Supplemental information

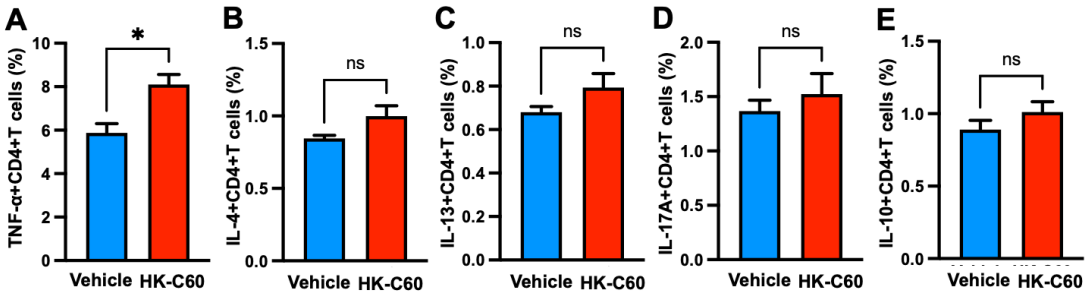
Supplementary Fig. 1. Cytokine production in antigen-dependently stimulated CD4+ T cells in co-culture with TPMs

A-E) The splenocytes were prepared from OT-II mice, then CD4+ T cells were isolated from the cell suspension by using magnetic sorting. The CD4+ T cells ($2.0x10^6$/mL) were cultured with TPMs ($1.0x10^7$/mL) the presence of $OVA_{323-339}$ peptide (100 ng/mL). The culture was further treated with HK-C60 ($5.0x10^8$ CFU/mL) or vehicle control (PBS) and incubated at 37℃ for 24 h. The cytokine producing cells in CD3+CD4+gate was analyzed by flow cytometry. The cumulative data were shown as mean +/- SEM of five samples in two independent experiments. Value of $*p < 0.05$ was regarded as significance. ns; not-significant.

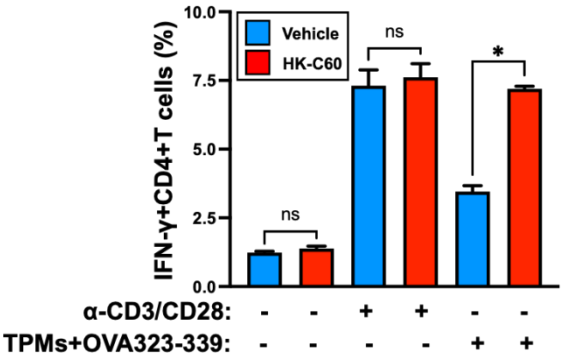
Supplementary Fig. 2. HK-C60 enhances CD4+ T cells activity with antigen presentation from macrophages

The splenic CD4+ T cells were isolated from OT-II mice by magnetic sorting. The CD4+ T cells ($2.0x10^6$/mL) were used for the stimulation with anti-CD3/CD28 microbeads ($4.0x10^5$/mL), co-culture with TPMs ($1.0x10^7$/mL) with $OVA_{323-339}$ peptide (100 ng/mL) or vehicle control (PBS). The cultures were further treated with HK-C60 ($5.0x10^8$/mL) or vehicle control (PBS). The cultures were incubated at 37℃ for 24 h, then IFN-γ+CD4+ T cells were analyzed by flow cytometry. IFN-γ production was detected in CD3+CD4+gate in the flow cytometry analysis. The cumulative data were shown as mean +/- SEM of three samples in three independent experiments. Value of $*p < 0.05$ was regarded as significance. ns; not-significant.

Supplementary Fig. 1



Supplementary Fig. 2



Supplemental protocol

In vitro CD4+ T cells stimulation

Splenic CD4+ T cells were isolated from OT-II mice. The CD4+ T cells ($2.0x10^6$/mL) were seeded in 96 well plate, then treated with Dynabeads™ Mouse T-Activator CD3/CD28 for T-Cell Expansion and Activation ($5.0x10^4$/mL, Thermo Fisher Scientific) or vehicle control (PBS). The cultures were further treated with HK-C60 ($5.0x10^8$/mL) or vehicle control (PBS). After incubation at 37℃ for 24 h, the IFN-γ production in CD4+ T cells were analyzed by flow cytometry.