

Design, Synthesis and Cholinesterase Inhibitory activity of New Dispiro Pyrrolidine Derivatives

Nadia Mohamed Yusoff ^a, Hasnah Osman ^a, Valentia Katemba ^b, Muhammad Solehin Abd Ghani ^a, Unang Supratman ^c, Mohammad Tasyriq Che Omar ^d, Vikneswaran Murugaiyah ^{e,f}, Xiang Ren ^g, Yvan Six ^{g,*} and Mohamad Nurul Azmi ^{a,*}

^a School of Chemical Sciences, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia

^b Institute of Pharmaceutical Science, King's College London, Franklin Wilkins Building, Stamford Street, London, SE1 9NH

^c Department of Chemistry, Faculty of Science and Mathematics, Universitas Padjadjaran, Jatinangor, 45363 West Java, Indonesia

^d Biological Section, School of Distance Education, Universiti Sains Malaysia, 11800 Penang, Malaysia

^e Centre for Drug Research, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia.

^f School of Pharmaceutical Sciences, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia

^g Laboratoire de Synthèse Organique (LSO), UMR 7652 CNRS/ENSTA/École Polytechnique, Institut Polytechnique de Paris, 91128 Palaiseau Cedex, France

*Corresponding authors: mnazmi@usm.my / yvan.six@polytechnique.edu

Supplementary information

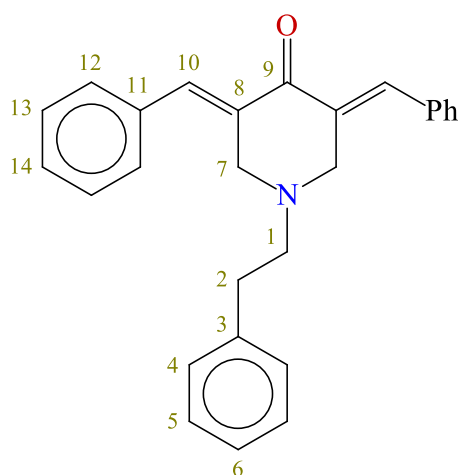
Characterisation data and NMR spectra for compounds **3**, **4**, **7a-h**

General information

All chemicals reagents were obtained from Acros Organic (USA), QRec (Asia), Sigma Aldrich Chemical Co. and E. Merck (Germany). Melting points were determined on a Stuart Scientific SMP10 or SMP40 melting point apparatus. Thin-layer chromatography (TLC) plates was performed on alumina plates pre-coated with silica gel (Merck 60 F254) in the solvent system acetone-hexane with two different ratios of 3:7 or 1:9. The progress of reactions was determined by the appearance of product and disappearance of reactant spots under UV radiation lamp ($\lambda_{\max} = 254 \text{ nm}$), respectively. All spectral data were obtained on the following instruments: infrared spectra were recorded on a Perkin Elmer 2000 FTIR spectrometer at wavenumbers ranging from 4000 to 600 cm^{-1} . ^1H (400.2 or 500 MHz) and ^{13}C (100.6 or 125 MHz) nuclear magnetic resonance spectra were recorded with an AVANCE 400 or an AVN 500 Bruker spectrometer (Bruker Biospin, Billerica, MA, U.S.A.). Data were analysed using NMRnotebook 2.80 (NMRTEC S.A.S.). Chemical shifts δ are reported in parts per million (ppm), referenced to the peak of tetramethylsilane, defined at $\delta = 0.00$ (^1H and ^{13}C NMR), or the residual solvent peak, defined at $\delta = 2.50$ (^1H NMR, DMSO- d_6), 7.26 (^1H NMR, CDCl_3), 39.5 (^{13}C NMR, DMSO- d_6) or 77.0 (^{13}C NMR, CDCl_3). Multiplicities are abbreviated as follows: s (singlet), d (doublet), t (triplet), q (quartet), quint (quintet), m (multiplet), br (broad). Coupling constants J are given in Hz and are rounded to the closest multiple of 0.5. Mass spectrometry experiments were performed using a Waters Xevo QTOF MS system or, alternatively, on a tims-TOF mass spectrometer (Bruker) in the *Laboratoire de Chimie Moléculaire* (Ecole polytechnique, CNRS, Palaiseau, France). The electrospray source was used in positive mode. Samples were prepared in acetonitrile with 0.1% of formic acid, introduced at a 5 $\mu\text{L}/\text{min}$ flow rate into the interface of the instrument. Capillary and end plate voltages were set at 4.5 kV and 0.7 kV. Nitrogen was used as the nebulizer and drying gas at 2.5 bar and 3 L/min, respectively, with a drying temperature of 180°C. Tuning mix (Agilent) was used for calibration. The elemental compositions of all ions were determined with the instrument software Data Analysis; the precision of mass measurement was less than 3 ppm.

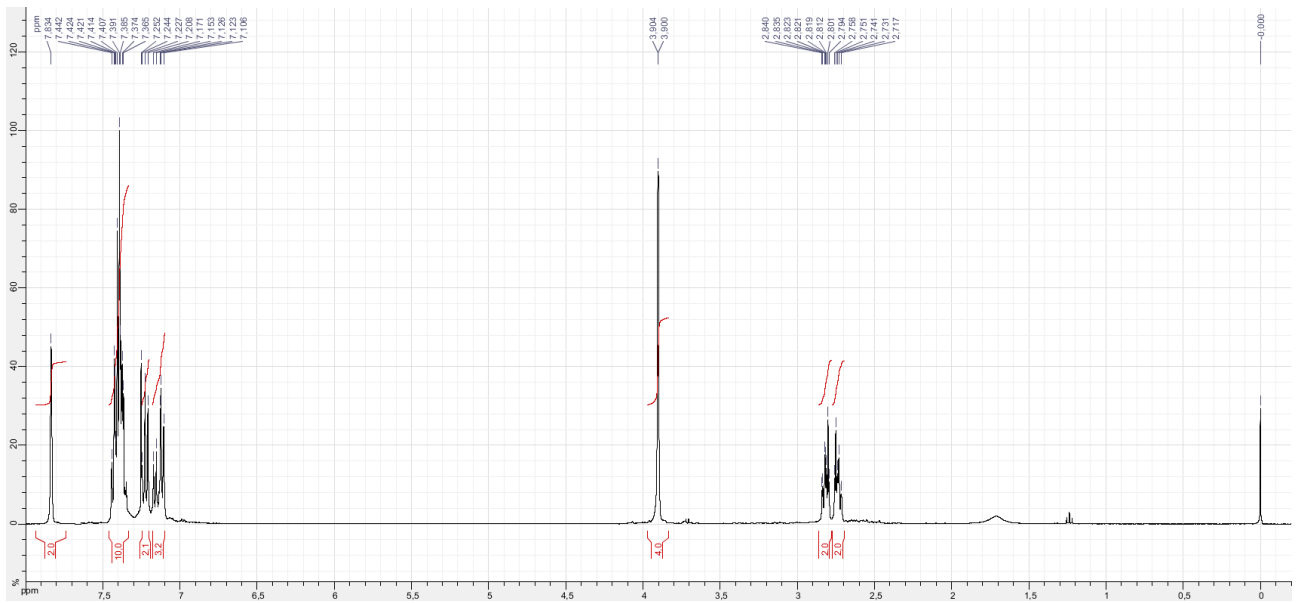
For the assays the chemicals and enzymes used in the cholinesterase inhibition investigation are as follows: Acetylthiocholine iodide (Sigma Aldrich, USA); 5,5'-Dithiobis-2-benzoic acid (Sigma Aldrich, USA); Acetylcholinesterase (AChE) from *Electrophorus electricus* (electric eel) (Sigma Aldrich, USA); Butyrylthiocholine iodide (Sigma Aldrich, USA); Butyrylcholinesterase (BChE) from equine serum (Sigma Aldrich, USA).

3,5-Dibenzylidene-1-phenylethyl-4-piperidinone (**3**)

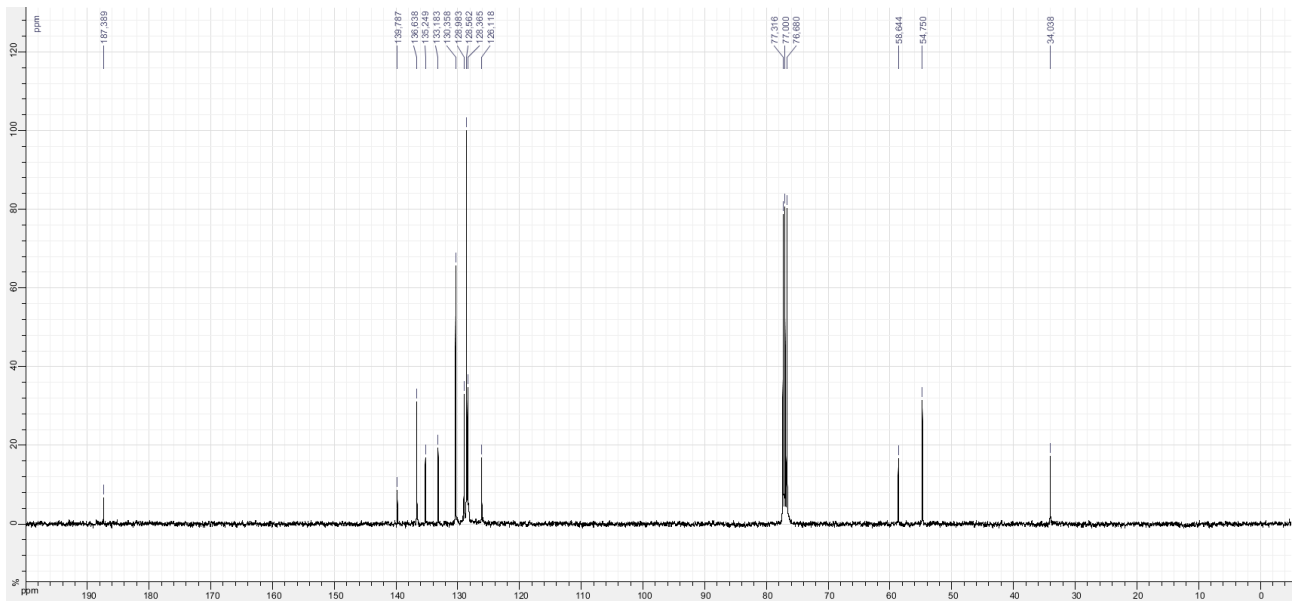


Yellow crystals. $R_f \approx 0.2$ [UV-active, EtOAc/petroleum ether 20%, orange spot (anisaldehyde)]. M.p. 88–92°C (EtOH) (Lit. 165 °C)¹. FTIR (ATR, neat): ν 3026 (w, C-H), 1669 (s, C=O), 1608 (s, C=C), 1181 (m, C-N) cm^{-1} . ¹H NMR (400 MHz, CDCl₃): δ 2.74 (2H, m, H1 or H2), 2.81 (2H, m, H1 or H2), 3.90 (4H, d, $J=1.5$ Hz, H7), 7.11 (2H, dd, $J=8.0, 1.5$ Hz, H4), 7.15 (1H, tt, $J=7.5, 1.5$ Hz, H6), 7.23 (2 H, br dd, $J=8.0, 7.5$ Hz, H5), 7.34-7.46 (10H, m, H12-H14), 7.83 (2H, t, $J=1.5$ Hz, H10). ¹³C NMR (100.6 MHz, CDCl₃): δ 34.0 (C2), 54.7 (C7), 58.6 (C1), 126.1 (C6), 128.4 (C4), 128.6 (C5), 128.6 (C13), 129.0 (C14), 130.4 (C12), 133.2 (C8), 135.2 (C11), 136.7 (C10), 139.8 (C3), 187.4 (C9). ¹H NMR (500 MHz, DMSO-*d*₆): δ 2.70 (2H, t, $J=7.5$ Hz, H1 or H2), 2.81 (2H, t, $J=7.5$ Hz, H1 or H2), 3.88 (4H, s, H7), 7.14-7.23 (5H, m, H4-H6), 7.42-7.51 (10H, m, H12-H14), 7.58 (2H, s, H10). ¹³C NMR (125.8 MHz, DMSO-*d*₆): δ 32.8 (C2), 54.0 (C7), 57.8 (C1), 125.8 (C6), 128.1 (C4), 128.5 (C5), 128.7 (C13), 129.2 (C14), 130.5 (C12), 133.8 (C8), 134.6 (C11), 134.8 (C10), 140.0 (C3), 186.9 (C9). HRMS (TOF-ES⁺): m/z 380.2016 (MH⁺ C₂₇H₂₆NO⁺ requires 380.2009).

1 - M.A. Ali, V.S. Lakshmi pathi, F. Beevi, R.S. Kumar, R. Ismail, T.S. Choon, A.C. Wei, Y.K. Yoon, A. Basiri, Antimycobacterial activity: Synthesis and biological evaluation of novel substituted (3E,5E)-3,5-diarylidene-1-phenethylpiperidine-4-one derivatives, Letters in Drug Design and Discovery 10 (2013) 471-476.

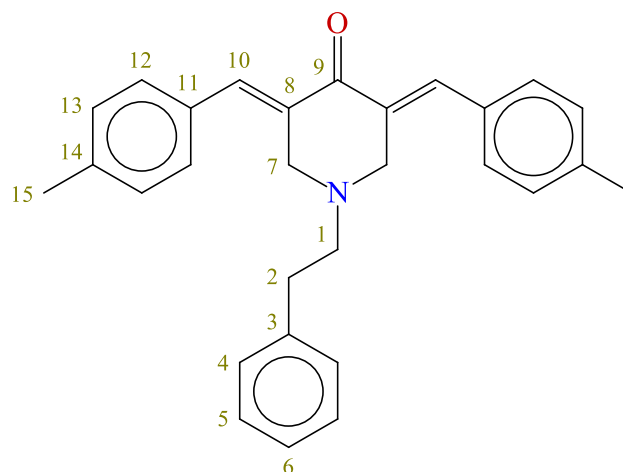


^1H NMR spectrum (CDCl_3 , 400 MHz).



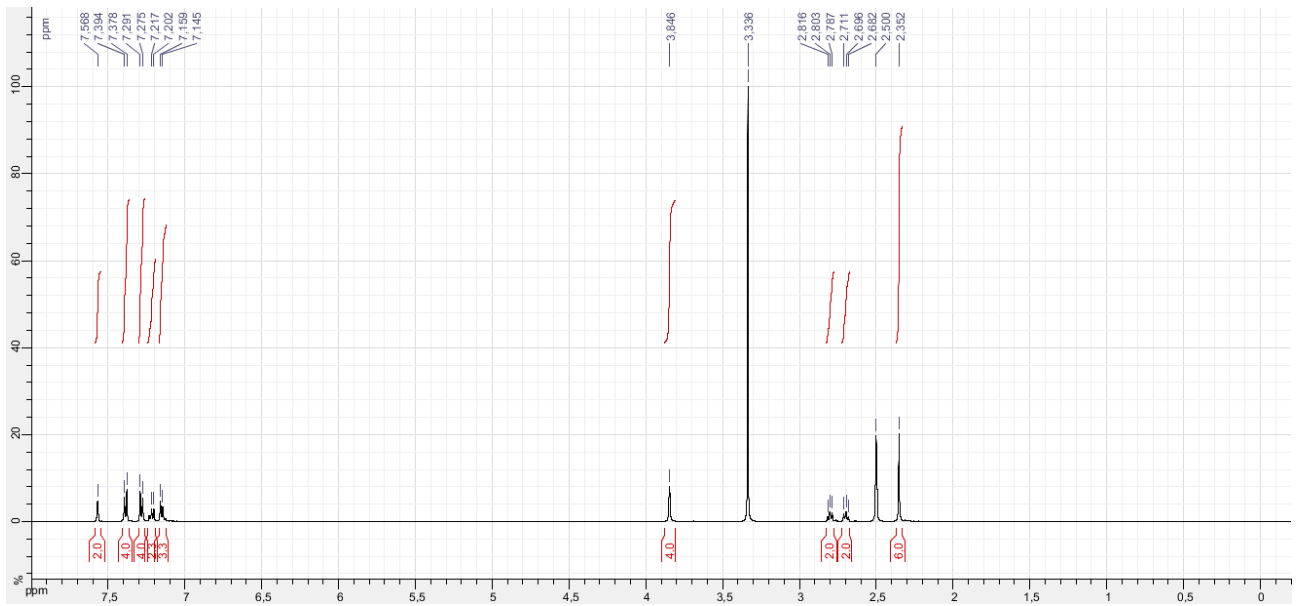
^{13}C NMR spectrum (CDCl_3 , 100.6 MHz).

3,5-Bis(4-methylbenzylidene)-1-phenylethyl-4-piperidinone (4)

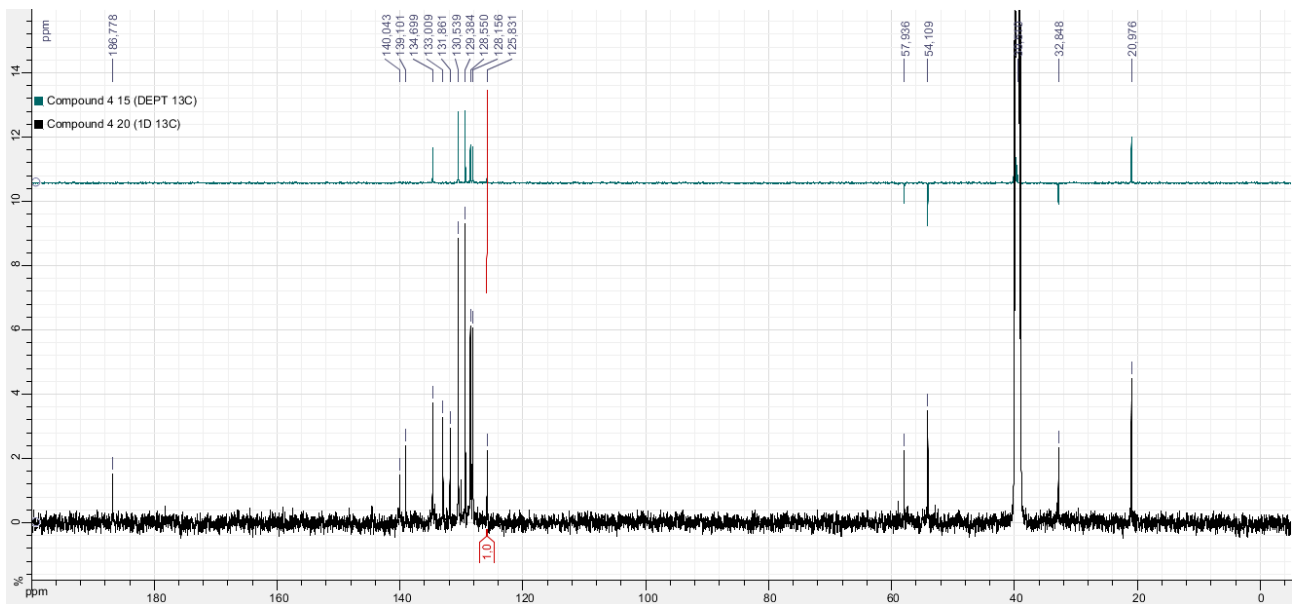


Yellow solid. $R_f \approx 0.78$ [UV-active, Acetone:hexane 10%]. M.p. 118–121 °C. FTIR (ATR, neat): ν 3024 (w, C–H), 1668 (s, C=O), 1606 (m, C=C), 1175 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO-d_6): δ 2.35 (6H, s, H15), 2.70 (2H, distorted t, $J=7.5$ Hz, H2), 2.80 (2H, distorted t, $J=7.5$ Hz, H1), 3.85 (4H, br s, H7), 7.14 (1H, tt, $J=7.5, 1.5$ Hz, H6), 7.15 (2H, d, $J=7.5$ Hz, H4), 7.22 (2H, br t, $J=7.5$ Hz, H5), 7.28 (4H, d, $J=8.0$ Hz, H13), 7.39 (4H, d, $J=8.0$ Hz, H12),² 7.57 (2H, br s, H10). ^{13}C NMR (125.8 MHz, DMSO-d_6): δ 21.0 (C15), 32.8 (C2), 54.1 (C7), 57.9 (C1), 125.8 (C6), 128.2 (C4), 128.5 (C5), 129.4 (C13), 130.5 (C12), 131.9 (C11), 133.0 (C8), 134.7 (C10), 139.1 (C14), 140.0 (C3), 186.8 (C9). HRMS (TOF- ES^+): m/z 408.2324 (MH^+ $\text{C}_{29}\text{H}_{30}\text{NO}^+$ requires 408.2322).

2 - The signals at 7.28 and 7.39 ppm constitute an AA'BB' system, which is described as a set of two doublets for simplification purposes

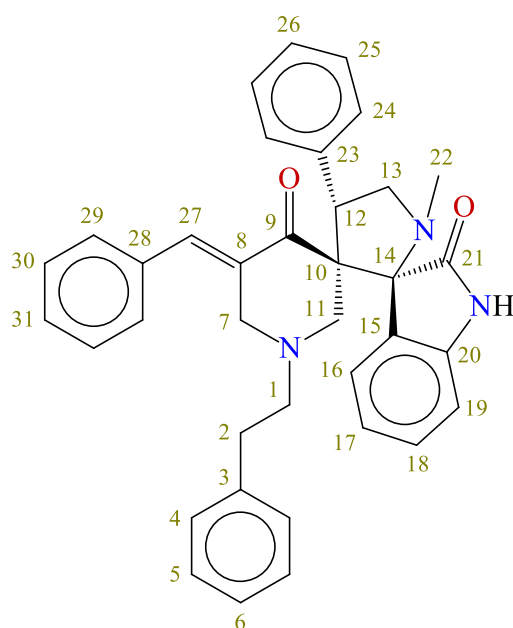


¹H NMR spectrum (DMSO-d₆, 500 MHz).



¹³C NMR and DEPT135 NMR spectra (DMSO-d₆, 125.8 MHz).

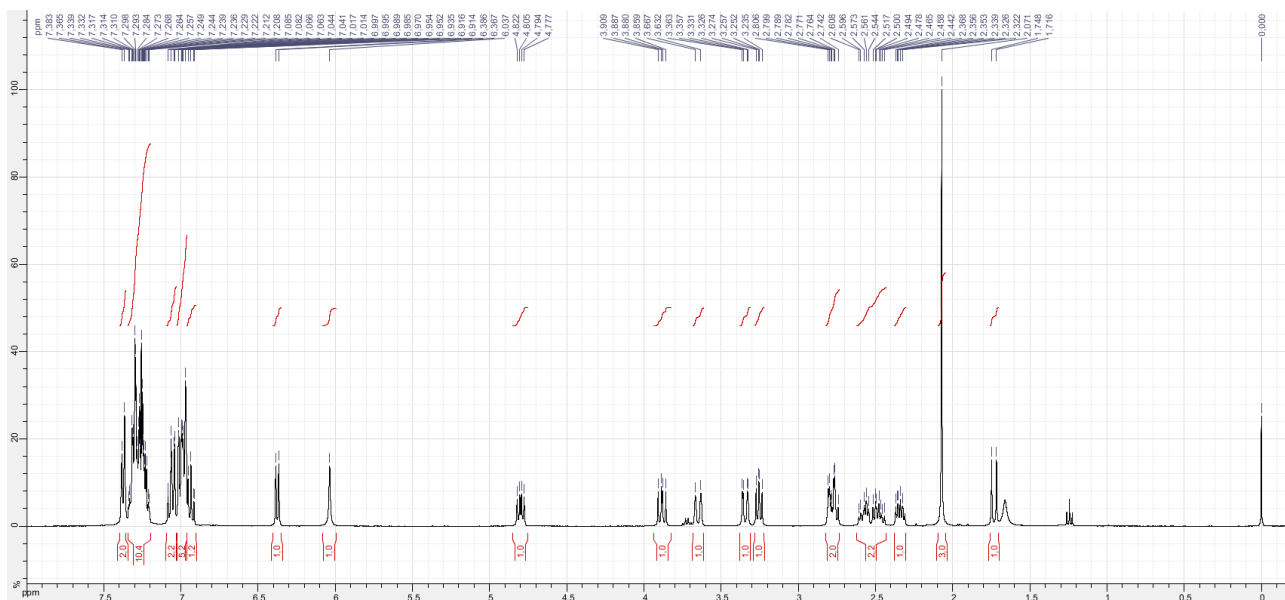
1-Methyl-4(phenyl)pyrrolo-(spiro[2.3]oxindole)-spiro[3.3]-5'-(phenylmethylidene)-1'-phenylethyl-4'-piperidinone (**7a**)



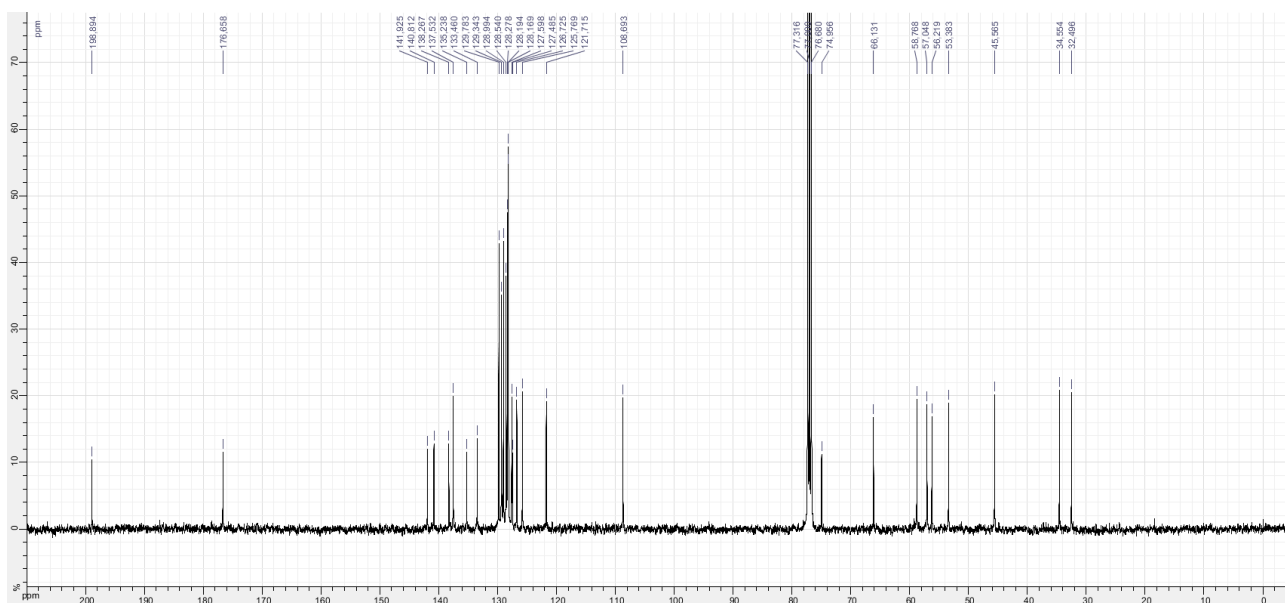
Pale yellow crystals. $R_f \approx 0.15$ [UV-active, EtOAc/petroleum ether 20%]. M.p. 200–202 °C (dec.) (EtOH). FTIR (ATR, neat): ν 3346 (w, N–H), 3027 (w, C–H), 1697 (s, C=O), 1599 (m, C=C), 1182 (m, C–N) cm^{-1} . ^1H NMR (400 MHz, CDCl_3): δ 1.73 (1H, d, $J=12.5$ Hz, H11a), 2.07 (3H, s, H22), 2.34 (1 H, ddd, $J= 12.0, 6.5, 5.0$ Hz, H1a), 2.53 (2H, AB part of an ABXY system, δ_A 2.48, δ_B 2.57, $J_{AB}= 14.0$ Hz, $J_{AX}= 6.5$ Hz, $J_{AY}= 9.0$ Hz, $J_{BX}= 5.0$ Hz, $J_{BY}= 7.0$ Hz, H2), 2.78 (ddd, $J= 12.0, 9.0, 7.0$ Hz, H1b), 2.78 (1H, dd, $J= 14.0, 2.0$ Hz, H7a), 3.25 (1H, dd, $J= 8.5, 7.0$ Hz, H13a), 3.34 (1H, dd, $J= 12.5, 2.0$ Hz, H11b), 3.65 (1 H, ddd, $J= 14.5, 2.0, 1.5$ Hz, H7b), 3.88 (1H, dd, $J= 11.5, 8.5$ Hz, H13b), 4.80 (1H, dd, $J= 11.5, 7.0$ Hz, H12), 6.04 (1H, br s, NH), 6.38 (1H, br d, $J= 7.5$ Hz, H19), 6.93 (1 H, td, $J= 7.5, 1.0$ Hz, H17), 6.97 (1H, dd, $J= 2.0, 1.5$ Hz, H27), 6.98 (2H, br dd, $J= 7.5, 2.0$ Hz, H29), 7.01 (2 H, br dd, $J= 8.0, 1.5$ Hz, H4), 7.05 (1H, br dd, $J= 7.5, 1.5$ Hz, H16), 7.07 (1H, ddd, $J= 8.0, 7.5, 1.5$ Hz, H18), 7.20–7.34 (9 H, m, H5–H6, H25–H26, H30–H31), 7.37 (2 H, br dd, $J= 7.5, 1.5$ Hz, H24). ^{13}C NMR (100.6 MHz, CDCl_3): δ 32.5 (C2), 34.6 (C22), 45.6 (C12), 53.4 (C7), 56.2 (C13), 57.0 (C11), 58.8 (C1), 66.1 (C10), 75.0 (C14), 108.7 (C19), 121.7 (C17), 125.8 (C6), 126.7 (C26), 127.5 (C15), 127.6 (C16), 128.17, 128.19 (C4, C25), 128.3, 128.5 (C5, C30), 129.0 (C18), 129.0 (C31), 129.3 (C24), 129.8 (C29), 133.5 (C8), 135.2 (C28), 137.5 (C27), 138.3 (C23), 140.8 (C3), 141.9 (C20), 176.7 (C21), 198.9 (C9). ^1H NMR (500 MHz, DMSO-d_6): δ 1.76 (1H, d, $J=12.5$ Hz, H11a), 1.99 (3H, s, H22), 2.30–2.55 (4H, m, H1, H2), 3.06 (1H, dd, $J= 15.0, 2.5$ Hz, H7a), 3.21 (1H, dd, $J= 8.5, 7.5$ Hz, H13a), 3.23 (1H, d, $J= 12.5$ Hz, H11b), 3.34 (1 H, br d, $J= 15.0$ Hz, H7b),³ 3.83 (1H, dd, $J= 11.0, 8.5$ Hz, H13b), 4.65 (1H, dd, $J= 11.0, 7.5$ Hz, H12), 6.64 (1H, br d, $J= 7.5$ Hz, H19), 6.85–6.90 (2H, m, H16, H17), 7.05 (1H, ddd, $J= 7.5, 6.0, 2.5$ Hz, H18), 7.09 (1H, m, H27), 7.11 (2H, d, $J= 7.5$ Hz, H29), 7.11 (2H, d, $J= 7.0$ Hz, H4), 7.11 (1H, t, $J= 7.0$ Hz, H26), 7.15 (1H, br t, $J= 7.5$ Hz, H6), 7.22 (2H, dd, $J= 7.5, 7.0$ Hz, H5), 7.30–7.36 (7H, m, H24, H25, H30, H31), 10.38 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO-d_6): δ 32.0 (C2), 34.1 (C22), 45.2 (C12), 53.9 (C7), 56.2, 56.3 (C11, C13), 59.1 (C1), 64.6 (C10), 75.2 (C14), 108.7 (C19), 120.7 (C17), 125.8 (C6), 126.75 (C16

3 - This signal was buried in the peak of water. However, its location and coupling pattern could be deduced from 2D NMR experiments and other relevant ^1H signals.

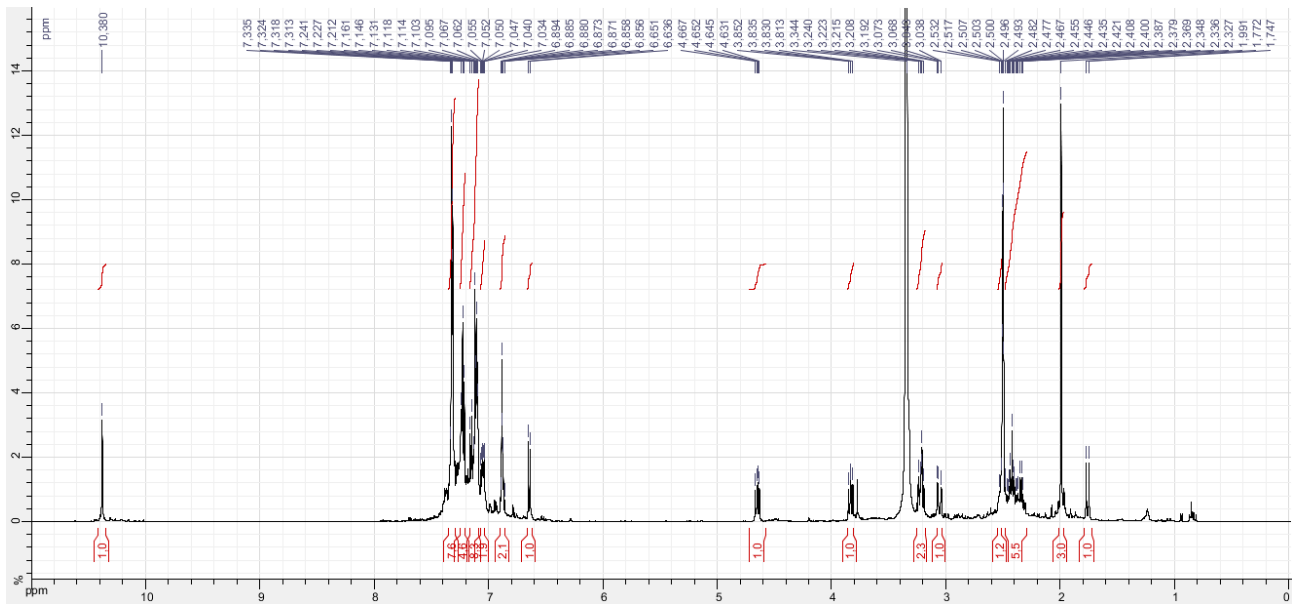
or C26), 126.85 (C15), 126.91 (C16 or C26), 128.18 128.20 (C4, C25), 128.46, 128.47 (C5, C30), 128.6 (C18), 128.9 (C31), 129.1 (C24), 130.0 (C29), 132.9 (C8), 134.5 (C28), 136.6 (C27), 138.4 (C23), 140.0 (C3), 143.4 (C20), 176.6 (C21), 198.2 (C9). HRMS (TOF-ES⁺): m/z 554.2801 (MH⁺ C₃₇H₃₆N₃O₂⁺ requires 554.2802).



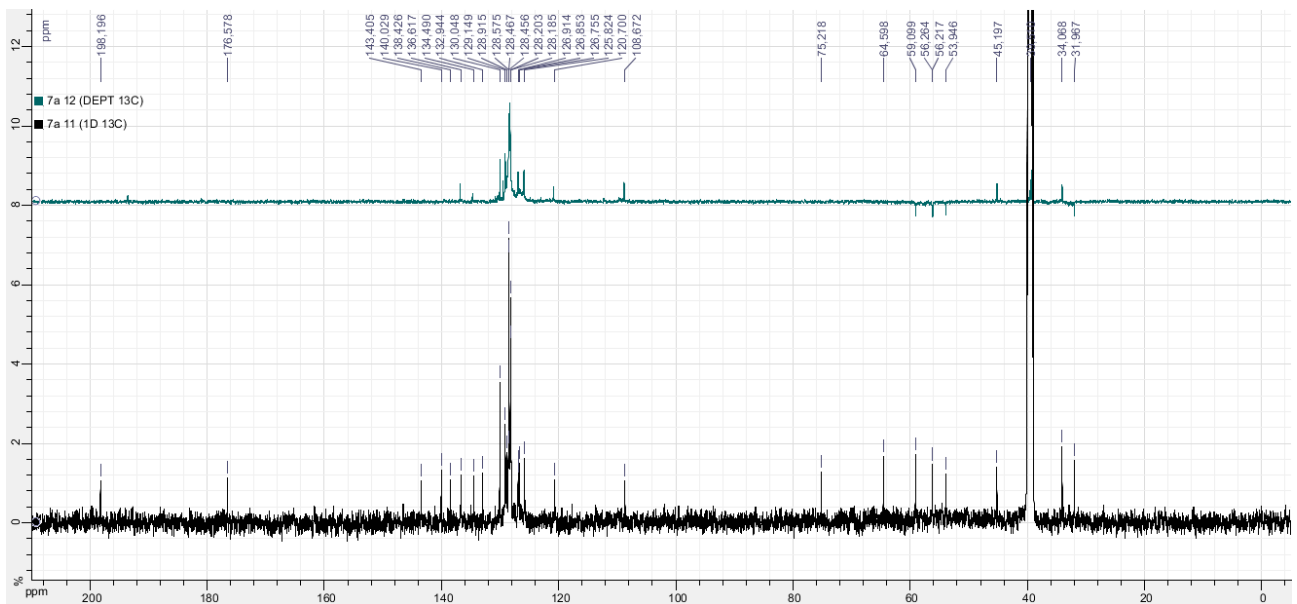
¹H NMR spectrum (CDCl₃, 400 MHz).



¹³C NMR spectrum (CDCl₃, 100.6 MHz).

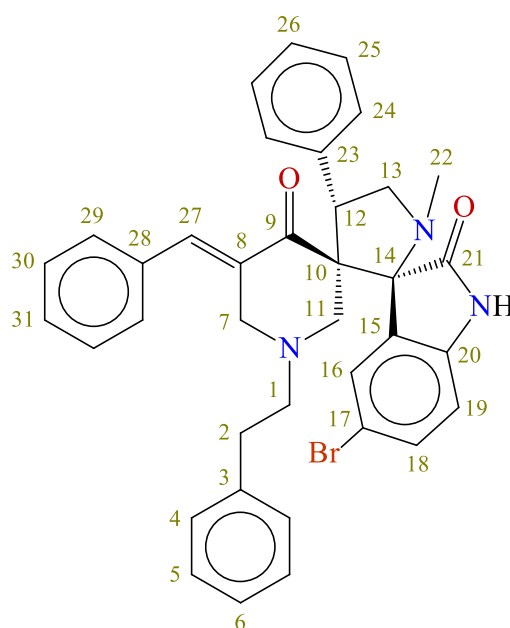


¹H NMR spectrum (DMSO-d₆, 500 MHz).

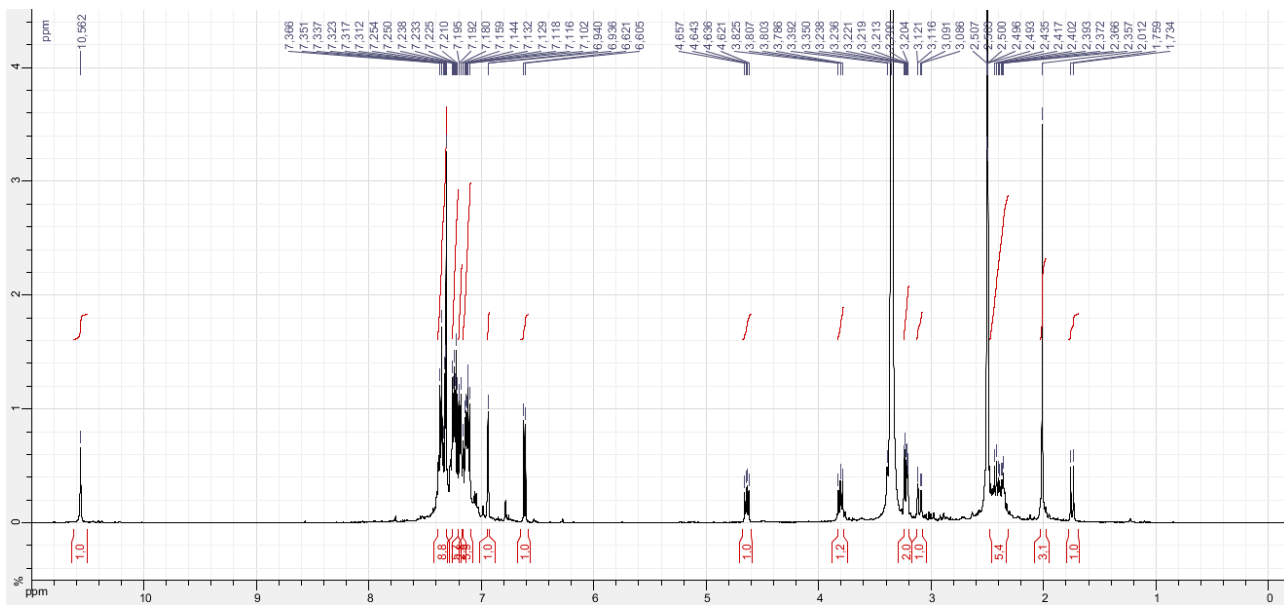


¹³C NMR and DEPT135 NMR spectra (DMSO-d₆, 125.8 MHz).

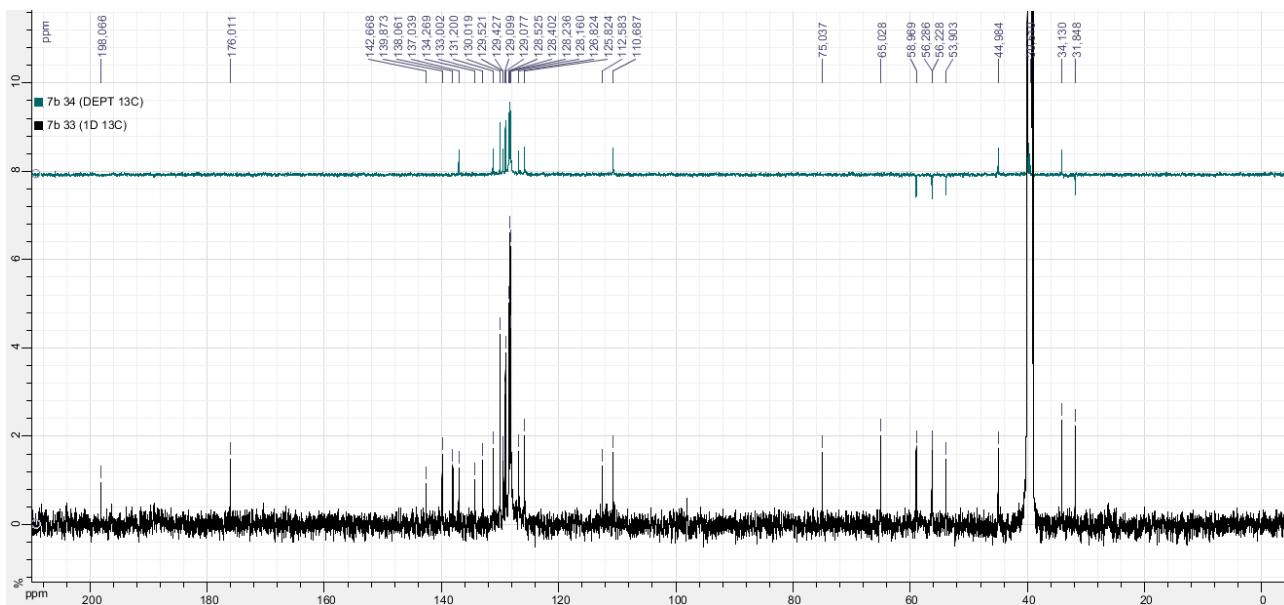
1-Methyl-4(phenyl)pyrrolo-(spiro[2.3]-5"-bromooxindole)-spiro[3.3]-5'-(phenylmethylidene)-1'-phenylethyl-4'-piperidinone (**7b**)



Pale yellow solid. $R_f \approx 0.62$ [UV-active, Acetone:hexane 10%]. M.p. 123-126 °C. FTIR (ATR, neat): ν 3387 (w, N-H), 3026 (w, C-H), 1711 (s, C=O), 1614 (m, C=C), 1181 (m, C-N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.75 (1H, d, $J=12.5$ Hz, H11a), 2.01 (3H, s, H22), 2.32–2.53 (4H, m, H1, H2), 3.10 (1H, dd, $J=15.0, 3.0$ Hz, H7a), 3.22 (1H, dd, $J=8.5, 7.5$ Hz, H13a), 3.22 (1H, dd, $J=12.5, 1.5$ Hz, H11b), 3.38 (1H, br d, $J=15.0$ Hz, H7b),³ 3.81 (1H, dd, $J=11.0, 8.5$ Hz, H13b), 4.64 (1H, dd, $J=11.0, 7.5$ Hz, H12), 6.61 (1H, d, $J=8.0$ Hz, H19), 6.94 (1H, d, $J=2.0$ Hz, H16), 7.11 (2H, br d, $J=7.5$ Hz, H4), 7.12 (1H, t, $J=7.5$ Hz, H26), 7.13 (1H, m, H27), 7.14 (1H, t, $J=7.5$ Hz, H6), 7.19 (2H, br dd, $J=8.0, 2.0$ Hz, H29), 7.23 (2H, t, $J=7.5$ Hz, H5), 7.24 (1H, dd, $J=8.0, 2.0$ Hz, H18), 7.30–7.38 (7H, m, H24, H25, H30, H31), 10.56 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 31.8 (C2), 34.1 (C22), 45.0 (C12), 53.9 (C7), 56.2, 56.3 (C11, C13), 59.0 (C1), 65.0 (C10), 75.0 (C14), 110.7 (C19), 112.6 (C17), 125.8 (C6), 126.8 (C26), 128.16 (C4), 128.24 (C25), 128.4 (C5), 128.5 (C30), 129.08 (C24), 129.10 (C31), 129.4 (C15), 129.5 (C16), 130.0 (C29), 131.2 (C18), 133.0 (C8), 134.3 (C28), 137.0 (C27), 138.1 (C23), 139.9 (C3), 142.7 (C20), 176.0 (C21), 198.1 (C9). HRMS (TOF-ES⁻) m/z 630.0966 ($[\text{M}-\text{H}]^- \text{C}_{37}\text{H}_{33}^{79}\text{BrN}_3\text{O}_2^-$ requires 630.1761), 632.0932 ($[\text{M}-\text{H}]^- \text{C}_{37}\text{H}_{33}^{81}\text{BrN}_3\text{O}_2^-$ requires 632.1741).

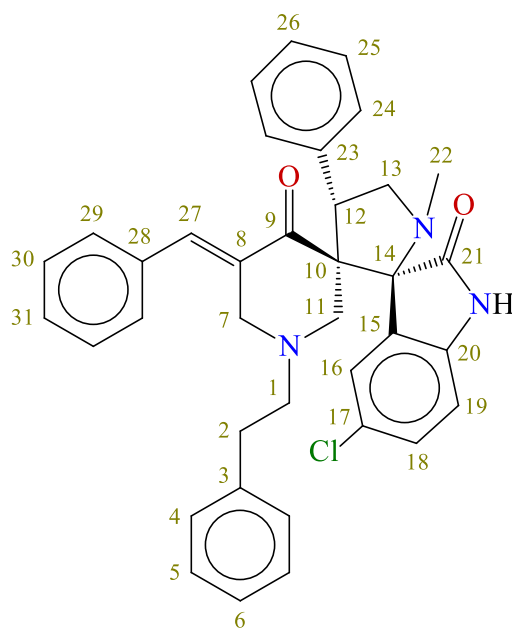


¹H NMR spectrum (DMSO-d₆, 500 MHz).

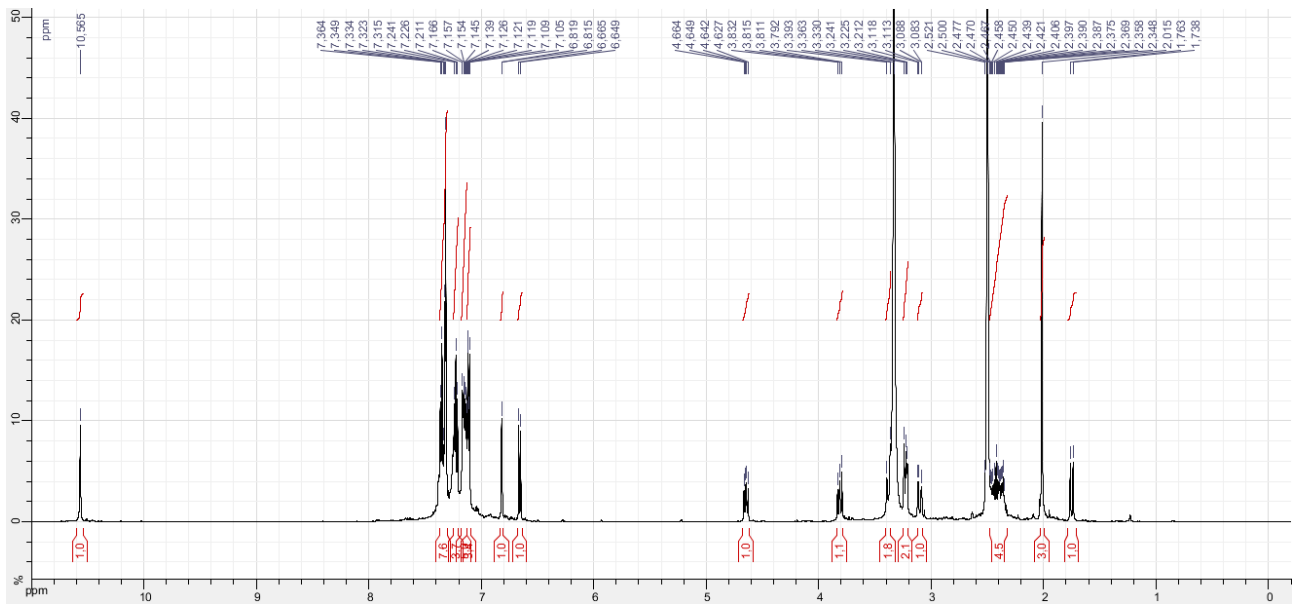


¹³C NMR and DEPT135 NMR spectra (DMSO-d₆, 125.8 MHz).

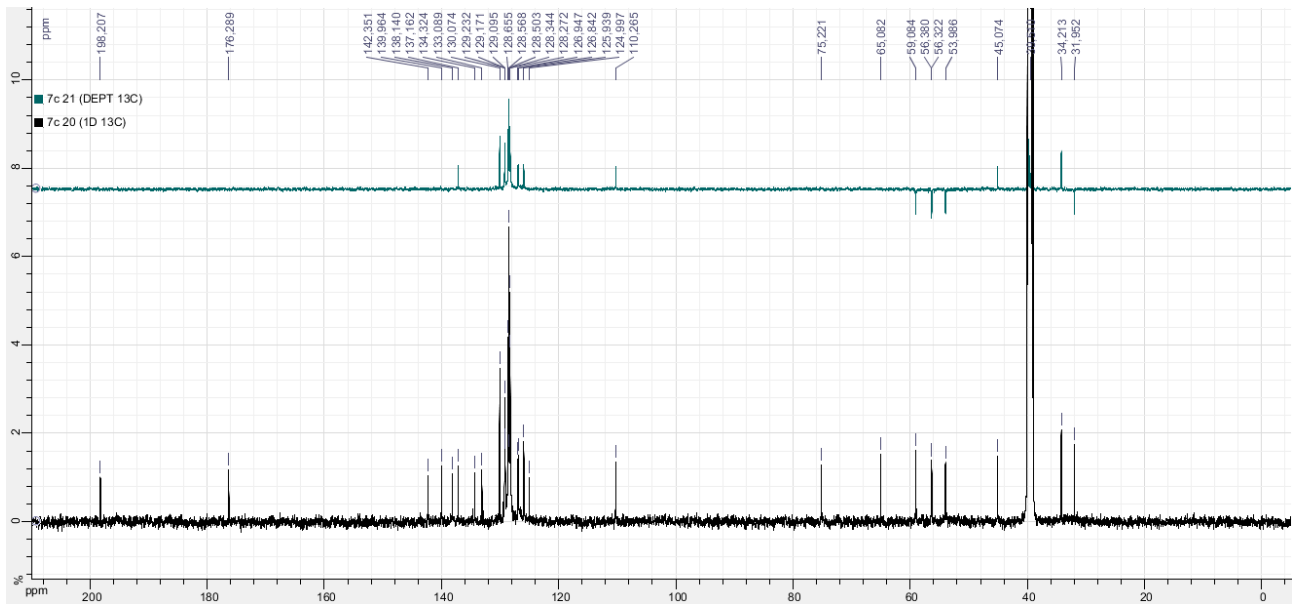
1-Methyl-4(phenyl)pyrrolo-(spiro[2.3]-5"-chlorooxindole)-spiro[3.3]-5'-(phenylmethylidene)-1'-phenylethyl-4'-piperidinone (**7c**)



Pale yellow solid. $R_f \approx 0.63$ [UV-active, Acetone:hexane 10%]. M.p. 131-134 °C. FTIR (ATR, neat): ν 3244 (w, N-H), 3027 (w, C-H), 1698 (s, C=O), 1615 (m, C=C), 1182 (m, C-N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.75 (1H, d, $J=12.5$ Hz, H11a), 2.01 (3H, s, H22), 2.32–2.53 (4H, m, H1, H2), 3.10 (1H, dd, $J=15.0, 2.5$ Hz, H7a), 3.23 (1H, dd, $J=8.5, 7.5$ Hz, H13a), 3.23 (1H, dd, $J=12.5, 1.5$ Hz, H11b), 3.38 (1H, br d, $J=15.0$ Hz, H7b), 3.81 (1H, dd, $J=11.0, 8.5$ Hz, H13b), 4.65 (1H, dd, $J=11.0, 7.5$ Hz, H12), 6.66 (1H, d, $J=8.0$ Hz, H19), 6.82 (1H, d, $J=2.0$ Hz, H16), 7.11 (2H, br d, $J=7.5$ Hz, H4), 7.12 (1H, dd, $J=8.0, 2.0$ Hz, H18), 7.14 (1H, m, H27), 7.15 (1H, t, $J=7.5$ Hz, H6), 7.13–7.18 (3H, m, H26, H29), 7.23 (2H, t, $J=7.5$ Hz, H5), 7.30–7.38 (7H, m, H24, H25, H30, H31), 10.56 (1H, s, NH). ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 32.0 (C2), 34.2 (C22), 45.1 (C12), 54.0 (C7), 56.3, 56.4 (C11, C13), 59.1 (C1), 65.1 (C10), 75.2 (C14), 110.3 (C19), 125.0 (C17), 125.9 (C6), 126.8 (C26), 126.9 (C16), 128.27 (C4), 128.34 (C25), 128.5 (C5), 128.6 (C18), 128.7 (C30), 129.1 (C15), 129.17 (C24), 129.23 (C31), 130.1 (C29), 133.1 (C8), 134.3 (C28), 137.2 (C27), 138.1 (C23), 140.0 (C3), 142.4 (C20), 176.3 (C21), 198.2 (C9). HRMS (TOF-ES $^+$): m/z 588.2390 (MH^+ $\text{C}_{37}\text{H}_{35}^{35}\text{ClN}_3\text{O}_2^+$ requires 588.2413).

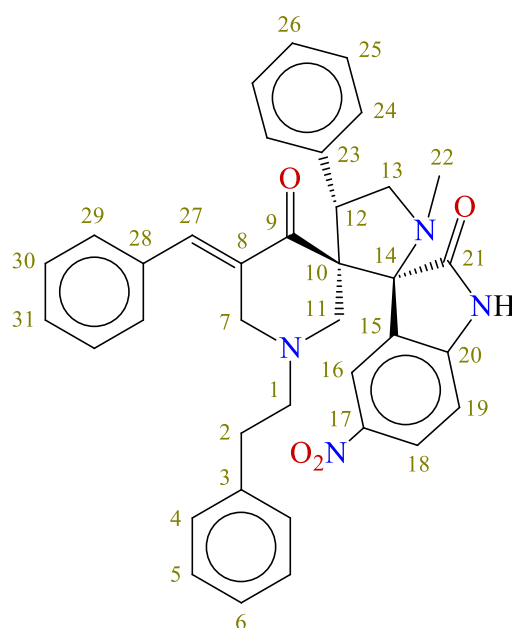


¹H NMR spectrum (DMSO-d₆, 500 MHz).

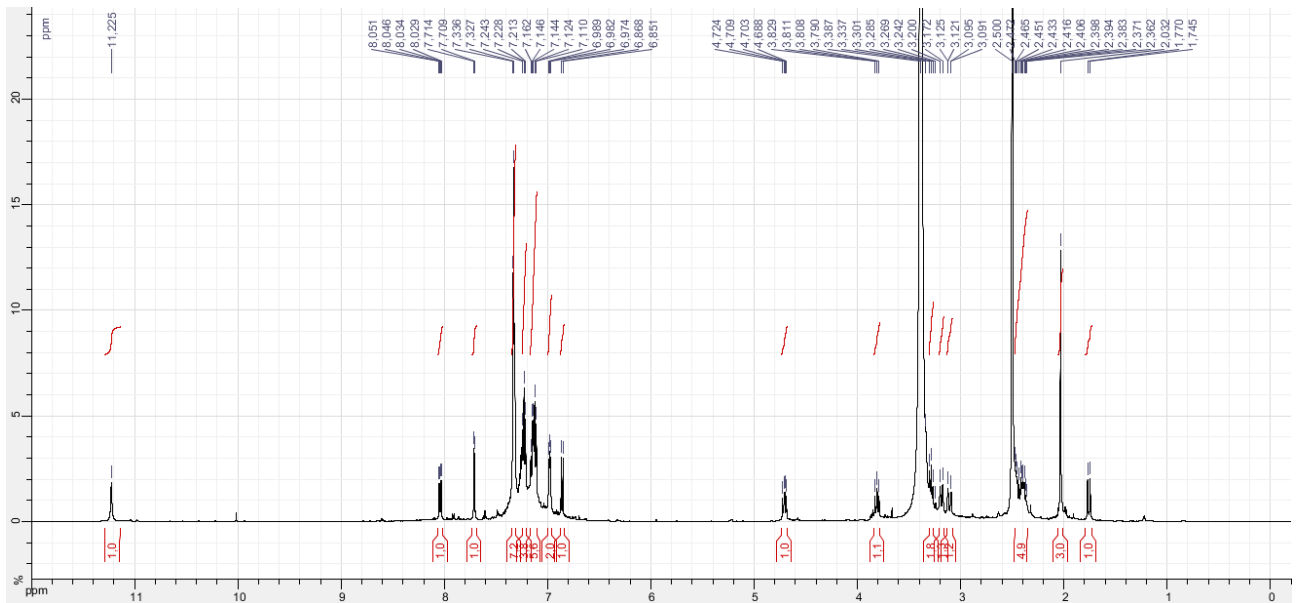


¹³C NMR and DEPT135 NMR spectra (DMSO-d₆, 125.8 MHz).

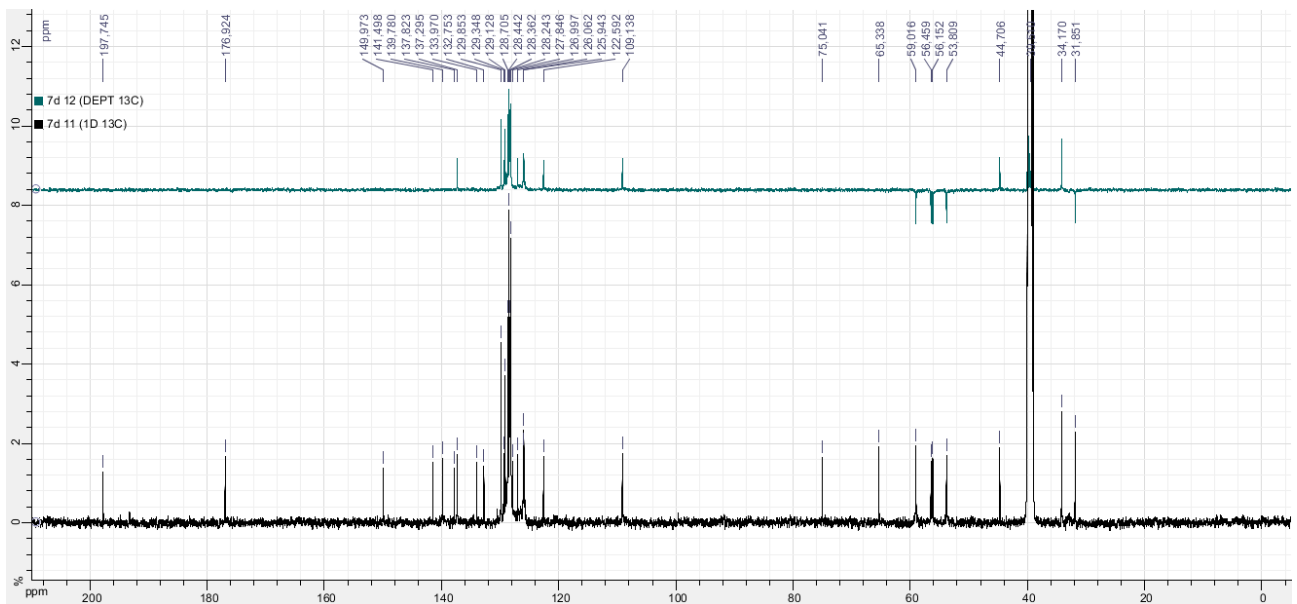
1-Methyl-4(phenyl)pyrrolo-(spiro[2.3']-5'-nitrooxindole)-spiro[3.3']-5'-(phenylmethylidene)-1'-phenylethyl piperidin-4'-one (**7d**)



Dark brown solid. $R_f \approx 0.47$ [UV-active, Acetone:hexane 10%]. M.p. 127–130 °C. FTIR (ATR, neat): ν 3248 (w, N–H), 3026 (w, C–H), 1718 (s, C=O), 1599 (m, C=C), 1182 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.76 (1H, d, $J=12.5$ Hz, H11a), 2.03 (3H, s, H22), 2.35–2.52 (4H, m, H1, H2), 3.11 (1H, dd, $J=15.0, 2.5$ Hz, H7a), 3.19 (1H, dd, $J=12.5, 1.5$ Hz, H11b), 3.29 (1H, dd, $J=8.5, 7.5$ Hz, H13a), 3.35 (1H, br d, $J=15.0$ Hz, H7b),³ 3.81 (1H, dd, $J=10.5, 8.5$ Hz, H13b), 4.71 (1H, dd, $J=10.5, 7.5$ Hz, H12), 6.86 (1H, d, $J=8.5$ Hz, H19), 6.98 (2H, m, H29), 7.12 (2H, d, $J=7.0$ Hz, H4), 7.14 (1H, m, H27), 7.14 (1H, t, $J=8.0$ Hz, H26), 7.15 (1H, t, $J=8.0$ Hz, H6), 7.23 (2H, dd, $J=8.0, 7.0$ Hz, H5), 7.31–7.35 (7H, m, H24, H25, H30, H31), 7.71 (1H, d, $J=2.5$ Hz, H16), 8.04 (1H, dd, $J=8.5, 2.5$ Hz, H18), 11.22 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 31.9 (C2), 34.2 (C22), 44.7 (C12), 53.8 (C7), 56.2, 56.5 (C11, C13), 59.0 (C1), 65.3 (C10), 75.0 (C14), 109.1 (C19), 122.6 (C16), 125.9 (C6), 126.1 (C18), 127.0 (C26), 127.8 (C15), 128.2 (C4), 128.36 (C25), 128.44 (C5), 128.7 (C30), 129.1 (C24), 129.3 (C31), 129.9 (C29), 132.8 (C8), 134.0 (C28), 137.3 (C27), 137.8 (C23), 139.8 (C3), 141.5 (C17), 150.0 (C20), 176.9 (C21), 197.7 (C9). HRMS (TOF-ES⁺): m/z 599.2764 (MH^+ $\text{C}_{37}\text{H}_{35}\text{N}_4\text{O}_4^+$ requires 599.2653).

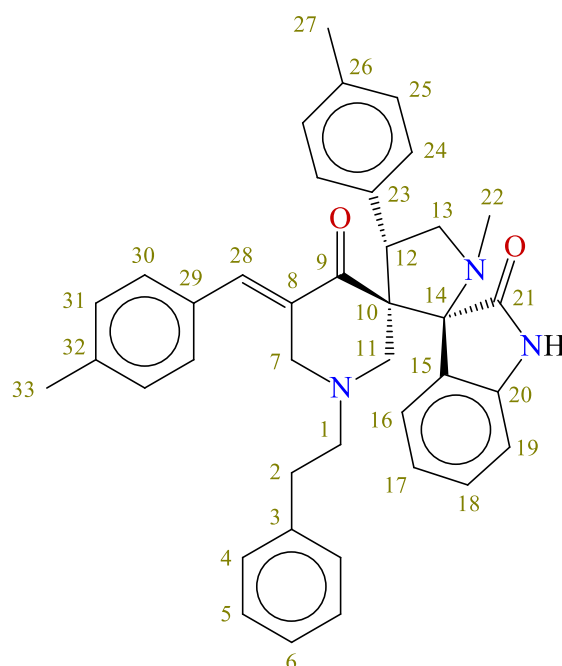


¹H NMR spectrum (DMSO-d₆, 500 MHz).

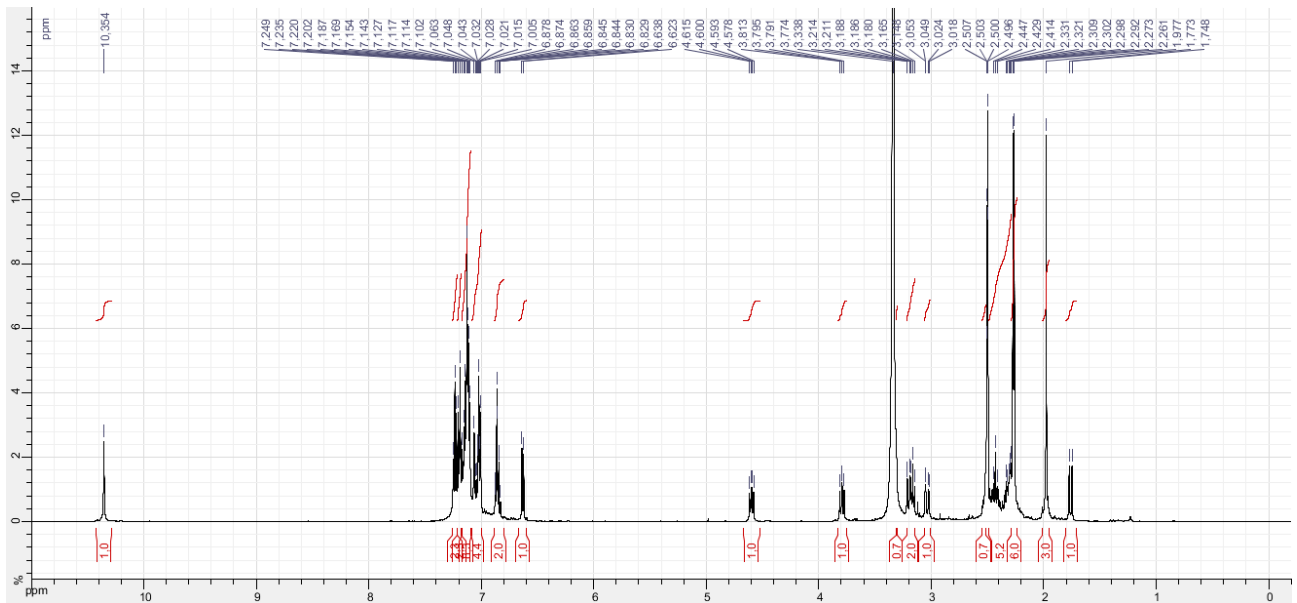


¹³C NMR and DEPT 135 NMR spectra (DMSO-d₆, 125.8 MHz).

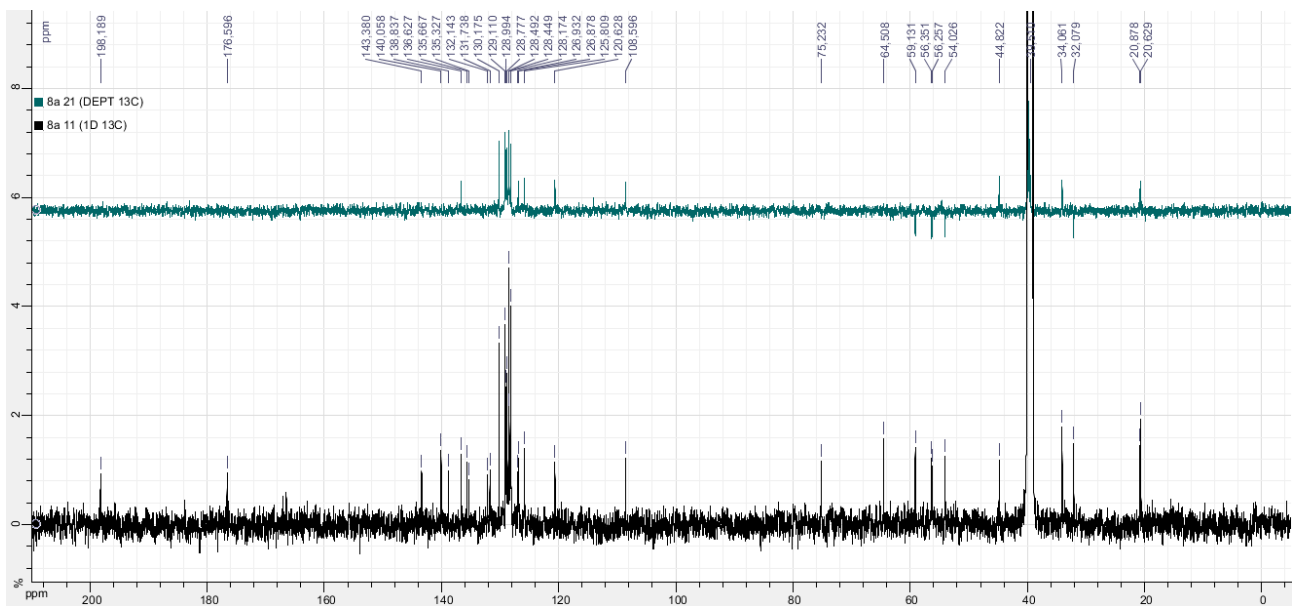
1-Methyl-4-(4-methylphenyl)pyrrolo-(spiro[2.3]oxindole)-spiro[3.3']-5'-(4-methylphenyl
methylidene)-1'-phenylethyl-4'-piperidinone (**7e**)



Pale yellow solid. $R_f \approx 0.40$ [UV-active, Acetone:hexane 10%]. M.p. 192–195 °C. FTIR (ATR, neat): ν 3397 (w, N–H), 3023 (w, C–H), 1694 (s, C=O), 1605 (m, C=C), 1177 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.76 (1 H, d, $J = 12.5$ Hz, H11a), 1.98 (3 H, s, H22), 2.26 (3 H, s, H27), 2.27 (3 H, s, H33), 2.28–2.55 (4H, m, H1, H2), 3.04 (1 H, dd, $J = 15.0, 2.5$ Hz, H7a), 3.16 (1 H, dd, $J = 8.5, 7.5$ Hz, H13a), 3.20 (1 H, dd, $J = 12.5, 1.5$ Hz, H11b), 3.33 (1 H, dd, $J = 15.0, 2.0$ Hz, H7b),³ 3.79 (1H, dd, $J = 11.0, 8.5$ Hz, H13b), 4.60 (1 H, dd, $J = 11.0, 7.5$ Hz, H12), 6.63 (1 H, br d, $J = 7.5$ Hz, H19), 6.82–6.89 (2 H, m, H16, H17), 7.01 (2 H, d, $J = 8.0$ Hz, H30), 7.03 (1 H, ddd, $J = 7.5, 7.0, 2.0$ Hz, H18), 7.06 (1 H, dd, $J = 2.5, 2.0$ Hz, H28), 7.11 (2 H, br d, $J = 8.0$ Hz, H25), 7.12 (2 H, br d, $J = 7.0$ Hz, H4), 7.14 (2 H, br d, $J = 8.0$ Hz, H31), 7.15 (1 H, t, $J = 8.0$ Hz, H6), 7.19 (2 H, br d, $J = 8.0$ Hz, H24), 7.23 (2 H, dd, $J = 8.0, 7.0$ Hz, H5), 10.35 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 20.6 (C27), 20.9 (C33), 32.1 (C2), 34.1 (C22), 44.8 (C12), 54.0 (C7), 56.3, 56.4 (C11, C13), 59.1 (C1), 64.5 (C10), 75.2 (C14), 108.6 (C19), 120.6 (C17), 125.8 (C6), 126.88 (C16), 126.93 (C15), 128.2 (C4), 128.5 (C18), 128.4 (C5), 128.8 (C25), 129.0 (C31), 129.1 (C24), 130.2 (C30), 131.7 (C29), 132.1 (C8), 135.3 (C23), 135.7 (C26), 136.6 (C28), 138.8 (C32), 140.1 (C3), 143.4 (C20), 176.6 (C21), 198.2 (C9). HRMS (TOF-ES⁺): m/z 582.3081 (MH^+ $\text{C}_{39}\text{H}_{40}\text{N}_3\text{O}_2^+$ requires 582.3115), 604.2894 (MNa^+ $\text{C}_{39}\text{H}_{39}\text{N}_3\text{NaO}_2^+$ requires 604.2935).

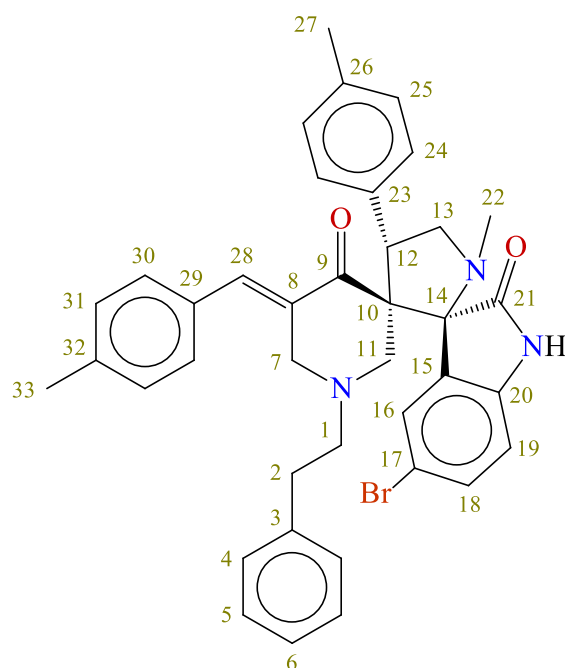


¹H NMR spectrum (DMSO-d₆, 500 MHz).

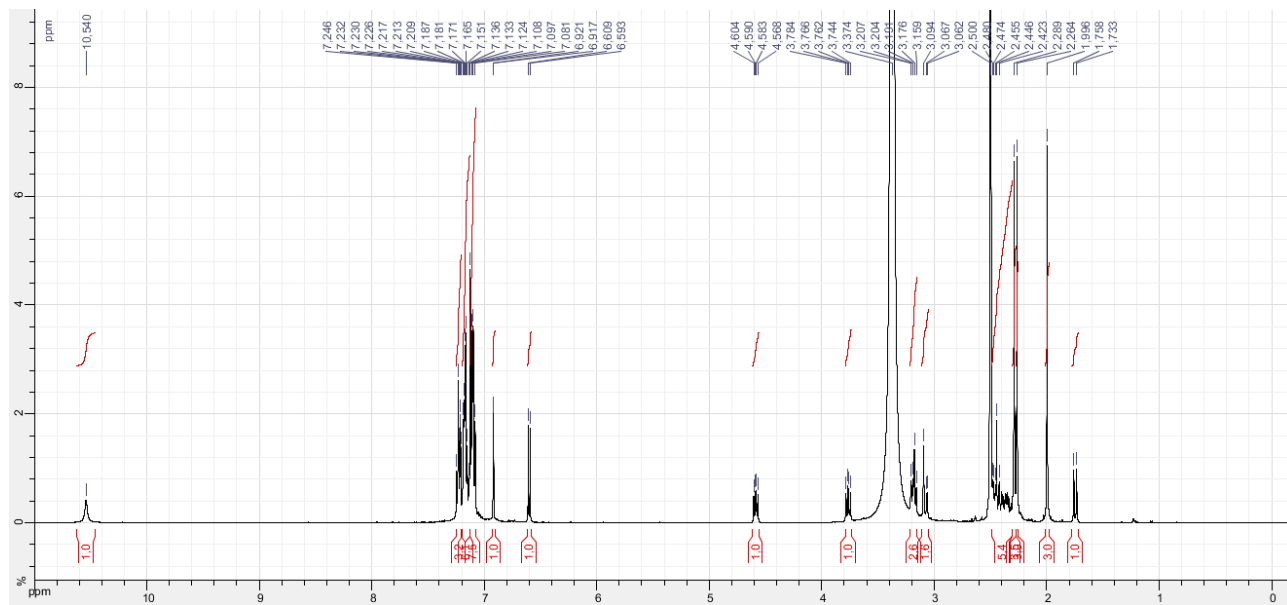


¹³C NMR and DEPT135 NMR spectra (DMSO-d₆, 125.8 MHz).

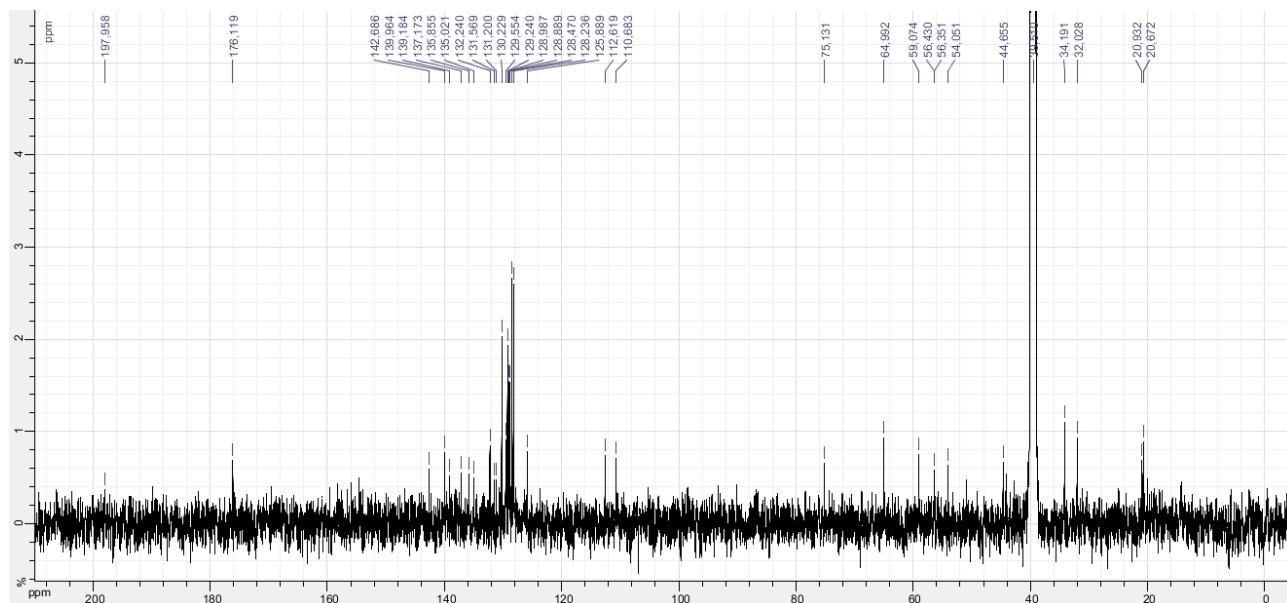
1-Methyl-4-(4-methylphenyl)pyrrolo-(spiro[2.3"]-5"-bromooxindole)-spiro[3.3']-5'-(4-methylphenyl methylidene)-1'-phenylethyl-4'-piperidinone (**7f**)



Pale yellow solid. $R_f \approx 0.66$ [UV-active, Acetone:hexane 10%]. M.p. 205–208 °C. FTIR (ATR, neat): ν 3347 (w, N–H), 3024 (w, C–H), 1690 (s, C=O), 1604 (m, C=C), 1180 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.75 (1 H, d, J = 12.5 Hz, H11a), 2.00 (3 H, s, H22), 2.26 (3 H, s, H27), 2.29 (3 H, s, H33), 2.29–2.53 (4H, m, H1, H2), 3.08 (1 H, dd, J = 15.0, 2.5 Hz, H7a), 3.17 (1 H, dd, J = 8.5, 7.0 Hz, H13a), 3.19 (1 H, dd, J = 12.5, 1.5 Hz, H11b), 3.37 (1 H, br d, J = 15.0 Hz, H7b),³ 3.76 (1H, dd, J = 11.0, 8.5 Hz, H13b), 4.59 (1 H, dd, J = 11.0, 7.0 Hz, H12), 6.60 (1 H, d, J = 8.0 Hz, H19), 6.92 (1 H, d, J = 2.0 Hz, H16), 7.09 (2 H, d, J = 8.0 Hz, H30), 7.09–7.12 (1 H, m, H28), 7.12 (2 H, br d, J = 7.0 Hz, H4), 7.12 (2 H, br d, J = 8.0 Hz, H25), 7.15 (1 H, t, J = 8.0 Hz, H6), 7.17 (2 H, br d, J = 8.0 Hz, H31), 7.18 (2 H, br d, J = 8.0 Hz, H24), 7.22 (1 H, dd, J = 8.0, 2.0 Hz, H18), 7.23 (2 H, dd, J = 8.0, 7.0 Hz, H5), 10.54 (1H, br s, NH) ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 20.7 (C27), 20.9 (C33), 32.0 (C2), 34.2 (C22), 44.7 (C12), 54.1 (C7), 56.351, 56.430 (C11, C13), 59.1 (C1), 65.0 (C10), 75.1 (C14), 110.7 (C19), 112.6 (C17), 125.9 (C6), 128.2 (C4), 128.5 (C5), 128.9 (C25), 129.0 (C31), 129.2 (C24), 129.6, 129.6 (C15, C16), 130.2 (C30), 131.2 (C18), 131.6 (C29), 132.2 (C8), 135.0 (C23), 135.9 (C26), 137.2 (C28), 139.2 (C32), 140.0 (C3), 142.7 (C20), 176.1 (C21), 198.0 (C9). HRMS (TOF-ES⁺): m/z 660.2328 (MH^+ $\text{C}_{39}\text{H}_{39}^{79}\text{BrN}_3\text{O}_2^+$ requires 660.2221), 662.2327 (MH^+ $\text{C}_{39}\text{H}_{39}^{81}\text{BrN}_3\text{O}_2^+$ requires 662.2200).

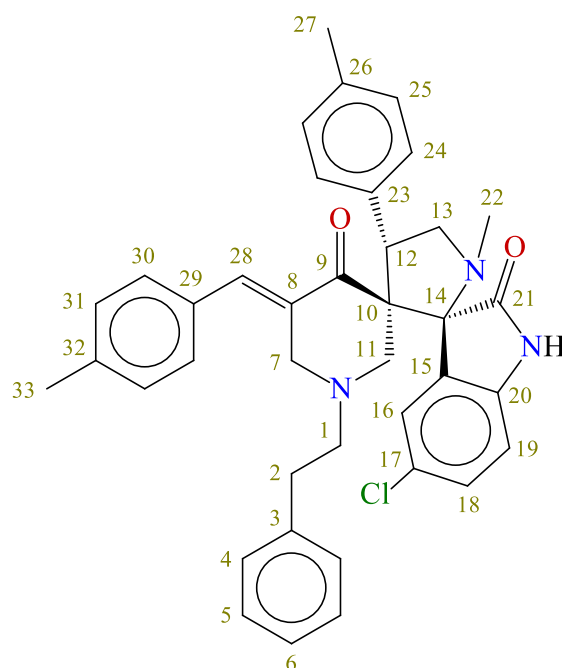


¹H NMR spectrum (DMSO-d₆, 500 MHz).



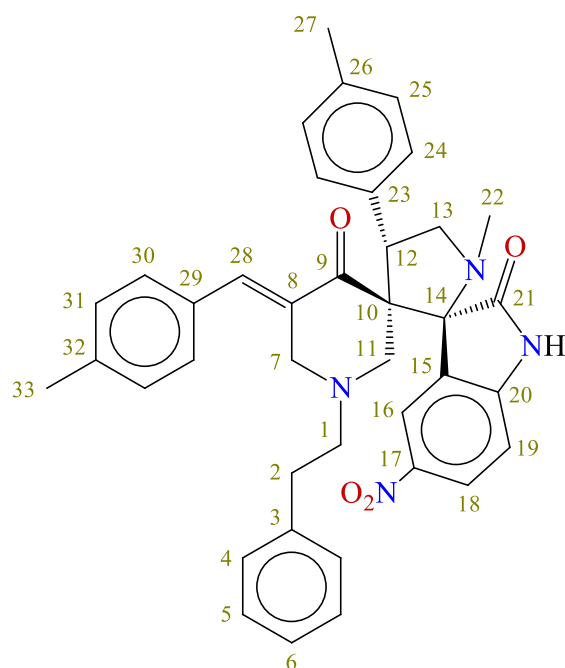
¹³C NMR spectrum (DMSO-d₆, 125.8 MHz).

1-Methyl-4-(4-methylphenyl)pyrrolo-(spiro[2.3']-5''-chlorooxindole)-spiro[3.3']-5'-(4-methylphenylmethylidene)-1'-phenylethyl-4'-piperidinone (**7g**)

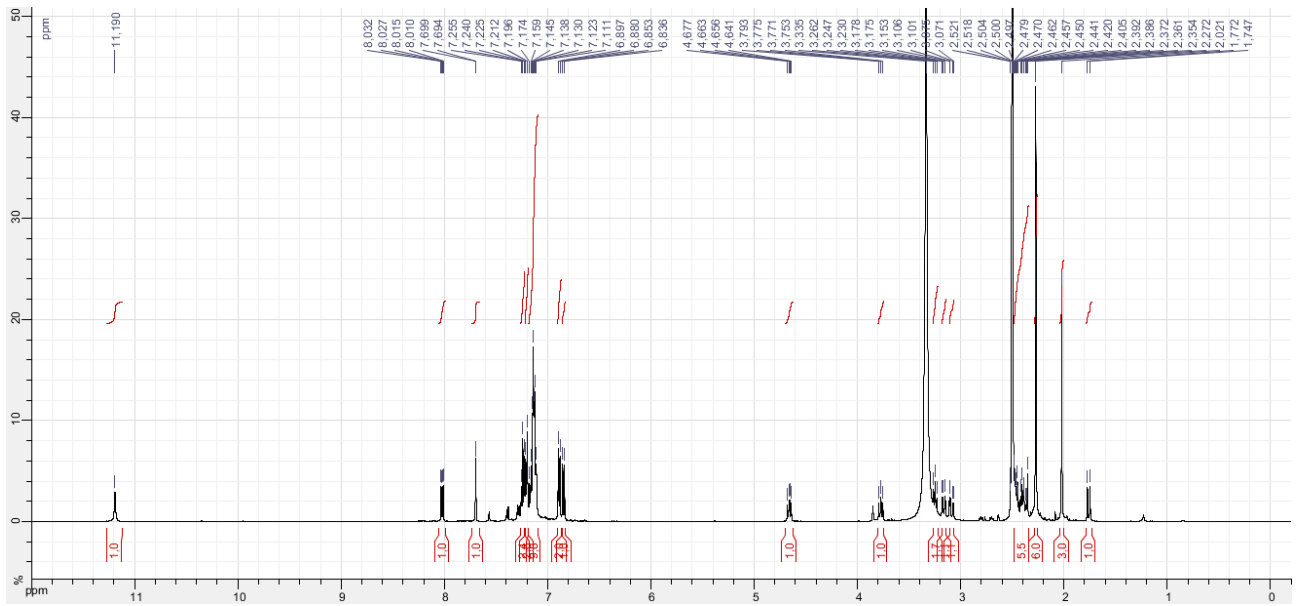


Pale yellow solid. $R_f \approx 0.66$ [UV-active, Acetone:hexane 10%]. M.p. 186–189 °C. FTIR (ATR, neat): ν 3180 (w, N–H), 3025 (w, C–H), 1691 (s, C=O), 1607 (m, C=C), 1182 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO- d_6): δ 1.75 (1 H, d, $J=12.5$ Hz, H11a), 2.00 (3 H, s, H22), 2.26 (3 H, s, H27), 2.29 (3 H, s, H33), 2.31–2.52 (4H, m, H1, H2), 3.08 (1 H, dd, $J=15.0, 2.5$ Hz, H7a), 3.18 (1 H, dd, $J=8.5, 7.5$ Hz, H13a), 3.20 (1 H, dd, $J=12.5, 1.5$ Hz, H11b), 3.37 (1 H, br d, $J=15.0$ Hz, H7b), 3.77 (1H, dd, $J=11.0, 8.5$ Hz, H13b), 4.59 (1 H, dd, $J=11.0, 7.5$ Hz, H12), 6.64 (1 H, d, $J=8.5$ Hz, H19), 6.80 (1 H, d, $J=2.0$ Hz, H16), 7.06 (2 H, d, $J=8.0$ Hz, H30), 7.09 (1 H, dd, $J=8.5, 2.0$ Hz, H18), 7.11 (1 H, m, H28), 7.12 (2 H, br d, $J=7.0$ Hz, H4), 7.12 (2 H, br d, $J=7.5$ Hz, H25), 7.15 (1 H, t, $J=8.0$ Hz, H6), 7.17 (2 H, br d, $J=8.0$ Hz, H31), 7.19 (2 H, br d, $J=7.5$ Hz, H24), 7.23 (2 H, dd, $J=8.0, 7.0$ Hz, H5), 10.53 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO- d_6): δ 20.6 (C27), 20.9 (C33), 32.0 (C2), 34.1 (C22), 44.6 (C12), 54.0 (C7), 56.3, 56.4 (C11, C13), 59.0 (C1), 64.9 (C10), 75.2 (C14), 110.1 (C19), 124.8 (C17), 125.8 (C6), 126.8 (C16), 128.2 (C4), 128.3 (C18), 128.4 (C5), 128.8 (C25), 128.9 (C31), 129.1 (C15), 129.2 (C24), 130.1 (C30), 131.5 (C29), 132.2 (C8), 135.0 (C23), 135.8 (C26), 137.1 (C28), 139.1 (C32), 139.9 (C3), 142.3 (C20), 176.2 (C21), 198.1 (C9). HRMS (TOF-ES $^+$): m/z 616.2735 (MH $^+$ C $_{39}$ H $_{39}$ ^{35}Cl N $_3$ O $_2^+$ requires 616.2726), 638.2595 (MNa $^+$ C $_{39}$ H $_{38}$ ^{35}Cl N $_3$ NaO $_2^+$ requires 638.2545).

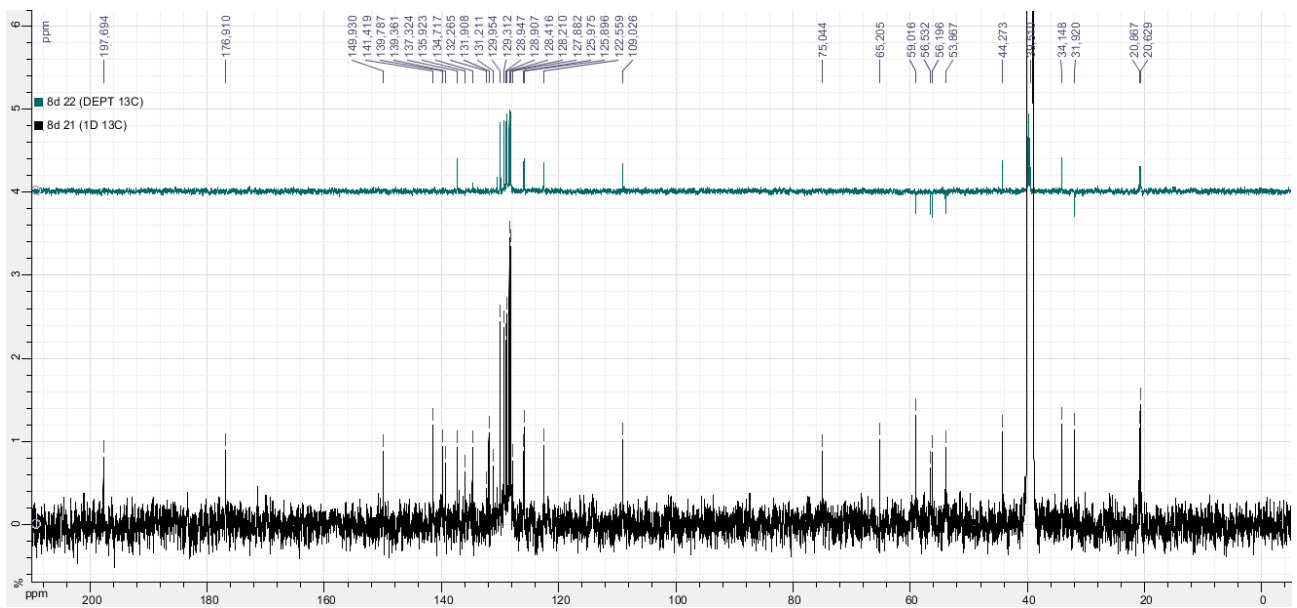
1-Methyl-4-(4-methylphenyl)pyrrolo-(spiro[2.3"]-5"-nitrooxindole)-spiro[3.3']-5'-(4-methylphenylmethylidene)-1'-phenylethyl-4'-piperidinone (**7h**)



Dark yellow solid. $R_f \approx 0.41$ [UV-active, Acetone:hexane 10%]. M.p. 191–194 °C. FTIR (ATR, neat): ν 3294 (w, N–H), 3028 (w, C–H), 1701 (s, C=O), 1601 (m, C=C), 1177 (m, C–N) cm^{-1} . ^1H NMR (500 MHz, DMSO-d_6): δ 1.76 (1 H, d, $J = 12.5$ Hz, H11a), 2.02 (3 H, s, H22), 2.27 (6 H, br s, H27, H33), 2.34–2.53 (4H, m, H1, H2), 3.09 (1 H, dd, $J = 15.0, 2.5$ Hz, H7a), 3.16 (1 H, dd, $J = 12.5, 1.5$ Hz, H11b), 3.25 (1 H, dd, $J = 8.5, 7.5$ Hz, H13a), 3.34 (1 H, br d, $J = 15.0$ Hz, H7b),³ 3.77 (1H, dd, $J = 11.0, 8.5$ Hz, H13b), 4.66 (1 H, dd, $J = 11.0, 7.5$ Hz, H12), 6.84 (1 H, d, $J = 8.5$ Hz, H19), 6.89 (2 H, d, $J = 8.0$ Hz, H30), 7.11 (1 H, m, H28), 7.13 (2 H, br d, $J = 7.0$ Hz, H4), 7.13 (2 H, br d, $J = 8.0$ Hz, H25), 7.14 (2 H, br d, $J = 8.0$ Hz, H31), 7.16 (1 H, t, $J = 8.0$ Hz, H6), 7.20 (2 H, br d, $J = 8.0$ Hz, H24), 7.24 (2 H, dd, $J = 8.0, 7.0$ Hz, H5), 7.70 (1 H, d, $J = 2.5$ Hz, H16), 8.02 (1 H, dd, $J = 8.5, 2.5$ Hz, H18), 11.19 (1H, br s, NH). ^{13}C NMR (125.8 MHz, DMSO-d_6): δ 20.6, 20.9 (C27, C33), 31.9 (C2), 34.1 (C22), 44.3 (C12), 53.9 (C7), 56.2, 56.5 (C11, C13), 59.0 (C1), 65.2 (C10), 75.0 (C14), 109.0 (C19), 122.6 (C16), 125.9 (C6), 126.0 (C18), 127.9 (C15), 128.2 (C4), 128.4 (C5), 128.91, 128.95 (C25, C31), 129.3 (C24), 130.0 (C30), 131.9 (C29), 132.3 (C8) 134.7 (C23), 135.9 (C26), 137.3 (C28), 139.4 (C32), 139.8 (C3), 141.4 (C17), 149.9 (C20), 176.9 (C21), 197.7 (C9). HRMS (TOF-ES⁺): m/z 627.3000 (MH^+ $\text{C}_{39}\text{H}_{39}\text{N}_4\text{O}_4^+$ requires 627.2966).



¹H NMR spectrum (DMSO-d₆, 500 MHz).



¹³C NMR and DEPT 135 NMR spectra (DMSO-d₆, 125.8 MHz).