Supporting Information

A shikonin-based self-assembled nanomedicine alleviates DSS-induced colitis involving HDC-associated histamine regulation and gut microbiota modulation

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Table S1.Disease Activity Index (DAI) score

Score	weight loss (%)	Stoll consistency	Bleeding
0	0	Normal	Normal
1	1-5	-	-
2	5-10	Mucoid stools	Moderate bleeding
3	10-20	-	-
4	>20	Diarrhea	Gross bleeding

Table S2.Primer sequences

Primer	FORWARD	REVERSE
β-actin	TGGCACCACACCTTCTACAATGAG	GAGGCATACAGGGACAGCACAG
TNF-α	ACGTGGAACTGGCAGAAGAGG	TGAGAAGAGGCTGAGACATAGGC
IL-1β	TCGCAGCAGCACATCAACAAG	TCCACGGGAAAGACACAGGTAG
IL-6	CGGAGAGGAGACTTCACAGAGG	TTCCACGATTTCCCAGAGAACATG
HDC	TCTACCTCCGACATGCCAACTC	CCGAAGGACCGAATCACAAACC
PKM2	TGCGGTGGCTCTGGATACAAAG	ACAGGATGTTCTCGTCACACTTCTC

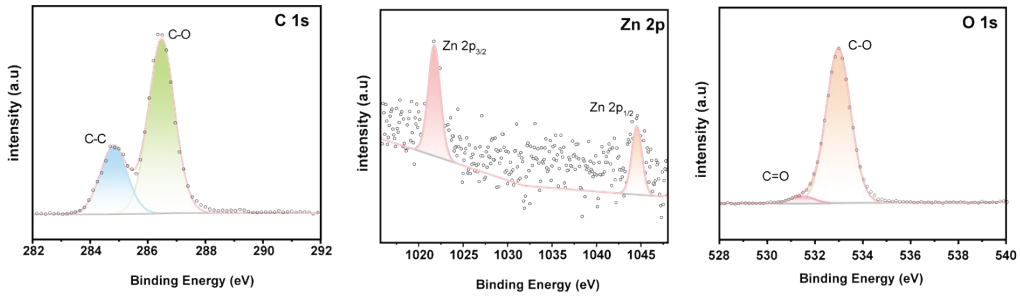

Figure S1. High-resolution XPS spectra of Zn-SHK-PEG NPs.

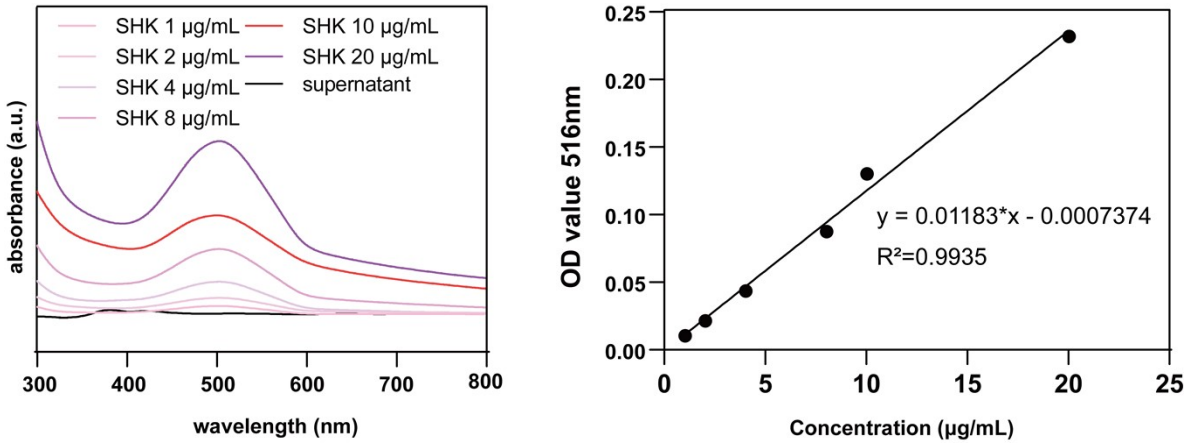

Figure S2. UV-Vis spectra and calibration curve of SHK at 516 nm for quantification of free SHK in the nanoparticle supernatant.

Sample	Zn Concentration	RSD
1 mg/mL shikonin-containing nanoparticle solution	207.57 mg/L	0.39%

Table S3. ICP-OES instrumental parameters used for zinc ion concentration analysis.

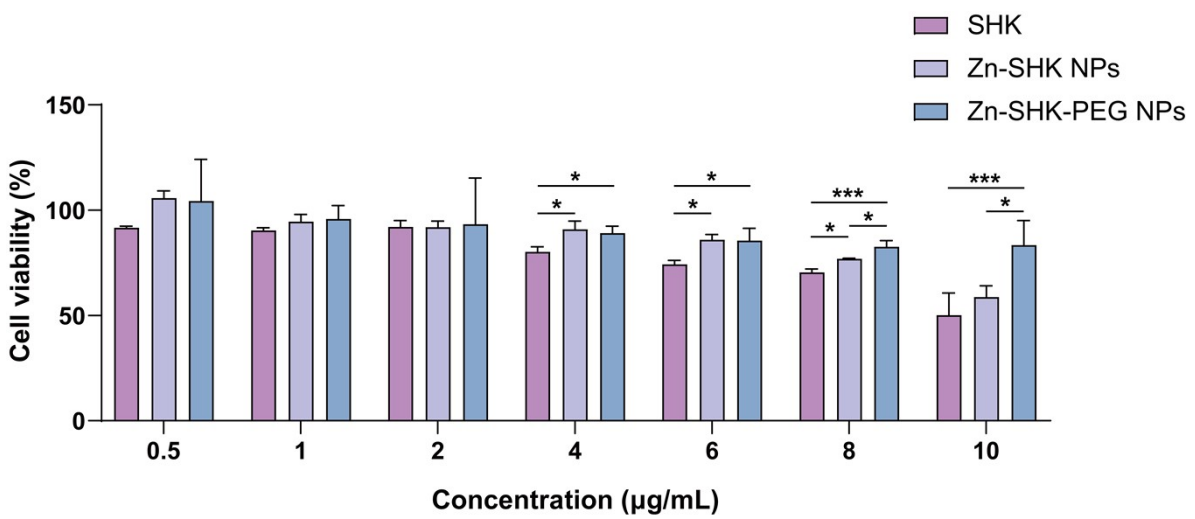

Figure S3. Cytotoxicity assays of SHK, Zn-SHK NPs and Zn-SHK-PEG NPs at different concentrations (at equivalent shikonin concentration) on Caco-2 cells, n=3.

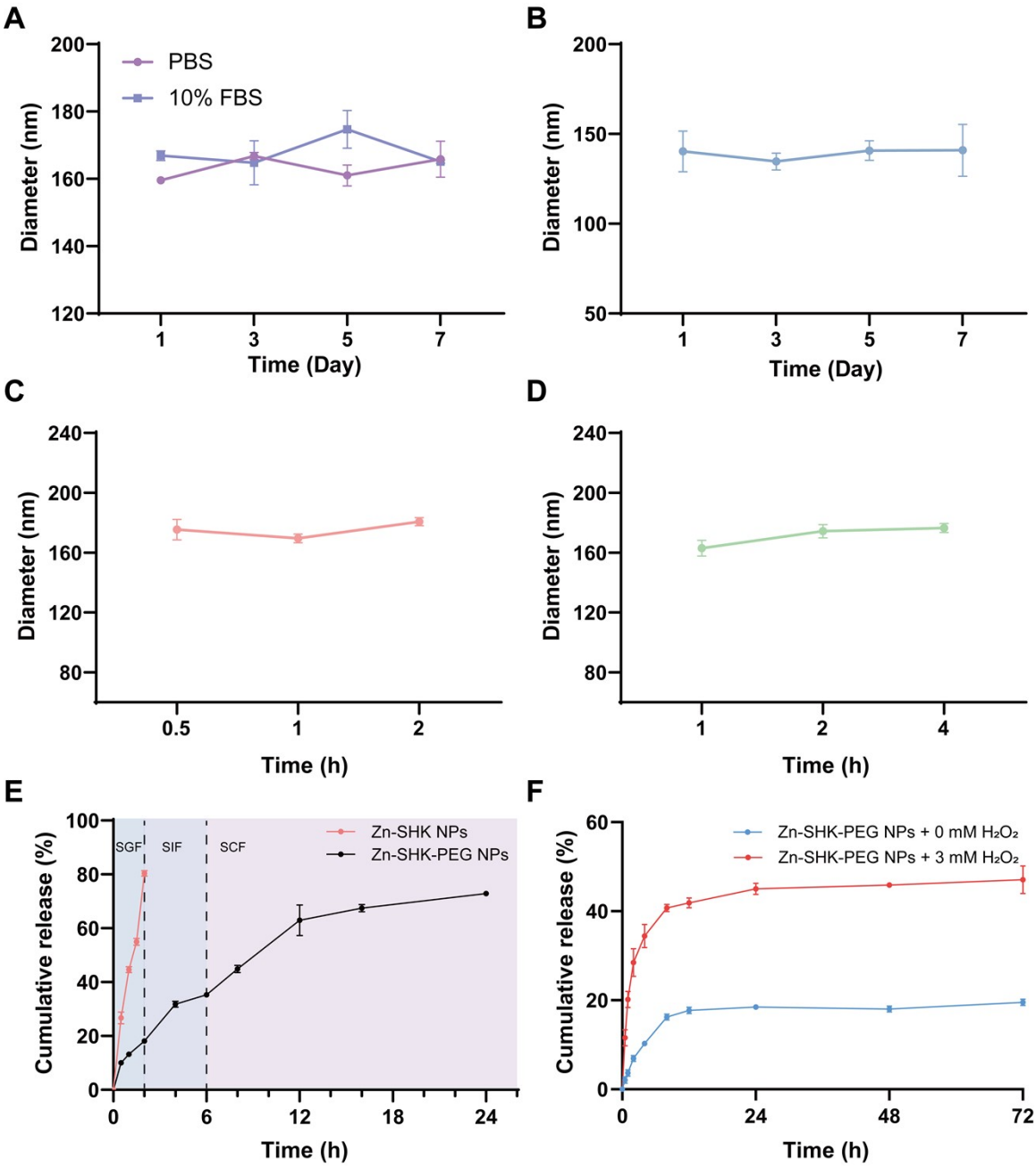

Figure S4. Colloidal stability and release profiles of Zn-SHK-PEG NPs under physiological gastrointestinal and inflammatory-mimicking conditions.

(A) Time-dependent hydrodynamic diameter of Zn-SHK-PEG NPs in PBS and 10% FBS, n=3.

(B) Time-dependent hydrodynamic diameter of Zn-SHK-PEG NPs in water, n=3.

(C) Hydrodynamic diameter of Zn-SHK-PEG NPs in simulated gastric fluid (SGF) within the estimated gastric retention time, n=3.

(D) Hydrodynamic diameter of Zn-SHK-PEG NPs in simulated intestinal fluid (SIF) within the estimated intestinal retention time, n=3.

(E) Simulated gastrointestinal release profiles of SHK from Zn-SHK NPs and Zn-SHK-PEG NPs, n=3.

(F) SHK release profiles from Zn-SHK-PEG NPs under physiological conditions and 3 mM H_2O_2-induced inflammatory-mimicking conditions, n=3.

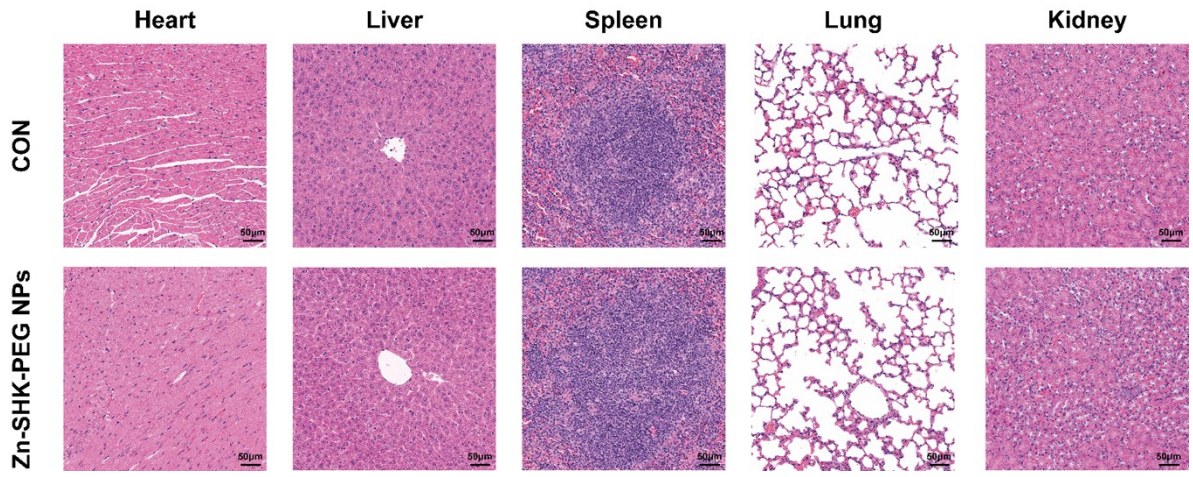

Figure S5. H&E analysis of hearts, livers, spleens, lungs, and kidneys tissues of mice from CON and Zn-SHK-PEG NPs, n=3.

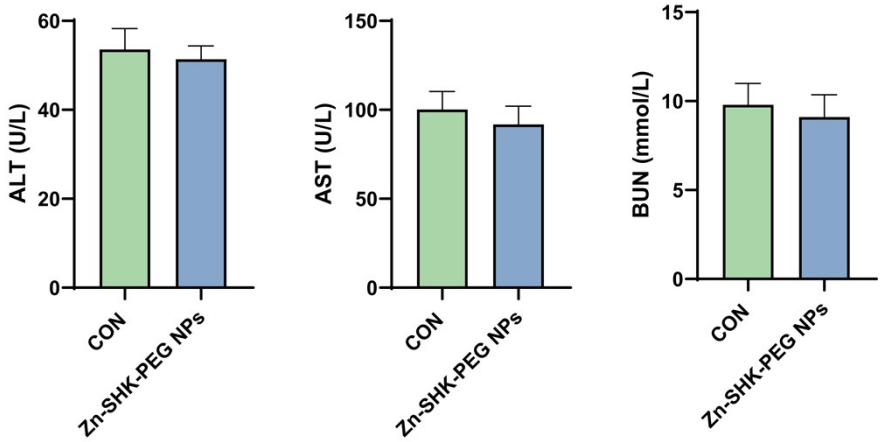

Figure S6. Serum biochemical analysis of mice after Zn-SHK-PEG NPs administration, n=3.