Supporting Information

***Rec. Nat. Prod*. 13:5 (2019) 429-433**

A New Ursane-type Triterpene from the Roots of *Salvia miltiorrhiza* Bunge

Le Quoc Hung[1,2], Phuong Thien Thuong[2] and Nguyen Huu Tung[1,*]

[1] *School of Medicine and Pharmacy, Vietnam National University, Hanoi (VNU); 144 Xuan Thuy St., Cau Giay, Hanoi, Vietnam*

[2]*National Institute of Medicinal Materials (NIMM); 3B Quang Trung St., Hoan Kiem district, Hanoi, Vietnam*

[*] Corresponding author: E- Mail: tunginpc@gmail.com (N. H. Tung), Phone +84-9787-45494

This supplementary information was re-prepared by the authors as the corrected version upon the decision of ACG PUBLICATIONS INTERNATIONAL ETHICS COMMITTE on December 04th, 2020 (Ref: 2020-ET-003)

Corrected on December 08, 2020

Table Contents	**Page**

S1: General Procedures

Optical rotations were measured with a DIP-360 digital polarimeter (JASCO, Easton, USA). NMR spectra were recorded on a JEOL ECX 400 FT-NMR spectrometer (JEOL, Japan) and Bruker Avance 500 NMR spectrometer (BrukerSpin, Germany) at room temperature using standard pulse program, with tetramethylsilane as the internal standard and chemical shift values were expressed in δ (ppm). ESI-MS experiments employed an Agilent 1260 TripleQuad-6420 LC-MS/MS (Agilent Technologies, USA). HR-ESI-MS experiments employed a JEOL AccuTOF^{TM} LC 1100 mass spectrometer (JEOL, Tokyo, Japan). Column chromatography was performed on silica gel 60 (230–400 mesh, Nacalai Tesque Inc., Kyoto, Japan) and YMC ODS-A gel (50 μm, YMC Co. Ltd., Kyoto, Japan). TLC was performed on Kieselgel 60 F_{254} and TLC Silica gel 60 RP-18 F_{254S} (Merck, Damstadt, Germany) plates. Spots were visualized by spraying with 1% $Ce(SO_4)_2$-10% aqueous H_2SO_4 solution, followed by heating.

S2: Extraction and Isolation

The air-dried danshen roots (550 g) were sliced and then extracted with EtOH-H_2O (80:20, v/v) (1.5 L × 3 times) at 40 ^oC under sonication. After removal of solvent, the obtained residue (122.3 g) was suspended in water (500 mL) and successively partitioned with hexane, EtOAc, and *n*-BuOH (each 500 mL × 3) to obtain soluble fractions of hexane (8.5 g), EtOAc (35.8 g), and BuOH (26.2 g).

The EtOAc portion was subjected to a silica gel column (Φ85 mm × 90 mm) with a stepwise gradient of hexane-EtOAc (5:1→0:1, v/v) to give seven fractions (F1 ~ F7). The fraction F5 (8.3 g) was further chromatographed over a silica gel column(Φ50 mm × 350 mm) with the eluent of $CHCl_3$-MeOH (15:1, v/v) to afford six sub fractions (F5.1~F5.6). The fraction F5.5 (210 mg) was then chromatographed on a reversed-phase C_{18} column (Φ20 mm × 400 mm) with MeOH-H_2O (7:2, v/v) to furnish **1** (11 mg). Likewise, the fraction F5.6 (370 mg) was chromatographed on a reversed-phase C_{18} column (Φ30 mm × 400 mm) with MeOH-H_2O (1:1, v/v) to yield **2** (9 mg).

S3: Antiproliferative Assay

Cell Culture and Sample Treatment

The HL-60 cell line was obtained from RIKEN BioResource Center Cell Bank. The cells were maintained in RPMI1640 medium. The medium was supplemented with 10% FBS and 1% penicillin–streptomycin and were then incubated at 37°C under 5% CO_2 in fully humidified conditions. For the cell treatment, DMSO concentrations in the cell culture medium did not exceed 0.2% (v/v) and the controls were always treated with the same amount of DMSO as used in the corresponding experiments.

MTT assay

Cell viability was determined by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. In brief, the cells were seeded in 96-well plates at a density of 1×10^4 cells/well for suspension cells. After incubation for 24 h, the cells were treated with each sample at various concentrations for 24 h. At the end of treatment, MTT solution was added to each well, and the cells were incubated for another 4 h. The precipitated MTT-formazan was dissolved with 0.04 N HCl-isopropanol, and the amount of formazan was measured at 595 nm using a microplate reader (iMark, BioRad, Tokyo, Japan). Cell viability was expressed as a percentage of the control culture.

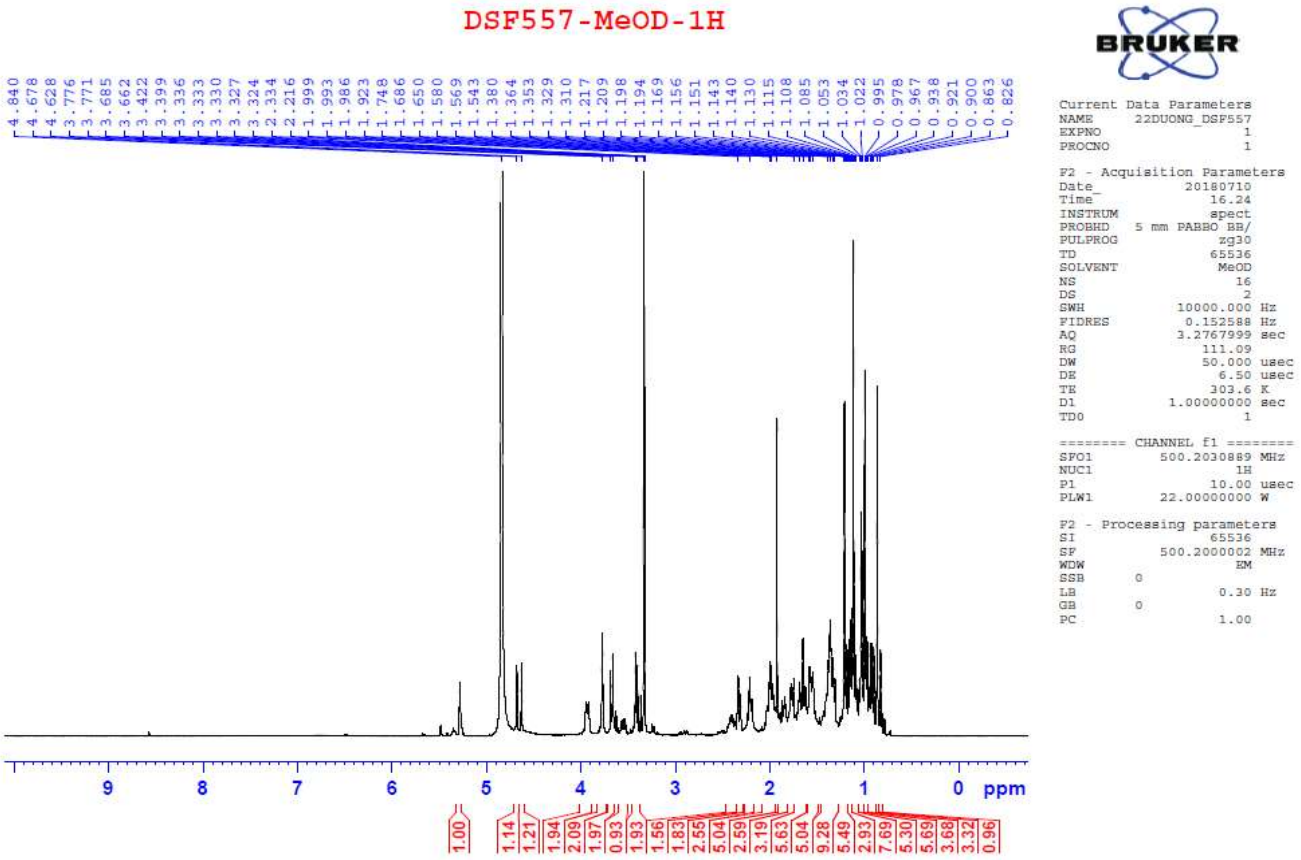

Figure S1: The ^{1}H NMR (500 MHz, CD_3OD) spectrum of **2**

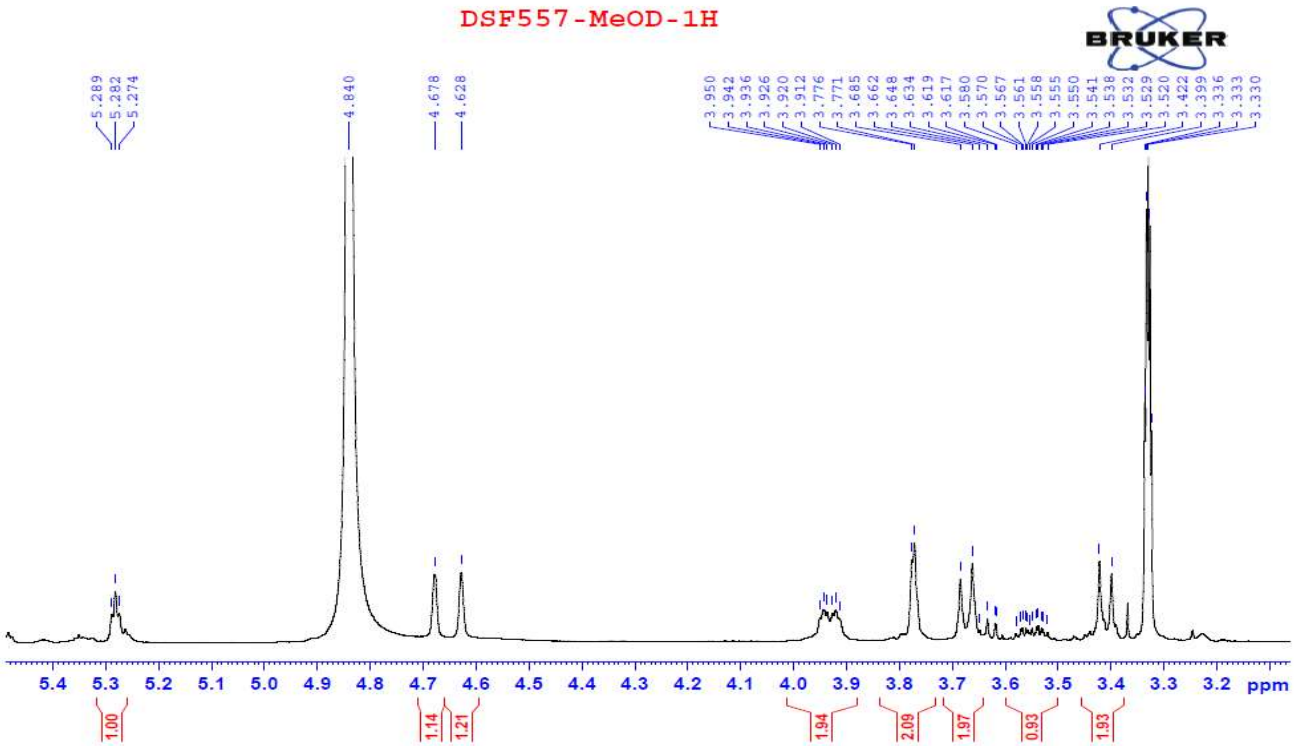

Figure S1: The ^{1}H NMR (500 MHz, CD_3OD) spectrum of **2** (expanded)

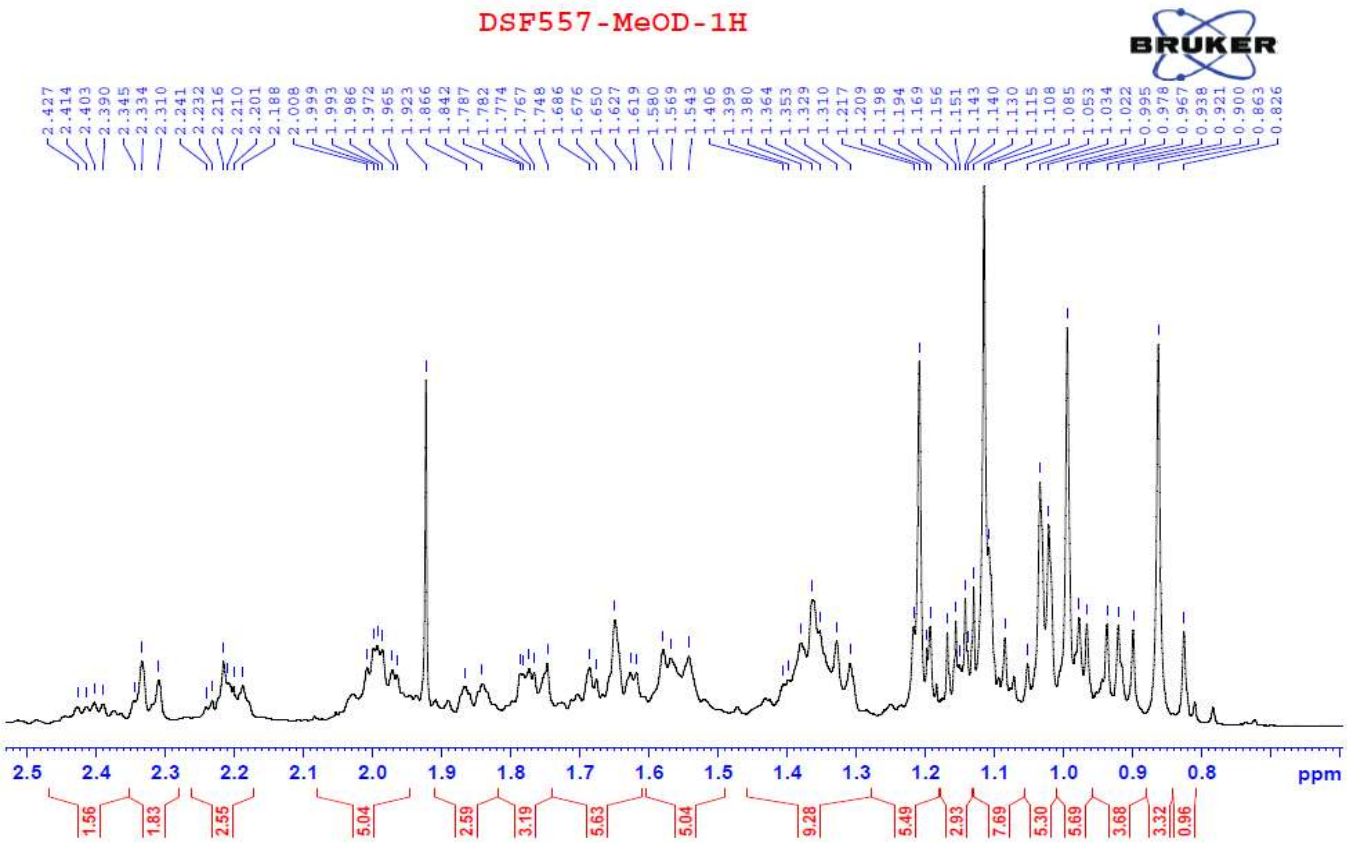

Figure S1: The ^{1}H NMR (500 MHz, CD_3OD) spectrum of **2** (expanded)

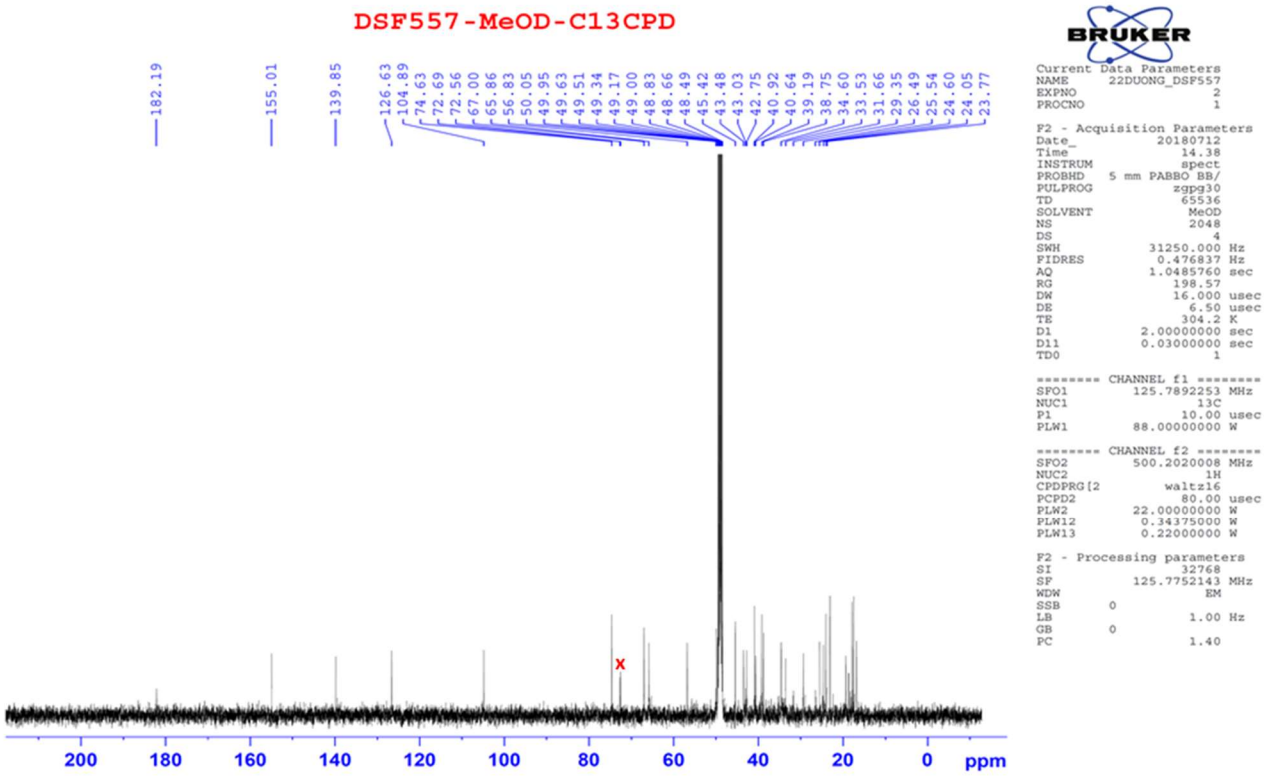

Figure S2: The ^{13}C NMR (125 MHz, CD_3OD) spectrum of **2**

(x: impurity peak is showed in the corrected version of spectrum)

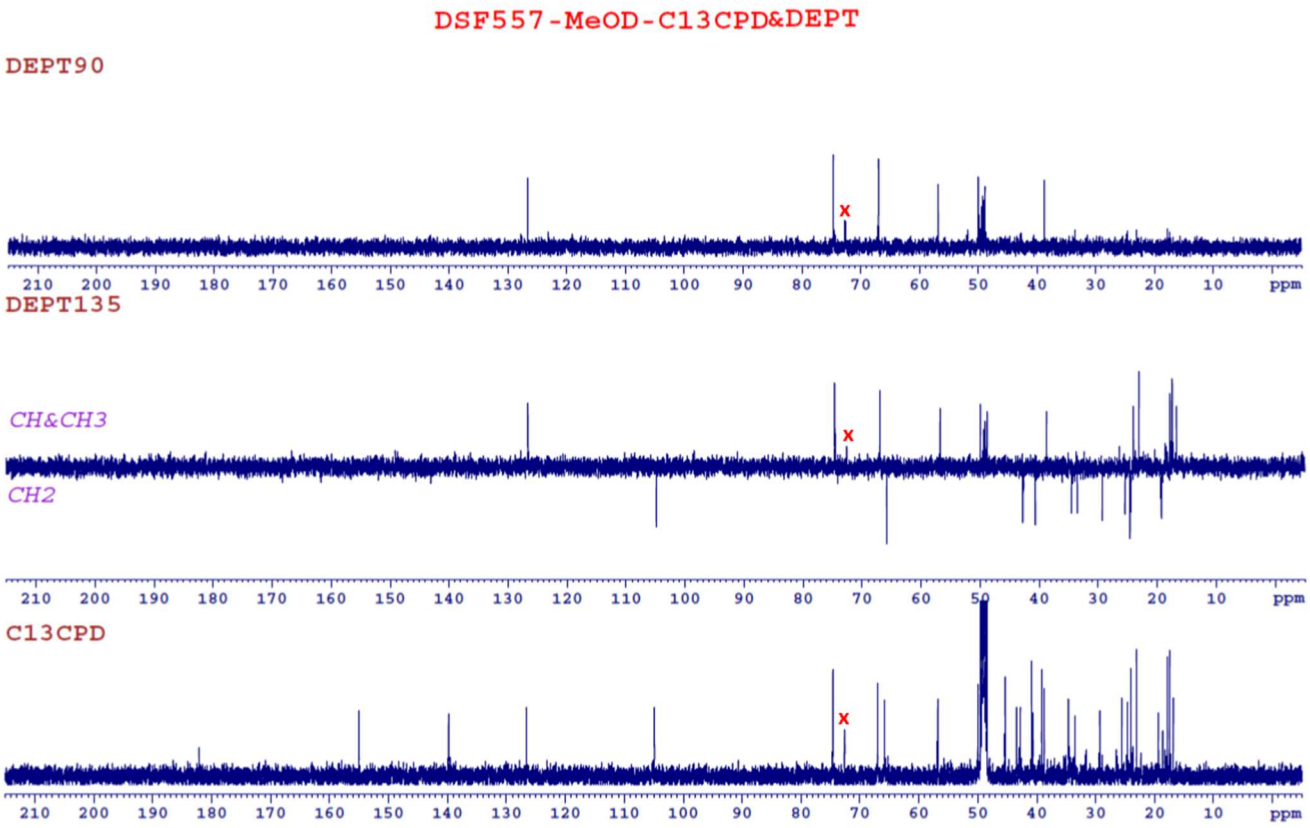

Figure S3: The DEPT spectrum of **2**

(x: impurity peak is showed in the corrected version of spectrum)

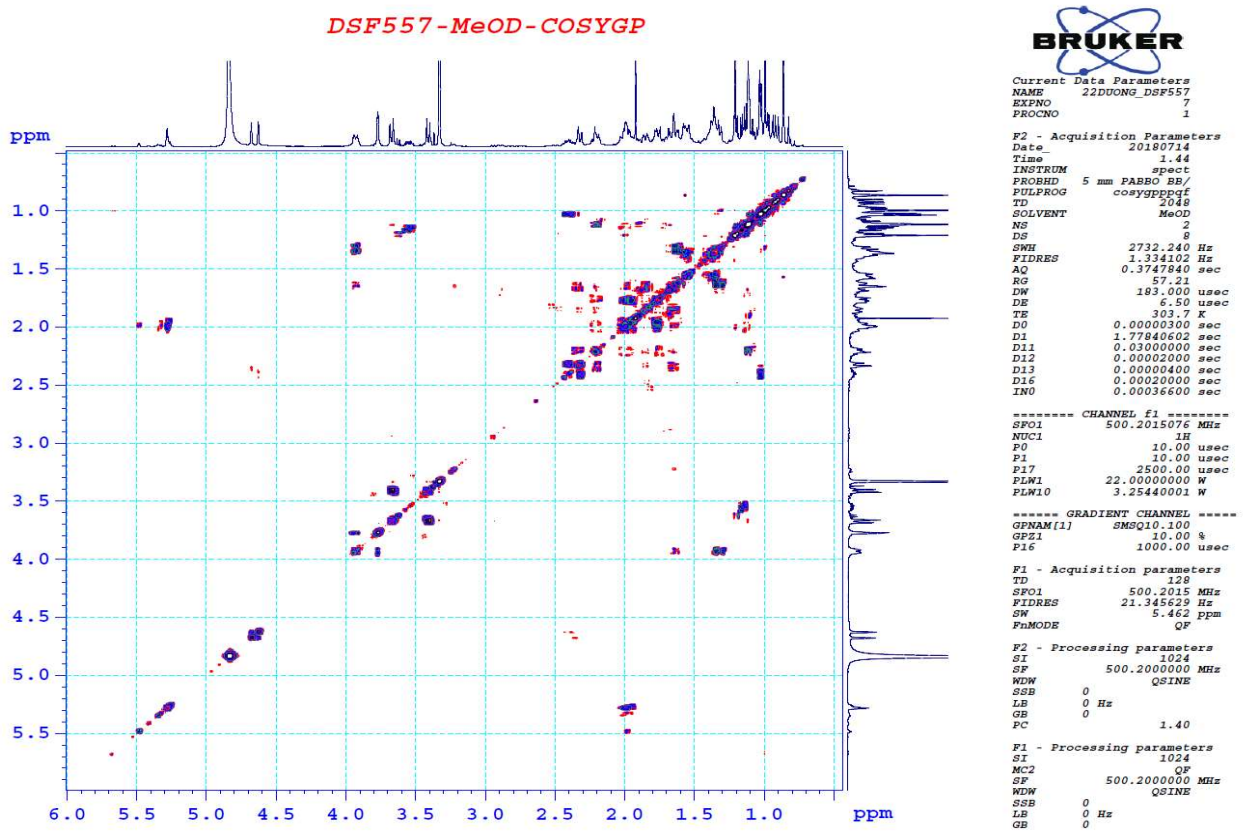

Figure S4: The H-H COSY spectrum of **2**

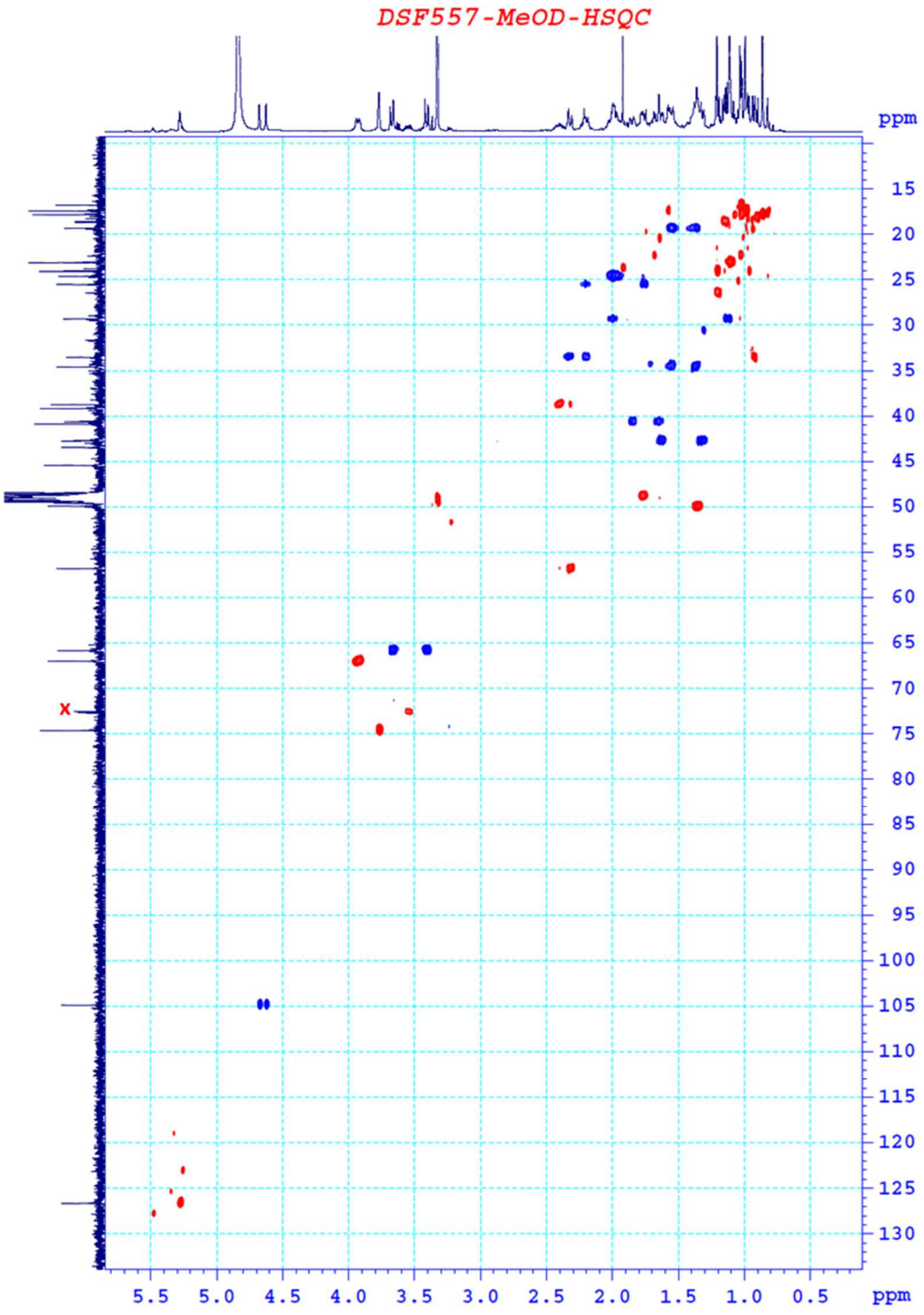

Figure S5: The HSQC spectrum of **2** (x: impurity peak)

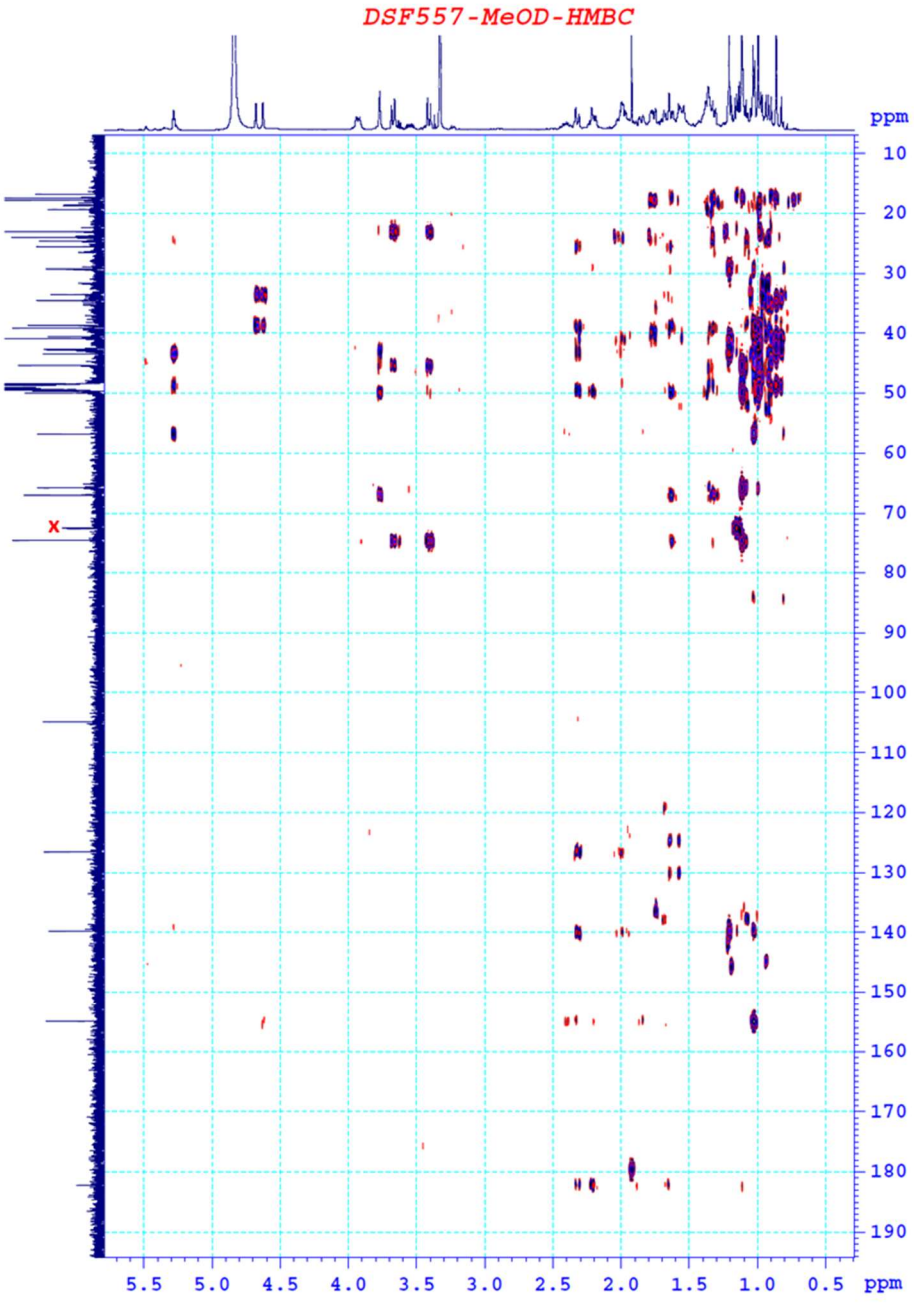

Figure S6: The HMBC spectrum of **2** (x: impurity peak)

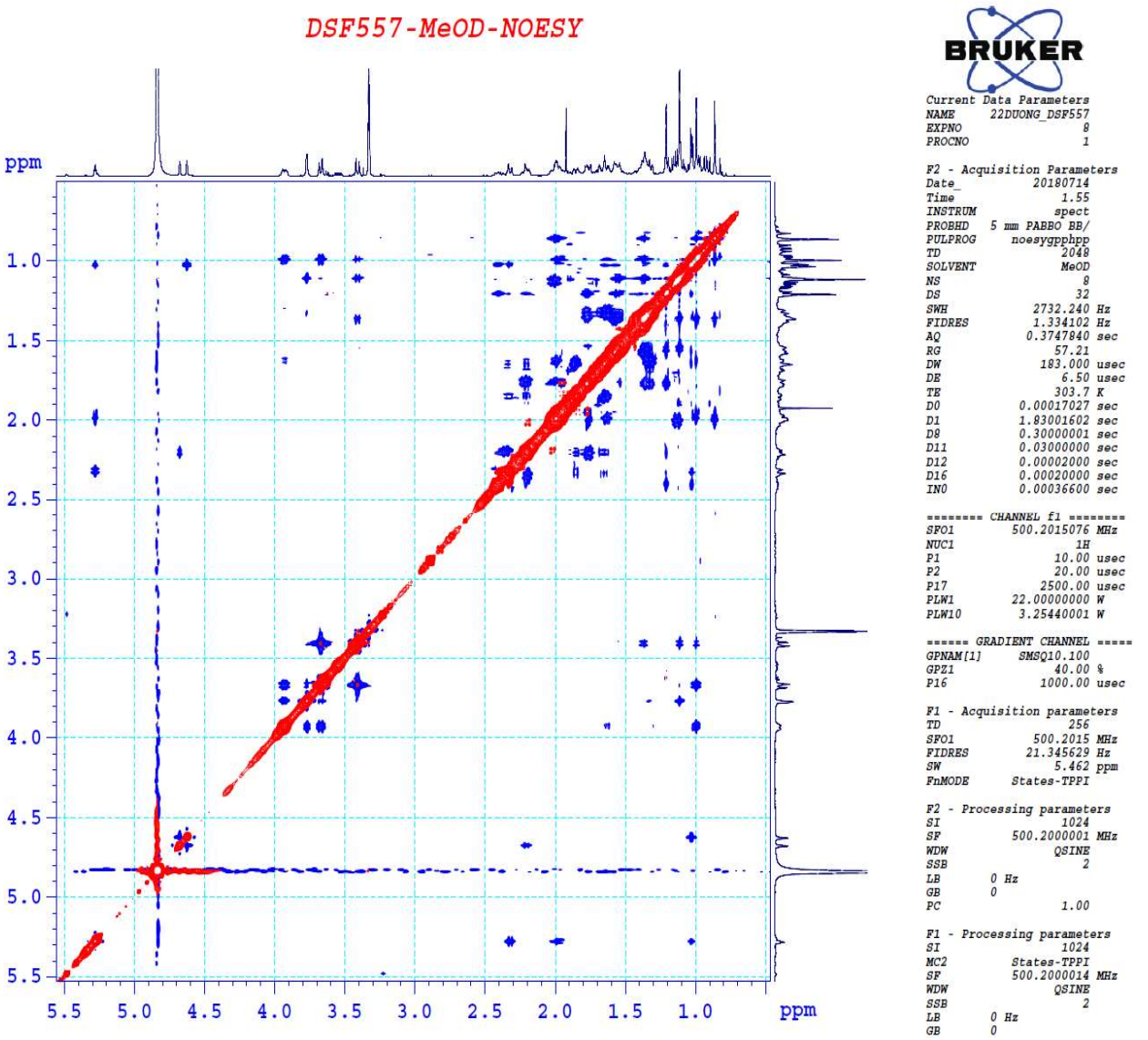

Figure S7: The NOESY spectrum of **2**

m/z, Calc.
$[M+H]^+$ 487.3431 (Calc. for $C_{30}H_{47}O_5$ 487.3423)

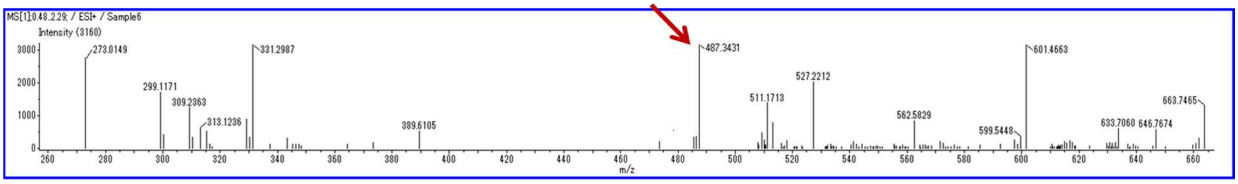

Figure S18: The HR-MS spectrum of **2**