Chlojaponilactone B Attenuates Lipopolysaccharide-Induced Inflammatory Responses by Suppressing TLR4-Mediated ROS Generation and NF-κB Signaling Pathway

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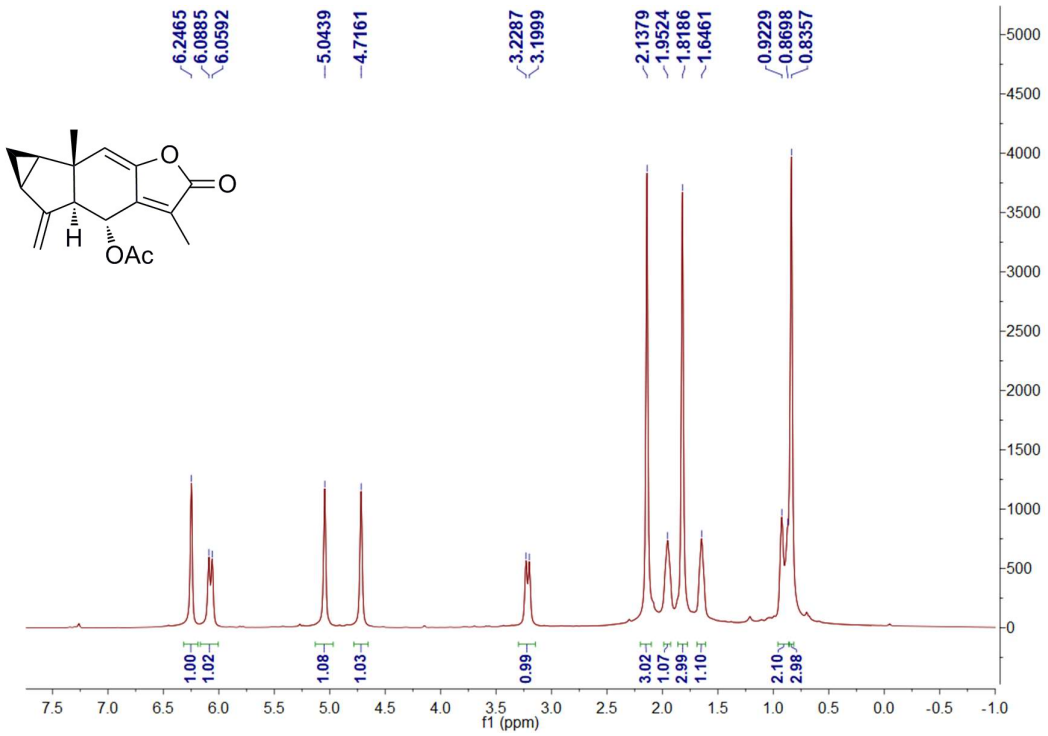
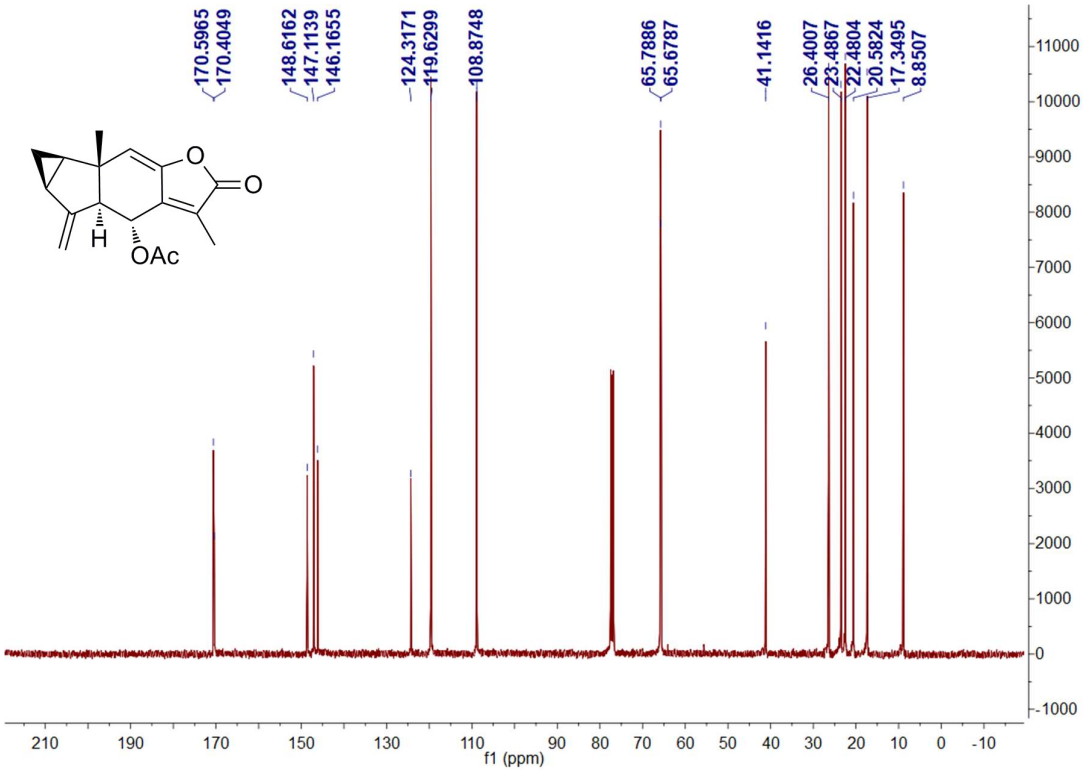


S1. ^{1}H NMR spectrum of chlojaponilactone B (**1**) ($CDCl_3$, 400 MHz)



S2. ^{13}C NMR spectrum of chlojaponilactone B (**1**) ($CDCl_3$, 100 MHz)

S3. *Cytotoxicity Test*

Cell viability was analyzed using the 3-(4,5-dimetrylthiazol)-2,5-diphenyltetrazolium bromide (MTT) assay. RAW 264.7 macrophages were seeded at a density of 5×10^3 cells/well in 96-well plates for 24 h. The cells were then treated with **1** or TAK-242 at various concentrations with or without LPS (1μg/ml) for 24h. An equal concentration of a solvent vehicle (DMSO, 0.5%) was included as the control. MTT (5 mg/mL in sterile PBS) solution was added to the culture (20 μL/well), followed by incubation for 4 h. Finally, the medium was removed, 100 μL DMSO was added to each well, and absorbance was measured at 490 nm by using a microplate reader (Molecular Devices, Sunnyvale, CA, USA).

S4.

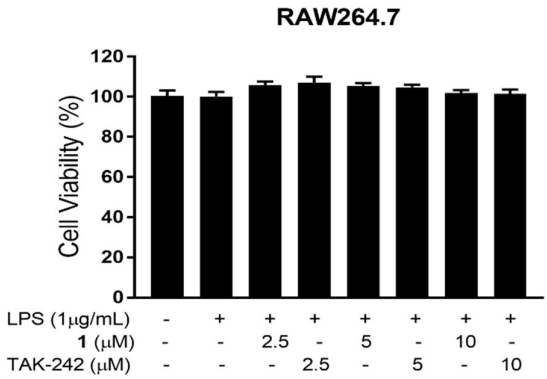


Figure S1. Determination of the cytotoxic effects of **1** and TAK-242 in LPS-induced RAW264.7 cells. Cells were incubated with increasing concentrations of **1** (2.5, 5 or 10 μM) or TAK-242 (2.5, 5 or 10 μM) and treated with LPS (1 μg/mL). The cell viability was examined with the MTT assay.