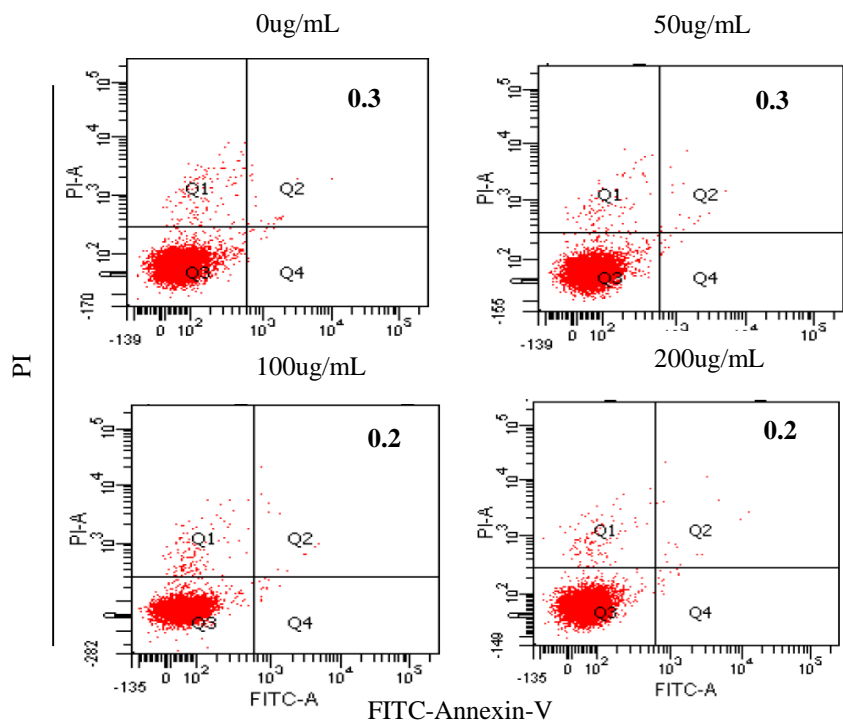
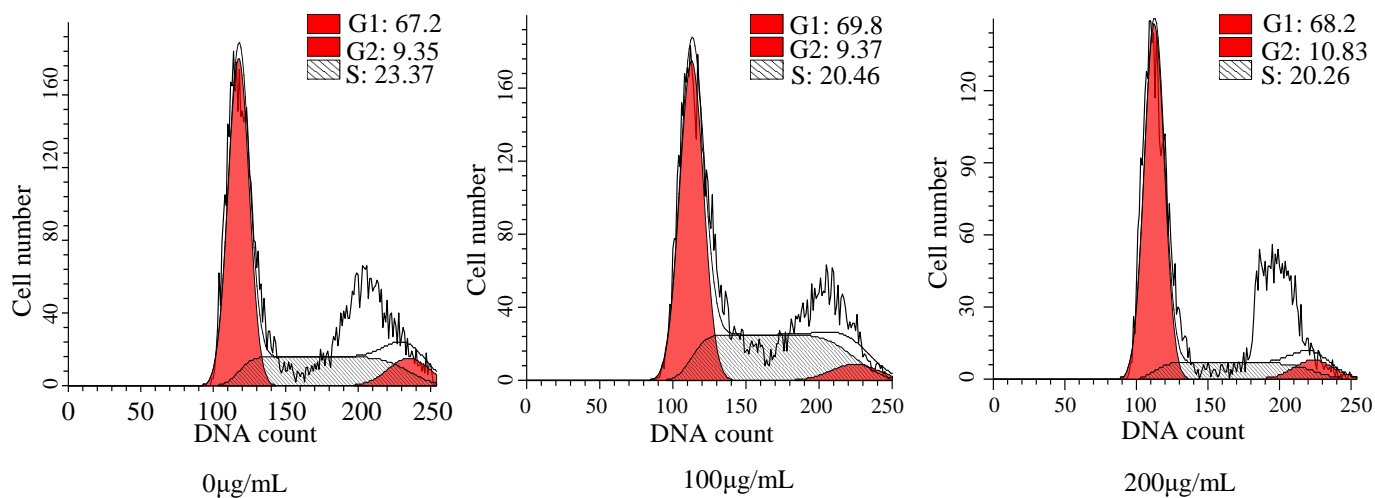
- (A) The tumor volume of mice in each group of the 4T1 model were measured and calculated at the indicated time points (n = 10).
- (B) Left: Representative photos of tumor; Right: the tumor weight of mice were measured and calculated at the end of assay (n = 10).
- (C) Left: Representative photos of spleen; Right: the spleen weight of mice were measured and calculated at the end of assay (n = 10). *p<0.05; **p<0.01; ##p<0.01. Error bars, SD.

GFI① - 2022-10-09 18-17-18+08-00-01.dx,RID1A

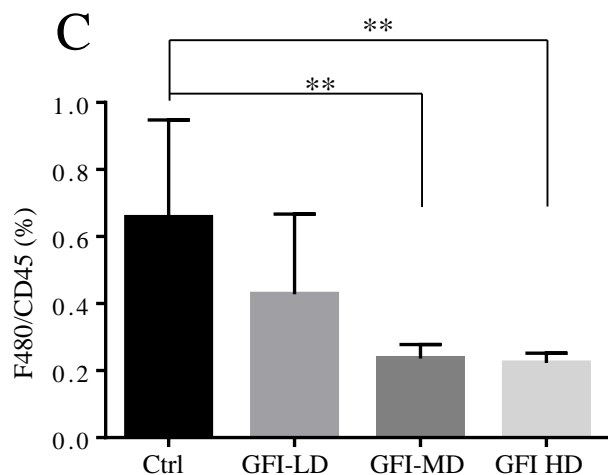
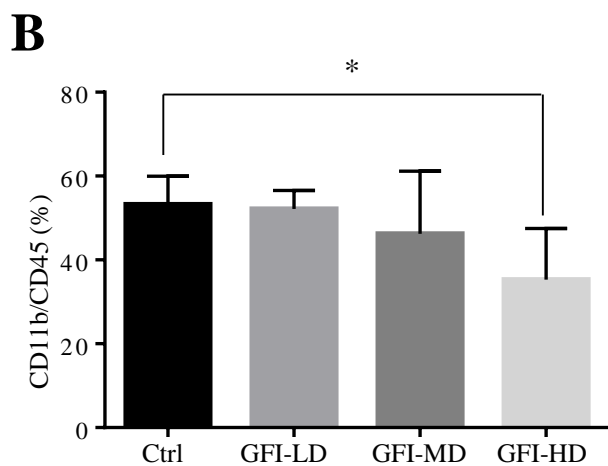
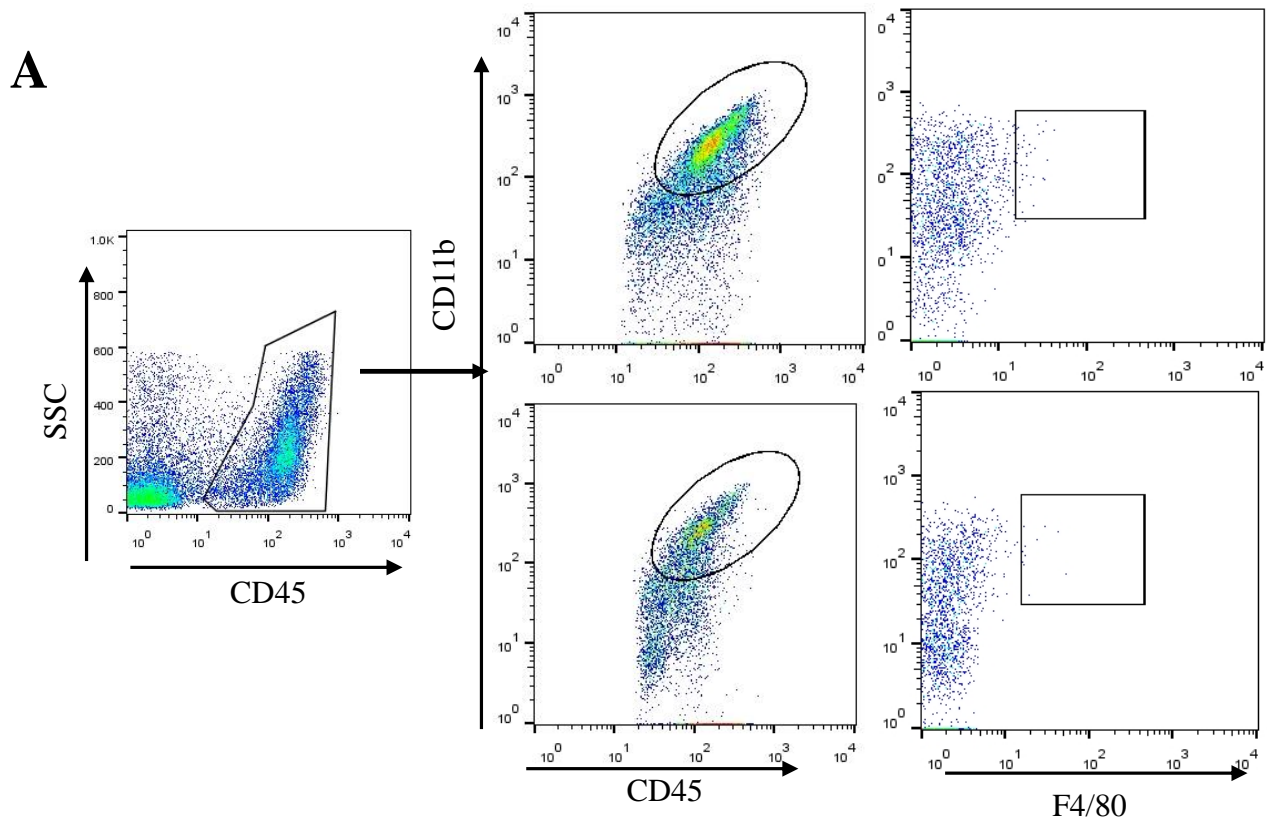


Supplementary Figure S2. Purification of bioactive components.

Chromatogram of the polysaccharides from the fruit bodies of *Grifola frondosa* sample

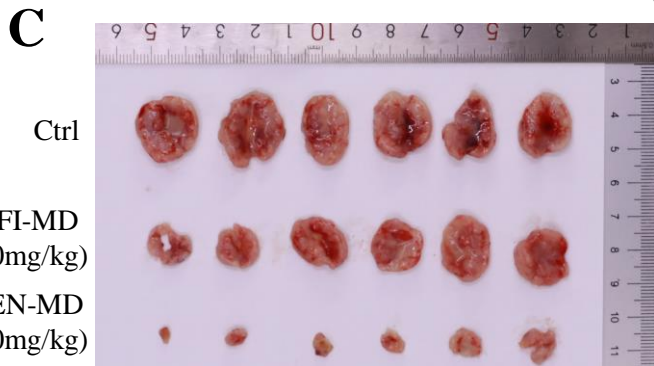
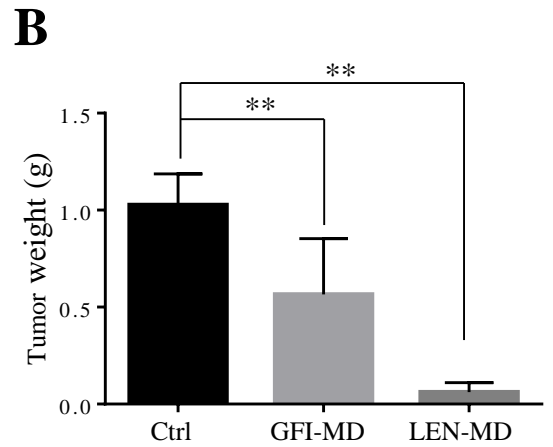
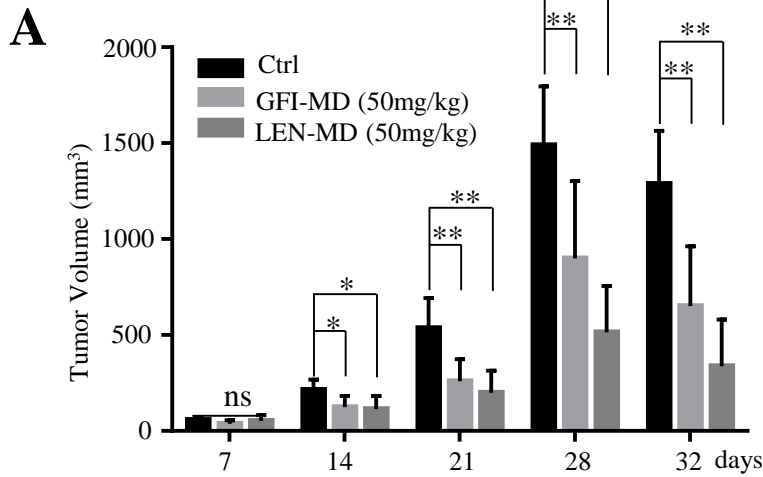
A**B****Supplementary Figure S3. Tumor cell flow cytometry.**

- (A) Representative FACS plots of cell apoptosis.
- (B) Histogram of cell cycle.

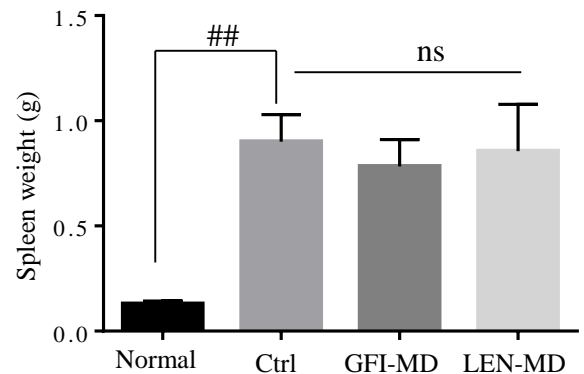
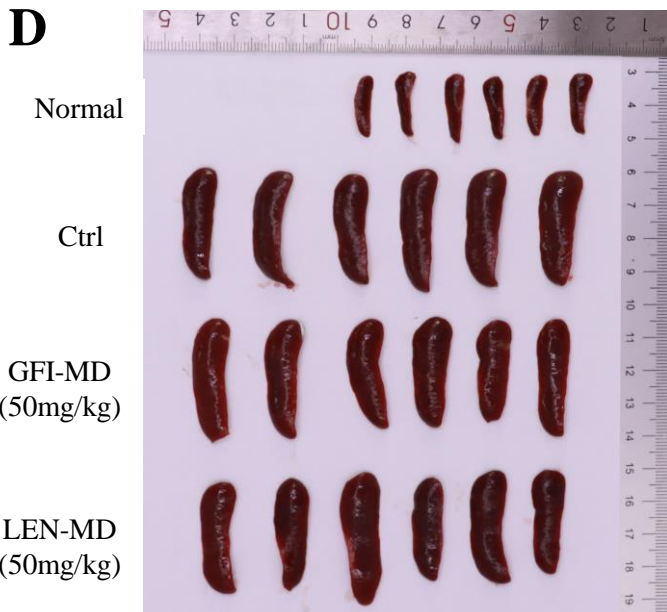


Supplementary Figure S4. Tumor cell flow cytometry.

- (A) Representative FACS plots illustrate the gating to assess F4/80 cells.
- (B) The alteration of CD11b⁺ myeloid cells in the tumor from the control or GFI-treated 4T1 murine model using FACS analysis.
- (C) The alteration of CD11b⁺ F4/80⁺ TAMs in the tumor from the control or GFI-treated 4T1 murine model using FACS analysis. *p<0.05; **p<0.01. Error bars, SD.



Sample name	Tumor weight (means ± SD, g)	Tumor-inhibition rate(means ± SEM, %)
Ctrl	1.026 ± 0.16	--
GFI-MD (50mg/kg)	0.566 ± 0.287**	44.83 ± 11.57
LEN-MD (50mg/kg)	0.061 ± 0.047****	93.99 ± 1.90



Supplementary Figure S5. Compared the antitumor effect of polysaccharide from *G. frondosa* and *L. edodes* in vivo

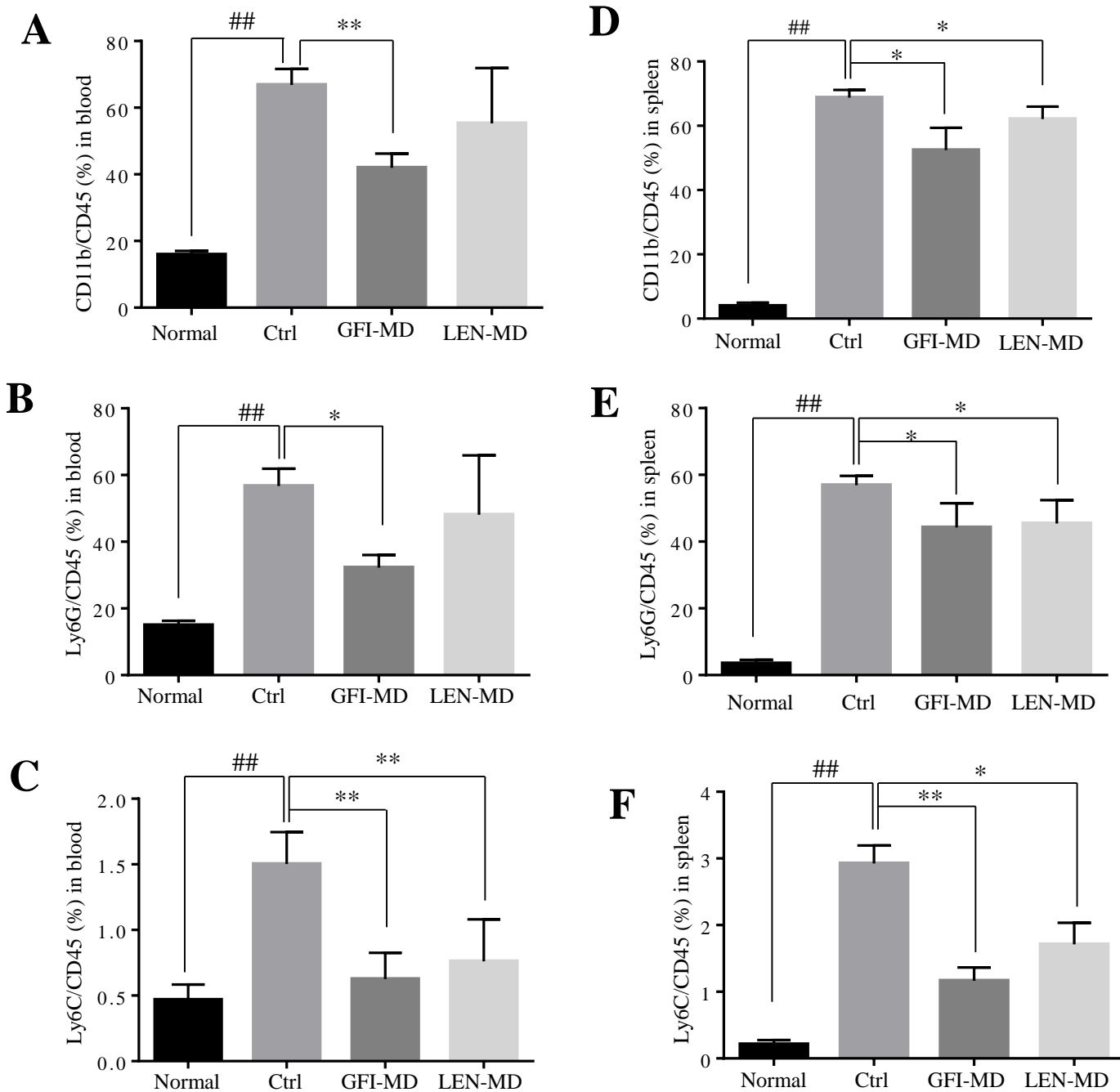
At one week tumor inoculation after 4T1 cells (0.5×10^5 cells/mouse) implantation into mice, the mice were given intraperitoneal injections of GFI (50mg/kg, GFI-MD), or lentinan (50mg/kg, LEN-MD) once every other day up to 35-day, with 100 μ L. Control group were given intraperitoneal injections of 0.9% saline.

(A) The tumor volumes of the mice were measured and calculated weekly (n = 6).

(B) The body weight of mice was weighed and calculated weekly (n = 6).

(C) Left: representative photos showing the effect of GFI on tumor growth inhibition in the 4T1 murine model (n = 6); Right: shown tumor weight and tumor-inhibition rate (means ± SEM) (n = 6).

(D) Left: representative photos of spleen in each group (n = 6); Right: shown spleen weight (n = 6). *p<0.05; **p<0.01. Error bars (no specifically indicated), SD.



Supplementary Figure S6. Polysaccharide from *G. frondosa* and *L. edodes* suppresses the enrichment of MDSCs in the peripheral blood and spleen.

After the mice were sacrificed, MDSCs were analyzed with cell surface markers (CD45⁺CD11b⁺) in peripheral blood and spleen from the control, or GFI treatment group, or LEN treatment group in 4T1 murine model using flow cytometry (n = 4).

- (A) GFI treatment significantly decreased CD11b⁺MDSCs enrichment in the peripheral blood as observed by using flow cytometry analysis; LEN treatment also reduced the percentage of CD11b⁺MDSCs in the peripheral blood, but not significantly.
- (B) Flow cytometry analyzed the density of PMN-MDSCs and M-MDSCs in the peripheral blood from the control or GFI-treated 4T1 murine model, staining with CD45, CD11b, Ly6G and Ly6C antibodies. GFI-MD treatment significantly decreased the percentage of PMN-MDSCs, but LEN-MD treatment did not.
- (C) GFI-MD and LEN-MD treatment significantly decreased the percentage of M-MDSCs in the peripheral blood of 4T1 murine model.
- (D) GFI treatment and LEN treatment significantly decreased CD11b⁺MDSCs enrichment in the spleen of 4T1 murine model.
- (E) GFI-MD and LEN-MD treatment significantly reduced the percentage of PMN-MDSCs in the spleen of 4T1 murine model.
- (F) GFI-MD and LEN-MD treatment significantly decreased the percentage of M-MDSCs in the spleen of 4T1 murine model.

*p<0.05; *p<0.01; #p<0.05; ##p<0.01; Error bars, SD.