# Supplementary Information

# Cytotoxic constituents from the sclerotia of *Poria cocos* against human lung adenocarcinoma cells by inducing mitochondrial apoptosis

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## 2.1. General experimental procedures

Infrared (IR) spectra were recorded on a Bruker IFS-66/S FT-IR spectrometer. Ultraviolet (UV) spectra were recorded with an Agilent 8453 UV-visible spectrophotometer (Agilent Technologies, CA, USA). Nuclear magnetic resonance (NMR) spectra were obtained from a Bruker AVANCE III 700 NMR spectrometer operating at 700 MHz ($^1$H) and 175 MHz ($^{13}$C) (Bruker), with chemical shifts given in ppm ($\delta$). LC/MS analysis was performed on an Agilent 1200 Series HPLC system equipped with a diode array detector, a 6130 Series ESI mass spectrometer, and an analytical Kinetex C18 100 Å column (100 mm × 2.1 mm i.d., 5 μm) (Phenomenex, Torrance, CA, USA). Preparative high-performance liquid chromatography (HPLC) was performed with a Waters 1525 Binary HPLC pump with a Waters 996 Photodiode Array (PDA) Detector (Waters Corporation, Milford, CT, USA). Semi-preparative HPLC was conducted with a Shimadzu Prominence HPLC System with SPD-20A/20AV Series Prominence HPLC UV-Vis Detectors (Shimadzu, Tokyo, Japan). Silica gel 60 (Merck, 230-400 mesh) and reversed-phase (RP)-C$_{18}$ silica gel (Merck, 230-400 mesh) were used for column chromatography. Merck precoated silica gel F$_{254}$ plates and RP-18 F$_{254s}$ plates were used for thin-layer chromatography (TLC). Spots on the TLC plates were detected under UV light or by heating after spraying with anisaldehyde-sulfuric acid.

## 2.2. Sample material

Sclerotia of *P. cocos* imported from China were purchased from Kyung-dong Herbal Medicine Market, Seoul, in January 2014. A voucher specimen of the material (PC1308) was identified by one of the authors (K. H. Kim) and deposited in the herbarium of the School of Pharmacy, Sungkyunkwan University, Suwon, Korea.

## 2.3. Extraction and isolation

Dried sclerotia of *P. cocos* (350.0 g) were extracted with 70% aqueous EtOH (each 2.0 L × 3 days) at room temperature. The extract was evaporated under reduced pressure with a rotavapor to yield the EtOH extract (23.3 g), which was suspended in distilled water (1.8 L) and MeOH (30.0 mL) and successively partitioned with hexane, dichloromethane (CH$_2$Cl$_2$), ethyl acetate (EtOAc), and *n*-butanol (BuOH). Four layers with increasing polarity were obtained: the hexane-soluble (6.5 g), CH$_2$Cl$_2$-soluble (2.3 g), EtOAc-soluble (2.7 g), and BuOH-soluble fractions (13.7 g). The hexane-soluble fraction (6.5 g) was separated by silica gel column chromatography (200 g, eluted with hexane/EtOAc [2:1]) to afford four fractions (H1-H4). Fraction H1 (4.3 g) was separated on an RP-C$_{18}$ column eluted with 95% MeOH, yielding 8 subfractions (H11-H18). Subfraction H17 (112.0 mg) was

separated by semi-preparative HPLC (87% MeOH) on a Phenomenex Luna HPLC phenyl-hexyl column (250 × 10 mm; flow rate: 2 mL/min), yielding compound **4** ($t_R$ 63.0 min, 1.7 mg). Fraction H2 (1.8 g) was chromatographed on an RP-$C_{18}$ column (70 g, eluted with 90% MeOH), generating 7 subfractions (H21-H27). Subfraction H26 (46.0 mg) was fractionated by preparative HPLC (95% MeOH) on an Agilent Eclipse C18 column (21.2 × 250 mm; flow rate: 5 mL/min), and 9 subfractions (H261-269) were obtained. Subfraction H265 (10.3 mg) was purified by semi-preparative HPLC (92% MeOH) with a Phenomenex Luna phenyl-hexyl column (250 × 10 mm; flow rate: 2 mL/min) to afford compound **2** ($t_R$ 20.5 min, 1.8 mg). On the same column, subfraction H268 (7.0 mg) was also separated by semi-preparative HPLC (87% MeCN), yielding compound **1** ($t_R$ 18.0 min, 1.0 mg). Fraction H3 (585.0 mg) was subjected to RP-$C_{18}$ column chromatography (25 g, eluted with 92% MeOH) to afford 5 subfractions (H31-35). Subfraction H33 (50.3 mg) was separated by semi-preparative HPLC (83% MeOH) with a Phenomenex Luna phenyl-hexyl column (250 × 10 mm; flow rate: 2 mL/min) to yield compound **3** ($t_R$ 32.5 min, 3.9 mg).

The EtOH extract of the sclerotia of *P. cocos* and the compounds isolated from it were prepared as stock solutions at concentrations of 100 mg/mL and 20 mM, respectively, in dimethyl sulfoxide (DMSO, Sigma, St. Louis, MO, USA) and stored at -80$^{\circ}$C until use.