Supplementary figures

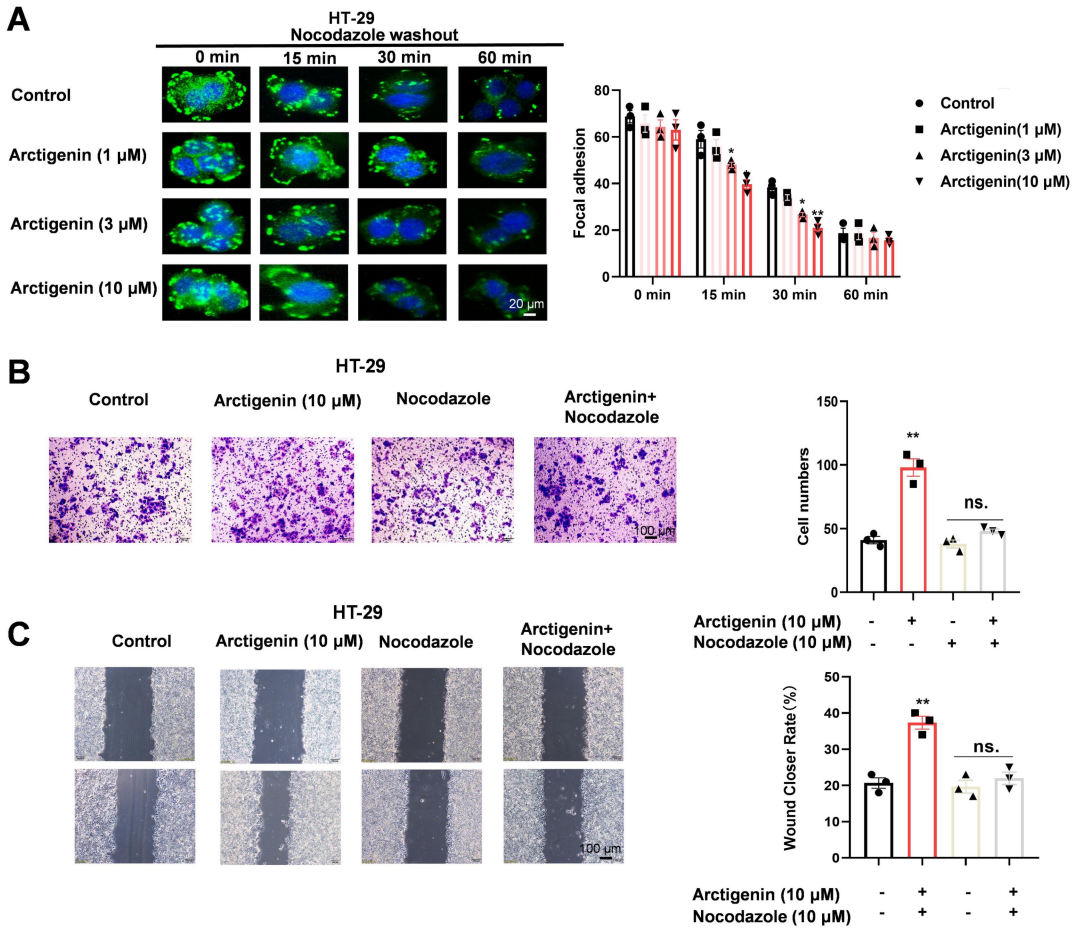

Fig. S1 Promotion of arctigenin on focal adhesion disassembly and its importance in acceleration of cell migration and wound healing in HT-29 cells. HT-29 cells were treated with arctigenin (10 μM) for 24 h in the presence or absence of norcodazole (10 μM). (A) Focal adhesion disassembly of HT-29 cells was assessed by confocal immunofluorescence using an antibody against vinculin (plotting scale: 20 μm). (B) Migration of HT-29 cells was measured using the transwell assay. (plotting scale: 100 μm). (C) Rate of wound healing of HT-29 cells was measured using the scratch assay. (plotting scale: 100 μm). The data are presented as means ± S.E.M.. Significant differences were indicated as follows: $^{*}P < 0.05$ and $^{**}P < 0.01$ compared to the Control group. $^{ns.}$ $P > 0.05$ compared to the nocodazole group, The results shown are representative of three independent experiments.

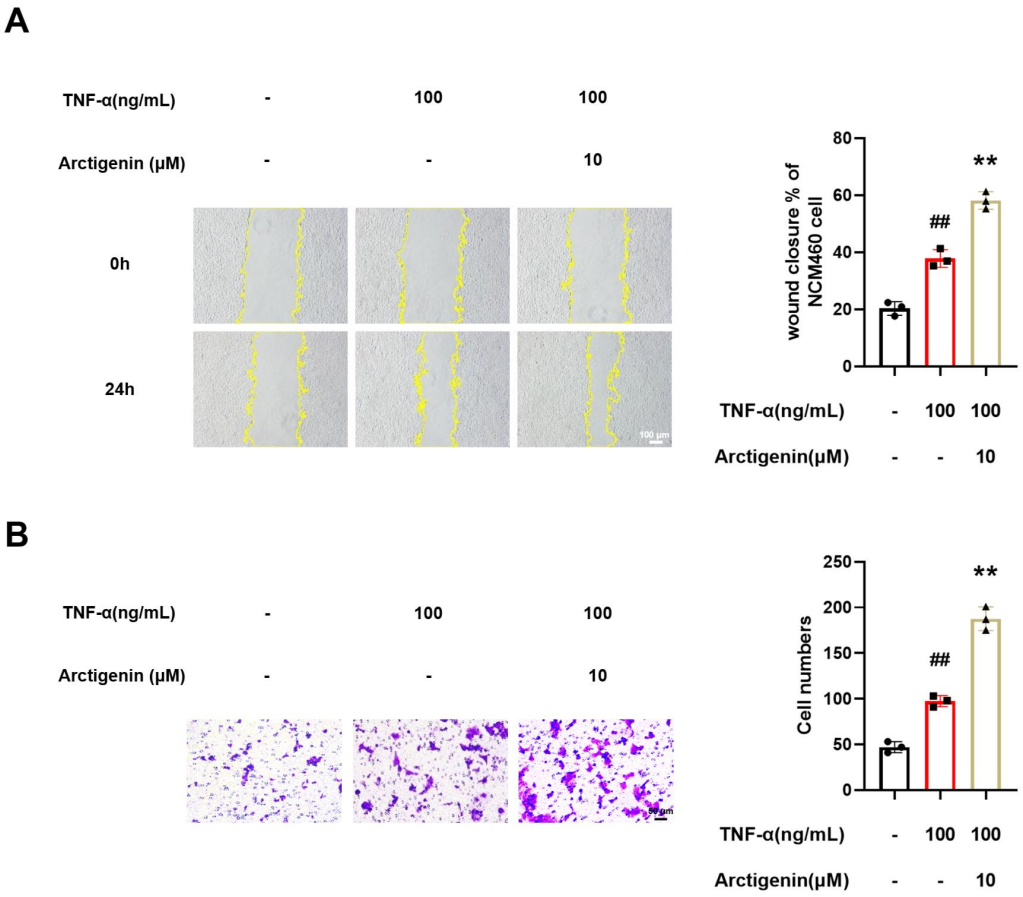

Fig. S2 Effects of arctigenin on wound healing and migration of NCM460 cells under TNF-α-induced inflammatory conditions (A) Wound healing rate of NCM460 cells was detected by scratch test. (B) Migrated cell number of NCM460 cells was detected by transwell migration assay. Data was showed as means ± SEM. $^{\#\#}p < 0.01$ vs. Control group; $^{**}p < 0.01$ vs. TNF-α group. The results are representative of three independent experiments.

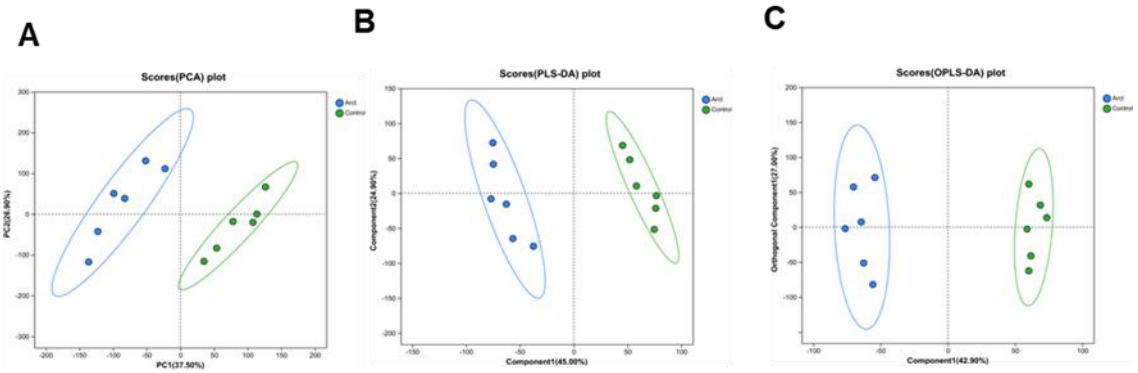

Fig. S3 Effect of arctigenin on the metabolomic profile of colon epithelial cells. NCM460 cells were treated with arctigenin (10 μM) for 24 h and pre-chilled 80% methanol was added to extract the cellular polar metabolites *via* repeated freeze-thaw cycles. (A-C) Cellular metabolites were identified using UPLC Q-TOF/MS, and the corresponding PCA, PLS-DA, and OPLS-DA plots were presented.

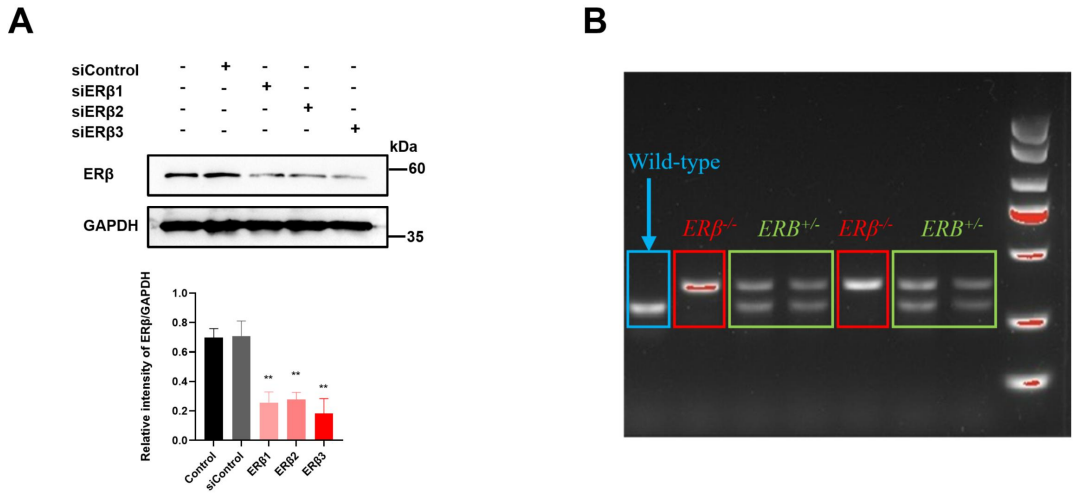

Fig. S4 Knockdown efficiency of siERβ in colon epithelial cells and genotype identification of ERβ$^{-/-}$ mice. (A) NCM460 cells were transfected with siControl, siERβ1, siERβ2, and siERβ3. The protein expression levels of ERβ in NCM460 cells were measured using western blotting. (B) The tail tissue samples were collected from ERβ knockout mouse. Wild-type mice, *ERβ*$^{-/-}$ mice, and *ERβ*$^{+/-}$ mice were identified by PCR and agarose gel electrophoresis.

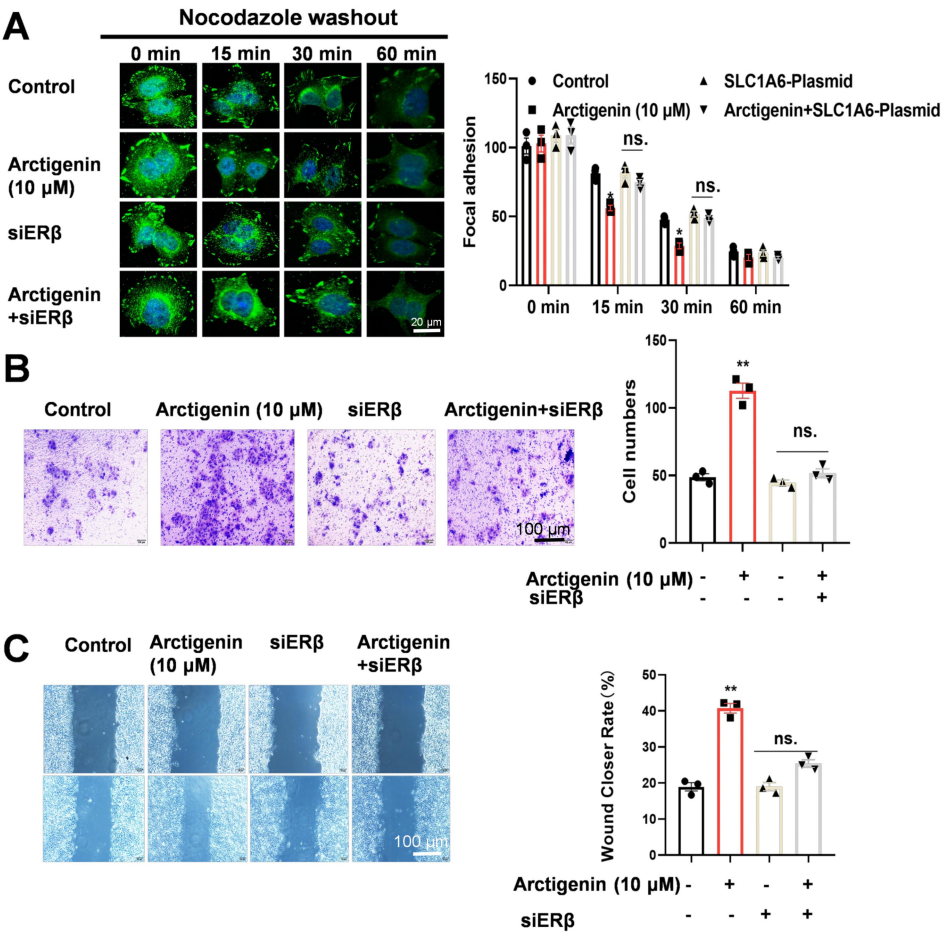

Fig. S5 Arctigenin accelerates focal adhesion disassembly, cell migration and wound healing of colonic epithelial cells in an ERβ-dependent manner. NCM460 cells were treated with arctigenin (10 μM) for 24 h in the presence or absence of siERβ. (A) Focal adhesion disassembly was assessed by confocal immunofluorescence using an antibody against vinculin (plotting scale: 20 μm). (B) The migration of NCM 460 cells was measured using the transwell assay (plotting scale: 100 μm). (C) Rate of wound healing of NCM 460 cells was measured using the scratch assay. (plotting scale: 100 μm). The data are presented as means ± S.E.M.. Significant differences were indicated as follows: $^{*}P < 0.05$ and $^{**}P < 0.01$ compared to the Control group. $^{ns.}$ $P > 0.05$ compared to the siERβ group , indicating no significant difference. The results shown are representative of three independent experiments.

Table 1 Human gene primer sequences

Gene	Primers	Sequence (5'-3')
SLC1A7	Forward	CCTCACCGTGGCGTACTACC
	Reverse	CTCTGCTCCGTGGTCTCCTTC
SLC1A6	Forward	CCTCCATGCCTCCTCCATTCTC
	Reverse	ACGGGTACAGTCTCCTCAAAGC
SLC1A3	Forward	TGCTGGGAAGATTGTGGAGATGG
	Reverse	AGAGGAGTGGCAAGACGATGAC
SLC1A2	Forward	ACTGGCTGCTGGACAGGATG
	Reverse	CGATGCTGGGAGTCAATGGTATC
SLC1A1	Forward	CTTGGGCATTGGGCAGATCATC
	Reverse	CACGGCACTCAGCACAATCAC
GLDH	Forward	CCATTGTACCCACGGCAGAGTTC
	Reverse	TCCATTGTGTATGCCAAGCCAGAG
GPT	Forward	CTGACGCTGGACGGCATGAAC
	Reverse	CGATGTTGGCACGGATGACCTC
GLS	Forward	GGCTGCGACACTGGCTAATGG
	Reverse	TGGAAAGCAAACTGCCCTGAGAAG
GAD1	Forward	TCCTCCTGGAAGTGGTGGACATAC
	Reverse	GCAACTGGTGTGGGTGATGAAAG
GOT1	Forward	CGTCAGTCTTTGCCGAGGTTCC
	Reverse	TATGCTCCCACTCCCAGGTTGAC
ASPH	Forward	AAGGTAGACGCTCGGAGGTTCG
	Reverse	GTCCTGCTCCATGCTGCTGTG
DDO	Forward	TGGTGTTCATTTGGTATCAGGTTGG
	Reverse	TCTTCAGCTCAGCCTCAGTCATC
CPS	Forward	CTACAGCCTCTATGCCAGTCTCG
	Reverse	CTCACCATCCACAGCCTCCTC
GAPDH	Forward	AATGGGCAGCCGTTAGGAAA
	Reverse	GCCCAATACGACCAAATCAGAG

Table 2 Mouse gene primer sequences

Gene	Primers	Sequence (5'-3')

TNF-α	Forward	ACCCTCACACTCACAAACCA
	Reverse	ACAAGGTACAACCCATCGGC
IL-6	Forward	CTCCCAACAGACCTGTCTATAC
	Reverse	CCATTGCACAACTCTTTTCTCA
IL-1β	Forward	CCAGATCCTGTCCAAACTAAGG
	Reverse	CTCTTTAGCATAGTAGTCCGCT
EGF	Forward	ACAGGAATGGAGGCTGTGAA
	Reverse	TGGTACTTCAGCCTGAGTGG
TFF3	Forward	CTGCAGGAGACAGAATGCAC
	Reverse	CTTGTGTTGGCTGTGAGGTC
Occludin	Forward	TCGCCATATTTGCCTGTGTG
	Reverse	CCAAAGAGCCCTGTCCCATA
GAPDH	Forward	GACATTTGAGAAGGGCCACAT
	Reverse	CAAAGAGGTCCAAAACAATCG