

Supplementary Information

Methods for determining the composition of jujube powder

Total sugar¹

Total sugar was determined using the phenol–sulfuric acid method. Jujube powder was suspended and diluted in deionized water. 1 mL of the sample suspension was mixed with 0.5 mL of 5% phenol solution and 2.5 mL of concentrated sulfuric acid. The mixture was then cooled and incubated at room temperature for 20 min. Afterward, 200 μ L of the sample was transferred to a 96-well plate. Absorbance was measured at 490 nm and compared to the standard curve of the D-glucose solution.

Reducing sugar²

Reducing sugar was determined using the 3,5-dinitrosalicylic acid (DNS) method. 2 mL of the diluted sample was combined with 1.5 mL of DNS reagent, mixed, and incubated in a boiling water bath for 5 min. After cooling to room temperature, absorbance was measured at 540 nm and compared to the standard curve of the D-glucose solution.

Uronic acid³

Uronic acid was measured using the 3-phenylphenol method. 1 mL of the sample was combined with 6 mL of sodium tetraborate in concentrated sulfuric acid (0.0125 mol/L) and incubated in a boiling water bath for 5 min. After cooling to room temperature, 100 μ L of 3-phenylphenol in 5 mg/mL sodium hydroxide solution (1.5 mg/mL) was added. The mixture was shaken for 5 min. Absorbance was read at 520 nm and then compared to the standard curve of the D-galacturonic acid solution.

Total phenol⁴

Total phenolic content was determined using the Folin-Ciocalteu method. 70 μ L of the sample was mixed with 300 μ L of Folin & Ciocalteu's phenol reagent and 230 μ L of 7.5% sodium carbonate solution. The mixture was incubated at room temperature for 1.5 h, and absorbance was read at 765 nm and compared to the standard curve of the gallic acid.

Total flavonoids⁵

2 mL of the sample was combined with 0.75 mL of 5% sodium nitrite solution. After 5 min of incubation, 0.5 mL of 10% aluminum nitrate was added, followed by an additional 6 min incubation. Then, 4 mL of 5% sodium hydroxide solution was added. The final mixture was diluted with deionized water to a total volume of 25 mL. Absorbance was read at 510 nm and compared to the standard curve of rutin to calculate total flavonoid content.

Protein⁶

The protein content was measured using the Bradford method. 20 μ L of the sample was mixed with 200 μ L of Coomassie brilliant blue G250 solution in a 96-well plate and incubated at room temperature for 5 min. Absorbance was read at 595 nm and compared to the standard curve of bovine serum albumin.

Fig. S1 Preparation procedure for jujube powder.

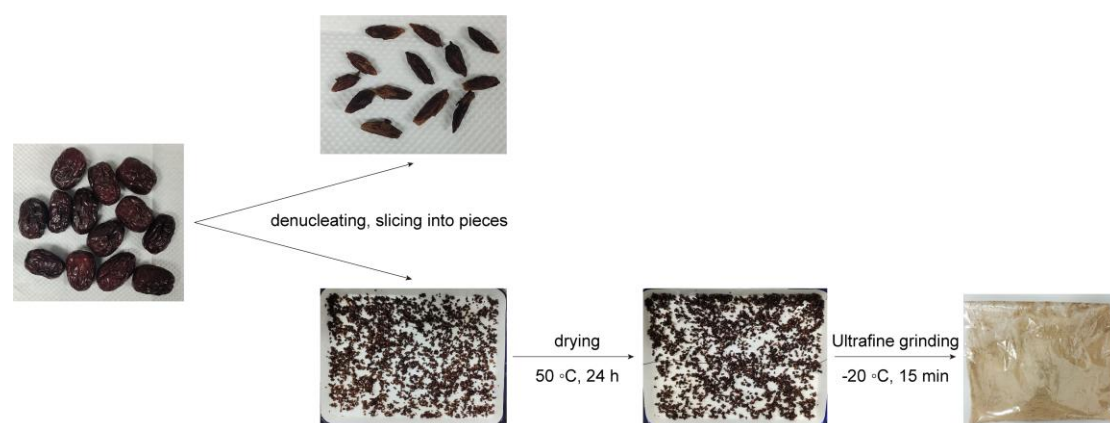


Table S1 Antibodies used in flow cytometry assays.

Target	Fluorochrome	Clone	Manufacturer
anti-mouse CD16/32	-	S17011E	Biolegend
Live-Dead dye	Zombie UV	-	Biolegend
CD45	APC/Fire 750	30-F11	Biolegend
CD19	APC/Fire 810	6D5	Biolegend
CD3	Spark Blue 550	17A2	Biolegend
CD4	FITC	RM4-5	Biolegend
CD8a	Spark PLUS UV 395	53-6.7	Biolegend
CD25	BV650	PC61	Biolegend
Foxp3	PE	MF-14	Biolegend
IL17A	APC	TC11-18H10.1	Biolegend
IFN γ	BV421	XMG1.2	Biolegend
IL4	PE/Cyanine7	11B11	Biolegend
CD11b	PE/Fire 640	M1/70	Biolegend
CD11c	BV711	N418	Biolegend
MHC II	PE/Dazzle 594	M5/114.15.2	Biolegend
Ly6G	bv785	1A8	Biolegend
Ly6C	BV605	HK1.4	Biolegend
F4	PE/Fire 810	BM8	Biolegend

Fig. S2 The relative abundance of Firmicutes and Bacteroidota.

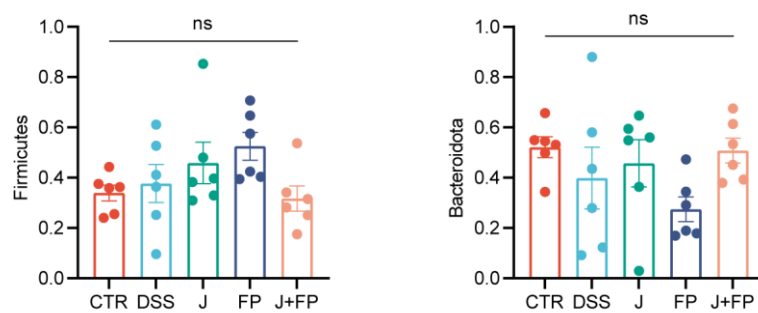


Fig. S3 Gating strategy of flow cytometry assays on innate immune cells.

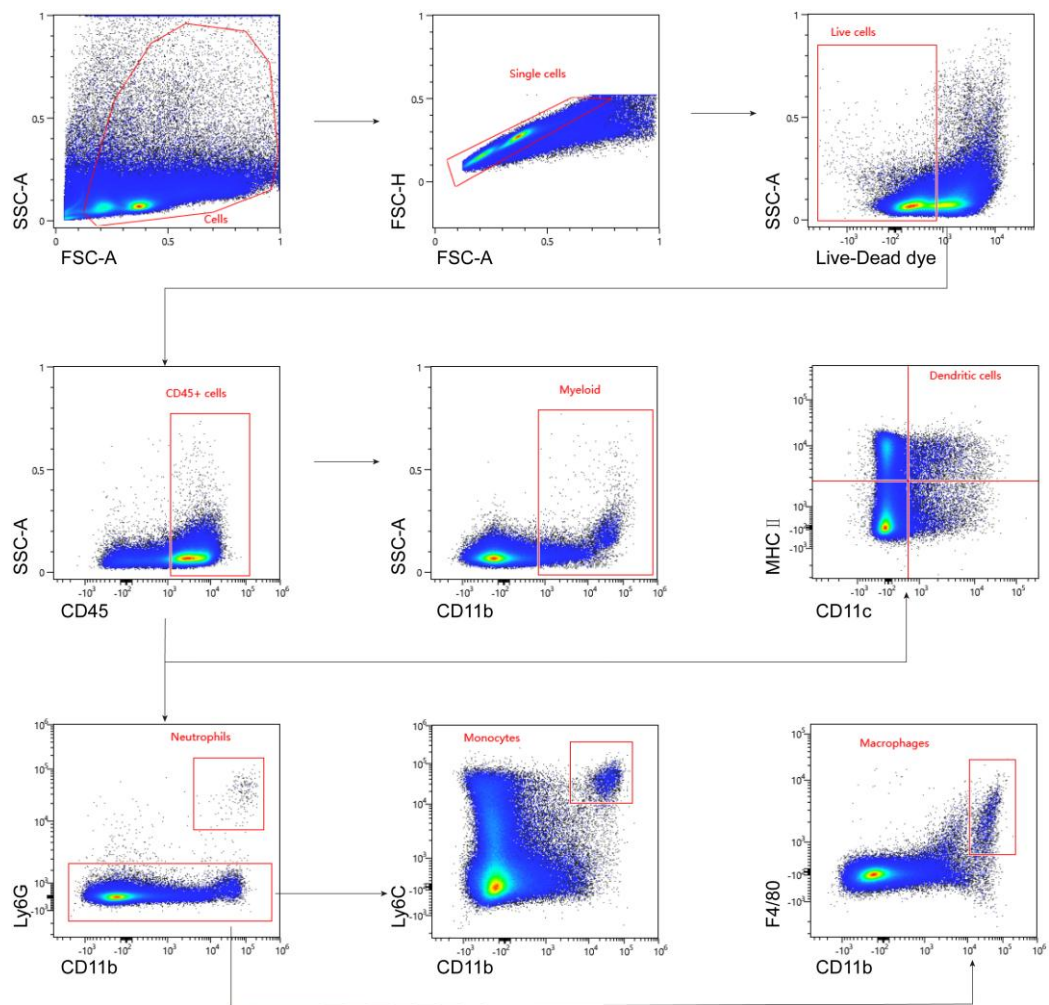


Fig. S4 Gating strategy of flow cytometry assays on adaptive immune cells.

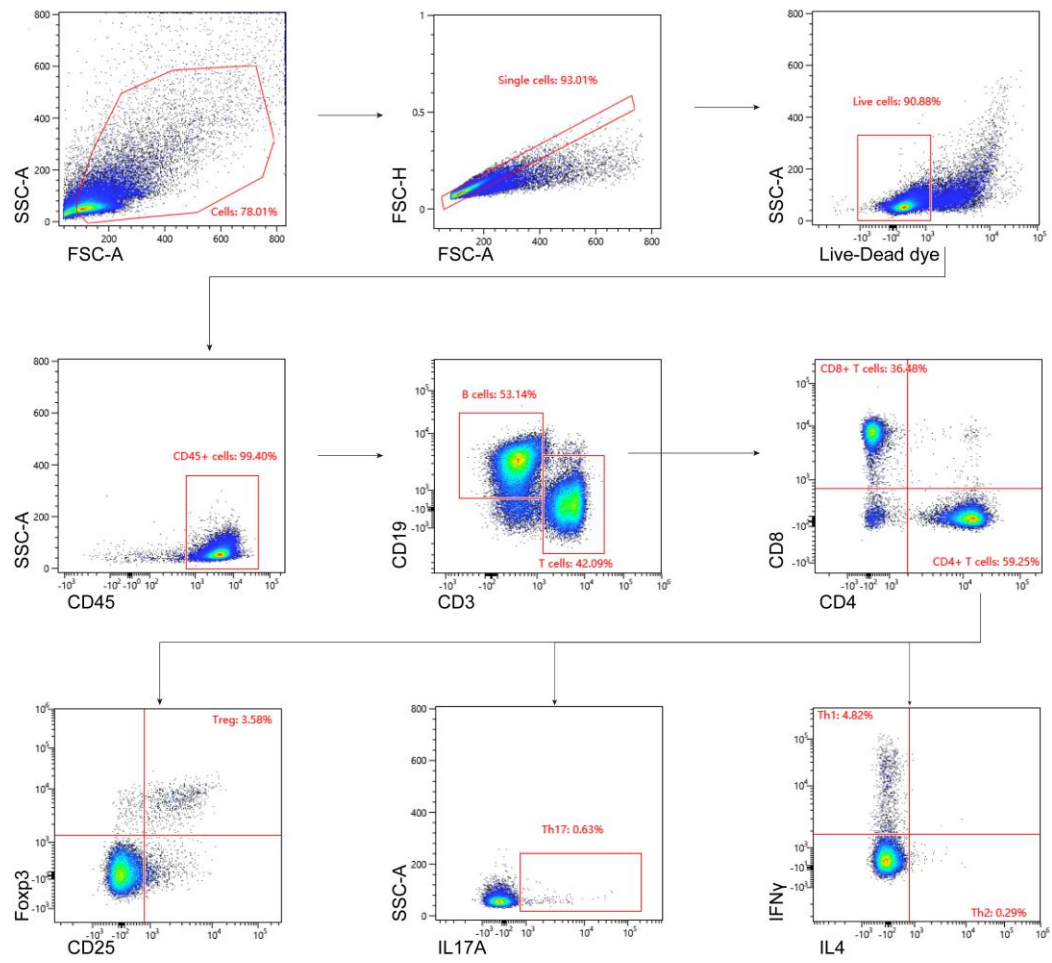
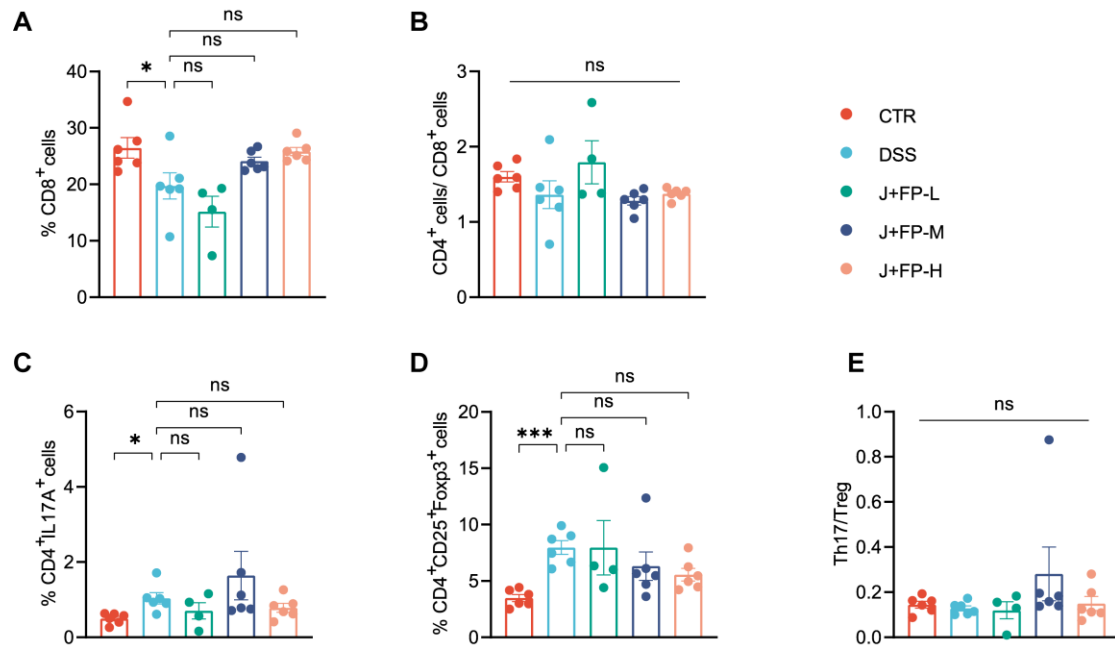
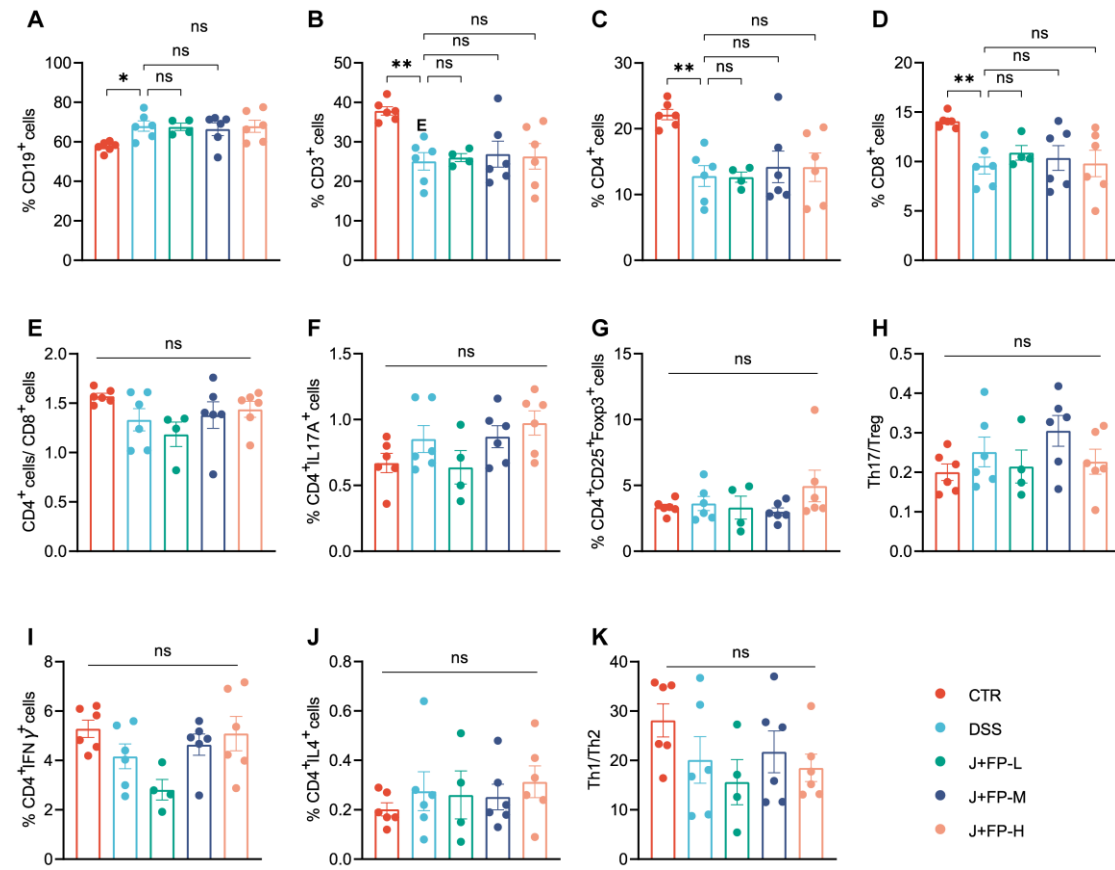


Fig. S5 Statistical results of flow cytometry assays on adaptive immune cells in the mesenteric lymph nodes.



(A) CD8⁺ T cells, (B) the ratio of CD4⁺ T cells to CD8⁺ T cells, (C) Th17 cells, (D) Tregs, and (E) the ratio of Th17 cells to Tregs in the mesenteric lymph nodes. Statistical analyses of CD8⁺ T cells were based on the proportion of cells to CD45⁺ T cells. Statistical analyses of Th17 and Treg cells were based on the proportion of cells to CD4⁺ T cells. ns, no significance, $p > 0.05$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$.

Fig. S6 Statistical results of flow cytometry assays on adaptive immune cells in the spleen.



(A) B cells, (B) T cells, (C) CD4⁺ T cells, (D) CD8⁺ T cells, (E) the ratio of CD4⁺ T cells to CD8⁺ T cells, (F) Th17 cells, (G) Tregs, (H) the ratio of Th17 cells to Tregs, (I) Th1 cells, (J) Th2 cells, and (K) the ratio of Th1 cells to Th2 cells in the spleen. Statistical analyses of B cells, T cells, CD4⁺ T cells, and CD8⁺ T cells were based on the proportion of cells to CD45⁺ T cells. Statistical analyses of Th17, Tregs, Th1 cells, and Th2 cells were based on the proportion of cells to CD4⁺ T cells. ns, no significance, $p > 0.05$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$.

Reference

1. M. Dubois, K. Gilles, J. Hamilton, P. Rebers and F. Smith, A Colorimetric Method for the Determination of Sugars, *Nature*, 1951, **168**, 167-167.
2. T. K. Ghose, Measurement of cellulase activities, *Pure Appl. Chem.*, 1987, **59**, 257-268.
3. I. Meseguer, V. Aguilar, M. J. Gonzalez and C. Martinez, Extraction and colorimetric quantification of uronic acids of the pectic fraction in fruit and vegetables, *J. Food Compost. Anal.*, 1998, **11**, 285-291.
4. V. L. Singleton and A. R. J. Joseph, Colorimetry of Total Phenolics with Phosphomolybdic-Phosphotungstic Acid Reagents, *Am. J. Enol. Vitic.*, 1965, **16**, 144-158.
5. Z. Jia, M. Tang and J. Wu, The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals, *Food Chem.*, 1999, **64**, 555-559.
6. M. M. Bradford, A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding, *Anal. Biochem.*, 1976, **72**, 248-254.