Supporting Information

Chemical Modification of Ovatodiolide Revealed a Promising Amino-prodrug with Improved Pharmacokinetic Profile

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Table of contents

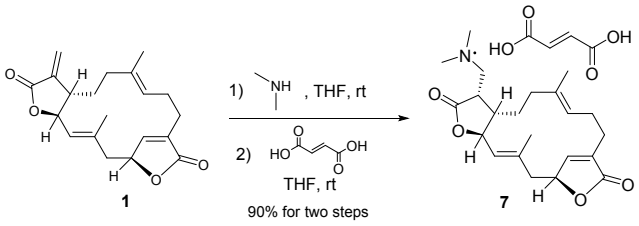
1. General Methods

All reactions were carried out under an argon atmosphere with dry solvents under anhydrous conditions, unless otherwise noted. Tetrahydrofuran (THF) were distilled immediately before use from sodium-benzophenone ketyl. Methylene chloride (DCM), Toluene were distilled from calcium hydride and stored under an argon atmosphere. Reagents were purchased at the highest commercial quality and used without further purification, unless otherwise stated. Solvents for chromatography were used as supplied by Tianjin Reagents chemical. Reactions were monitored by thin layer chromatography (TLC) carried out on 0.25 mm Qingdao silica gel plates (GF-254) using UV light as visualizing agent, 5% phosphomolybdic acid/EtOH (PMA). Qingdao silica gel (60, particle size 0.040-0.063 mm) was used for flash column chromatography. NMR spectra were recorded on Bruker AV-400 instrument and calibrated by using residual undeuterated solvent ($CDCl_3$ δ_H = 7.26 ppm, δ_C = 77.00 ppm, DMSO-d_6 δ_H = 2.50 ppm, δ_C = 39.52 ppm) as internal references. The following abbreviations are used to designate multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, quint = quintet, br = broad. IR spectra were recorded on a Bruker Tensor 27 instrument. High-resolution mass spectra (HRMS) were detected by Varian 7.0T FTMS. Optical rotations were recorded on an Insmark IP 120 digital polarimeter. HPLC data was recorded on UltiMate™ 3000 RSLCnano.

2. Experimental Procedures

2.1 Organic Synthesis Experimental Procedures

Synthesis of DMA-ovatodiolide (7)



To a solution of ovatodiolide (**1**) (1.00 g, 3.04 mmol, 1.0 equiv) in THF (15.0 mL) was added dimethylamine (2.0 M in THF, 15.2 ml, 30.4 mmol, 10.0 equiv). The resulting mixture was stirred at room temperature for 3 h. The mixture was concentrated under reduced pressure. Without purifying, the resulting solid was dissolved in dry THF (20 ml). Then fumaric acid (353 mg, 3.04 mmol, 1.0 equiv) was added to the solution. The resulting mixture was stirred at room temperature for 4 h, concentrated under reduced pressure, filtered, washed with DCM (3 × 15 mL) to give DMA-ovatodiolide (**7**) (1.20 g, 90%, white solid).

[α] $^{22}_{D}$ = +25.2° (c = 0.14, MeOH)

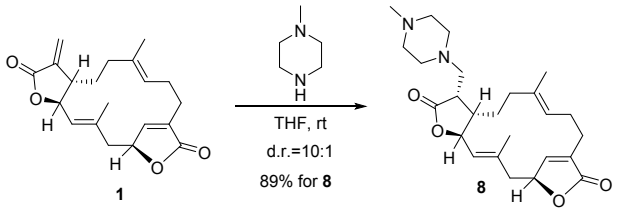
IR (neat) 2948, 2850, 1773, 1742, 1441, 1333, 1197, 1105, 1078, 983, 880, 646 cm^{-1}

^{1}H NMR (400 MHz, DMSO) δ 7.46 (s, 1H), 6.60 (s, 2H), 5.38 – 5.10 (m, 2H), 4.87 (d, *J* = 10.8 Hz, 2H), 3.18 – 2.90 (m, 1H), 2.74 – 2.53 (m, 3H), 2.45 – 2.27 (m, 4H), 2.21 (s, 6H), 2.14 – 2.00 (m, 3H), 1.90 – 1.74 (m, 2H), 1.72 (s, 3H), 1.50 (s, 3H), 1.06 (t, *J* = 13.2 Hz, 1H)

^{13}C NMR (101 MHz, DMSO) δ 177.2, 173.0, 166.2, 149.2, 134.6, 134.1, 132.4, 132.2, 126.3, 124.6, 78.8, 77.8, 54.5, 44.6, 39.6, 39.4, 38.8, 35.5, 24.3, 23.1, 22.8, 18.8, 14.5

HRMS-ESI (m/z): calcd. for $C_{22}H_{32}O_4N$ $[M+H]^+$: 374.2331, found 374.2328.

Synthesis of 8



To a solution of ovatodiolide (**1**) (1.00 g, 3.04 mmol, 1.0 equiv) in THF (15.0 mL) was added N-methyl piperazine (1.69 ml, 15.2 mmol, 5.0 equiv). The resulting mixture was stirred at room temperature for 4 h. The mixture was concentrated under reduced pressure, and then purified by column chromatography on silica gel (1.2% MeOH in DCM-2.5% MeOH in DCM) to give **8** (1.10 g,

89%, white solid).

R_f = 0.6 (DCM/MeOH=10/1, UV& I_2, PMA)

[α] $^{23}_D$ = - 44.3° (c = 0.56, $CHCl_3$)

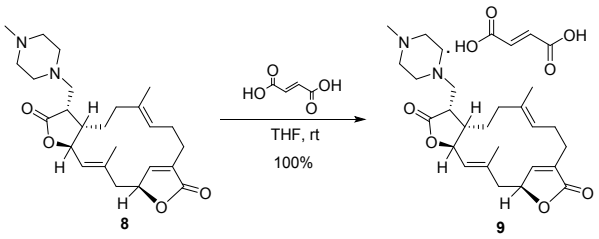
IR (neat) 2929, 2874, 2842, 2798, 1753, 1666, 1450, 1091, 980, 960, 883, 861 cm^{-1}

^{1}H NMR (400 MHz, $CDCl_3$) δ 6.98 (s, 1H), 5.35 (d, *J* = 10.7 Hz, 1H), 5.11 (s, 1H), 4.84 (dd, *J* = 17.8, 8.8 Hz, 2H), 3.07 (s, 1H), 2.90 (dd, *J* = 14.3, 3.5 Hz, 1H), 2.85 – 2.75 (m, 1H), 2.63 (dd, *J* = 12.6, 5.0 Hz, 2H), 2.59 – 2.47 (m, 4H), 2.47 – 2.36 (m, 4H), 2.29 (s, 3H), 2.27 – 2.18 (m, 3H), δ 2.17 – 2.07 (m, 2H), 2.07 – 1.87 (m, 3H), 1.75 (s, 3H), 1.65 (s, 3H), 1.22 – 1.10 (m, 1H)

^{13}C NMR (101 MHz, $CDCl_3$) δ 178.1, 173.2, 146.9, 135.9, 134.9, 131.0, 127.6, 124.4, 78.9, 78.4, 55.3, 53.6, 46.2, 40.3, 40.2, 38.9, 36.2, 25.1, 23.7, 23.2, 19.5, 15.5

HRMS-ESI (m/z): calcd. for $C_{25}H_{37}O_4N_2$ $[M+H]^+$: 429.2753, found 429.2750.

Synthesis of NMP-ovatodiolide (9)



To a solution of **8** (1.00 g, 2.30 mmol, 1.0 equiv) in THF (25 mL) was added fumaric acid (270.8 mg, 2.30 mmol, 1.0 equiv). The resulting mixture was stirred at room temperature for 5 h, concentrated under reduced pressure to give NMP-ovatodiolide (**9**) (1.27 g, 100%, white solid) .

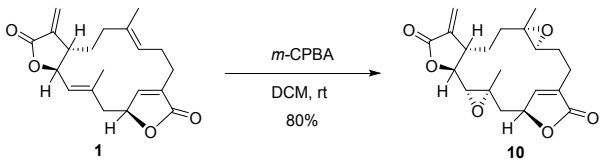
[α] $^{23}_D$ = - 16.3° (c = 0.15, MeOH)

IR (neat) 2922, 2851, 1750 cm^{-1}

^{1}H NMR (400 MHz, DMSO) δ 7.47 (d, *J* = 0.9 Hz, 1H), 6.55 (s, 2H), 5.76 (s, 1H), 5.29 (d, *J* = 10.7 Hz, 1H), 5.22 (d, *J* = 1.4 Hz, 1H), 4.88 (d, *J* = 10.6 Hz, 2H), 3.16 – 3.03 (m, 1H), 2.69 (dd, *J* = 14.2, 3.4 Hz, 2H), 2.66 – 2.52 (m, 5H), 2.47 (s, 1H), 2.44 – 2.34 (m, 4H), 2.33 (s, 3H), 2.32 – 2.24 (m, 1H), 2.19 – 1.97 (m, 4H), 1.88 – 1.74 (m, 2H), 1.73 (d, *J* = 0.7 Hz, 3H), 1.59 (s, 3H), 1.14 – 0.96 (m, 1H)

^{13}C NMR (101 MHz, DMSO) δ 177.2, 173.2, 167.1, 149.1, 134.7, 134.6, 132.5, 131.7, 126.4, 124.6, 79.0, 77.9, 54.9, 54.1, 52.8, 44.7, 38.1, 35.5, 24.2, 23.1, 22.6, 18.8, 15.0

Synthesis of diepoxyovatodiolide (10)



1 10

To a solution of ovatodiolide (**1**) (9.50 g, 27.7 mmol, 1.0 equiv) in DCM (300 mL) was added *m*-CPBA (24.7 g, 121.8 mmol, 85% by weight, 4.4 equiv). The resulting mixture was stirred at room temperature for 2 days. The mixture was poured into saturated aqueous $Na_2S_2O_3$ solution (100 mL), and the organic layer was separated. The aqueous layer was extracted with CH_2Cl_2 (3 × 150 mL), and the combined organic layer was washed with saturated $NaHCO_3$ (2 × 40 mL), brine (30 mL), dried over Na_2SO_4, filtered and concentrated under reduced pressure, and then purified by column chromatography on silica gel (100% EtOAc in Hexane) to give diepoxyovatodiolide (**10**) (8.00 g, 80%, white solid).

R_f = 0.38 (Hexane/EtOAc =1/1, UV& I_2, PMA)

[α] $^{22}_D$ = - 22.5° (c = 0.52, $CHCl_3$)

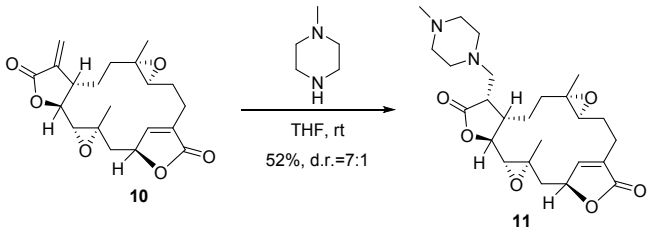
IR (neat) 2930, 1754, 1643, 1435, 1390, 1326, 1263, 1201, 1117, 1082, 1040, 986, 957, 838, 639 cm^{-1}

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.34 (s, 1H), 6.29 (s, 1H), 5.79 (s, 1H), 5.17 (s, 1H), 3.87 (d, *J* = 9.3 Hz, 1H), 2.84 (d, *J* = 9.3 Hz, 1H), 2.57 (dd, *J* = 12.7, 7.3 Hz, 3H), 2.48 (dt, *J* = 8.2, 4.3 Hz, 2H), 2.17 – 1.90 (m, 4H), 1.62 – 1.35 (m, 3H), 1.34 (s, 3H), 1.32 (s, 3H)

^{13}C NMR (101 MHz, $CDCl_3$) δ 173.02, 169.35, 148.56, 137.60, 133.71, 124.49, 81.58, 78.74, 77.48, 77.16, 76.84, 61.97, 60.17, 58.59, 55.99, 41.65, 41.05, 33.01, 29.83, 25.96, 21.87, 19.77, 17.29

HRMS-ESI (m/z): calcd. for $C_{20}H_{24}O_6Na$ $[M+Na]^+$: 383.1471, found 383.1470.

Synthesis of 11



10 11

To a solution of diepoxyovatodiolide (**10**) (7.47 g, 20.7 mmol, 1.0 equiv) in THF (100 mL) was added N-methyl piperazine (11.5 ml, 103.6 mmol, 5.0 equiv). The resulting mixture was stirred at room temperature overnight. The mixture was concentrated under reduced pressure, and then purified via column chromatography on silica gel (5% MeOH in DCM-6.7% MeOH in DCM) to give **11** (5.00 g, 52%, white solid) with d.r. = 7:1 based on crude ^{1}H NMR.

R_f = 0.5 (DCM/MeOH=15/1, UV& I_2, PMA)

[α] $^{22}_{D}$ = - 64.8° (c = 0.53, $CHCl_3$)

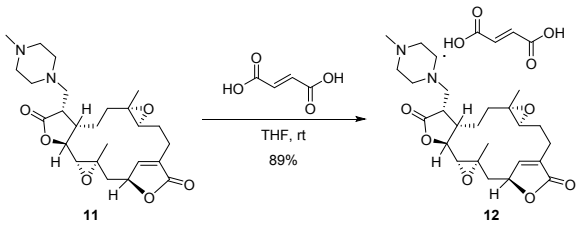
IR (neat) 3084, 2979, 2937, 1761, 1740, 1640, 1461, 1288, 1167, 1072, 1038, 1012, 996, 903 cm^{-1}

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.20 (s, 1H), 5.14 (s, 1H), 3.80 (d, *J* = 10.0 Hz, 1H), 3.12 (ddd, *J* = 12.4, 7.2, 5.5 Hz, 1H), 3.02 (d, *J* = 9.9 Hz, 1H), 2.71 – 2.51 (m, 6H), 2.50 – 2.31 (m, 6H), 2.28 (s, 3H), 2.12 (dd, *J* = 15.4, 3.0 Hz, 1H), 2.00 (t, *J* = 12.7 Hz, 4H), 1.79 – 1.53 (m, 1H), 1.34 (s, 3H), 1.12 (d, *J* = 7.9 Hz, 3H), 1.12 – 1.01 (m, 1H), 0.45 (t, *J* = 13.8 Hz, 1H), 0.28 (s, 2H)

^{13}C NMR (101 MHz, $CDCl_3$) δ 177.2, 172.9, 147.3, 134.7, 81.8, 78.4, 77.5, 77.2, 76.8, 59.1, 57.0, 55.5, 54.3, 46.2, 41.8, 39.7, 37.0, 36.6, 25.6, 25.4, 23.3, 22.1, 20.0, 18.8, 16.9, 16.6

HRMS-ESI (m/z): calcd. for $C_{25}H_{37}O_6N_2$ $[M+H]^+$: 461.2652, found 461.2650.

Synthesis of NMP-diepoxyovatodiolide (12)



To a solution of **11** (3.00 g, 6.50 mmol, 1.0 equiv) in THF (30 mL) was added fumaric acid (1.40 g, 12.3 mmol, 1.9 equiv). The resulting mixture was stirred at room temperature for 3 h. The solution was filtered, washed with THF (3×15 mL), Et_2O (3×10 mL) to give NMP-diepoxyovatodiolide (**12**) (3.30 g, 89%, white solid).

[α] $^{22}_{D}$ = - 39.5° (c = 0.5, MeOH)

IR (neat) 2956, 2929, 2835, 1751, 1703, 1458, 1390, 1145, 1103, 981, 645 cm^{-1}

^{1}H NMR (400 MHz, DMSO) δ 7.95 (s, 1H), 6.57 (s, 2H), 5.32 (s, 1H), 3.99 (d, *J* = 9.7 Hz, 1H), 3.18 (s, 1H), 2.99 (d, *J* = 9.9 Hz, 1H), 2.80 – 2.45 (m, 9H), 2.46 – 2.18 (m, 8H), 2.04 – 1.60 (m, 6H), 1.33 (s, 3H), 1.31 (s, 3H), 1.13 (t, *J* = 11.2 Hz, 1H), 0.90 (t, *J* = 13.4 Hz, 1H)

^{13}C NMR (101 MHz, DMSO) δ 176.2, 173.1, 167.2, 149.8, 134.6, 131.5, 80.7, 78.9, 61.1, 58.4, 58.2, 57.3, 53.7, 53.2, 43.8, 40.7, 38.7, 36.2, 34.9, 24.2, 22.1, 21.8, 18.9, 15.5

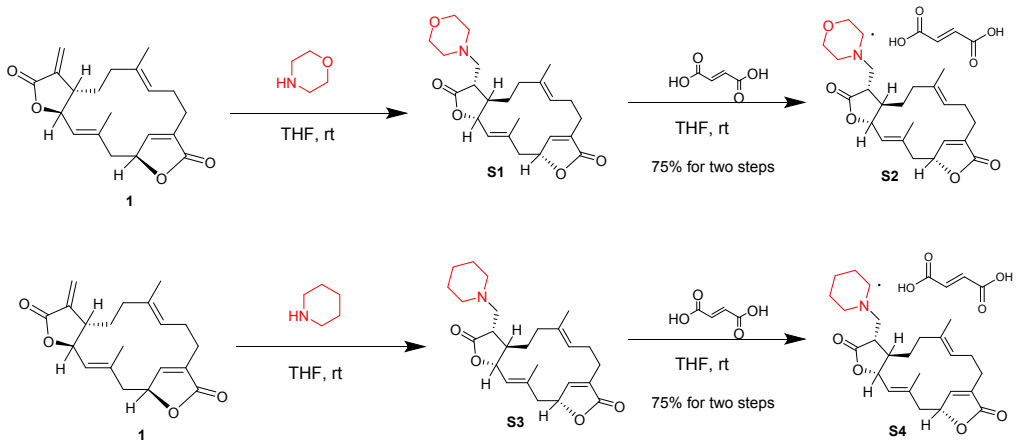
2.2 Biological Experimental Procedures

2.2.1 Release Studies of Prodrug to Active Drug

A 20 μL volume of DMA-ovatodiolide (**7**), NMP-ovatodiolide (**9**) and NMP-diepoxyovatodiolide (**12**) solution was placed in 980 μL of PBS at pH 7.4 (1.25 mM). The tubes were then incubated in a bath incubator at 37 °C. Samples were removed in 10 min, and the concentration of DMA-ovatodiolide

(**7**), NMP-ovatodiolide (**9**), NMP-diepoxyovatodiolide (**12**), ovatodiolide (**1**) and diepoxyovatodiolide (**10**) was analyzed by HPLC.

2.2.1.1 Study the Effect of Release of Prodrug



Scheme S1. Synthesis of MPL-ovatodiolide (**S2**) and PPD-ovatodiolide (**S4**).

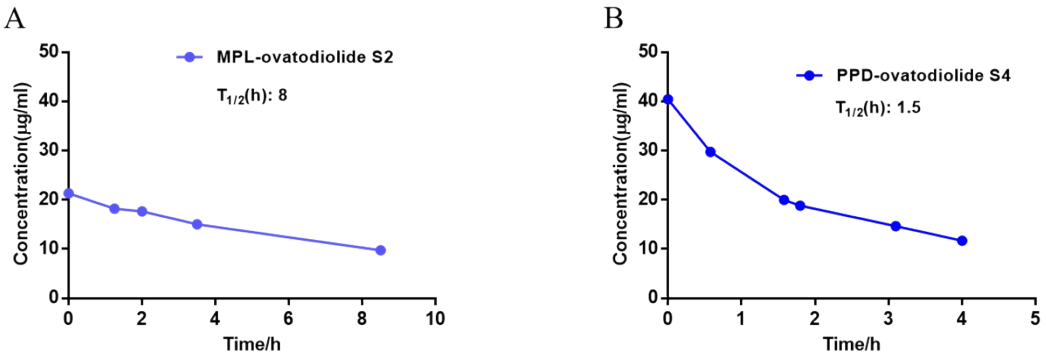
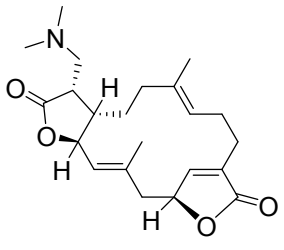


Figure S1. The stability of compounds in PBS buffer. A) MPL-ovatodiolide (**S2**), B) PPD-ovatodiolide (**S4**)

2.2.1.2 Calculate the C-N Bond Energy



S5

Computational Methods

The bond energies of Compoud **S1**, **S3**, **S5** and **8** were calculated with the Gaussian09 program[1]. Geometries of all molecular species were optimized using B3LYP[2-5] method and the 6-

31+G* basis set. Vibrational frequency calculations were carried out to ensure that the optimized geometries are indeed associated with local minima on the potential energy surfaces and to determine the thermal corrections to the Gibbs free energies. Single-point energy calculations were performed with the MP2 method and the 6-31+G* basis set.

Results

The bond energies of Compoud **S1**, **S3**, **S5** and **8** are 76.9, 76.6, 71.4 and 77.1 kcal/mol, respectively.

2.2.2 Materials and Experimental Procedure of compounds towards human liver microsomes

The HPLC−MS/MS system consist of an UltiMate 3000 High Performance Liquid Chromatograph (Dian Company) and QTRAP 5500 tandem triple quadrupole mass spectrometry (AB Sciex Company). All solvents and chemicals were of HPLC grade and purchased from TEDIA (USA). The whole incubation volume was 50 mL, including 0.2 mg/mL liver microsomes, 5 mM $MgCl_2$ and 1 μM ovatodiolide (**1**). The methanol ratio in the incubation system was less than 1% and the DMSO ratio was less than 0.1% when the appropriate amount of human liver microsome protein, ovatodiolide (**1**) and $MgCl_2$ were added to the cold 50 mM Tris-citric acid buffer (pH = 7.4). After incubation at 37 °C in water bath for 3 min, 1 μL of 50 mM NADPH (41.65 mg/mL) was added to initiate the reaction. After incubation for 0, 15, 30 and 60 min, 1000 mL methanol solution containing 1 ng/mL buspirone hydrochloride was added to the incubation system to stop the reaction. The system was swirled for 3 minutes and centrifuged for 12000 rpm for 5 minutes. Negative control group was set up. After incubation at 37 °C in water bath for 3 minutes, NADPH was not added to the incubation system. The residual amount of ovatodiolide (**1**) was detected after 2 μL injection of resultant solution, and three samples were parallel. NMP-ovatodiolide (**9**), diepoxyovatodiolide (**10**) and NMP-diepoxyovatodiolide (**12**) were followed the above procedure**.**

2.2.3 The pharmacokinetic study

The HPLC−MS/MS system consist of an UltiMate 3000 High Performance Liquid Chromatograph (Dian Company) and QTRAP 5500 tandem triple quadrupole mass spectrometry (AB Sciex Company). All solvents and chemicals were of HPLC grade and purchased from TEDIA (USA). Male SD rats (250～260 g) were supplied by Jinan Pengyue Laboratory Animal Breeding Co., Ltd. (Jinan, China). The experimental protocol was approved by the Nankai University Ethics Committee for the use of experimental animals and conformed to the Guide for Care and Use of Laboratory Animals. Rats were housed at 22 ± 2 °C and 55 ± 5% relative humidity under a 12 h light–dark cycle. They were fasted for 12 h before drug administration, and water was freely available. NMP-diepoxyovatodiolide (**12**) aqueous solution was p.o. (100 mg/kg) or i.v. (20 mg/kg) administered (n = 3). The blood samples were collected from the orbital veins at the setting time. All the biological specimens were stored at −80 °C. PK data were generated using noncompartmental analysis. DMA-ovatodiolide (**7**) were followed the above procedure**.**

2.2.4 Materials and experimental procedure of the tissue distribution study

The HPLC−MS/MS system consist of an UltiMate 3000 High Performance Liquid Chromatograph (Dian Company) and QTRAP 5500 tandem triple quadrupole mass spectrometry (AB Sciex Company). All solvents and chemicals were of HPLC grade and purchased from TEDIA (USA). The experimental protocol was approved by the Nankai University Ethics Committee for the use of experimental animals and conformed to the Guide for Care and Use of Laboratory Animals. Male SD rats (250～260 g) were supplied by Jinan Pengyue Laboratory Animal Breeding Co., Ltd. (Jinan, China). Rats were housed at 22 ± 2 °C and 55 ± 5% relative humidity under a 12 hours light−dark cycle. They were fasted for 12 hours before drug administration. NMP-ovatodiolide (**9**) aqueous solution and NMP-diepoxyovatodiolide (**12**) aqueous solution was p.o. (100 mg/kg) administered (n = 3) respectively. The blood samples were collected from the orbital veins at the setting time. All the biological specimens were stored at −20 °C. Tissue distribution data were generated using noncompartmental analysis.

2.2.5 Mechanism of action of diepoxyovatiodiade study

2.2.5.1 Spleen cell isolation

C57BL/6J mice aged 6-8 weeks were sacrificed, and their spleens were harvested. Single-cell suspensions were obtained through grinding and filtering with 70 μm cell strainers. After erythrocyte lysis and washing, the cell pellets were re-suspended with complete RPMI 1640 medium (supplemented with 10% fetal bovine serum and 1% penicillin and streptomycin). The experimental protocol was approved by the Nankai University Ethics Committee for the use of experimental animals and conformed to the Guide for Care and Use of Laboratory Animals.

2.2.5.2 Cytotoxicity assay

Isolated spleen cells were seeded into 96-well plates (2×10^6 per well). Different concentrations of Diepoxyova and 6 μM of ovatodiolide were added into corresponding wells. The plates were incubated at 37℃ with 5% CO_2 for 48 h. Then 10 μL of CCK-8 was added into each well, followed by 2 hour's incubation at 37℃. Shake the plates and read the OD value at 450 nm. The viabilities of the cells were calculated.

2.2.5.3 T cell differentiation and flow cytometry analysis

Isolated spleen cells were seeded into Anti-mouse CD3-precoated 24-well plates (2.5×10^6 per well). For Th1, the complete medium was supplemented with anti-mouse CD28 (1 μg/mL), rmIL-2 (2 ng/mL), rmIL-12 (5 ng/mL) and anti-mouse IL-4 (20 μg/mL). For Th2, the complete medium was supplemented with anti-mouse CD28 (1 μg/mL), rmIL-2 (2 ng/mL), rmIL-4 (20 ng/mL) and anti-mouse IFN-γ (20 μg/mL). Different concentrations of diepoxyovatodiolide and 6 μM of ovatodiolide were added into corresponding wells. The plates were incubated at 37℃ with 5% CO_2 for 48 h. The cells were collected, washed with PBS, and then fixed and permeabilized. Anti-mouse CD4-FITC and IFN-γ-APC antibodies were used to identify Th1. Anti-mouse CD4-FITC and IL-4-APC antibodies were used to identify Th2. The staining was proceeded in room temperature for 30 minutes in dark. At last, the stained cells were washed for two times and resuspended in PBS for flow cytometry analysis.

Results

In the cytotoxicity assay, the cytotoxic effects of ovatodiolide and diepoxyovatodiolide against spleen cells were assessed. It turned out that high concentration of these two compounds would significantly impair the viability of spleen cells (Fig. S3A). Thus, we chose 6 μM for ovatodiolide and 2.5, 5, 10 μM for diepoxyovatodiolide in subsequent assays as those doses did not showed cytotoxicity towards splenocyte.

Spleen cells were cultured with or without ovatodiolide and diepoxyovatodiolide for 48 hours. Flow cytometry analysis results revealed that 6 μM ovatodiolide significantly reduced the percentage of Th2 ($CD4^+IL\text{-}4^+$), compared with that of control group (8.48% vs 3.81%, Fig. S2C). Different concentrations of diepoxyovatodiolide displayed same effects with dose dependency (Fig. S2B, C). Similar results were obtained for Th1 ($CD4^+IFN\text{-}\gamma^+$) that ovatodiolide and diepoxyovatodiolide prevented spleen cells from differentiating into Th1(Fig. S2D). Those outcomes demonstrated that diepoxyovatodiolide might share the same mechanism with ovatodiolide.

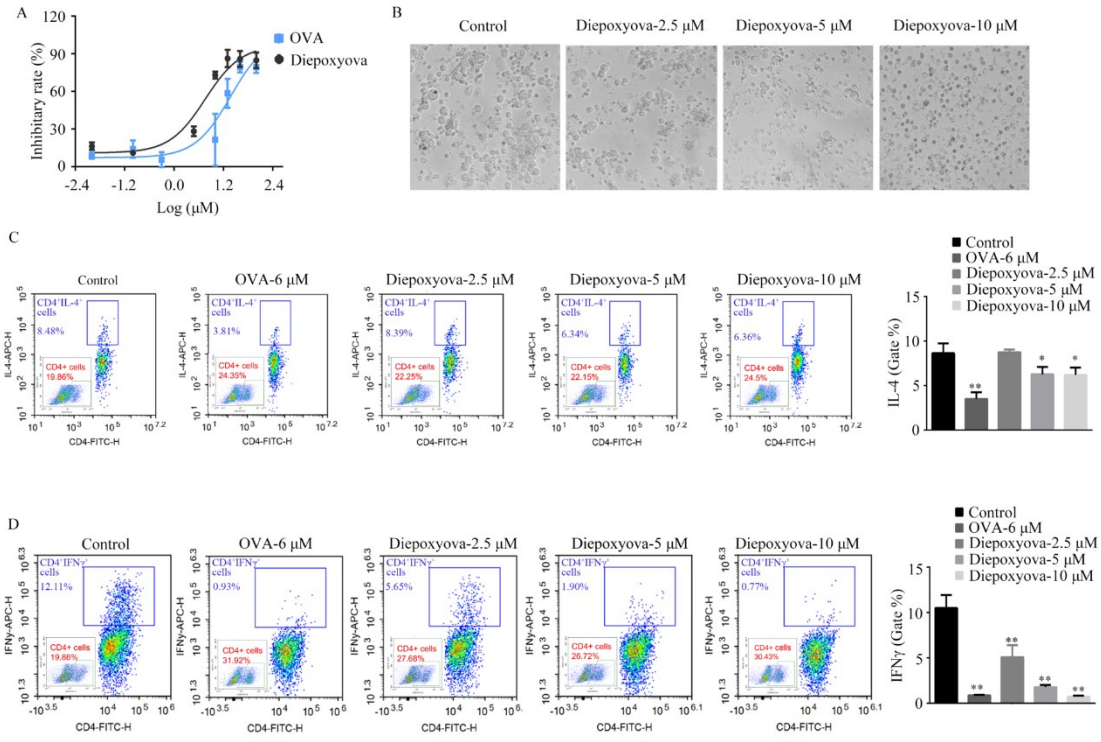


Figure S2: Impact of ovatodiolide and diepoxyova on spleen cells. A) Different concentrations of OVA and diepoxyova impaired the viability of spleen cells; B) Images of spleen cells cultured with or without diepoxyova polarizing into Th2. Diepoxyova suppressed Th2 activation and proliferation; C) Flow cytometry analysis of Th2 cells; D) Flow cytometry analysis of Th1 cells. The results shown represent the mean±SD values (n=3). OVA = ovatodiolide (**1**), Diepoxyova = diepoxyovatodiolide (**10**). Dead cells were excluded by employing a proper FSC-SSC threshold. Gating on $CD4^+$ cells was performed following standard leukocyte size gating and doublet exclusion. Th1 cells were identified using IFNγ and Th2 cells were identified using IL-4. $CD4^+$

cells and IFNγ$^+$ or IL-4$^+$ cells were sorted according to unstained controls.

2.2.6 Experimental procedure of the autoimmune hepatitis study

2.2.6.1 Animal test

Concanavalin A（ConA）(batch numbers: SLBX7517) purchased from SIGMA (USA); sodium chloride injection (batch numbers: 1809261902) purchased from Shijiazhuang Four Pharmaceutical Group Co., Ltd.; Mouse TNF alpha ELISA kit (batch numbers: GR3278815-1) purchased from Abcam; Mouse Interferon gamma ELISA kit（IFNG）(batch numbers: GR3280886-2) purchased from Abcam; Alanine aminotransferase assay kit (batch numbers: 20181005148) purchased from Shanghai Rongsheng Biopharmaceutical Co., Ltd. The experimental protocol was approved by the Nankai University Ethics Committee for the use of experimental animals and conformed to the Guide for Care and Use of Laboratory Animals.

C57BL/6J mice (20 ± 2 g) were supplied by Beijing Huafukang Laboratory Animal Technology Co., Ltd. (Beijing, China). Mice were housed at 22 ± 2 °C and 55 ± 5% relative humidity under a 12 hours light−dark cycle. They were fasted for 12 hours before drug administration.

Animals were randomly divided into groups according to their body weight and given for 14 consecutive days. One hour after the last administration, all groups except control group were injected ConA solution into tail vein to establish acute liver injury model. The dosage of ConA solution was 10 mg/kg. Blood was collected from canthus within 2 hours after injection and serum was separated. The content of TNF-alpha and IFN-gamma in serum were detected by ELASA kit. Blood was taken again after injection for 10 hours and serum was separated. Serum alanine aminotransferase (ALT) activity was detected by ELISA kit.

Test groups: NMP-diepoxyovatodiolide (**12**) (dissolved in saline) was administered orally once a day.

The model control group was administered orally the same equal volume of saline as test groups.

Control group: Ten mices of the same batch were randomly selected as control group on the day of blood collection, and the same volume of saline was injected into tail vein.

The medicine dosage of test groups: NMP-diepoxyovatodiolide (**12**) 400 mg/kg group; NMP-diepoxyovatodiolide (**12**) 100 mg/kg group; NMP-diepoxyovatodiolide (**12**) 25 mg/kg group; NMP-diepoxyovatodiolide (**12**) 5 mg/kg group; There were ten mices in each group, a total of 60. Mice's state was observed every day, body weight was measured every three days.

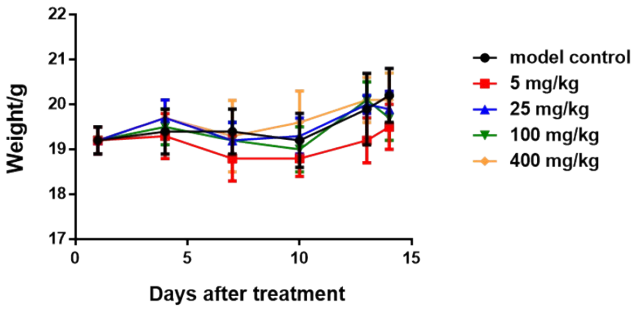


Figure S3: The weight of C57 mice after treated by NMP-diepoxyovatodiolid(**12**). The results

shown represent the mean±SD values (n=10)

2.2.6.2 CD69, CD64 stain and quantify the proportion of activated Th1 and Th2 cells in liver

A part of mice liver tissue was collected, grinded, filtered with 200 mesh sieve and centrifugated. Hepatic lymphocytes were obtained by density gradient centrifugation with 40% and 70% percoll solution. Then a part of the cells were stained by PE anti-mouse CD69 and FITC anti-mouse CD4, analyzed by flow cytometry. Other part of the cells were stained by FITC anti-mouse CD4， PE anti-mouse IFN-γ and APC anti-mouse IL-4, analyzed by flow cytometry to quantify the proportion of activated Th1 and Th2 cells in liver.

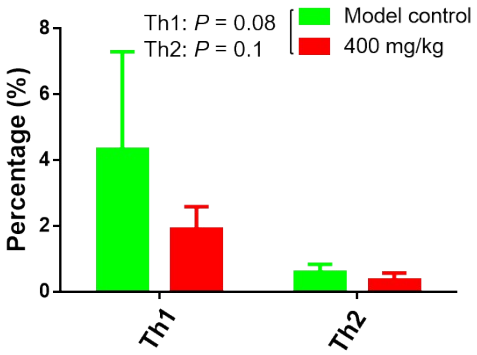
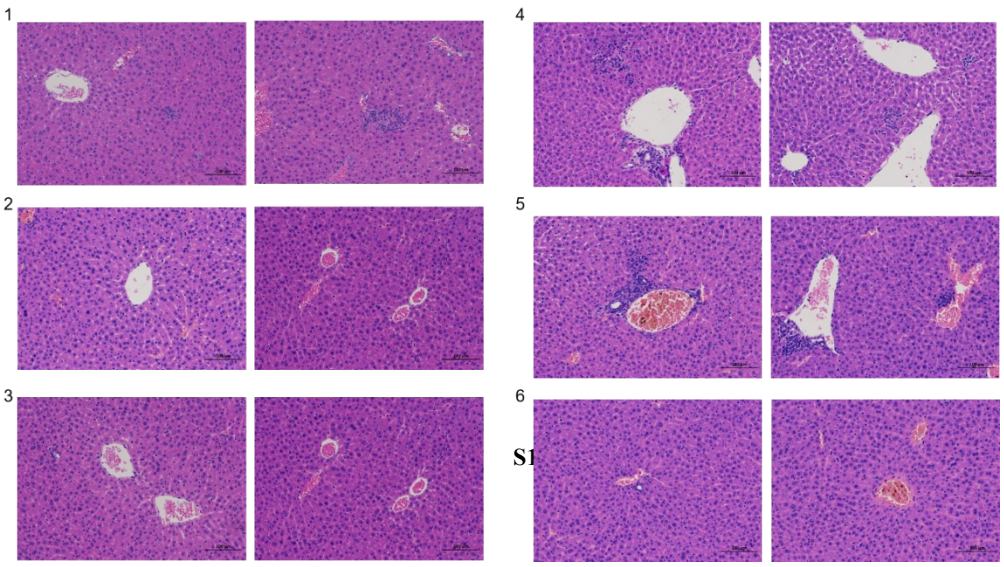


Figure S4: The percentage of activated Th1 and Th2 in liver tissue. The results shown represent the mean±SD values (n=6).

2.2.6.3 Histopathological Analysis

Another part of mice liver tissue was collected for histological analysis, the tissue sections were stained with hematoxylin and eosin (H&E).



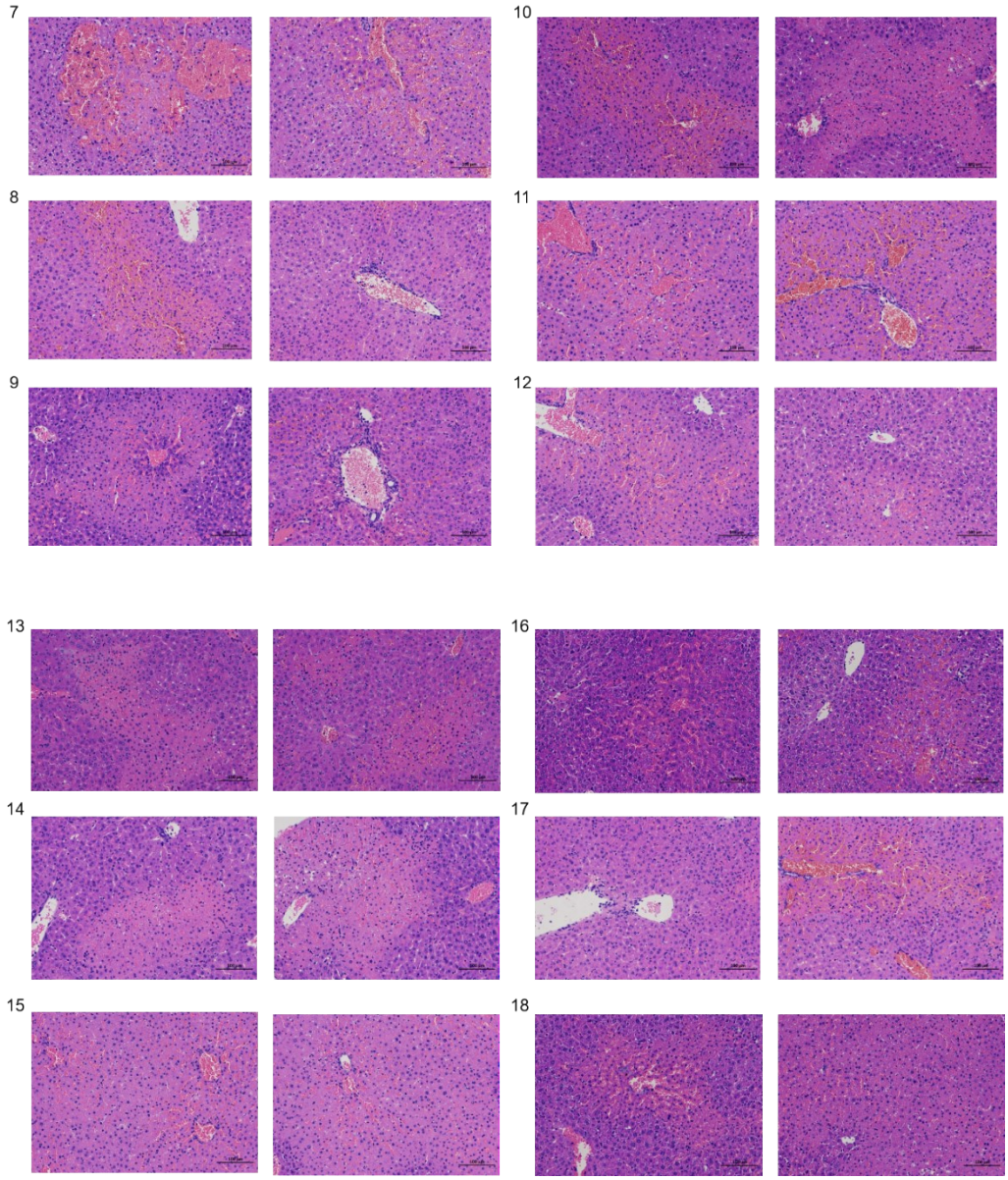


Figure S5: H&E stained histopathologic images of liver. Control group: 1 - 6; Model control group: 7 - 12; 400 mg/kg NMP-diepoxyovatodiolide treated group: 13 - 18

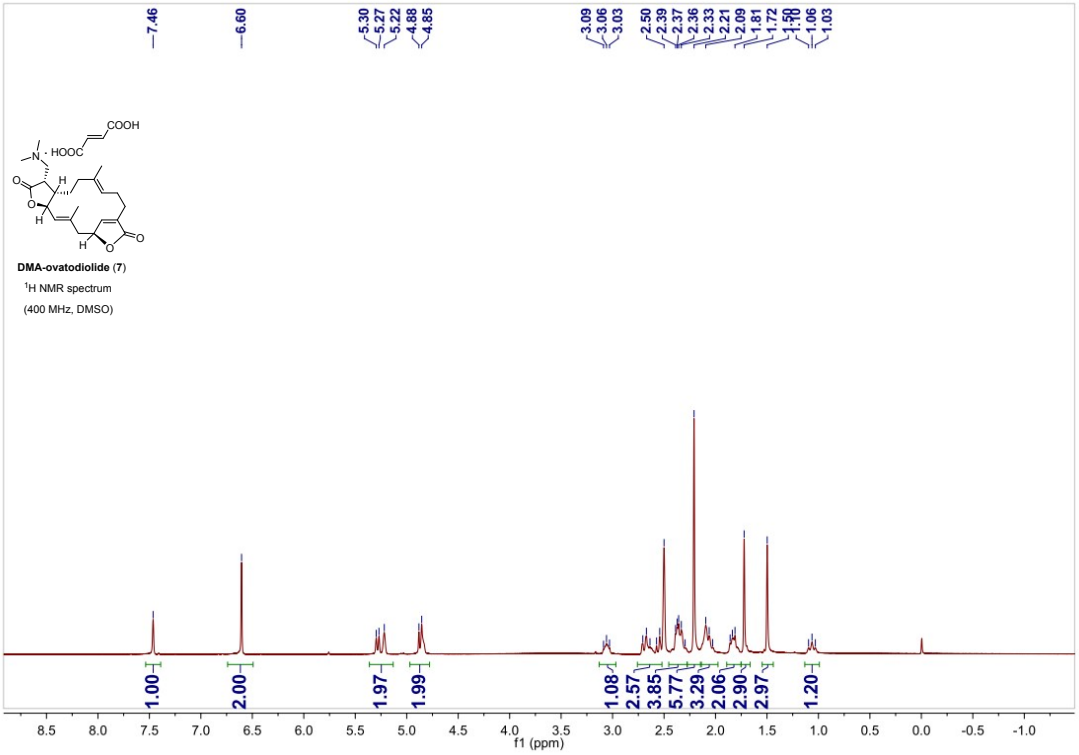
Table S1: Pathological score

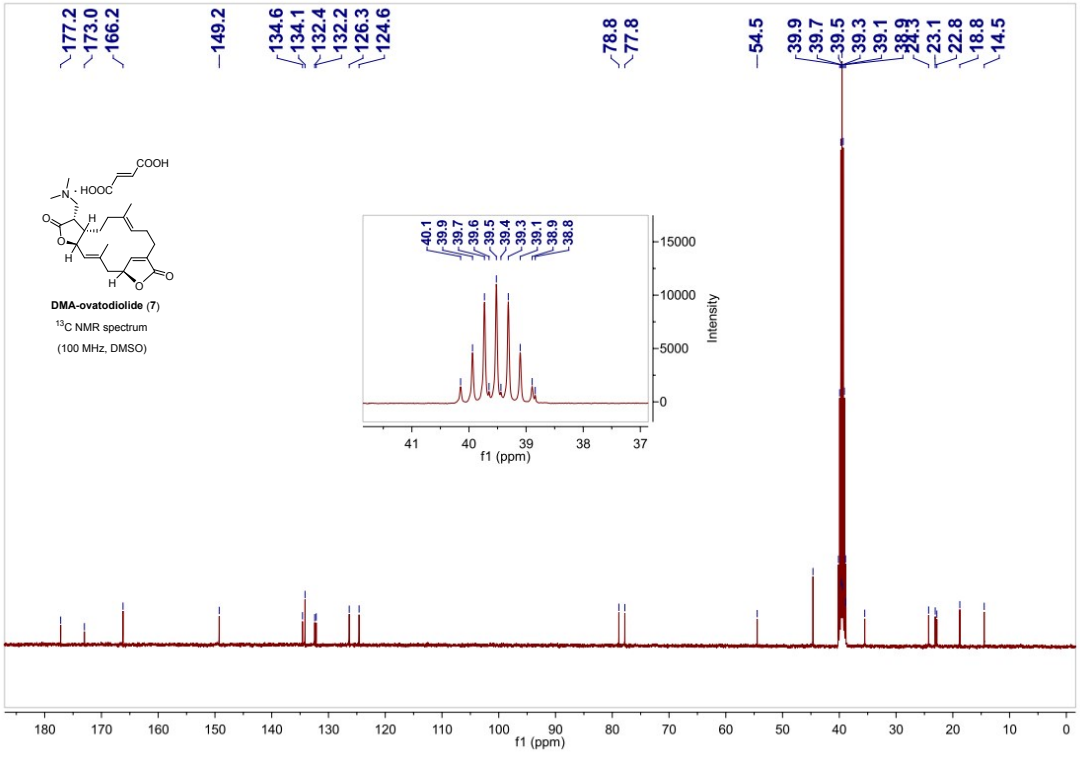
Sample Number	Necrosis	Inflammation	Congestion	Steatosis
1	0	1	0	0
2	0	0	0	0
3	0	0	0	0
4	1	1	0	0
5	1	1	0	0

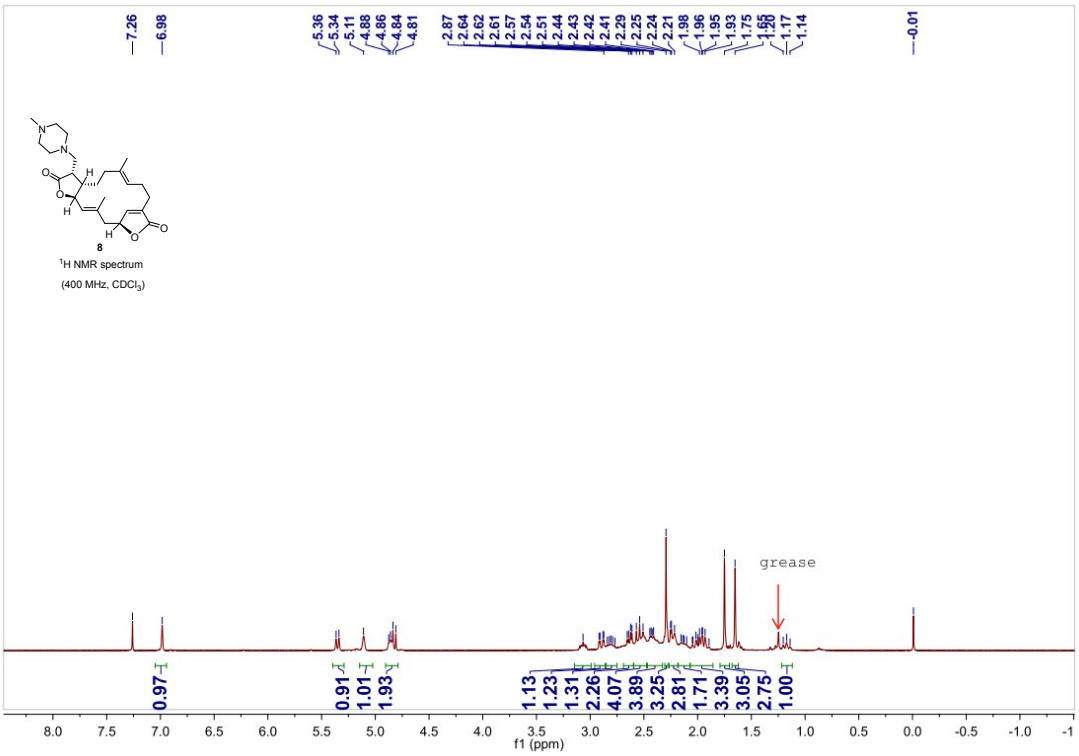
6	0	0	0	0
7	1	0	4	2
8	3	1	4	1
9	1	1	2	0
10	2	0	3	0
11	2	0	3	1
12	3	0	4	1
13	2	0	2	0
14	2	0	2	0
15	2	0	3	0
16	2	0	3	0
17	2	1	3	0
18	2	0	1	0

The degree of histopathological changes was divided into five grades. 0: no or very few changes; 1: mild or small changes; 2: moderate changes; 3: severe or multiple changes; 4: extremely severe or large changes.

3 ^{1}H and ^{13}C NMR Spectra

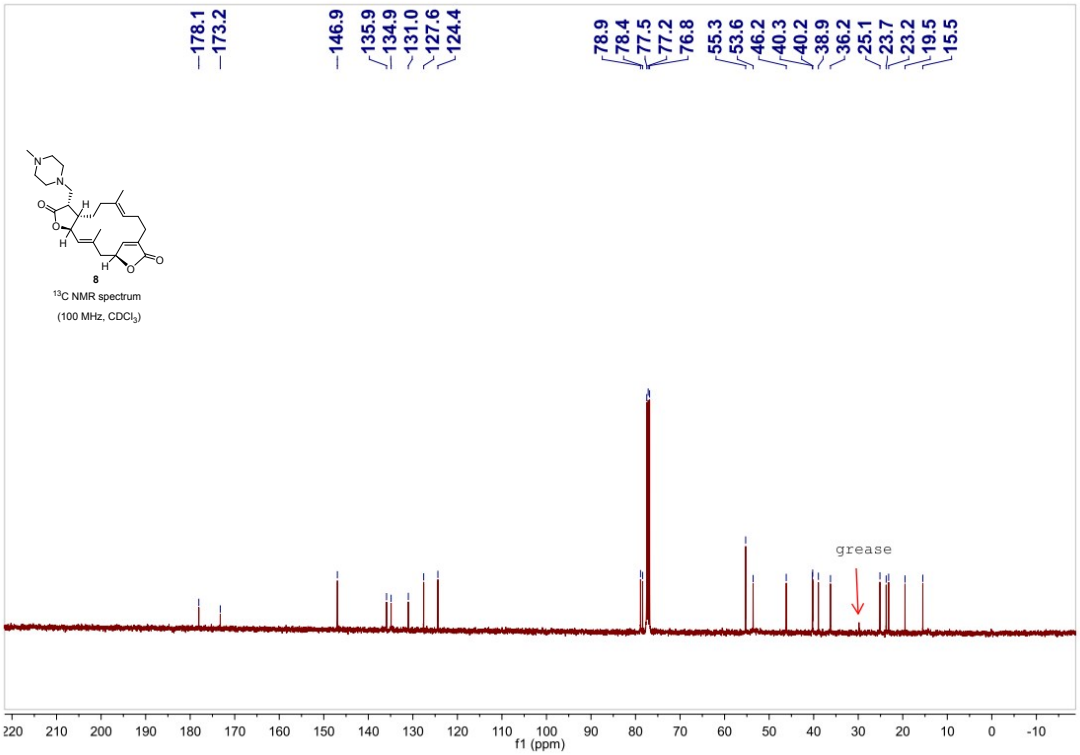






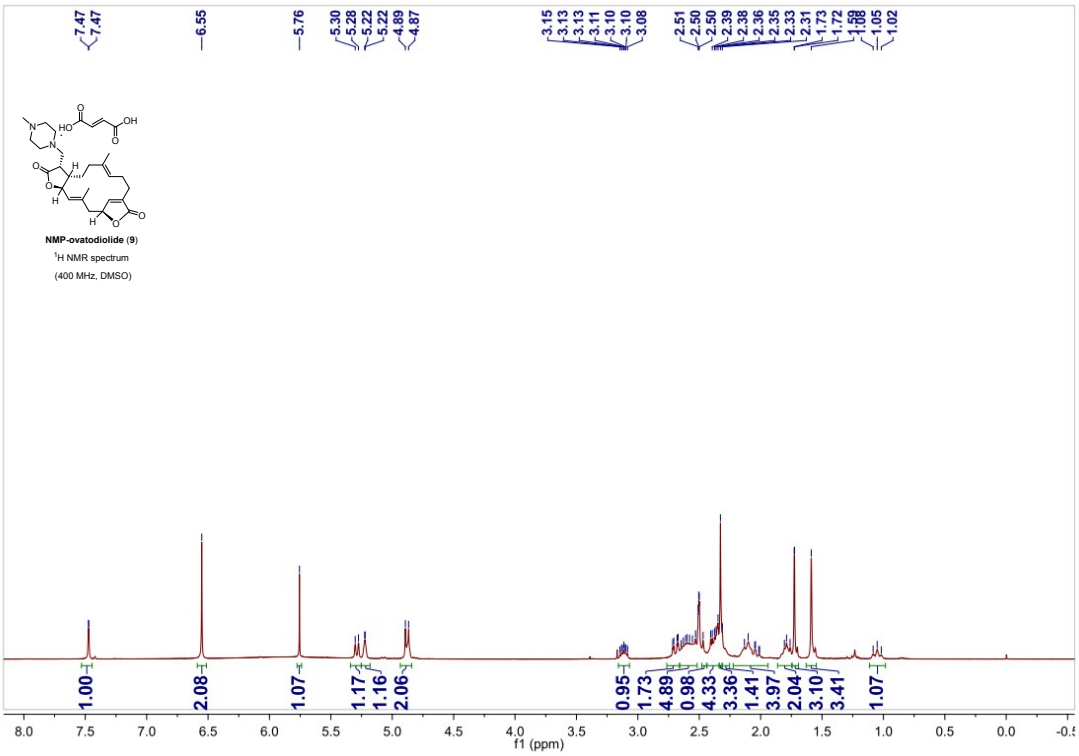
8

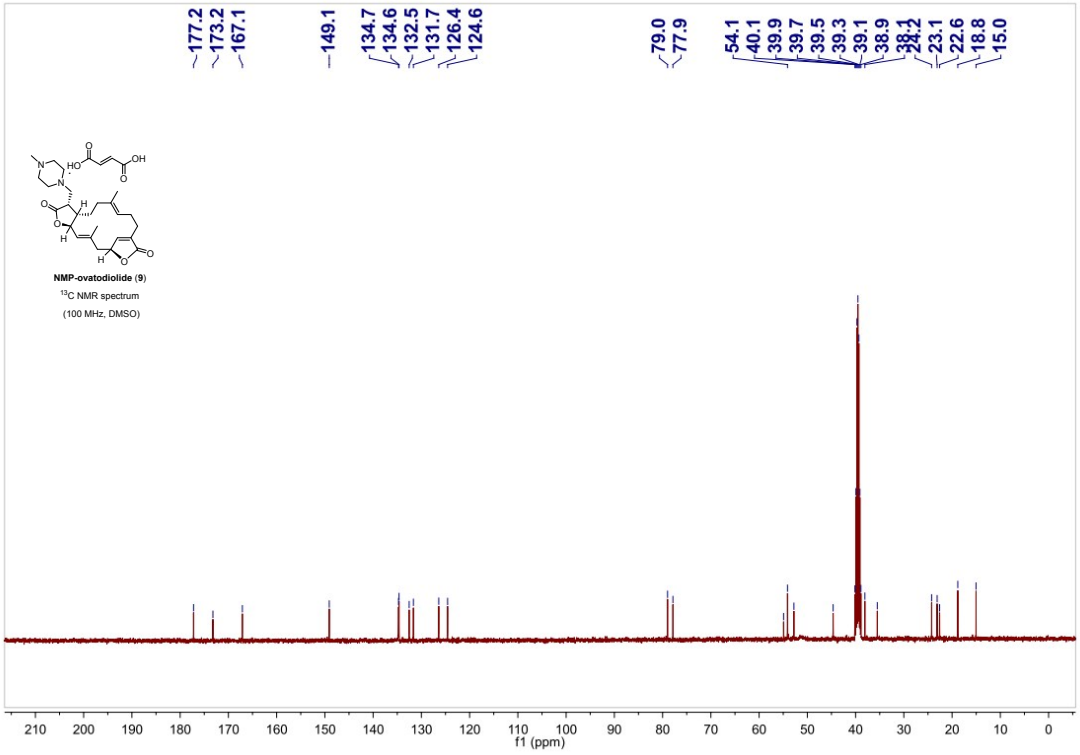
^{1}H NMR spectrum
(400 MHz, $CDCl_3$)

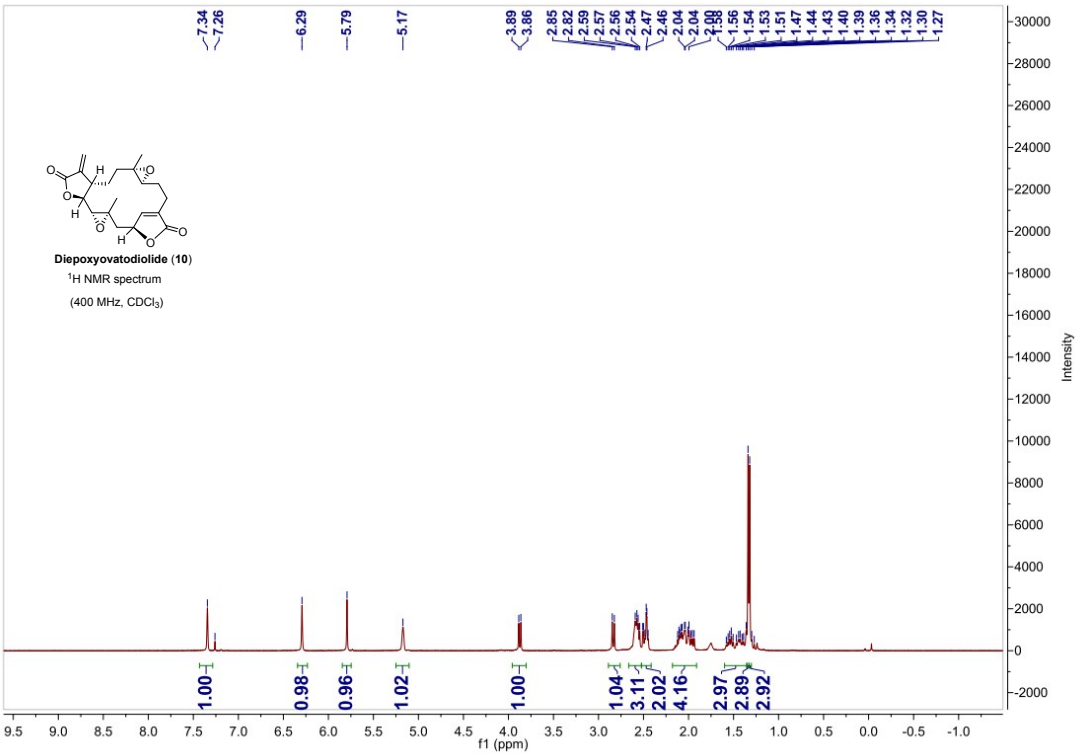


8

^{13}C NMR spectrum
(100 MHz, $CDCl_3$)


NMP-ovatodiolide (9)
1H NMR spectrum
(400 MHz, DMSO)
f1 (ppm)

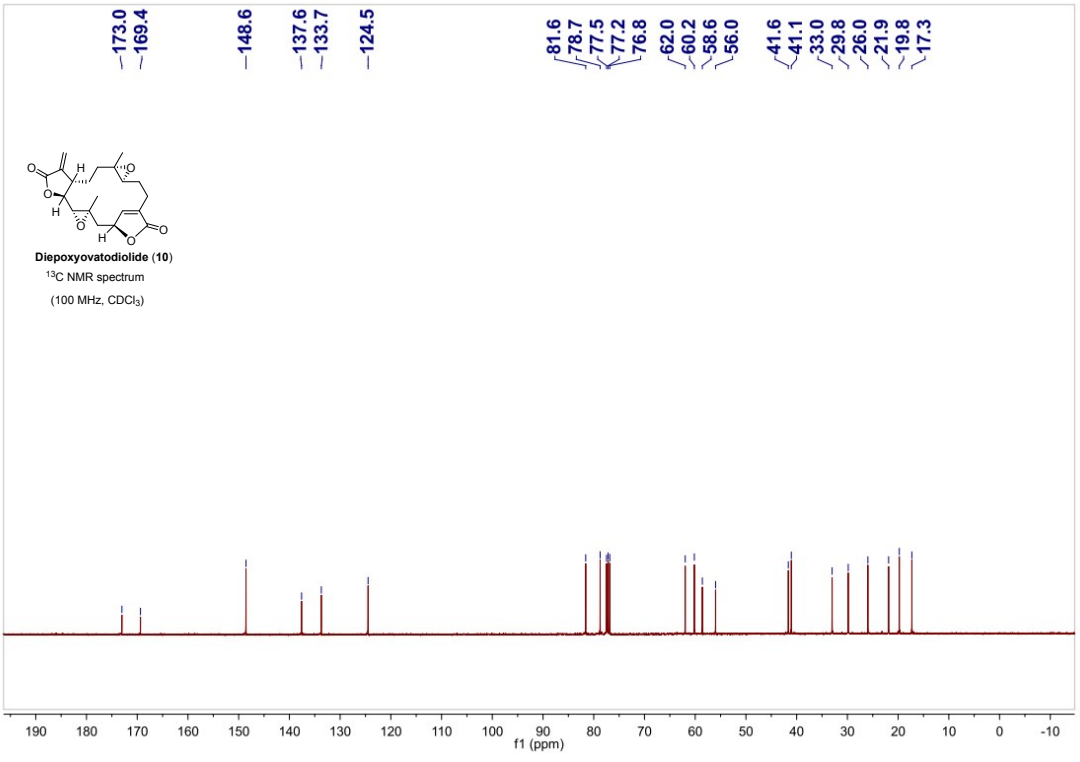

NMP-ovatodiolide (9)
13C NMR spectrum
(100 MHz, DMSO)
f1 (ppm)



Diepoxyovatodiolide (**10**)

^{1}H NMR spectrum

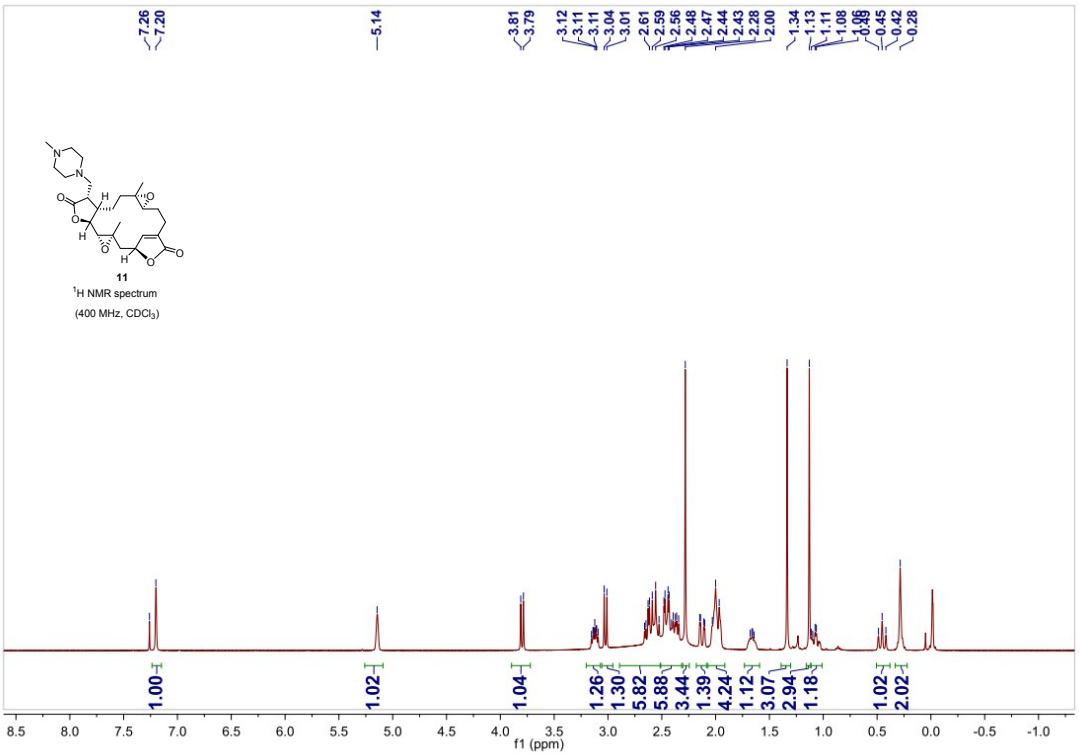
(400 MHz, $CDCl_3$)

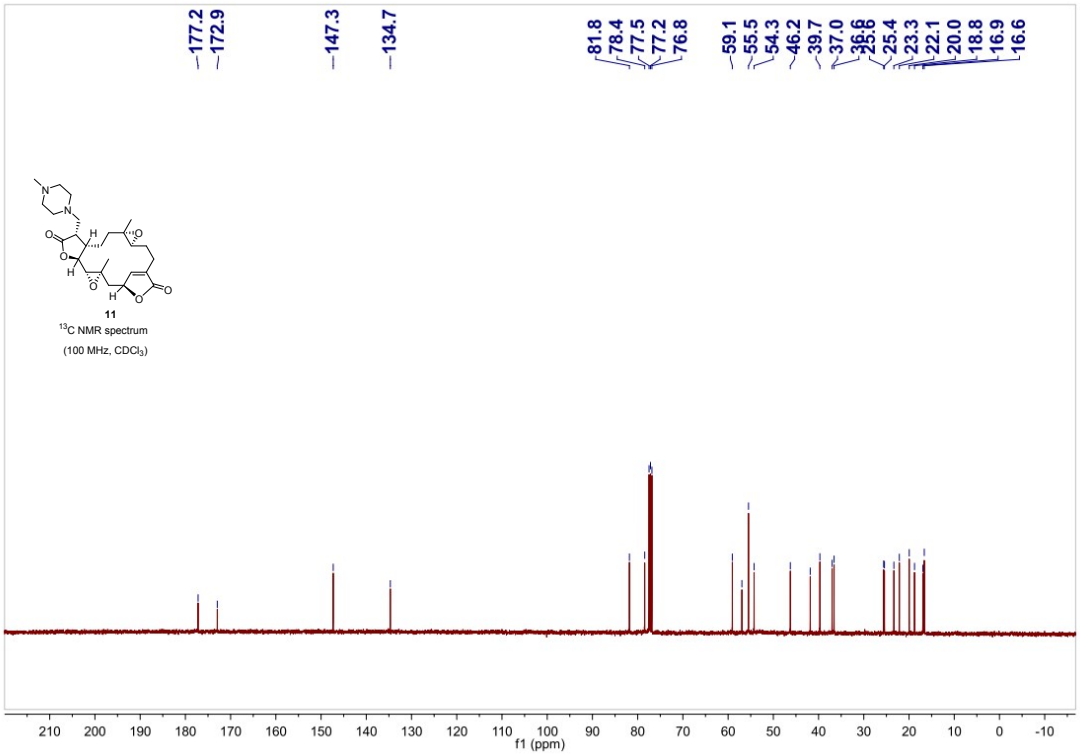


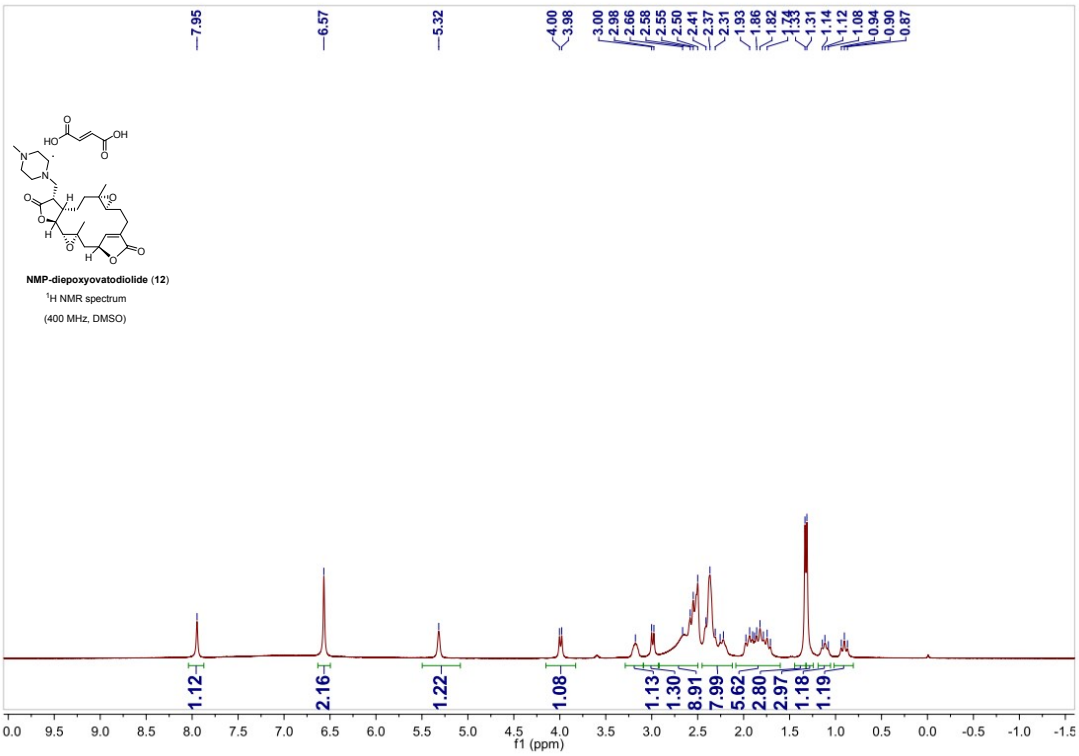
Diepoxyovatodiolide (**10**)

^{13}C NMR spectrum

(100 MHz, $CDCl_3$)


11
1H NMR spectrum
(400 MHz, CDCl3)
f1 (ppm)

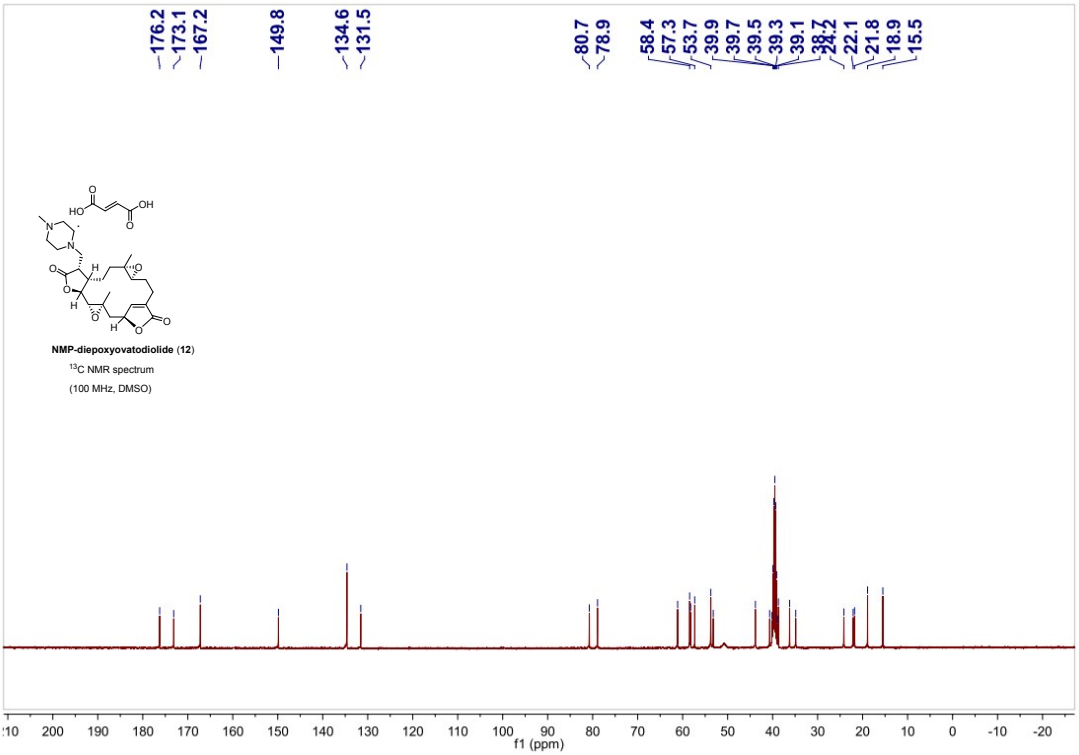

11
13C NMR spectrum
(100 MHz, CDCl3)
f1 (ppm)



NMP-diepoxyovatodiolide (**12**)

^{1}H NMR spectrum

(400 MHz, DMSO)

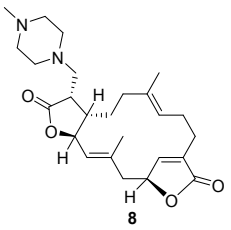


NMP-diepoxyovatodiolide (**12**)

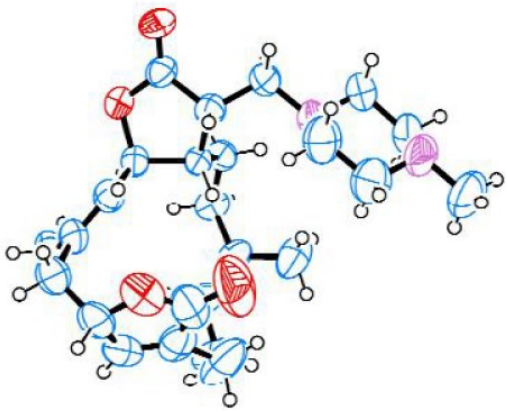
^{13}C NMR spectrum

(100 MHz, DMSO)

4 Crystallographic Data



8



ORTEP of 8 (CCDC 1886771)

Crystal data and structure refinement.

Identification code	P20181010d_sq
Empirical formula	$C_{25}H_{36}N_2O_4$
Formula weight	428.56
Temperature/K	294.15
Crystal system	monoclinic
Space group	$P2_1$
a/Å	13.93601(6)
b/Å	13.66097(8)
c/Å	14.35128(7)
α/°	90
β/°	92.6127(4)
γ/°	90
Volume/Å^3	2729.35(2)
Z	4
ρ_{calc}g/cm^3	1.043
μ/mm^{-1}	0.562
F(000)	928.0
Crystal size/mm^3	0.34 × 0.28 × 0.26
Radiation	CuKα (λ = 1.54184)
2Θ range for data collection/°	6.164 to 158.442

Index ranges	$-17 \leqslant h \leqslant 17$, $-16 \leqslant k \leqslant 15$, $-18 \leqslant l \leqslant 18$
Reflections collected	65502
Independent reflections	11249 [R_{int} = 0.0277, R_{sigma} = 0.0233]
Data/restraints/parameters	11249/1/566
Goodness-of-fit on F^2	1.063
Final R indexes [I>=2σ (I)]	R_1 = 0.0441, wR_2 = 0.1363
Final R indexes [all data]	R_1 = 0.0448, wR_2 = 0.1373
Largest diff. peak/hole / e $Å^{-3}$	0.28/-0.19
Flack parameter	0.01(7)

Fractional Atomic Coordinates (×10^4) and Equivalent Isotropic Displacement Parameters ($Å^2$×10^3). U_{eq} is defined as 1/3 of of the trace of the orthogonalised U_{IJ} tensor.

Atom	*x*	*y*	*z*	U(eq)
O1	12209.0(15)	7860(2)	1637.9(19)	95.2(7)
O2	11744(3)	8146(5)	3055(2)	163.7(18)
O3	8985.5(13)	5695.2(13)	1031.8(11)	65.9(4)
O4	8722.0(17)	4381.3(14)	1908.0(16)	83.5(5)
O5	2937.8(18)	2193(3)	949(3)	120.4(10)
O6	3084(4)	2121(4)	2514(4)	193(2)
O7	6369.2(13)	4103.6(13)	1094.9(11)	67.9(4)
O8	6523.8(19)	5420.4(15)	1994.2(17)	89.4(6)
N3	5799.5(15)	3456.0(16)	4271.5(13)	63.2(4)
N4	4877(2)	2916(2)	5943.5(17)	85.2(7)
C26	3202(3)	1755(4)	1752(4)	116.8(14)
C27	3693(2)	815(3)	1558(2)	84.2(8)
C28	3702(2)	750(3)	643(2)	82.7(8)
C29	3252(2)	1617(3)	169(3)	96.7(10)
C30	3871(2)	2251(3)	-418(2)	83.2(8)
C31	4887.6(18)	2357(2)	-27.4(16)	64.8(5)
C32	5093.0(15)	2969.2(18)	673.4(15)	58.4(5)
C33	6046.6(14)	3082.3(16)	1172.9(14)	53.6(4)
C34	6052.6(13)	2887.4(14)	2229.0(13)	48.1(4)
C35	5855.7(15)	3912.7(16)	2612.5(15)	55.0(4)
C36	6288.7(18)	4575.8(18)	1905.6(17)	63.6(5)
C37	7008.0(15)	2444.1(18)	2585.0(16)	60.0(5)
C38	7149.3(17)	1389(2)	2264(2)	69.7(6)
C39	6364.9(19)	694.3(18)	2545.5(18)	66.6(5)
C40	5803(2)	247(2)	1916(2)	76.8(7)
C41	4954(3)	-412(3)	2108(3)	101.5(11)
C42	4060(3)	171(3)	2324(3)	104.5(12)

C43	5597(3)	1699(3)	-463(2)	86.5(8)
C44	6247(3)	596(3)	3578(2)	90.6(9)
C45	6200.3(19)	4138.9(19)	3610.4(17)	65.3(5)
C46	4773(2)	3579(3)	4357(2)	83.3(8)
C47	4398(3)	2834(4)	5032(2)	98.1(10)
C48	5891(3)	2813(3)	5847(2)	89.0(9)
C49	6279(2)	3558(3)	5189.1(19)	80.7(7)
C50	4510(5)	2203(4)	6586(3)	125.3(16)
N1	9049.6(18)	6415.5(18)	4258.9(13)	71.9(5)
N2	9628(4)	7093(2)	6082.6(18)	116.3(13)
C1	11792(2)	8419(4)	2253(2)	99.2(11)
C2	11416.1(19)	9295(3)	1803(2)	86.4(9)
C3	11610(2)	9224(2)	911(2)	82.2(7)
C4	12118.3(19)	8301(3)	742(2)	82.2(8)
C5	11615(2)	7608(3)	41(2)	90.7(9)
C6	10557.1(19)	7446(2)	204.2(17)	70.3(6)
C7	10283.8(17)	6857(2)	880.2(17)	64.7(5)
C8	9273.5(15)	6717.8(17)	1163.2(14)	55.5(4)
C9	9124.9(14)	6928.3(15)	2205.7(13)	49.0(4)
C10	9279.1(16)	5906.3(16)	2634.1(15)	58.8(5)
C11	8955.0(17)	5219.0(17)	1860.4(17)	61.6(5)
C12	8139.6(14)	7365.2(18)	2358.0(15)	57.1(4)
C13	8006.2(15)	8397.7(19)	1969.8(18)	63.1(5)
C14	8725.8(18)	9139.0(18)	2352.6(19)	66.2(6)
C15	9272(2)	9637(2)	1784(3)	79.2(7)
C16	10008(3)	10399(3)	2041(4)	113.9(14)
C17	10942(3)	10038(4)	2379(4)	135(2)
C18	9883(3)	8017(3)	-429(2)	93.8(9)
C19	8754(3)	9268(3)	3386(3)	101.7(11)
C20	8799(2)	5684(2)	3540.5(17)	68.9(6)
C21	10048(3)	6374(4)	4592(3)	104.0(11)
C22	10247(4)	7164(4)	5310(3)	118.7(15)
C23	8647(4)	7130(3)	5726(2)	114.1(15)
C24	8423(3)	6322(3)	5033.5(19)	91.9(9)
C25	9821(7)	7869(4)	6763(3)	162(3)

Anisotropic Displacement Parameters ($Å^2×10^3$). The Anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2a^{*2}U_{11}+2hka^*b^*U_{12}+\ldots]$.

Atom	U_{11}	U_{22}	U_{33}	U_{23}	U_{13}	U_{12}
O1	68.4(11)	111.1(18)	105.6(16)	23.1(14)	-1.0(10)	7.4(11)

O2	152(3)	260(6)	77.0(16)	25(2)	-11.2(17)	-19(3)
O3	79.9(10)	64.5(10)	53.7(8)	-10.4(7)	6.1(7)	-10.1(8)
O4	108.7(15)	50.7(10)	92.0(13)	-9.3(8)	14.9(11)	-2.6(9)
O5	74.4(13)	130(2)	159(3)	5(2)	30.5(15)	15.6(14)
O6	236(5)	184(4)	170(4)	-37(3)	129(4)	5(4)
O7	79.3(10)	66.6(10)	57.8(8)	14.5(7)	3.1(7)	-16.3(8)
O8	115.6(16)	53.2(11)	98.5(14)	13.9(9)	-5.3(11)	-18.7(10)
N3	69.8(10)	66.2(11)	53.6(9)	-3.2(8)	4.0(7)	-3.0(9)
N4	112.8(19)	79.7(16)	65.2(12)	-4.9(10)	25.4(12)	-10.7(13)
C26	104(3)	120(3)	131(3)	-17(3)	63(3)	-16(2)
C27	64.4(13)	98(2)	90.4(18)	-5.9(15)	10.0(12)	-27.3(14)
C28	79.0(15)	81.8(19)	86.3(18)	-16.7(14)	-6.0(13)	-19.3(14)
C29	61.3(14)	110(3)	116(2)	-11(2)	-26.5(15)	-7.0(15)
C30	83.7(17)	85.8(19)	77.2(15)	-4.1(14)	-26.4(13)	-0.7(14)
C31	68.9(12)	70.8(14)	53.9(10)	0.8(10)	-4.1(9)	0.8(10)
C32	55.4(10)	63.4(13)	56.2(10)	0.5(9)	-0.7(8)	4.3(8)
C33	52.6(9)	57.0(11)	51.5(9)	1.8(8)	4.2(7)	-2.7(8)
C34	45.1(8)	47.7(10)	51.5(9)	3.7(7)	3.8(6)	-2.5(7)
C35	60.7(10)	48.5(11)	55.8(10)	2.7(8)	2.2(8)	-1.1(8)
C36	70.0(12)	54.2(13)	66.2(12)	11.5(9)	-2.8(9)	-6.6(9)
C37	49.6(9)	63.6(13)	66.6(11)	7.0(10)	-1.3(8)	1.5(8)
C38	60.3(11)	67.5(15)	82.2(15)	11.2(11)	11.6(10)	17.0(10)
C39	76.5(13)	50.3(12)	73.6(13)	12.0(10)	9.7(10)	16.0(10)
C40	99.9(19)	55.3(14)	75.7(15)	6.9(11)	7.6(13)	7.6(12)
C41	137(3)	63.9(19)	102(2)	16.4(16)	-8(2)	-23.0(18)
C42	104(2)	120(3)	90(2)	12(2)	14.6(17)	-47(2)
C43	91.8(18)	93(2)	74.7(16)	-20.8(15)	8.1(13)	3.4(15)
C44	112(2)	87(2)	73.3(16)	15.9(15)	8.0(15)	-4.7(17)
C45	77.3(13)	55.1(12)	63.1(12)	-4.5(9)	-0.9(10)	-8.2(10)
C46	74.9(15)	104(2)	70.8(15)	0.7(14)	5.7(12)	7.9(15)
C47	86.0(19)	125(3)	85.1(19)	-11.0(19)	21.4(15)	-15.8(19)
C48	120(2)	85(2)	61.9(14)	6.0(13)	-0.5(14)	-2.3(17)
C49	91.7(17)	87.8(19)	61.9(13)	0.1(12)	-5.2(12)	-11.4(15)
C50	181(5)	109(3)	90(2)	5(2)	49(3)	-31(3)
N1	91.7(14)	75.0(14)	48.7(9)	2.0(8)	0.6(9)	-2.8(11)
N2	201(4)	92(2)	52.6(12)	2.7(12)	-27.9(17)	-20(2)
C1	72.1(16)	149(4)	75.0(17)	9.8(19)	-8.4(13)	-21(2)
C2	58.6(13)	102(2)	99(2)	-28.4(17)	3.3(12)	-19.1(13)
C3	78.6(15)	73.4(17)	94.3(19)	15.1(14)	0.9(13)	-16.2(13)
C4	60.5(12)	96(2)	92.4(18)	1.8(15)	29.3(12)	-10.9(12)

C5	79.7(17)	101(2)	94.5(19)	-14.1(17)	42.5(15)	-9.1(15)
C6	72.1(13)	78.7(16)	62.0(11)	-13.2(11)	22.8(10)	-8.3(11)
C7	61.7(11)	70.2(14)	63.2(12)	-7.5(10)	12.8(9)	3.9(10)
C8	58.7(10)	59.5(12)	48.5(9)	-3.1(8)	4.2(7)	-2.2(8)
C9	51.2(9)	48.7(10)	47.0(9)	-3.6(7)	1.9(7)	-1.0(7)
C10	64.9(11)	54.2(12)	56.8(11)	-0.5(8)	-1.1(8)	3.4(9)
C11	67.5(12)	51.6(12)	66.1(12)	-8.8(9)	7.2(9)	1.6(9)
C12	49.5(9)	62.1(12)	60.0(10)	-2.0(9)	6.3(8)	-1.2(8)
C13	51.8(10)	60.5(13)	76.5(13)	-3.5(10)	-3.9(9)	6.3(9)
C14	65.8(12)	53.2(12)	79.0(14)	-12.7(10)	-6.1(10)	13.8(9)
C15	70.5(14)	56.0(14)	111(2)	-13.8(13)	3.3(13)	1.6(10)
C16	91(2)	67(2)	185(4)	-28(2)	10(2)	-7.9(15)
C17	88(2)	152(4)	164(4)	-91(4)	1(2)	-21(2)
C18	104(2)	115(3)	62.6(14)	17.2(15)	8.4(13)	-14.0(19)
C19	126(3)	91(2)	86(2)	-28.4(17)	-10.2(18)	13(2)
C20	89.7(16)	61.4(13)	55.5(11)	5.6(9)	2.3(10)	-2.4(11)
C21	105(2)	128(3)	77.3(18)	0.9(19)	-16.5(16)	5(2)
C22	130(3)	139(4)	83(2)	4(2)	-33(2)	-23(3)
C23	185(5)	100(3)	58.2(15)	-9.9(15)	24(2)	-21(3)
C24	128(3)	92(2)	56.8(13)	0.2(13)	15.9(15)	-16.3(19)
C25	281(8)	124(4)	77(2)	-13(2)	-46(3)	-33(4)

Bond Lengths.

Atom	**Atom**	**Length/Å**	**Atom**	**Atom**	**Length/Å**
O1	C1	1.321(5)	C39	C40	1.319(4)
O1	C4	1.421(4)	C39	C44	1.505(4)
O2	C1	1.214(5)	C40	C41	1.522(5)
O3	C8	1.463(3)	C41	C42	1.522(7)
O3	C11	1.358(3)	C46	C47	1.514(5)
O4	C11	1.192(3)	C48	C49	1.505(4)
O5	C26	1.336(7)	N1	C20	1.466(3)
O5	C29	1.452(5)	N1	C21	1.451(5)
O6	C26	1.220(6)	N1	C24	1.450(4)
O7	C33	1.472(3)	N2	C22	1.438(7)
O7	C36	1.339(3)	N2	C23	1.438(7)
O8	C36	1.205(3)	N2	C25	1.458(5)
N3	C45	1.460(3)	C1	C2	1.448(6)
N3	C46	1.450(4)	C2	C3	1.323(5)
N3	C49	1.456(3)	C2	C17	1.484(5)
N4	C47	1.446(5)	C3	C4	1.472(5)

N4	C48	1.433(5)	C4	C5	1.528(5)
N4	C50	1.450(4)	C5	C6	1.519(4)
C26	C27	1.487(6)	C6	C7	1.329(4)
C27	C28	1.317(5)	C6	C18	1.497(5)
C27	C42	1.480(6)	C7	C8	1.495(3)
C28	C29	1.489(5)	C8	C9	1.547(3)
C29	C30	1.508(5)	C9	C10	1.537(3)
C30	C31	1.506(4)	C9	C12	1.522(3)
C31	C32	1.329(3)	C10	C11	1.508(3)
C31	C43	1.495(4)	C10	C20	1.520(3)
C32	C33	1.489(3)	C12	C13	1.525(4)
C33	C34	1.538(3)	C13	C14	1.511(3)
C34	C35	1.534(3)	C14	C15	1.329(4)
C34	C37	1.529(3)	C14	C19	1.492(4)
C35	C36	1.507(3)	C15	C16	1.495(5)
C35	C45	1.521(3)	C16	C17	1.455(7)
C37	C38	1.529(4)	C21	C22	1.509(6)
C38	C39	1.516(4)	C23	C24	1.508(5)

Bond Angles.

Atom	**Atom**	**Atom**	**Angle/°**	**Atom**	**Atom**	**Atom**	**Angle/°**
C1	O1	C4	109.7(3)	N4	C48	C49	112.2(3)
C11	O3	C8	111.33(16)	N3	C49	C48	109.9(3)
C26	O5	C29	109.9(3)	C21	N1	C20	113.5(3)
C36	O7	C33	110.64(16)	C24	N1	C20	110.3(2)
C46	N3	C45	112.9(2)	C24	N1	C21	110.2(3)
C46	N3	C49	109.0(2)	C22	N2	C25	111.6(5)
C49	N3	C45	110.7(2)	C23	N2	C22	108.4(3)
C47	N4	C50	111.2(3)	C23	N2	C25	110.9(5)
C48	N4	C47	108.8(2)	O1	C1	C2	109.9(3)
C48	N4	C50	112.0(4)	O2	C1	O1	120.1(5)
O5	C26	C27	109.7(4)	O2	C1	C2	130.0(5)
O6	C26	O5	123.1(6)	C1	C2	C17	118.6(4)
O6	C26	C27	127.1(6)	C3	C2	C1	106.6(3)
C28	C27	C26	105.6(4)	C3	C2	C17	134.8(4)
C28	C27	C42	133.0(4)	C2	C3	C4	110.1(3)
C42	C27	C26	121.3(4)	O1	C4	C3	103.7(3)
C27	C28	C29	112.3(3)	O1	C4	C5	110.6(3)
O5	C29	C28	102.4(3)	C3	C4	C5	115.4(3)
O5	C29	C30	108.7(3)	C6	C5	C4	114.1(2)

C28	C29	C30	118.2(3)	C7	C6	C5	120.9(3)
C31	C30	C29	113.5(2)	C7	C6	C18	124.5(2)
C32	C31	C30	120.6(2)	C18	C6	C5	114.6(3)
C32	C31	C43	124.8(2)	C6	C7	C8	125.4(2)
C43	C31	C30	114.5(2)	O3	C8	C7	110.01(19)
C31	C32	C33	125.9(2)	O3	C8	C9	104.73(16)
O7	C33	C32	109.31(18)	C7	C8	C9	114.11(18)
O7	C33	C34	104.54(16)	C10	C9	C8	101.28(16)
C32	C33	C34	114.90(16)	C12	C9	C8	111.92(16)
C35	C34	C33	101.68(16)	C12	C9	C10	114.16(17)
C37	C34	C33	111.30(16)	C11	C10	C9	103.85(17)
C37	C34	C35	114.14(17)	C11	C10	C20	112.2(2)
C36	C35	C34	102.93(17)	C20	C10	C9	117.70(19)
C36	C35	C45	113.13(19)	O3	C11	C10	109.05(19)
C45	C35	C34	117.98(18)	O4	C11	O3	122.0(2)
O7	C36	C35	110.3(2)	O4	C11	C10	129.0(2)
O8	C36	O7	121.3(2)	C9	C12	C13	113.95(18)
O8	C36	C35	128.3(3)	C14	C13	C12	114.79(19)
C38	C37	C34	113.27(19)	C15	C14	C13	120.6(3)
C39	C38	C37	113.94(19)	C15	C14	C19	124.0(3)
C40	C39	C38	121.4(2)	C19	C14	C13	115.3(3)
C40	C39	C44	123.1(3)	C14	C15	C16	127.6(4)
C44	C39	C38	115.5(3)	C17	C16	C15	116.1(4)
C39	C40	C41	126.3(3)	C16	C17	C2	117.4(4)
C42	C41	C40	112.2(3)	N1	C20	C10	111.5(2)
C27	C42	C41	114.7(3)	N1	C21	C22	109.9(4)
N3	C45	C35	111.61(19)	N2	C22	C21	112.3(4)
N3	C46	C47	110.3(3)	N2	C23	C24	112.0(4)
N4	C47	C46	111.6(3)	N1	C24	C23	109.1(3)

Torsion Angles.

A	**B**	**C**	**D**	**Angle/°**	**A**	**B**	**C**	**D**	**Angle/°**
O1	C1	C2	C3	1.0(3)	C46	N3	C49	C48	58.1(4)
O1	C1	C2	C17	-178.2(3)	C47	N4	C48	C49	57.3(4)
O1	C4	C5	C6	69.5(4)	C48	N4	C47	C46	-56.5(4)
O2	C1	C2	C3	-176.4(4)	C49	N3	C45	C35	168.7(2)
O2	C1	C2	C17	4.3(6)	C49	N3	C46	C47	-58.0(3)
O3	C8	C9	C10	28.6(2)	C50	N4	C47	C46	179.7(4)
O3	C8	C9	C12	-93.5(2)	C50	N4	C48	C49	-179.4(3)
O5	C26	C27	C28	0.6(4)	N1	C21	C22	N2	57.1(5)

O5	C26	C27	C42	-179.8(3)	N2	C23	C24	N1	-59.8(4)
O5	C29	C30	C31	79.7(3)	C1	O1	C4	C3	1.7(3)
O6	C26	C27	C28	-176.0(5)	C1	O1	C4	C5	-122.7(3)
O6	C26	C27	C42	3.6(6)	C1	C2	C3	C4	0.1(3)
O7	C33	C34	C35	29.04(19)	C1	C2	C17	C16	-130.8(5)
O7	C33	C34	C37	-92.9(2)	C2	C3	C4	O1	-1.0(3)
N3	C46	C47	N4	58.2(4)	C2	C3	C4	C5	120.2(3)
N4	C48	C49	N3	-59.2(4)	C3	C2	C17	C16	50.2(7)
C26	O5	C29	C28	2.5(4)	C3	C4	C5	C6	-47.9(4)
C26	O5	C29	C30	-123.3(3)	C4	O1	C1	O2	176.0(4)
C26	C27	C28	C29	1.1(3)	C4	O1	C1	C2	-1.7(3)
C26	C27	C42	C41	-148.3(3)	C4	C5	C6	C7	-76.9(4)
C27	C28	C29	O5	-2.2(3)	C4	C5	C6	C18	101.3(3)
C27	C28	C29	C30	117.1(3)	C5	C6	C7	C8	173.7(2)
C28	C27	C42	C41	31.2(5)	C6	C7	C8	O3	119.0(3)
C28	C29	C30	C31	-36.3(4)	C6	C7	C8	C9	-123.6(3)
C29	O5	C26	O6	174.7(5)	C7	C8	C9	C10	-91.8(2)
C29	O5	C26	C27	-2.1(4)	C7	C8	C9	C12	146.2(2)
C29	C30	C31	C32	-78.6(4)	C8	O3	C11	O4	178.8(2)
C29	C30	C31	C43	98.9(3)	C8	O3	C11	C10	-2.7(2)
C30	C31	C32	C33	174.6(2)	C8	C9	C10	C11	-29.8(2)
C31	C32	C33	O7	121.6(3)	C8	C9	C10	C20	-154.4(2)
C31	C32	C33	C34	-121.3(3)	C8	C9	C12	C13	-68.3(2)
C32	C33	C34	C35	-90.8(2)	C9	C10	C11	O3	21.4(2)
C32	C33	C34	C37	147.31(19)	C9	C10	C11	O4	-160.3(3)
C33	O7	C36	O8	179.3(2)	C9	C10	C20	N1	-54.1(3)
C33	O7	C36	C35	-2.4(3)	C9	C12	C13	C14	-57.7(3)
C33	C34	C35	C36	-29.79(19)	C10	C9	C12	C13	177.39(18)
C33	C34	C35	C45	-155.12(18)	C11	O3	C8	C7	106.0(2)
C33	C34	C37	C38	-70.8(2)	C11	O3	C8	C9	-17.0(2)
C34	C35	C36	O7	21.2(2)	C11	C10	C20	N1	-174.5(2)
C34	C35	C36	O8	-160.6(3)	C12	C9	C10	C11	90.7(2)
C34	C35	C45	N3	-57.3(3)	C12	C9	C10	C20	-34.0(3)
C34	C37	C38	C39	-57.5(3)	C12	C13	C14	C15	122.6(3)
C35	C34	C37	C38	174.85(18)	C12	C13	C14	C19	-58.4(3)
C36	O7	C33	C32	106.0(2)	C13	C14	C15	C16	179.3(3)
C36	O7	C33	C34	-17.5(2)	C14	C15	C16	C17	80.5(5)
C36	C35	C45	N3	-177.5(2)	C15	C16	C17	C2	51.3(7)
C37	C34	C35	C36	90.2(2)	C17	C2	C3	C4	179.2(3)
C37	C34	C35	C45	-35.2(3)	C18	C6	C7	C8	-4.4(4)

C37	C38	C39	C40	116.8(3)	C19	C14	C15	C16	0.4(5)
C37	C38	C39	C44	-60.3(3)	C20	N1	C21	C22	178.7(3)
C38	C39	C40	C41	-175.8(3)	C20	N1	C24	C23	-175.7(3)
C39	C40	C41	C42	79.2(4)	C20	C10	C11	O3	149.5(2)
C40	C41	C42	C27	61.7(4)	C20	C10	C11	O4	-32.2(4)
C42	C27	C28	C29	-178.5(3)	C21	N1	C20	C10	-68.8(3)
C43	C31	C32	C33	-2.6(4)	C21	N1	C24	C23	58.1(4)
C44	C39	C40	C41	1.1(5)	C22	N2	C23	C24	58.2(4)
C45	N3	C46	C47	178.5(2)	C23	N2	C22	C21	-56.8(5)
C45	N3	C49	C48	-177.1(3)	C24	N1	C20	C10	167.0(3)
C45	C35	C36	O7	149.6(2)	C24	N1	C21	C22	-57.0(4)
C45	C35	C36	O8	-32.2(4)	C25	N2	C22	C21	-179.3(4)
C46	N3	C45	C35	-68.8(3)	C25	N2	C23	C24	-178.9(4)

Hydrogen Atom Coordinates (Å×10^4) and Isotropic Displacement Parameters (Å^2×10^3).

Atom	***x***	***y***	***z***	**U(eq)**
H28	3959	224	329	99
H29	2688	1403	-209	116
H30A	3884	1973	-1039	100
H30B	3583	2896	-475	100
H32	4596	3362	870	70
H33	6512	2649	889	64
H34	5524	2446	2370	58
H35	5159	4014	2570	66
H37A	7529	2843	2369	72
H37B	7040	2462	3261	72
H38A	7178	1381	1590	84
H38B	7761	1152	2523	84
H40	5940	342	1294	92
H41A	4813	-824	1569	122
H41B	5124	-836	2633	122
H42A	3556	-283	2477	125
H42B	4204	571	2871	125
H43A	5726	1937	-1074	130
H43B	5339	1049	-511	130
H43C	6183	1688	-84	130
H44A	6075	-65	3721	136
H44B	5751	1032	3766	136
H44C	6841	761	3907	136
H45A	6896	4103	3660	78

H45B	6013	4801	3767	78
H46A	4447	3498	3751	100
H46B	4642	4234	4578	100
H47A	3713	2932	5087	118
H47B	4494	2181	4788	118
H48A	6025	2161	5619	107
H48B	6218	2885	6455	107
H49A	6175	4211	5428	97
H49B	6965	3462	5142	97
H50A	4659	1555	6378	188
H50B	3826	2274	6608	188
H50C	4800	2308	7197	188
H3	11449	9690	458	99
H4	12761	8452	530	99
H5A	11674	7871	-582	109
H5B	11941	6981	67	109
H7	10761	6501	1204	78
H8	8845	7138	779	67
H9	9626	7375	2450	59
H10	9972	5814	2747	71
H12A	8038	7378	3022	69
H12B	7653	6943	2067	69
H13A	8047	8372	1297	76
H13B	7366	8622	2102	76
H15	9187	9495	1152	95
H16A	10099	10805	1497	137
H16B	9753	10815	2518	137
H17A	10870	9762	2994	162
H17B	11371	10595	2452	162
H18A	10028	7895	-1066	141
H18B	9953	8703	-297	141
H18C	9234	7820	-330	141
H19A	8111	9289	3596	153
H19B	9077	9870	3548	153
H19C	9094	8730	3678	153
H20A	8108	5674	3427	83
H20B	8998	5042	3762	83
H21A	10462	6464	4074	125
H21B	10185	5738	4867	125
H22A	10910	7116	5541	142

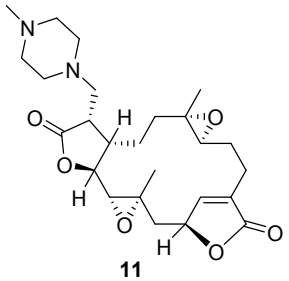
H22B	10160	7799	5017	142
H23A	8528	7757	5426	137
H23B	8222	7075	6240	137
H24A	8521	5690	5332	110
H24B	7758	6366	4809	110
H25A	9844	8487	6448	243
H25B	10427	7748	7089	243
H25C	9321	7881	7200	243

Solvent masks information.

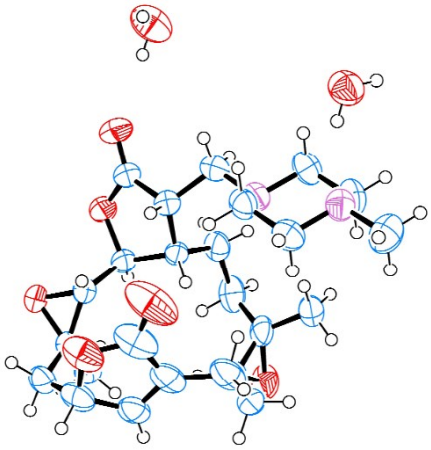
Number	X	Y	Z	Volume	Electroncount	Content
1	0.747	0.005	0.753	197	37	
2	0.253	0.505	0.247	197	37	

Crystal structure determination

Crystal Data for $C_{25}H_{36}N_2O_4$ (M =428.56 g/mol): monoclinic, space group $P2_1$ (no. 4), a = 13.93601(6) Å, b = 13.66097(8) Å, c = 14.35128(7) Å, β = 92.6127(4)°, V = 2729.35(2) Å^3, Z = 4, T = 294.15 K, μ(CuKα) = 0.562 mm^{-1}, *Dcalc* = 1.043 g/cm^3, 65502 reflections measured (6.164° ≤ 2Θ ≤ 158.442°), 11249 unique (R_{int} = 0.0277, R_{sigma} = 0.0233) which were used in all calculations. The final R_1 was 0.0441 (I > 2σ(I)) and wR_2 was 0.1373 (all data).



11



ORTEP of 11·2H$_2$O (CCDC 1886770)

Crystal data and structure refinement.

Identification code	P20180927a
Empirical formula	$C_{25}H_{40}N_2O_8$
Formula weight	496.59
Temperature/K	294.15
Crystal system	monoclinic
Space group	$P2_1$
a/Å	10.65993(6)
b/Å	11.44033(5)
c/Å	11.66689(6)
α/°	90
β/°	113.2519(6)
γ/°	90
Volume/Å^3	1307.249(13)
Z	2
ρ_{calc}g/cm^3	1.262
μ/mm^{-1}	0.773
F(000)	536.0
Crystal size/mm^3	0.28 × 0.24 × 0.22
Radiation	CuKα (λ = 1.54184)
2Θ range for data collection/°	8.248 to 157.924
Index ranges	-13 ⩽ h ⩽ 13, -14 ⩽ k ⩽ 14, -14 ⩽ l ⩽ 14
Reflections collected	31811
Independent reflections	5591 [R_{int} = 0.0238, R_{sigma} = 0.0117]
Data/restraints/parameters	5591/1/326
Goodness-of-fit on F^2	1.045
Final R indexes [I>=2σ (I)]	R_1 = 0.0320, wR_2 = 0.0908
Final R indexes [all data]	R_1 = 0.0323, wR_2 = 0.0911
Largest diff. peak/hole / e Å^{-3}	0.16/-0.14
Flack parameter	0.03(3)

Fractional Atomic Coordinates (×10^4) and Equivalent Isotropic Displacement Parameters (Å^2×10^3) for. U_{eq} is defined as 1/3 of of the trace of the orthogonalised U_{IJ}tensor.

Atom	*x*	*y*	*z*	U(eq)
O1	3185.6(15)	3567.6(11)	896.0(14)	57.6(3)
O2	2692(2)	2344.4(14)	2128(2)	84.3(6)
O3	5523.6(15)	4517.8(12)	471.3(15)	58.1(4)
O4	7137.0(16)	6470(2)	3429.1(15)	79.7(6)
O5	5926(2)	6425(3)	4608.2(19)	119.8(11)
O6	1345(2)	8798.8(19)	-123(2)	93.6(7)

N1	1929.9(16)	5498.4(15)	3906.1(15)	50.9(4)
N2	1911.2(18)	6827.8(17)	6003.2(16)	56.6(4)
C1	2907(2)	3327.1(17)	1903(2)	57.9(5)
C2	2934(2)	4440.7(16)	2614.0(19)	50.3(4)
C3	2706.3(16)	5389.8(14)	1612.8(16)	42.0(3)
C4	3386.7(18)	4825.9(15)	809.9(17)	45.2(4)
C5	4901.8(18)	5022.3(16)	1252.2(18)	46.4(4)
C6	5538.8(18)	5781.4(16)	604.8(18)	46.5(4)
C7	6959.3(19)	6270(2)	1307(2)	59.4(5)
C8	7110(2)	7116(2)	2352(2)	64.4(6)
C9	5995(2)	7979(2)	2120(2)	60.6(5)
C10	5412(2)	7825(2)	2926(2)	67.3(6)
C11	6142(3)	6857(3)	3759(2)	80.9(8)
C12	4246(3)	8444(3)	3068(3)	81.2(8)
C13	3355(3)	9156(2)	1943(4)	90.8(10)
C14	2583(2)	8377(2)	847(2)	66.0(6)
C15	1266(2)	7820(2)	644(2)	57.8(5)
C16	917(2)	6676(2)	-38.0(19)	65.1(6)
C17	1189.7(18)	5626.1(19)	828(2)	56.6(5)
C18	4709(2)	6425.0(18)	-576.4(18)	52.7(4)
C19	539(2)	8084(2)	1491(2)	65.6(6)
C20	1946(2)	4399.6(19)	3270(2)	58.8(5)
C21	3141.8(19)	5637(2)	5059(2)	56.0(5)
C22	3107(2)	6780(2)	5700(2)	60.5(5)
C23	691(2)	6695(3)	4860(2)	65.4(6)
C24	728(2)	5560(2)	4214(2)	61.8(5)
C25	1863(3)	7912(3)	6649(3)	81.1(7)
O7	1401(2)	852(3)	3369(2)	96.3(7)
O8	1466.1(18)	4951.9(18)	7515.2(17)	75.2(5)

Anisotropic Displacement Parameters ($Å^2×10^3$). The Anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2a^{*2}U_{11}+2hka^*b^*U_{12}+\ldots]$.

Atom	U_{11}	U_{22}	U_{33}	U_{23}	U_{13}	U_{12}
O1	64.1(8)	39.7(7)	77.4(9)	-13.2(6)	36.8(7)	-9.5(6)
O2	110.4(15)	38.9(7)	126.3(16)	3.0(9)	71.0(13)	-6.4(8)
O3	59.6(8)	44.0(7)	82.9(9)	3.1(6)	41.1(7)	9.5(6)
O4	49.1(8)	113.7(15)	61.0(8)	14.8(9)	5.4(7)	-16.8(9)
O5	82.3(13)	203(3)	65.1(10)	26.0(15)	20.0(9)	-37.5(16)
O6	105.1(14)	92.3(14)	110.2(14)	60.4(12)	71.2(12)	58.0(12)
N1	45.7(8)	56.9(9)	52.2(8)	2.0(7)	21.5(6)	0.4(7)

N2	58.0(9)	61.2(10)	51.5(8)	3.8(7)	22.7(7)	7.1(8)
C1	60.8(11)	38.6(9)	82.8(13)	-2.3(9)	37.4(10)	-4.6(8)
C2	51.4(9)	42.8(9)	60.5(10)	-1.1(8)	26.2(8)	-2.5(7)
C3	35.0(7)	35.4(8)	54.3(9)	-5.3(6)	16.2(7)	-4.6(6)
C4	43.8(8)	37.5(8)	55.1(9)	-4.0(7)	20.3(7)	-3.0(6)
C5	41.2(8)	42.6(8)	56.7(9)	3.5(7)	20.8(7)	5.4(7)
C6	41.2(8)	41.4(8)	60.2(10)	3.2(7)	23.7(8)	3.3(7)
C7	37.3(8)	64.2(12)	76.2(13)	9.2(1)	22.0(8)	0.5(8)
C8	39.2(9)	79.7(15)	67.1(12)	4.2(1)	13.1(8)	-17.8(9)
C9	56.1(11)	59.0(11)	68.3(12)	-10.2)	26.3(9)	-21.1(9)
C10	57.4(11)	80.6(15)	64.5(12)	-2411)	24.7(10)	-33.8(11)
C11	60.9(13)	119(2)	49.1(10)	-0.2(13)	6.9(9)	-34.3(14)
C12	84.5(17)	84.9(17)	93.0(17)	-42.8(15)	55.0(15)	-41.5(15)
C13	108(2)	42.9(11)	165(3)	-18.8(15)	101(2)	-12.0(12)
C14	73.3(14)	55.7(11)	88.8(15)	21.4(11)	53.4(13)	22.9(10)
C15	56.1(11)	61.6(12)	61.1(11)	20.0(9)	28.9(9)	26.8(9)
C16	44.8(9)	91.9(17)	49.0(9)	-1.5(10)	8.3(8)	17.3(10)
C17	35.8(8)	59.7(11)	69.2(12)	-14.9(9)	15.4(8)	-6.4(8)
C18	53.3(10)	49.1(9)	56.5(10)	3.4(8)	22.5(8)	0.1(8)
C19	64.7(13)	65.8(13)	79.2(14)	6.5(11)	42.2(11)	12.4(10)
C20	60.6(11)	52.5(11)	71.6(12)	0.4(9)	35.1(10)	-8.4(9)
C21	42.8(9)	65.6(12)	60.1(10)	8.1(9)	20.7(8)	5.9(8)
C22	54.3(10)	69.3(13)	56.4(10)	1.6(9)	20.2(8)	-6.2(10)
C23	52.1(10)	85.6(16)	58.3(11)	3.7(11)	21.6(9)	15.8(11)
C24	41.9(9)	83.7(15)	59.1(11)	-3.8(10)	19.4(8)	-2.8(9)
C25	97.8(19)	71.4(15)	73.8(14)	-6.2(12)	33.5(14)	12.1(14)
O7	86.8(13)	121.9(19)	83.8(12)	-11.6(13)	37.5(11)	-31.1(13)
O8	71.2(10)	86.1(12)	71.0(10)	21.1(9)	31.0(8)	8.6(9)

Bond Lengths.

Atom	**Atom**	**Length/Å**	**Atom**	**Atom**	**Length/Å**
O1	C1	1.349(3)	C3	C4	1.535(2)
O1	C4	1.465(2)	C3	C17	1.535(2)
O2	C1	1.197(3)	C4	C5	1.506(2)
O3	C5	1.442(2)	C5	C6	1.480(3)
O3	C6	1.453(2)	C6	C7	1.515(3)
O4	C8	1.448(3)	C6	C18	1.504(3)
O4	C11	1.339(4)	C7	C8	1.515(4)
O5	C11	1.208(3)	C8	C9	1.486(4)

O6	C14	1.441(3)	C9	C10	1.327(3)
O6	C15	1.456(3)	C10	C11	1.476(4)
N1	C20	1.463(3)	C10	C12	1.496(4)
N1	C21	1.460(3)	C12	C13	1.519(5)
N1	C24	1.463(2)	C13	C14	1.509(4)
N2	C22	1.453(3)	C14	C15	1.473(3)
N2	C23	1.458(3)	C15	C16	1.501(4)
N2	C25	1.462(3)	C15	C19	1.507(3)
C1	C2	1.514(3)	C16	C17	1.522(3)
C2	C3	1.542(2)	C21	C22	1.514(3)
C2	C20	1.528(3)	C23	C24	1.509(4)

Bond Angles.

Atom	Atom	Atom	Angle/°	Atom	Atom	Atom	Angle/°
C1	O1	C4	110.40(14)	C18	C6	C7	114.54(16)
C5	O3	C6	61.49(12)	C8	C7	C6	115.86(17)
C11	O4	C8	109.5(2)	O4	C8	C7	109.4(2)
C14	O6	C15	61.12(14)	O4	C8	C9	103.7(2)
C21	N1	C20	112.19(17)	C9	C8	C7	117.05(18)
C21	N1	C24	108.10(15)	C10	C9	C8	110.3(2)
C24	N1	C20	110.63(17)	C9	C10	C11	107.1(2)
C22	N2	C23	108.98(17)	C9	C10	C12	131.1(3)
C22	N2	C25	111.7(2)	C11	C10	C12	121.8(2)
C23	N2	C25	111.10(19)	O4	C11	C10	109.3(2)
O1	C1	C2	110.02(15)	O5	C11	O4	122.0(3)
O2	C1	O1	120.6(2)	O5	C11	C10	128.7(3)
O2	C1	C2	129.4(2)	C10	C12	C13	114.9(2)
C1	C2	C3	102.37(16)	C14	C13	C12	111.16(19)
C1	C2	C20	112.37(16)	O6	C14	C13	119.4(2)
C20	C2	C3	117.86(16)	O6	C14	C15	59.92(15)
C4	C3	C2	101.26(13)	C15	C14	C13	123.9(2)
C17	C3	C2	112.93(15)	O6	C15	C14	58.96(15)
C17	C3	C4	110.49(15)	O6	C15	C16	114.05(19)
O1	C4	C3	104.72(14)	O6	C15	C19	114.70(17)
O1	C4	C5	106.82(14)	C14	C15	C16	118.91(18)
C5	C4	C3	115.44(14)	C14	C15	C19	120.8(2)
O3	C5	C4	114.98(16)	C16	C15	C19	116.0(2)
O3	C5	C6	59.64(11)	C15	C16	C17	113.22(16)
C6	C5	C4	123.98(16)	C16	C17	C3	114.53(16)
O3	C6	C5	58.88(11)	N1	C20	C2	112.08(16)
O3	C6	C7	113.01(15)	N1	C21	C22	111.34(17)

O3	C6	C18	114.28(16)	N2	C22	C21	110.33(18)
C5	C6	C7	120.16(17)	N2	C23	C24	110.90(18)
C5	C6	C18	122.19(15)	N1	C24	C23	111.21(19)

Torsion Angles.

A	B	C	D	Angle/°	A	B	C	D	Angle/°
O1	C1	C2	C3	21.6(2)	C8	O4	C11	O5	177.4(3)
O1	C1	C2	C20	148.98(18)	C8	O4	C11	C10	-1.7(3)
O1	C4	C5	O3	65.78(19)	C8	C9	C10	C11	1.2(2)
O1	C4	C5	C6	134.76(18)	C8	C9	C10	C12	-178.8(2)
O2	C1	C2	C3	-159.0(3)	C9	C10	C11	O4	0.3(3)
O2	C1	C2	C20	-31.5(4)	C9	C10	C11	O5	-178.7(3)
O3	C5	C6	C7	-100.21(19)	C9	C10	C12	C13	17.0(3)
O3	C5	C6	C18	100.8(2)	C10	C12	C13	C14	67.3(3)
O3	C6	C7	C8	-131.08(19)	C11	O4	C8	C7	-123.3(2)
O4	C8	C9	C10	-2.2(2)	C11	O4	C8	C9	2.4(2)
O6	C14	C15	C16	102.2(2)	C11	C10	C12	C13	-163.0(2)
O6	C14	C15	C19	-102.1(2)	C12	C10	C11	O4	-179.66(19)
O6	C15	C16	C17	165.34(17)	C12	C10	C11	O5	1.3(4)
N1	C21	C22	N2	59.7(2)	C12	C13	C14	O6	157.0(2)
N2	C23	C24	N1	-58.8(2)	C12	C13	C14	C15	85.3(3)
C1	O1	C4	C3	-19.6(2)	C13	C14	C15	O6	107.1(3)
C1	O1	C4	C5	103.30(18)	C13	C14	C15	C16	-150.7(2)
C1	C2	C3	C4	-31.34(17)	C13	C14	C15	C19	5.1(3)
C1	C2	C3	C17	86.82(18)	C14	O6	C15	C16	-110.4(2)
C1	C2	C20	N1	-177.31(18)	C14	O6	C15	C19	112.4(2)
C2	C3	C4	O1	31.36(16)	C14	C15	C16	C17	98.8(2)
C2	C3	C4	C5	-85.77(17)	C15	O6	C14	C13	-114.5(2)
C2	C3	C17	C16	171.40(16)	C15	C16	C17	C3	-68.3(2)
C3	C2	C20	N1	-58.7(2)	C17	C3	C4	O1	-88.54(17)
C3	C4	C5	O3	-178.27(14)	C17	C3	C4	C5	154.32(15)
C3	C4	C5	C6	-109.3(2)	C18	C6	C7	C8	95.7(2)
C4	O1	C1	O2	179.0(2)	C19	C15	C16	C17	-58.1(2)
C4	O1	C1	C2	-1.4(2)	C20	N1	C21	C22	179.82(16)
C4	C3	C17	C16	-76.0(2)	C20	N1	C24	C23	-179.42(18)
C4	C5	C6	O3	-101.26(19)	C20	C2	C3	C4	-155.16(16)
C4	C5	C6	C7	158.52(17)	C20	C2	C3	C17	-37.0(2)
C4	C5	C6	C18	-0.5(3)	C21	N1	C20	C2	-75.3(2)
C5	O3	C6	C7	112.41(19)	C21	N1	C24	C23	57.4(2)
C5	O3	C6	C18	-114.23(17)	C22	N2	C23	C24	58.0(3)
C5	C6	C7	C8	-64.8(2)	C23	N2	C22	C21	-58.1(2)

C6	O3	C5	C4	116.21(17)	C24	N1	C20	C2 163.93(18)
C6	C7	C8	O4	77.0(2)	C24	N1	C21	C22 -57.9(2)
C6	C7	C8	C9	-40.5(3)	C25	N2	C22	C21 178.76(19)
C7	C8	C9	C10	118.4(2)	C25	N2	C23	C24 -178.5(2)

Hydrogen Atom Coordinates (Å×10^4) and Isotropic Displacement Parameters (Å^2×10^3).

Atom	*x*	*y*	*z*	U(eq)
H2	3859	4537	3254	60
H3	3177	6114	1994	50
H4	2927	5084	-58	54
H5	5422	4933	2152	56
H7A	7579	5620	1654	71
H7B	7245	6663	714	71
H8	7975	7537	2580	77
H9	5741	8546	1499	73
H12A	4609	8960	3784	97
H12B	3678	7868	3244	97
H13A	2712	9614	2156	109
H13B	3923	9693	1714	109
H14	3166	7918	544	79
H16A	1447	6595	-545	78
H16B	-41	6682	-595	78
H17A	795	4938	331	68
H17B	727	5747	1387	68
H18A	5194	6435	-1117	79
H18B	4556	7213	-378	79
H18C	3848	6038	-988	79
H19A	690	8886	1751	98
H19B	886	7585	2210	98
H19C	-422	7949	1051	98
H20A	1033	4236	2660	71
H20B	2208	3768	3874	71
H21A	3951	5614	4872	67
H21B	3195	4993	5617	67
H22A	3926	6852	6458	73
H22B	3087	7428	5156	73
H23A	628	7343	4304	79
H23B	-112	6710	5057	79
H24A	742	4910	4754	74

H24B	-91	5492	3456	74
H25A	2656	7961	7416	122
H25B	1056	7919	6825	122
H25C	1846	8568	6129	122
H7C	1744	1316	3001	144
H7D	2021	774	4097	144
H8A	1669	5378	7016	113
H8B	636	5113	7356	113

Crystal structure determination

Crystal Data for $C_{25}H_{40}N_2O_8$ (M =496.59 g/mol): monoclinic, space group $P2_1$ (no. 4), a = 10.65993(6) Å, b = 11.44033(5) Å, c = 11.66689(6) Å, β = 113.2519(6)°, V = 1307.249(13) Å^3, Z = 2, T = 294.15 K, μ(CuKα) = 0.773 mm^{-1}, *Dcalc* = 1.262 g/cm^3, 31811 reflections measured (8.248° ≤ 2Θ ≤ 157.924°), 5591 unique (R_{int} = 0.0238, R_{sigma} = 0.0117) which were used in all calculations. The final R_1 was 0.0320 (I > 2σ(I)) and wR_2 was 0.0911 (all data).

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