### Supporting information for

# Synthesis of protectin D1: A potent anti-inflammatory and proresolving lipid mediator

Marius Aursnes,<sup>a</sup> Jørn E. Tungen,<sup>a</sup> Anders Vik,<sup>a</sup> Jesmond Dalli<sup>b</sup> and Trond V. Hansen<sup>\*,a</sup>

<sup>a</sup>School of Pharmacy, Department of Pharmaceutical Chemistry, University of Oslo, PO Box 1068 Blindern, N-0316 Oslo, Norway <sup>b</sup>Center for Experimental Therapeutics and Reperfusion Injury, Department of Anesthesiology, Perioperative and Pain Medicine, Harvard Institutes of Medicine, Brigham and Women's Hospital and Harvard Medical School, Boston, Massachusetts, 02115, USA \*E-mail: t.v.hansen@farmasi.uio.no

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### **General Information**

Unless stated otherwise, all commercially available reagents and solvents were used as received without any further purification. The stated yields are based on isolated material. Thin layer chromatography was performed on silica gel 60 F<sub>254</sub> aluminum-backed plates fabricated by Merck. Flash column chromatography was performed on silica gel 60 (40-63 µm) produced by Merck. NMR spectra were recorded on either a Bruker DRX500, Bruker AVII400 or a Bruker DPX300 spectrometer at 500 MHz, 400 MHz or 300 MHz respectively for <sup>1</sup>H NMR and at 126 MHz, 100 MHz or 75 MHz respectively for <sup>13</sup>C NMR. Coupling constants (J) are reported in hertz and chemical shifts are reported in parts per million ( $\delta$ ) relative to the central residual protium solvent resonance in <sup>1</sup>H NMR (CDCl<sub>3</sub> =  $\delta$  7.27, DMSO- $d_6 = \delta$  2.50 and MeOD- $d_4 = \delta$  3.31) and the central carbon solvent resonance in <sup>13</sup>C NMR (CDCl<sub>3</sub> =  $\delta$  77.00 ppm, DMSO- $d_6 = \delta$  39.43 and MeOD- $d_4 = \delta$  49.00). Mass spectra were recorded at 70 eV on Waters Prospec Q spectrometer using EI, ES or CI as the methods of ionization. High resolution mass spectra were recorded on Waters Prospec Q spectrometer using EI or ES as the methods of ionization. Optical rotations were measured using a 1 mL cell with a 1.0 dm path length on a Perkin Elmer 341 polarimeter. HPLC analyses were performed on an Agilent Technologies 1200 Series instrument with a diode array detector set at 254 nm and equipped with a C<sub>18</sub> stationary phase (Eclipse XDB-C18 5 $\mu$ m 4.6 × 150 mm), applying the conditions stated. The UV/Vis spectra from 190-900 nm were recorded using a Biochrom Libra S32PC spectrometer using quartz cuvettes. IR spectra ( $4000 - 600 \text{ cm}^{-1}$ ) were obtained on a Perkin-Elmer Spectrum BX series FT-IR spectrophotometer. Diastereomeric ratios or yields reported in this paper have not been validated by calibration, see reference Hudlicky and Wernerova for discussions and guidelines.<sup>1</sup>

#### (*S*,*Z*)-*tert*-Butyldimethyl(oct-5-en-1-yn-3-yloxy)silane (5).



2,6-Lutidine (843 mg, 7.98 mmol, 3.0 equiv.) and TBSOTf (843 mg, 3.19 mmol, 1.2 equiv.) were added to a solution of known<sup>2</sup> (3*S*)-oct-5*Z*-en-1-yn-3-ol (**25**) (330 mg, 2.66 mmol, 1 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (26 mL) at -78 °C.The reaction was stirred at that temperature for 4 h before it was quenched with saturated aq. NH<sub>4</sub>Cl (15 mL). The layers were separated and the aqueous layer was extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 x 20 mL). The combined organic layers were dried (Na<sub>2</sub>SO<sub>4</sub>), before concentrated *in vacuo*. The crude product was purified by column chromatography on silica (hexanes/EtOAc 98:2) to afford the title compound **5** as a colourless oil. Yield: 513 mg (81%); TLC (hexanes/EtOAc 95:5, CAM stain):  $R_{\rm f} = 0.66$ ;  $[\alpha]_D^{20} = 21.5$  (c = 0.07, CHCl<sub>3</sub>); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  5.57 – 5.36 (m, 2H), 4.34 (td, J = 6.6, 2.1 Hz, 1H), 2.45 (t, J = 6.9 Hz, 2H), 2.38 (d, J = 2.1 Hz, 1H), 2.07 (p, J = 7.1 Hz, 2H), 0.97 (t, J = 7.5 Hz, 3H), 0.91 (s, 9H), 0.14 (s, 3H), 0.11 (s, 3H); <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>)  $\delta$  134.6, 123.8, 85.6, 72.2, 63.0, 36.7, 25.9 (3C), 20.9, 18.4, 14.4, -4.5, -4.9.

#### (S)-1-(4-Isopropyl-2-thioxothiazolidin-3-yl)ethan-1-one (9a).



Nagao's chiral auxiliary **9a** was prepared from commercially available (*S*)-4-isopropylthiazolidine-2-thione (**26**) by using the procedure of Nagao *et al.*<sup>3</sup> All spectroscopic and physical data were in full agreement with those reported in the literature.<sup>3</sup> Yield: 7.88 g (87%);  $[\alpha]_D^{20} = 434.2$  (c = 0.26, CHCl<sub>3</sub>); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  5.14 (ddd, *J* = 7.6, 6.2, 1.2 Hz, 1H), 3.50 (dd, *J* = 11.5, 8.0 Hz, 1H), 3.02 (dd, *J* = 11.5, 1.2 Hz, 1H), 2.77 (s, 3H), 2.48 - 2.25 (m, 1H), 1.06 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.9 Hz, 3H); <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  203.3, 170.8, 71.4, 30.9, 30.5, 27.0, 19.2, 17.9.

#### (S)-1-(4-Phenyl-2-thioxothiazolidin-3-yl)ethan-1-one (9b).



To (*S*)-4-Phenyl-1,3-thiazolidine-2-thione (**27**) (1.00 g, 5.12 mmol, 1.0 equiv.) in THF (25 mL), 60% NaH dispersion in mineral oil (147 mg, 6.15 mmol, 1.0 equiv.) was slowly added at 0 °C. The reaction mixture was stirred for 15 min at 0 °C, and acetyl chloride (482 mg, 6.15 mmol, 1.2 equiv.) was added dropwise. The reaction mixture was stirred for 30 min at 0 °C, upon which it was warmed to room temperature and allowed to stir for 2 h. The reaction was quenched with saturated NH<sub>4</sub>Cl (15 mL) and the layers were separated. The aqueous layer was extracted with EtOAc (2 x 20 mL) and the combined organic layers were dried (Na<sub>2</sub>SO<sub>4</sub>), filtered, and concentrated *in vacuo*. The crude product was purified by column chromatography on silica (hexanes/EtOAc 95:05) to afford the title compound **9b** as a yellow oil. Yield: 1.07 g (83%); TLC (hexanes/EtOAc 80:20, KMnO<sub>4</sub>)

stain):  $R_{\rm f} = 0.18$ ;  $[\alpha]_D^{20} = 323.5$  (c = 0.18, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  7.45 – 7.31 (m, 5H), 6.27 (dd, J = 8.1, 1.5 Hz, 1H), 3.96 (dd, J = 11.2, 8.2 Hz, 1H), 3.10 (dd, J = 11.2, 1.5 Hz, 1H), 2.83 (s, 3H); <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>)  $\delta$  202.7, 170.6, 139.2, 129.1 (2C), 128.6, 125.5 (2C), 69.5, 36.7, 27.2. HRMS (EI+): Exact mass calculated for C<sub>11</sub>H<sub>11</sub>NOS<sub>2</sub> [M]<sup>+</sup>: 237.0282, found 237.0286.

#### (S)-1-(4-Benzyl-2-thioxothiazolidin-3-yl)ethan-1-one (9c).



Nagao's chiral auxiliary **9c** was prepared from commercially available (*S*)-4-benzylthiazolidine-2-thione (**28**) by using the procedure of Jensen *et al.*<sup>4</sup> All spectroscopic and physical data were in full agreement with those reported in the literature.<sup>4</sup> Yield: 979 mg (81%);  $[\alpha]_D^{20} = 253$  (c = 0.35, CHCl<sub>3</sub>); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  7.39 – 7.23 (m, 5H), 5.45 – 5.31 (m, 1H), 3.39 (ddd, *J* = 11.5, 7.3, 1.1 Hz, 1H), 3.22 (dd, *J* = 13.3, 3.9 Hz, 1H), 3.04 (dd, *J* = 13.2, 10.5 Hz, 1H), 2.89 (dd, *J* = 11.6, 0.8 Hz, 1H), 2.80 (s, 3H); <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  201.7, 170.8, 136.6, 129.6 (2C), 129.0 (2C), 127.3, 68.3, 36.8, 31.9, 27.2.

#### Potassium (1E,3E)-5-oxopenta-1,3-dien-1-olate (15).



Glutaconaldehyde potassium salt **15** was prepared from commercially available pyridinium-1-sulfonate (**14**) according to the procedure of Becher.<sup>5</sup> All spectroscopic and physical data were in full agreement with those reported in the literature.<sup>5</sup> Yield: 28.5 g (55%); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>)  $\delta$  8.67 (d, *J* = 9.2 Hz, 2H), 7.04 (t, *J* = 12.9 Hz, 1H), 5.10 (dd, *J* = 13.0, 9.1 Hz, 2H); <sup>13</sup>C NMR (75 MHz, DMSO-*d*<sub>6</sub>)  $\delta$  184.4 (2C), 159.8, 106.2 (2C).

#### (2*E*,4*E*)-5-Bromopenta-2,4-dienal (8).



Bromopentadienal **8** was prepared by the bromination of glutaconaldehyde potassium salt **15** according to the protocol reported by Duhamel *et al.*<sup>6</sup> All spectroscopic and physical data were in agreement with those reported in the literature.<sup>6</sup> Yield: 8.1 g (75%); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>)  $\delta$  9.58 (d, *J* = 7.8 Hz, 1H), 7.07 – 6.90 (m, 3H), 6.24 – 6.11 (m, 1H); <sup>13</sup>C NMR (75 MHz, CDCl<sub>3</sub>)  $\delta$  193.3, 147.9, 135.7, 132.0, 120.0.

#### (R,4E,6E)-7-Bromo-3-hydroxy-1-((S)-4-phenyl-2-thioxothiazolidin-3-yl)hepta-4,6-dien-1-one (16b).



To a solution of *N*-acetylthiazolidinethione **9b** (200 mg, 0.73 mmol, 1.0 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (7.3 mL), TiCl<sub>4</sub> (1M in CH<sub>2</sub>Cl<sub>2</sub> 139 mg, 0.73 mmol, 1.0 equiv.) was added at -78 °C and stirred for 5 min. Diisopropylethylamine (113 mg, 0.88 mmol, 1.2 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (1.8 mL) was added and the solution was stirred for 30 min at -78 °C, whereupon the freshly prepared aldehyde **8** (106 mg, 0.66 mmol, 0.9 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (1.8 mL) was added dropwise. The mixture was stirred for 1 h at -78 °C, then quenched with half saturated ammonium chloride (10 mL) and warmed to room temperature. The layers were separated and the aq. layer was extracted with CH<sub>2</sub>Cl<sub>2</sub> (2 x 20 mL). The combined organic layers were dried (Na<sub>2</sub>SO<sub>4</sub>) and filtered, before concentrated *in vacuo*. The diastereomeric ratio (4.5:1) on the crude product was determined by HPLC analysis (Eclipse XDB-C18, MeOH/H<sub>2</sub>O 7:3, 1.0 mL/min, *t<sub>r</sub>*(minor) = 8.23 min and *t<sub>r</sub>*(major) = 10.29 min). The crude product was purified by column chromatography on silica (hexanes/EtOAc 7:3) to afford the title compound **16b** as a yellow oil. Yield: 141 mg (54%); TLC (hexanes/EtOAc 1:1, KMnO<sub>4</sub> stain): *R*<sub>f</sub> = 0.24;  $[\alpha]_D^{20}$  = 189.5 (c = 0.12, CHCl<sub>3</sub>); <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  7.45 – 7.30 (m, 5H), 6.70 (dd, *J* = 13.6, 10.7 Hz, 1H), 6.34 (d, *J* = 13.5 Hz, 1H), 6.26 – 6.18 (m, 2H), 5.77 (dd, *J* = 15.3, 5.5 Hz, 1H), 3.10 (d, *J* = 10.7 Hz, 1H), 2.92 (bs, *J* = 4.3 Hz, 1H); <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>)  $\delta$  202.5, 172.4, 139.0, 136.7, 134.7, 129.3 (2C), 128.8, 128.1, 125.5 (2C), 109.7, 69.7, 67.9, 45.8, 36.9; HRMS (TOF ES+): Exact mass calculated for C<sub>16</sub>H<sub>16</sub><sup>79</sup>BrNO<sub>2</sub>S<sub>2</sub>Na [*M*+Na]<sup>+</sup>: 419.9703, found 419.9711.

#### (R,4E,6E)-1-((S)-4-Benzyl-2-thioxothiazolidin-3-yl)-7-bromo-3-hydroxyhepta-4,6-dien-1-one (16c).



To a solution of N-acetylthiazolidinethione 9c (200 mg, 0.80 mmol, 1.0 equiv.) in CH<sub>2</sub>Cl<sub>2</sub>(7.9 mL), TiCl<sub>4</sub> (1M in CH<sub>2</sub>Cl<sub>2</sub>, 151 mg, 0.80 mmol, 1.0 equiv.) was added at -78 °C and stirred for 5 min. Diisopropylethylamine (189 mg, 1.46 mmol, 1.2 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (2.0 mL) was added and the solution was stirred for 30 min at -78 °C, whereupon the freshly prepared aldehyde 8 (116 mg, 0.72 mmol, 0.9 equiv.) in CH<sub>2</sub>Cl<sub>2</sub> (2.0 mL) was added dropwise. The mixture was stirred for 1 h at -78 °C, then quenched with half saturated ammonium chloride (10 mL) and warmed to room temperature. The layers were separated and the aq layer was extracted with  $CH_2Cl_2$  (2 x 20 mL). The combined organic layers were dried ( $Na_2SO_4$ ) and filtered, before concentrated in vacuo. The diastereomeric ratio (9.8:1) on the crude product was determined by HPLC analysis (Eclipse XDB-C18, MeOH/H<sub>2</sub>O 7:3, 1.0 mL/min,  $t_r$ (minor) = 16.60 min and  $t_r$ (major) = 21.08 min). The crude product was purified by column chromatography on silica (hexanes/EtOAc 8:2) to afford the title compound 16c as a yellow oil. Yield: 232 mg (79%); TLC (hexanes/EtOAc 7:3, KMnO<sub>4</sub> stain):  $R_{\rm f} = 0.16$ ;  $[\alpha]_D^{20} = 116.8$  (c = 0.11, CHCl<sub>3</sub>); <sup>1</sup>H NMR  $(400 \text{ MHz}, \text{CDCl}_3) \delta 7.38 - 7.26 \text{ (m, 5H)}, 6.72 \text{ (dd, } J = 13.6, 10.8 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.5 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.5 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.6 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.6 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.6 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.36 \text{ (d, } J = 13.6 \text{ Hz}, 1\text{H}), 6.27 \text{ (dd, } J = 13.6 \text{ Hz}, 1\text{H}), 6.28 \text{ Hz}, 1\text{Hz}, 1\text{H}), 6.28 \text{ Hz}, 1\text{Hz}, 1\text{Hz},$ = 15.3, 10.8 Hz, 1H), 5.81 (dd, J = 15.3, 5.5 Hz, 1H), 5.40 (ddd, J = 10.8, 7.1, 4.1 Hz, 1H), 4.75 - 4.67 (m, 1H), 3.70 (dd, J = 17.7, 2.9 Hz, 1H), 3.42 (dd, J = 11.6, 7.2 Hz, 1H), 3.29 (dd, J = 17.8, 8.9 Hz, 1H), 3.23 (dd, J = 13.5, 4.2 Hz, 1H), 3.05 (dd, J = 13.2, 10.4 Hz, 1H), 2.92 (d, J = 11.6 Hz, 1H), 2.87 (bs, 1H); <sup>13</sup>C NMR (101 MHz, 1H); <sup>13</sup>C NMR (110 MHz, 1H); <sup>13</sup>C NMR (110 MHz, 1H); <sup>13</sup>C NMR ( CDCl<sub>3</sub>)  $\delta$  201.5, 172.4, 136.7, 136.4, 134.8, 129.6 (2C), 129.1 (2C), 128.1, 127.5, 109.7, 68.4, 68.0, 45.5, 37.0, 32.3; HRMS (TOF ES+): Exact mass calculated for  $C_{17}H_{18}^{79}BrNO_2S_2Na [M+Na]^+$ : 433.9860, found 433.9867.

#### Methyl 7-hydroxyhept-4-ynoate (18).



To 4-pentynoic acid (11) (5.00 g, 51 mmol, 1.0 equiv.) in dry HMPA (100 mL), n-BuLi (44.9 mL, 2.5 M, 2.2 equiv.) was added at 0 °C. The reaction mixture was stirred for 1 h before ethylene oxide (10) in THF (25 mL, 2.5-3.3 M) was added slowly at the above-mentioned temperature. The cooling bath was removed and the reaction mixture was allowed to warm up to ambient temperature and then stirred for 24 h. The reaction mixture was cooled to 0 °C before water (150 mL) was added. Next, concentrated aq. HCl was added dropwise until the mixture was acidic to litmus. The reaction mixture was extracted with Et<sub>2</sub>O (3 x 100 mL) and EtOAc (2 x 100 mL), the combined organic phase was dried (MgSO<sub>4</sub>), filtered and concentrated in vacuo. The crude, HMPAcontaminated carboxylic acid was dissolved in dry MeOH (250 mL) and a few drops of concentrated sulphuric acid. The reaction mixture was refluxed for 12 h before it was allowed to cool to ambient temperature. Next, solid NaHCO<sub>3</sub> (5.00 g) and saturated aq. NaHCO<sub>3</sub> (15 mL) were added and the reaction mixture was concentrated in *vacuo*. To the mixture was added aq. NaHCO<sub>3</sub> (50 mL), and the aqueous phase was extracted with  $E_{12}O$  (4 x 100 mL). The organic phases were combined and dried (Na<sub>2</sub>SO<sub>4</sub>), filtered and concentrated in vacuo. The crude product was purified by column chromatography on silica (hexanes/EtOAc 7:3) to afford the title compound 18 as a pale yellow oil. Yield: 4.36 g (55% for two steps starting from 11); TLC (hexanes/EtOAc 5:5, KMnO<sub>4</sub> stain):  $R_{\rm f} = 0.25$ ; Spectroscopic and physical data were in full agreement with those reported in the literature.<sup>7</sup> <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  3.69 (s, 3H), 3.65 (q, J = 6.2 Hz, 2H), 2.54 – 2.45 (m, 4H), 2.42 – 2.37 (m, 2H); <sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ 172.7, 80.6, 77.9, 61.4, 51.9, 33.8, 23.3, 14.9.

#### Methyl (Z)-7-hydroxyhept-4-enoate (19).



Methyl ester **18** (2.00 g, 12.81 mmol) was dissolved in 30% EtOAc in hexanes (100 mL), and the flask was evacuated and filled with argon gas two times. Lindlar's catalyst (400 mg) was added and the reaction flask was again evacuated and filled with hydrogen gas. The reaction mixture was stirred for 12 h, filtrated through a pad of Celite and then concentrated *in vacuo* to give a pale yellow oil. Yield of **19**: 1.96 g (97%); TLC (hexanes/EtOAc 5:5, KMnO<sub>4</sub> stain):  $R_f = 0.25$ ; Spectroscopic and physical data were in full agreement with those reported in the literature.<sup>7 1</sup>H NMR (400 MHz, DMSO- $d_6$ )  $\delta$  5.45 – 5.33 (m, 2H), 4.47 (t, J = 5.3 Hz, 1H), 3.58 (s, 3H), 3.38 (td, J = 6.8, 5.3 Hz, 2H), 2.37 – 2.32 (m, 2H), 2.29 – 2.23 (m, 2H), 2.16 (q, J = 6.7 Hz, 2H). <sup>13</sup>C NMR (101 MHz, DMSO)  $\delta$  172.8, 128.9, 127.7, 60.5, 51.2, 33.4, 30.7, 22.4.

#### (Z)-(7-Methoxy-7-oxohept-3-en-1-yl)triphenylphosphonium iodide (7).



To a solution of methyl ester **19** (1.50 g, 9.49 mmol) in dry  $CH_2Cl_2$  (100 mL), triphenylphosphine (3.82 g, 14.58 mmol) and imidazole (1.00 g, 14.70 mmol) were added. The reaction flask was placed in a cooling bath (ice, water, salt) for 15 min before iodide (3.74 g, 14.74 mmol) was added in one portion with rapid stirring. The

reaction mixture was stirred for 15 min before the cooling bath was removed and the reaction mixture stirred at ambient temperature for another 35 min. A saturated solution of aq. Na<sub>2</sub>SO<sub>3</sub> (10 mL) was added, and the aqueous phase was extracted with CH<sub>2</sub>Cl<sub>2</sub> (2 x 100 mL), dried (Na<sub>2</sub>SO<sub>4</sub>), filtrated and concentrated *in vacuo* to a viscous yellow oil which was run through a short sillica column (8% EtOAc in hexanes) to yield a pale yellow oil, which was dissolved in dry MeCN (100 mL). Triphenylphosphine (4.25 g, 16.22 mmol) was added and the reaction mixture was refluxed for 12 h, cooled, concentrated *in vacuo* and purified by flash chromography (CH<sub>2</sub>Cl<sub>2</sub> until all triphenylphosphine was out of the column followed by 5% MeOH in CH<sub>2</sub>Cl<sub>2</sub>) to yield a viscous clear oil. Yield: 3.97 g (79% over two steps from **19**); TLC (CH<sub>2</sub>Cl<sub>2</sub>/MeOH 95:5, KMnO<sub>4</sub> stain):  $R_f = 0.28$ ; All spectroscopic and physical data were in full agreement with those reported in the literature.<sup>7 1</sup>H NMR (400 MHz, DMSO-*d*<sub>6</sub>)  $\delta$  7.93 – 7.74 (m, 15H), 5.49 – 5.36 (m, 2H), 3.69 – 3.58 (m, 2H), 3.56 (s, 3H), 2.37 – 2.26 (t, *J* = 7.2 Hz, 4H), 2.12 (q, *J* = 7.0 Hz, 2H).

### Authentic protectin D1 (2).

Authentic protectin D1 (2) was formed by using endogenous murine self-resolving exudates as previously reported.<sup>8</sup> HPLC and LC/MS-MS analyses were performed as reported by Serhan and co-workers.<sup>9,10</sup>

























Figure S-10<sup>13</sup>C-NMR spectrum of compound 21.







Figure S-14 <sup>13</sup>C-NMR spectrum of compound 23.













Figure S-19 UV-Vis chromatogram of methyl ester 24.











Area Percent Report

Sorted By	:	Siqnal			
Multiplier	:	1.0000			
Dilution	:	1.0000			
Sample Amount		1.00000	[ng/ul]	ínot used	in calc.)
Use Multiplier &	Dilution	Factor with	h ISTDs	,	
Gioval 1. Wind 1					
Signai I: YWDI A,	wavelen	gth-204 nm			
Peak RetTime Type # [min]	Width [min]	Area mAU *s	Heiqht [mAU ]	Area %	
1 16.601 BB	0.5653	2356.82593	63.10266	9.2898	
2 21.083 VB	0.6936	2.30132e4	503.34839	90.7102	
Totals :		2.53700e4	566.45105		

#### Figure S-23 HPLC chromatogram of aldol 16c.







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Figure S-27 HPLC chromatograms for matching experiments. Authentic protectin D1 (2) from self-resolving peritoneal inflammatory exudates matched synthetic material obtained after saponification of ester 24. Selected ion chromarograms (m/z 359-153) depicting (A) Authentic protectin D1 (2), marked as PD1, obtained from mice injected with *Escherichia coli* (10<sup>5</sup> CFU) and exudates collected at 12 hr. (B) Synthetic material obtained after saponification of ester 24. (C) Coinjection of authentic protectin D1 (2) from self-resolving inflammatory exudates with synthetic material obtained after saponification of ester 24. Figures (A)-(C) are representative HPLC chromatograms (n = 4).



Figure S-28. Matching MS-MS spectra for authentic protectin D1 (2) and synthetic material 24. (A) authentic protectin D1 (2) obtained from mice injected with *Escherichia coli* and exudates collected at 12 hr. MS-MS spectrum for peak at  $T_R = 13.2$  min; representative MS-MS spectra (n=4 mice) (B) MS-MS spectrum of synthetic material ( $T_R = 13.2$  min) after saponification of ester 24. Representative MS-MS spectra (d = 4).



Figure S-30. Matching MS-MS spectra for authentic protectin D1 (2) and synthetic 2. (A) authentic protectin D1 (2) obtained from mice injected with *Escherichia coli* and exudates collected at 12 hr. MS-MS spectrum for peak at  $T_R = 13.2$  min; representative MS-MS spectra (n=4 mice) (B) MS-MS spectrum of synthetic material protectin D1 (2) ( $T_R = 13.2$  min). Representative MS-MS spectra (d = 4).

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