Supplementary data

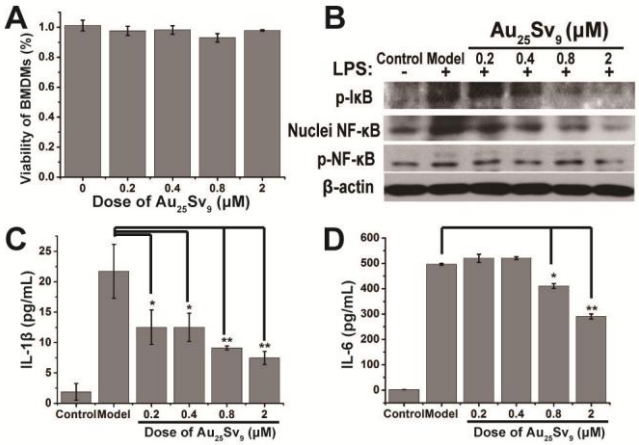

Fig. S1. Effects of $Au_{25}Sv_9$ on LPS-induced expression of inflammatory cytokines and signaling pathways in BMDMs. (A) Cell viability of BMMs in the presence of different dose of $Au_{25}Sv_9$ was measured using the CCK-8 assay, the data is presented as mean ± Standard deviation of triplicate experiments. (B) The LPS-induced activation of NF-κB in the presence or absence of $Au_{25}Sv_9$ was detected by Western blotting (phosphorylation of IκB and nuclear translocation of NF-κB), β-actin was used as the loading control. Data was from two independent experiments and one representative result is shown here. (C and D) The LPS-induced secretion of IL-1β and IL-6 in BMMs in the presence or absence of $Au_{25}Sv_9$ was detected by ELISA. The data is presented as mean ± Standard deviation of triplicate experiments, *P < 0.05, **P < 0.01, compared to the LPS only group.

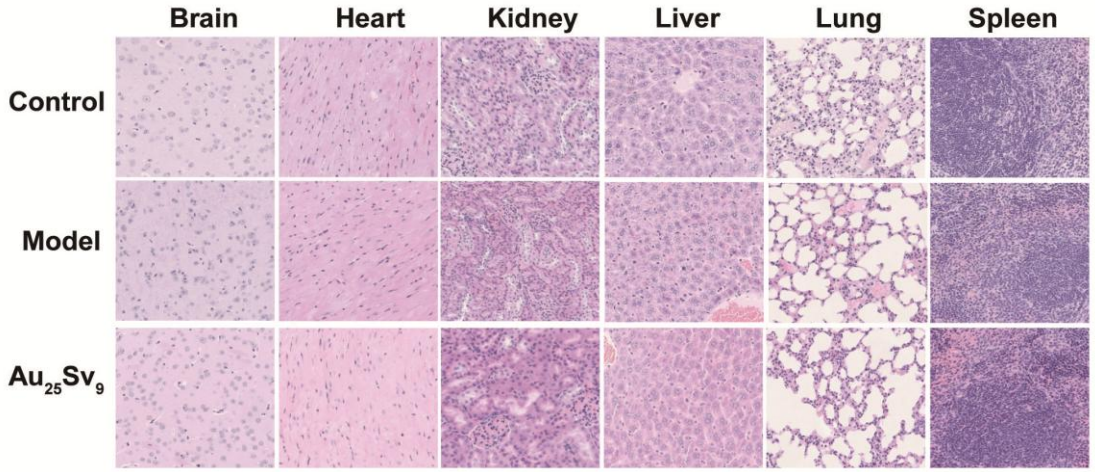

Fig. S2. Representative histopathological images of main organs from each group of CIA mice treated with vehicle or $Au_{25}Sv_9$, and normal mice.

Table S1. Effect of $Au_{25}Sv_9$ cluster on hematology index of mice

Index	**Control**	**$Au_{25}Sv_9$**
WBC（10^9/L）	4.02 ±1.88	4.26 ±1.76
RBC（10^12/L）	12.16 ±0.34	12.12 ±0.81
HGB（g/L）	178.60 ±3.21	174.80 ±9.14
HCT（%）	55.34 ±1.52	54.70 ±3.12
MCV（fL）	45.56 ±0.50	45.24 ±0.45
MCH（pg)	14.70 ±0.21	14.46 ±0.23
MCHC（g/L）	322.80 ±3.70	319.60 ±3.13

Table S2. Effect of $Au_{25}Sv_9$ cluster on biochemistry index of mice

Index	**Control**	**$Au_{25}Sv_9$**
ALT(U/L)	30 ±3	29 ±4
AST(U/L)	129 ±37	154 ±63
TP(g/L)	59 ±1	56 ±2
ALB(g/L)	22 ±0.5	21 ±1
ALP(U/L)	183 ±17	151 ±23
UREA(mM)	9 ±3	8 ±2
CREA(μM)	9 ±2	8 ±2

Supplementary method

The bone marrow-derived macrophages (BMDMs) were isolated from the tibiae and femora of 4-weeks old C57/BL6 mice as described previously [1, 2]. The isolated cells were collected by centrifugation (1200 rpm), and then treated with red blood cell lysis buffer (Beyotime, Haimen, China) for 5 min on room temperature. Next, the primary bone marrow cells were harvested by centrifugation (1200 rpm) and washed

once with α-minimum essential medium (α-MEM; Hyclone Laboratories, Logan, UT, USA). The purified cells were suspended in α-MEM medium containing 10% fetal bovine serum (FBS), 1% penicillin, 1% streptomycin, and supplemented with 30ng/mL M-CSF (R&D Systems, Minneapolis, MN, USA), and cultured for 3 days to obtain BMDMs. To investigate the effects of the gold cluster on LPS-induced activation of NF-κB in naïve macrophages, BMDMs were treated with LPS (1 μg/mL) for 24 h after treatment for 1 h with or without the $Au_{25}Sv_9$ clusters, and the activation of NF-κB was detected by Western bloting. The secretion of pro-inflammatory cytokines was determined by ELISA assay.

References

[1] Han SB, Lee JK. Anti-inflammatory effect of Trichostatin-A on murine bone marrow-derived macrophages. Arch Pharm Res. 2009;32:613-24.

[2] Suzuki E, Sugiyama C, Umezawa K. Inhibition of inflammatory mediator secretion by (-)-DHMEQ in mouse bone marrow-derived macrophages. Biomedicine & pharmacotherapy = Biomedecine & pharmacotherapie. 2009;63:351-8.