

Supplementary information

Supplementary figures

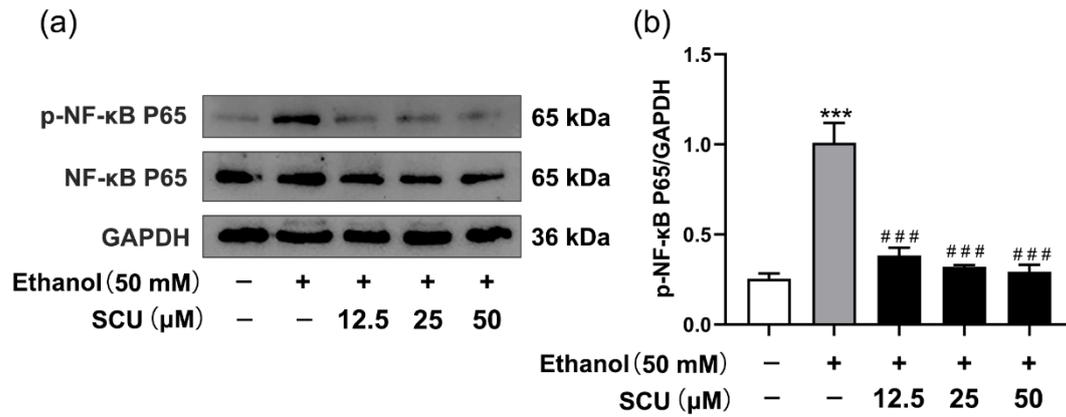


Fig. S1 Scutellarin inhibited NF- κB p65 activation in ethanol-treated RAW264.7 cells. (a) The protein level of p-NF- κB p65 was analyzed utilizing western blot. (b) Quantification analysis of p-NF- κB p65 protein expression in RAW264.7 cells in (a). Values are demonstrated as mean \pm standard deviation (SD), n=3. *** P<0.001 versus the control group; ### P < 0.001 versus the ethanol group. SCU: scutellarin; GAPDH: glyceraldehyde-3-phosphate dehydrogenase; p-NF- κB : phosphorylated nuclear factor- κB .

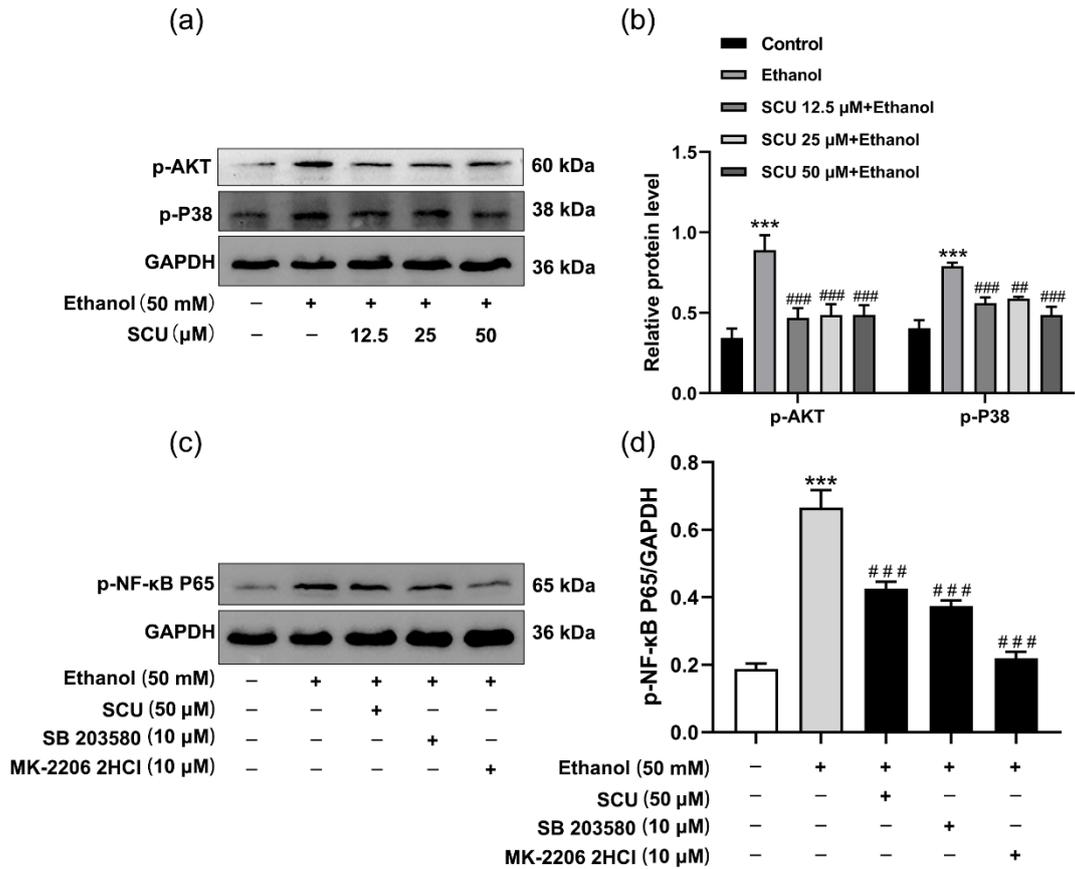


Fig. S2 Inhibition of p-AKT and p-p38 by scutellarin involved in NF- κ B signaling pathway in ethanol-treated RAW264.7 cells. (a) The protein levels of p-AKT and p-P38 were analyzed utilizing western blot in RAW264.7 cells. (b) Quantification analysis of p-AKT and p-P38 protein expression in (a). (c) The protein level of p-NF- κ B P65 was analyzed by western blot in RAW264.7 cells. (d) Quantification analysis of protein expression in (a). Values are demonstrated as mean \pm standard deviation (SD), $n=3$. *** $P<0.001$ versus control group; ## $P<0.01$ and ### $P<0.001$ versus ethanol group. SCU: scutellarin; p-AKT: phosphorylated protein kinase B; GAPDH: glyceraldehyde-3-phosphate dehydrogenase; p-NF- κ B: phosphorylated nuclear factor- κ B.

Supplementary tables

Table S1 Antibody dilution ratio for western blot

Antibody	Dilution ratio
Nrf2	1:1000
CYP2E1	1:2000
HO-1	1:2000
GAPDH	1:10 000
p38	1:1000
p-p38	1:1000
p-AKT	1:1000
AKT	1:1000
p-NF- κ B p65	1:1000
NF- κ B p65	1:1000
I κ B α	1:1000
p-ERK1/2	1:1000
ERK1/2	1:1000
p-JNK	1:1000
JNK	1:1000
Caspase-1	1:1000
NLRP3	1:1000
ASC	1:500
β -Tubulin	1:1000

Nrf2: nuclear factor erythroid 2-related factor 2; CYP2E1: cytochrome P450 family 2 subfamily E member 1; HO-1: heme oxygenase-1; GAPDH: glyceraldehyde-3-phosphate dehydrogenase; p-NF- κ B: phosphorylated nuclear factor- κ B; p-AKT: phosphorylated protein kinase B; I κ B α : inhibitor of NF- κ B- α ; p-ERK1/2: phosphorylated extracellular signal-regulated kinase 1/2; JNK: c-Jun N-terminal kinase; NLRP3: nucleotide-binding oligomerization domain (NOD)-like receptor protein 3; ASC: apoptosis-associated speck-like protein containing a caspase recruitment domain.

Table S2 Primer sequences for RT-qPCR

Name	Forward primer sequence	Reverse primer sequence
<i>TNF-α</i>	5'-GACGTGGAAGCTGGCAGAAGA-3'	5'-GGCTACAGGCTTGTCACCTCG-3'
<i>Il-1β</i>	5'-AGGAGAACCAAGCAACGACA-3'	5'-CTCTGCTTGTGAGGTGCTGA-3'
<i>Il-6</i>	5'-TGCCTTCTTGGGACTGATGC-3'	5'-GCAAGTGCATCATCGTTGTTC-3'
<i>β-Actin</i>	5'-GCCATGTACGTAGCCATCCA-3'	5'-ACGCACGATTTCCCTCTCAG-3'

RT-qPCR: reverse transcription quantitative real-time polymerase chain reaction; IL: interleukin; TNF- α : tumor necrosis factor- α .