Supporting Information

Chlorizidine, A Cytotoxic 5*H*-Pyrrolo[2,1-*a*]isoindol-5-one-Containing Alkaloid from a Marine *Streptomyces* sp.

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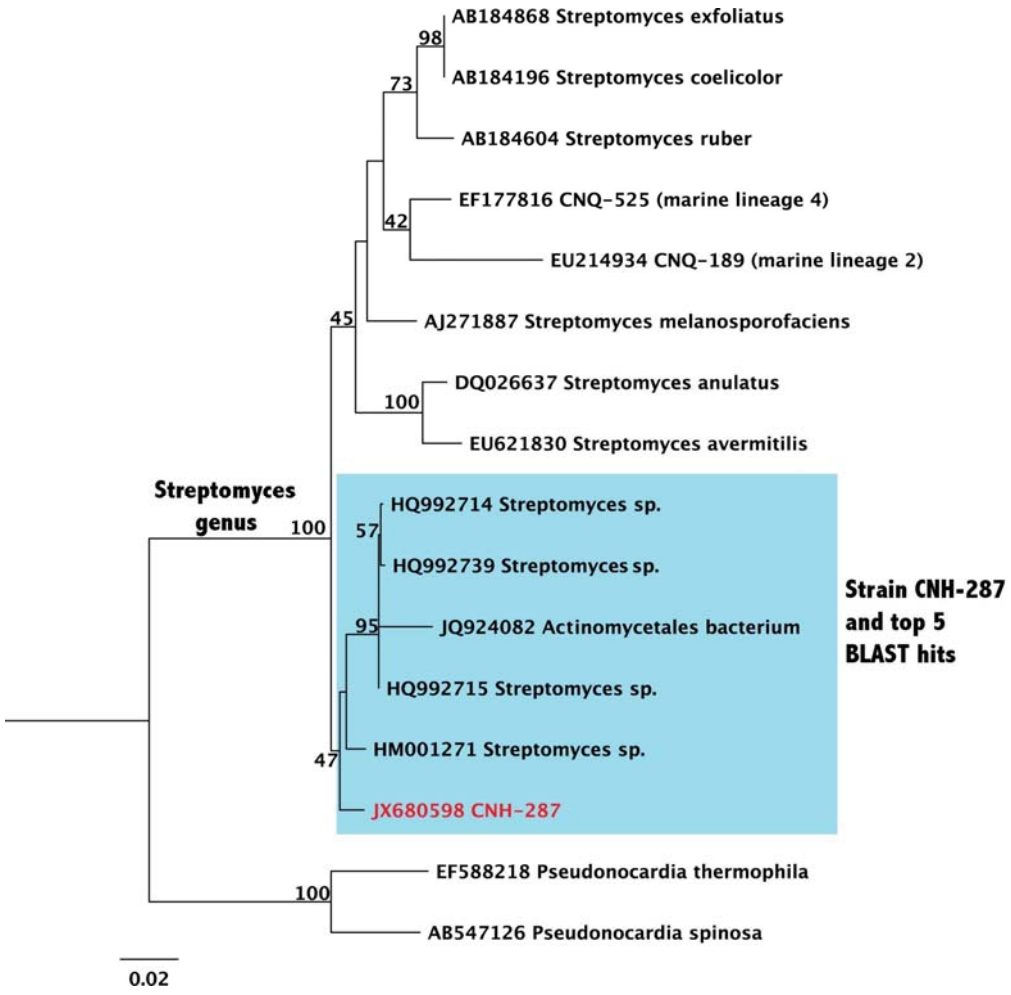
General. Pyridine was distilled from calcium hydride prior to use, and dimethylformamide (DMF) was dried prior to use by eluting through a column of activated alumina. All other reagents and solvents were purchased commercially and were used without further purification. Organic extracts were dried with sodium sulfate. Reactions were analyzed by TLC (Merck® silica gel 60 F_{254}) or, when appropriate, an analytical 1090 Series HP system (0.7 mL min^{-1}) with UV detection (254 nm) using a C18(2) Phenomenex® Luna column (5 µm, 100 x 4.6 mm). Semi-preparative RP-HPLC purification was performed on a C8(2) Phenomenex® Luna (5 µm, 250 x 10 mm), C18(2) Phenomenex® Luna (5 µm, 250 x 10 mm) or Kromasil® Diol (5 µm, 250 x 10 mm) column with UV detection (254 nm). ^{1}H NMR spectra were recorded at 500 MHz or 600 MHz in DMSO-d_6 (residual solvent referenced to 2.50 ppm), CD_3CN (residual solvent referenced to 1.94 ppm), or benzene-d_6 (residual solvent referenced to 7.16 ppm) on a Varian Inova 500 MHz or Bruker 600 MHz NMR spectrometer. ^{13}C NMR spectra were recorded at 125 MHz in DMSO-d_6 (referenced to 39.5 ppm), CD_3CN (referenced to 1.2 ppm) or benzene-d_6 (referenced to 128.0 ppm) on a Varian Inova 500 MHz NMR spectrometer.

Phylogeny of strain CNH-287. The identity of strain CNH-287 was determined by phylogenetic analysis of the 16S rRNA gene sequence. The sequence from this strain was used as a query in a BLASTn search at the NCBI database. The top hits were all members of the *Streptomyces* genus. Twenty-two of the top BLAST hits were from strains that shared 99% or greater identity with the CNH-287 sequence. Of these twenty-two strains, all that had a specified location were cultured from the marine environment, suggesting that strain CNH-287 may be part of a previously undescribed marine *Streptomyces* lineage.

A phylogeny of strain CNH-287 was built using the top five BLAST hits for this strain and a number of diverse *Streptomyces* sequences, including two from described marine *Streptomyces* lineages. Two outgroups were used that fell into the genus *Pseudonocardia*, members of the Pseudonocardiaceae which clade closely with streptomycetes in overall actinobacterial phylogenies.[1] Sequences were aligned using MUSCLE,[2] and a maximum likelihood phylogeny built using raxmlGUI.[3] GTR+G was used as a substitution model with 100 thorough bootstraps.

(1) Stackebrandt, E.; Rainey, F. A.; Ward-Rainey, N. L. *Int. J. Syst. Bacteriol.* **1997**, *47*, 479-491.
(2) Edgar, R. C. *Nucleic Acids Res.* **2004**, *32*, 1792-1797.
(3) Silvestro, D.; Michalak, I. *Org. Divers. Evol.* **2011**.

Figure S1. Phylogenetic tree for CNH-287.



Cultivation of strain CNH-287. The strain was cultured in 2.8 L Fernbach flasks (20 x 1 L) in a seawater-based medium (per 1 L seawater: 10 g of starch, 4 g of yeast extract, 2 g of peptone, 1 g of $CaCO_3$, 40 mg of $Fe_2(SO_4)_3 \cdot 4H_2O$, 100 of mg KBr) and shaken at 230 rpm at 27 °C.

Isolation and purification of chlorizidines A (1) and B (7). After one day of a 20 L cultivation, sterilized Amberlite XAD-18 resin (20 g L^{-1}) was added. The culture and resin were shaken for six days. The resin was filtered through cheesecloth, washed with deionized water, and eluted with acetone. The acetone was removed under reduced pressure, and the resulting aqueous layer was extracted with EtOAc (3 x 400 mL). The combined extracts were concentrated to yield 1.7 g of crude extract. The extract was fractionated by column chromatography on silica gel (20 g), eluting with a step gradient of isooctane, EtOAc, and CH_3OH. The 1:1 isooctane/EtOAc fraction contained chlorizidine A (**1**). This sample was

further fractionated on C18 column (3 g) under vacuum, eluting with a step gradient of CH_3CN and water. Fractions 4:1 and 9:1 CH_3CN/water were purified by reversed-phase HPLC (80% CH_3CN in water, t_R = 16 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to afford chlorizidine A (**1**) (50.0 mg) as a yellow film. In a separate 20 L culture, resin was instead added at the end of the cultivation (day seven). The crude extract (2.0 g) was fractionated by column chromatography on silica gel (20 g), again eluting with a step gradient of isooctane, EtOAc, and CH_3OH. The 3:2 isooctane/EtOAc fraction contained chlorizidine A (**1**) and chlorizidine B (**7**). Chlorizidine A (**1**) was purified as before to yield 10.0 mg. Chlorizidine B (**7**) was purified by reversed-phase HPLC (70% CH_3CN in water, t_R = 16.5 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to give 16.0 mg as a rapidly decomposing white solid.

Chlorizidine A (1): UV/vis (CH_3CN) λ_{max} = 212 (31800), 261 (47400), 340 (7300), 356 (8500), 409 (2300); $[\alpha]_D$ –35 (*c* 0.50, CH_3CN); IR (film) $\tilde{\nu}$ = 3119, 1721, 1626 cm^{-1}; ^1H NMR see Table 1; ^{13}C NMR (CD_3CN) δ 163.9, 163.0, 157.5, 136.8, 135.5, 132.7, 116.9, 113.3, 113.0, 109.7, 108.2, 106.0, 105.9, 101.9, 99.3, 53.0, 32.5, 25.4; HRESI-FT-MS (Orbit-Trap-MS): *m/z* $(M+H)^+$ calcd for $C_{18}H_{11}{}^{35}Cl_4N_2O_3$ 442.9524, found 442.9511.

Chlorizidine B (7): UV/vis (CH_3CN) λ_{max} = 299; ^1H NMR see Table S1; HRESI-FT-MS (Orbit-Trap-MS): *m/z* $(M+H)^+$ calcd for $C_{17}H_{13}{}^{35}Cl_4N_2O_2$ 416.9731, found 416.9724.

Diacetyl chlorizidine (2): To a solution of chlorizidine A (**1**) (9.0 mg, 0.020 mmol) in dry CH_3CN (2.0 mL) at room temperature was added dry Et_3N (85 µL, 0.61 mmol) followed by acetic anhydride (30 µL, 0.30 mmol). The mixture was stirred at rt for 3 h, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The crude material was filtered on a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was purified by reversed-phase HPLC (85% CH_3CN in water, t_R = 11 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to yield 5.7 mg (54%) of **2** as a yellow film: UV/vis (CH_3CN) λ_{max} = 205 (26000), 267 (38400), 303 (2200), 316 (2200), 330 (1400), 414 (1600); $[\alpha]_D$ –140 (*c* 0.10, CH_3CN); IR (film) $\tilde{\nu}$ = 3449, 1767, 1628 cm^{-1}; ^1H NMR see Table S2; ^{13}C NMR (CD_3CN) δ 168.7, 168.0, 167.2, 165.7, 157.9, 155.3, 154.9, 148.1, 147.5, 135.5, 134.9, 131.4, 125.8, 125.2, 117.7, 117.3, 114.6, 113.5, 113.0, 109.3, 109.2, 106.2, 100.1, 100.0, 53.0, 33.8, 33.6, 24.9, 24.8, 20.7, 20.3, 20.0, 19.6; HRESI-FT-MS (Orbit-Trap-MS): *m/z* $(M+Na)^+$ calcd for $C_{22}H_{14}{}^{35}Cl_4N_2O_5Na$ 548.9555, found 548.9556.

Dimethyl chlorizidine (3): To a solution of chlorizidine A (**1**) (8.0 mg, 0.018 mmol) in dry acetone (3.0 mL) at room temperature was added potassium carbonate (50 mg, 0.36 mmol) followed by dimethyl sulfate (21 µL, 0.18 mmol). The mixture was stirred at rt overnight, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The

crude material was filtered through a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was purified by reversed-phase HPLC (95% CH_3CN in water, t_R = 14 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to give 7.5 mg (87%) of **3** as a yellow film: UV/vis (CH_3CN) λ_{max} = 214 (32700), 267 (56900), 330 (4900), 343 (5200), 408 (2100); $[\alpha]_D$ –77 (*c* 0.10, CH_3CN); IR (film) $\tilde{\nu}$ = 3421, 2945, 2854, 1750, 1615, 1581 cm^{-1}; ^1H NMR see Table S2; ^{13}C NMR (DMSO-d_6) δ 165.2, 164.1, 159.8, 158.7, 158.5, 137.5, 136.1, 132.1, 120.0, 116.2, 112.6, 107.4, 105.0, 100.9, 100.1, 99.2, 62.8, 61.8, 57.1, 52.7, 33.0, 25.2; HRESI-FT-MS (Orbit-Trap-MS) *m/z* $(M+H)^+$ calcd for $C_{20}H_{15}{}^{35}Cl_4N_2O_3$ 470.9837, found 470.9833.

Diisobutyryl chlorizidine (4): To a solution of chlorizidine A (**1**) (4.5 mg, 0.010 mmol) in dry CH_3CN (1.5 mL) at room temperature was added dry Et_3N (27 µL, 0.20 mmol) followed by isobutyryl chloride (10 µL, 0.10 mmol). The mixture was stirred at rt for 3 h, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The crude material was filtered on a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was purified by reversed-phase HPLC (95% CH_3CN in water, t_R = 12 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to give 4.2 mg (72%) of **4** as a yellow film: UV/vis (CH_3CN) λ_{max} = 205 (34000), 267 (53500), 303 (3700), 316 (3700), 330 (3300), 414 (2300); $[\alpha]_D$ –100 (*c* 0.10, CH_3CN); IR (film) $\tilde{\nu}$ = 2979, 2938, 1627 cm^{-1}; ^1H NMR Table S2; ^{13}C NMR (DMSO-d_6) δ 174.3, 174.2, 173.7, 173.2, 171.5, 157.9, 157.8, 155.7, 155.1, 155.0, 148.6, 148.3, 147.6, 135.4, 135.1, 135.0, 134.9,131.5, 131.4, 131.3, 125.4, 125.2, 125.0, 119.0, 118.4, 117.7, 117.2, 114.4, 113.6, 113.5, 113.4, 113.2, 112.9, 109.5, 109.3, 109.2, 109.0, 106.5, 106.4, 100.3, 100.2, 100.1, 54.4, 53.5, 53.3, 33.8, 33.7, 33.4, 33.3, 33.2, 32.9, 32.7, 24.9, 24.8, 24.4, 19.0, 18.6, 18.5, 18.3, 17.8, 17.7, 17.5; HRESI-FT-MS (Orbit-Trap-MS) *m/z* $(M+Na)^+$ calcd for $C_{26}H_{22}{}^{35}Cl_4N_2O_5Na$ 605.0181, found 605.0174.

Dipivaloyl chlorizidine (5): To a solution of chlorizidine A (**1**) (4.5 mg, 0.010 mmol) in dry CH_3CN (1.5 mL) at room temperature was added dry Et_3N (27 µL, 0.20 mmol) followed by pivaloyl chloride (12 µL, 0.10 mmol). The mixture was stirred at rt for 3 h, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The crude material was filtered on a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was purified by reversed-phase HPLC (97% CH_3CN in water, t_R = 13 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to give 4.7 mg (77%) of **5** as a yellow film: UV/vis (CH_3CN) λ_{max} = 205 (36000), 267 (56700), 303 (4300), 316 (4300), 330 (2400), 414 (2400); $[\alpha]_D$ –57 (*c* 0.10, CH_3CN); IR (film) $\tilde{\nu}$ = 3401, 2976, 1761, 1627 cm^{-1}; ^1H NMR Table S2; ^{13}C NMR (DMSO-d_6) δ 175.7, 175.4, 173.6, 158.0, 157.9, 156.4, 155.7, 155.4, 149.3, 149.0, 147.9, 135.4, 135.3, 135.2, 134.8, 131.4, 131.3, 124.2, 124.1, 118.7,

118.1, 117.8, 117.3, 117.2, 114.1 113.7, 113.6, 113.5, 113.2, 113.0, 109.7, 109.6, 109.3, 109.2, 109.1, 107.1, 106.7, 106.6, 100.6, 100.5, 55.0, 54.3, 53.8, 26.8, 26.7, 26.5, 26.4, 26.3, 24.6; HRESI-FT-MS (Orbit-Trap-MS) *m/z* $(M+Na)^+$ calcd for $C_{28}H_{26}{}^{35}Cl_4N_2O_5Na$ 633.0494, found 633.0487.

Dibenzoyl chlorizidine (6): To a solution of chlorizidine A (**1**) (8.0 mg, 0.018 mmol) in dry CH_3CN (3 mL) at room temperature was added dry Et_3N (50 µL, 0.36 mmol) followed by benzoyl chloride (21 µL, 0.18 mmol). The mixture was stirred at rt for 3 h, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The crude material was filtered on a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was purified by reversed-phase HPLC (90% CH_3CN in water, t_R = 14 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) to give 5.0 mg (42%) of **6** as a yellow film: UV/vis (CH_3CN) λ_{max} = 233 (50000), 267 (63700), 303 (4200), 316 (3900), 330 (2300), 414 (2300); $[\alpha]_D$ –74 (*c* 0.050, CH_3CN); IR (film) $\tilde{\nu}$ = 3485, 3058, 2963, 1754, 1629 cm^{-1}; ^1H NMR Table S2; ^{13}C NMR (benzene-d_6) δ 164.4, 164.1, 163.9, 163.4, 162.1, 157.9, 157.7, 157.5, 155.9, 155.1, 155.0, 149.9, 149.5, 148.8, 136.0, 135.9, 135.0, 134.7, 134.6, 134.4, 134.3, 134.2, 133.5, 131.3, 131.2, 131.1, 131.0, 130.9, 130.7, 130.6, 130.5, 130.4, 129.3, 129.2, 129.0, 128.8, 128.7, 128.6, 128.5, 128.4, 128.2, 128.1, 128.0, 127.1, 126.6, 125.9, 120.6, 119.9, 118.9, 118.2, 118.1, 114.8, 114.1, 112.1, 112.0, 111.8, 111.7, 108.3, 108.2, 108.1, 107.7, 107.5, 101.2, 54.6, 54.1, 53.8, 34.4, 34.0, 33.8, 25.1, 24.7; HRESI-FT-MS (Orbit-Trap-MS) *m/z* $(M+H)^+$ calcd for $C_{32}H_{19}{}^{35}Cl_4N_2O_5$ 651.0048, found 651.0033.

Diacetyl chlorizidine B (8): To a solution of chlorizidine B (**7**) (16 mg, 0.039 mmol) in dry CH_3CN (5.0 mL) at room temperature was added dry Et_3N (85 µL, 0.78 mmol) followed by acetic anhydride (36 µL, 0.39 mmol). The mixture was stirred at rt for 3 h, then diluted with water (10 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were dried, filtered, and concentrated. The crude material was filtered on a short plug of silica gel with EtOAc. The organic layer was concentrated, and the product was first purified by reversed-phase HPLC (80% CH_3CN in water, t_R = 12 min, C18(2) Phenomenex® Luna, 2.5 mL min^{-1}) and then purified by normal phase HPLC (85% isooctane in isopropanol, t_R = 15 min, Kromasil® 60-5-Diol, 2.5 mL min^{-1}) to furnish 6.0 mg (31%) of **8**: UV/vis (CH_3CN) λ_{max} = 300 (23500); $[\alpha]_D$ –75 (*c* 0.10, CH_3CN); IR (film) $\tilde{\nu}$ 3131, 2989, 1767, 1625 cm^{-1}; ^1H NMR Table S1; ^{13}C NMR (DMSO-d_6) δ 169.3, 167.7, 149.5, 149.4, 135.3, 131.8, 128.2, 122.7, 116.9, 114.9, 113.0, 109.1, 109.0, 107.3, 106.4, 99.9, 53.9, 33.8, 24.7, 20.6, 20.0; HRESI-FT-MS (Orbit-Trap-MS): *m/z* $(M+H)^+$ calcd for $C_{21}H_{17}{}^{35}Cl_4N_2O_4$ 500.9942, found 500.9935.

Thioester 9: To a solution of chlorizidine A (**1**) (15.4 mg, 0.0347 mmol) and potassium carbonate (40 mg, 0.29 mmol) in dry DMF (2.0 mL) at room temperature was added *N*-acetylcysteamine (20 μL, 0.19 mmol) via syringe. The mixture was stirred at 60 °C for 1 h, then diluted with water (5 mL) and a saturated aq. NH_4Cl solution (5 mL) and extracted with EtOAc (3 x 5 mL). The combined extracts were washed with brine (4 mL), dried, filtered, and concentrated. The product was purified by reversed-phase HPLC (55% CH_3CN in water, 0.2% TFA, t_R = 27 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}) to give 10.2 mg (52%) of **9**: UV/vis (CH_3CN/water) λ_{max} = 223, 266, 360, 417; $[\alpha]_D$ –19 (*c* 0.38, CH_3CN); IR (film) $\tilde{\nu}$ = 3352, 1728, 1626 cm^{-1}; ^1H and 2D NMR Table S3; HRMS (ESI-TOF) *m/z* $(M+H)^+$ calcd for $C_{22}H_{20}{}^{35}Cl_4N_3O_4S$ 561.9929, found 561.9925.

Amide 10: To a solution of chlorizidine A (**1**) (5.3 mg, 0.012 mmol) in dry CH_3CN (2.0 mL) at room temperature was added benzylamine (30 μL, 0.27 mmol) via syringe. The mixture was stirred at rt for 3 h and then concentrated. The product was purified by reversed-phase HPLC (70% CH_3CN in water, 0.2% TFA, t_R = 20 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}), collecting the eluent in a vial containing an aq. 0.2 M $NaHCO_3$ solution. After concentration, the white solid was triturated with CH_3CN. The solution concentrated onto C18 silica (100 mg) and fractionated on a C18 SPE cartridge (100 mg), eluting with water and then with CH_3CN. The organic fraction was concentrated to give 2.2 mg (33%) of **10**: UV/vis (CH_3CN/water) λ_{max} = 239, 307; $[\alpha]_D$ +2 (*c* 0.34, CH_3CN); IR (film) $\tilde{\nu}$ = 3237, 1613 cm^{-1}; ^1H and 2D NMR Table S4; HRMS (ESI-TOF) *m/z* $(M+H)^+$ calcd for $C_{25}H_{20}{}^{35}Cl_4N_3O_3$ 550.0259, found 550.0264.

Ester 11: To chlorizidine A (**1**) (1.3 mg, 0.0029 mmol) and K_2CO_3 (10 mg) was added dry MeOH (1.0 mL) at room temperature. The solution was stirred at rt overnight, then diluted with a saturated aq. NH_4Cl solution (4 mL) and extracted with EtOAc (2 x 2 mL). The combined extracts were washed with brine (1 mL), dried, filtered, and concentrated. The product was purified by reversed-phase HPLC (65% CH_3CN, 0.2% TFA, t_R = 30 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}) to give 0.4 mg (29%) of **11**: HRMS (ESI-TOF) *m/z* $(M–H)^–$ calcd for $C_{19}H_{13}{}^{35}Cl_4N_2O_4$ 472.9629, found 472.9646. The complete characterization of **11** was not possible due to its instability.

Diacetylated thioester 12: To a solution of thioester **9** (3.0 mg, 0.0053 mmol) in dry pyridine (1.0 mL) at room temperature was added acetic anhydride (0.10 mL). The mixture was stirred at rt for 4 h, CH_3OH (1 mL) was added, and the solution was concentrated. The product was purified by reversed-phase HPLC (70% CH_3CN, 0.2% TFA, t_R = 16 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}) to give 1.8 mg (52%) of **12**: UV/vis (CH_3CN/water) λ_{max} = 219, 267, 321, 417; $[\alpha]_D$ –4 (*c* 0.17, CH_3CN); IR (film) $\tilde{\nu}$ = 1761, 1684, 1626 cm^{-1}; ^1H NMR (600 MHz, CD_3CN) δ 10.02 (br s), 9.95 (br s), 7.12 (s), 7.09 (s), 6.46 (s), 5.86 (s),

5.83 (s) 4.04 (m), 3.35 (m), 3.08 (m), 2.58 (m), 2.42 (m), 2.36 (s), 2.32 (s), 2.29 (s), 2.22 (s), 1.87 (s), 1.85 (s); HRMS (ESI-TOF) *m/z* $(M+H)^+$ calcd for $C_{26}H_{24}{}^{35}Cl_4N_3O_6S$ 646.0140, found 646.0130.

Triacetylated amide 13: To a solution of chlorizidine A (**1**) (5.0 mg, 0.011 mmol) in dry CH_3CN (2.0 mL) at room temperature was added benzylamine (30 μL, 0.27 mmol). The mixture was stirred at rt for 5 h and then concentrated. To a solution of the crude product in dry pyridine (1 mL) was added acetic anhydride (0.20 mL). The mixture was stirred at rt overnight, CH_3OH (1 mL) was added, and the solution was concentrated. The product was purified by reversed-phase HPLC (70% CH_3CN, 0.2% TFA, t_R = 24 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}) to give 3.4 mg (45%) of **13**: UV/vis (CH_3CN/water) λ_{max} = 235, 282; $[\alpha]_D$ –17 (*c* 0.29, CH_3CN); IR (film) $\tilde{\nu}$ = 1773, 1684 cm^{-1}; ^1H NMR (600 MHz, CD_3CN) δ 7.34-7.22 (m), 7.14-7.03 (m), 7.08 (s), 6.32 (s), 6.30 (s), 5.88 (s), 5.85 (s), 5.64 (dd, *J* = 8.8, 5.5 Hz), 5.49 (dd, *J* = 8.8, 6.4 Hz), 4.39 (d, *J* = 5.8 Hz), 4.36 (d, *J* = 6.4 Hz), 4.33 (d, *J* = 5.8 Hz), 4.30 (d, *J* = 5.8 Hz), 4.26 (d, *J* = 6.2 Hz), 3.03 (m), 2.92 (m), 2.44 (m), 2.33 (s), 2.27 (s) 2.15 (s), 1.71 (s); HRMS (ESI-TOF) *m/z* $(M+H)^+$ calcd for $C_{31}H_{26}{}^{35}Cl_4N_3O_6$ 676.0576, found 676.0595.

Triacetylated ester 14: To a solution of diacetyl chlorizidine A (**2**) (7.9 mg, 0.015 mmol) in dry CH_3OH (2.0 mL) at room temperature was added an aqueous solution of NaOH (0.10 mL, 0.05 M). The mixture was stirred at rt overnight, then diluted with a saturated aq. NH_4Cl solution (4 mL) and extracted with EtOAc (3 x 2 mL). The combined extracts were washed with brine (2 x 1 mL), dried, filtered, and concentrated. To a solution of the crude product in dry pyridine (1 mL) was added acetic anhydride (0.20 mL). The mixture was stirred at rt overnight, CH_3OH (1 mL) was added, and the solution was concentrated. The product was purified by reversed-phase HPLC (70% CH_3CN, 0.2% TFA, t_R = 25 min, C8(2) Phenomenex® Luna, 3 mL min^{-1}) to give 3.4 mg (38%) of **14**: UV/vis (CH_3CN/water) λ_{max} = 238, 287; $[\alpha]_D$ –21 (*c* 0.28, CH_3CN); IR (film) $\tilde{\nu}$ = 1773, 1684 cm^{-1}; ^1H NMR (600 MHz, CD_3CN) δ 7.18 (s), 7.09 (s), 6.29 (s), 5.89 (s), 5.68 (dd, *J* = 9.4, 5.5 Hz), 5.57 (dd, *J* = 8.8, 5.8 Hz), 3.71 (s), 3.59 (s), 3.06 (m), 2.94 (m), 2.46 (m), 2.34 (s), 2.32 (s), 2.31 (s), 2.29 (s); HRMS (ESI-TOF) *m/z* $(M+H)^+$ calcd for $C_{25}H_{21}{}^{35}Cl_4N_2O_7$ 601.0103, found 601.0091.

Figure S2. HMBC correlations for chlorizidine A (**1**)

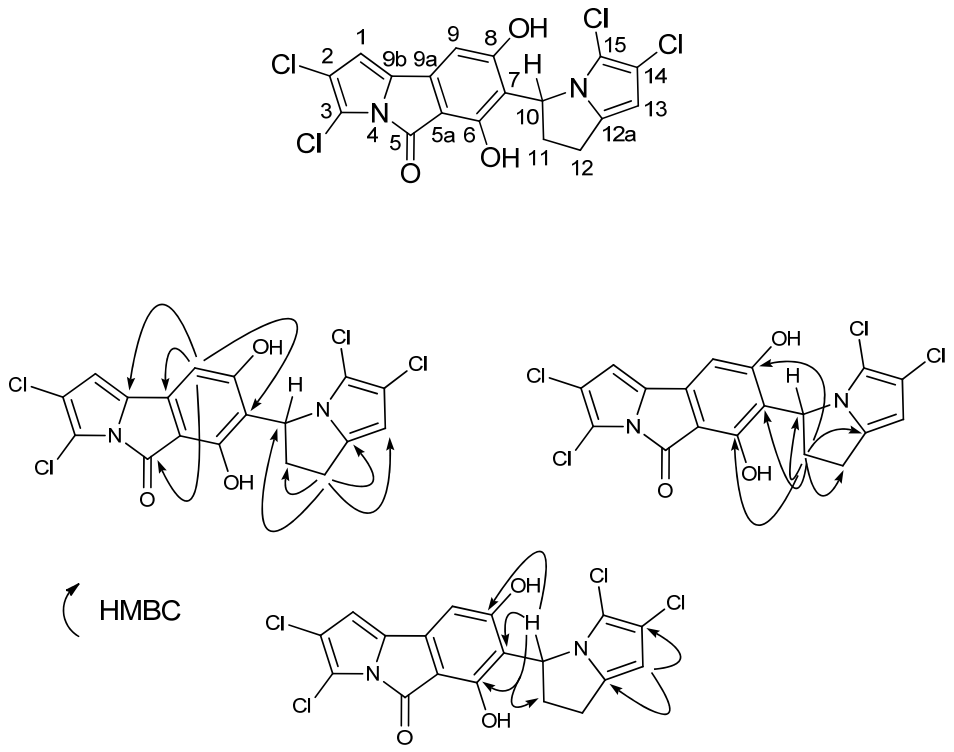
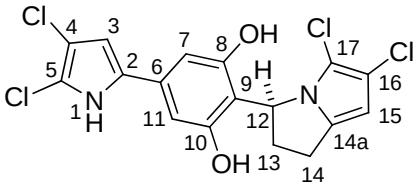
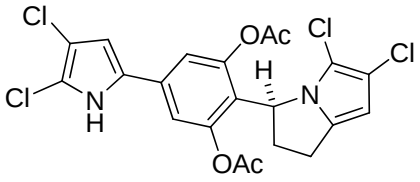


Table S1. Selected NMR spectral data for **7** and **8**



7

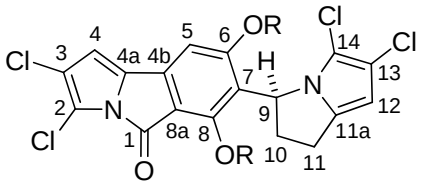


8

position	δ_H (**7**), mult. (*J*, Hz)[a] CD_3CN	δ_H, (**8**), mult. (*J*, Hz)[a] DMSO-d_6
1	10.06, br s	12.42,br s
3	6.45, d (3.0)	6.79, d (3.0)
7	6.51, br s[b]	7.35, s[b]
11	6.51, br s[b]	7.40, s[b]
12	5.84, dd (9.5, 6.5)	5.59, dd (6.5, 5.5)
13	2.80, ddd (17.5, 14.0, 5.0)	2.84, ddd (18.5, 10.0, 4.5)
	2.55, ddd (17.5, 10.0, 6.0)	2.25, dd (12.0, 6.0)
14	3.06, ddd (14.5, 10.0, 5.0)	2.99, ddd (19.0, 14.5, 6.0)
	2.88, ddd (14.5, 10.0, 6.0)	2.88, ddd (17.5, 9.5, 5.5)
15	5.79, s	5.95, s
Ac		2.39, s
		1.94, s

[a] 500 MHz. [b] H-7 and H-11 are not distinguishable due to the pseudo-symmetry of **7** and **8**.

Table S2. Selected NMR spectral data for **2-6**

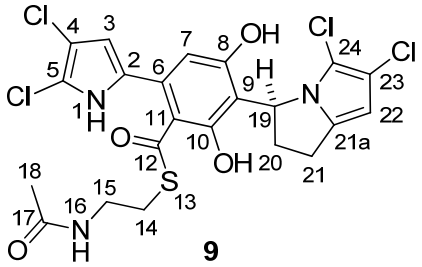


2, R = Ac
3, R = Me
4, R = CO*i*-Pr
5, R = Piv
6, R = Bz

position	δ_H (**2**) mult. (*J*, Hz)[a] DMSO-d_6	δ_H (**3**) mult. (*J*, Hz)[a] DMSO-d_6	δ_H (**4**) mult. (*J*, Hz)[a] DMSO-d_6	δ_H (**5**) mult. (*J*, Hz)[a] DMSO-d_6	δ_H (**6**) mult. (*J*, Hz)[a] benzene- d_6
1	6.80, s 6.79, s	7.17, s 7.10, s	6.80, s 6.78, s	6.80, s 6.79, s	5.48, s 5.43, s 5.40, s
9	7.42, s 7.36, s	6.67, s	7.45, s 7.42, s 7.33, s	7.44, s 7.43, s 7.30, s	6.43, s 6.38, s 6.35, s
10	5.70, br s 5.75, dd (6.0, 5.0)	5.82, s	5.63, t (8.5) 5.61, t (8.5) 5.58, t (6.0)	5.48, br s 5.46, t (8.5) 5.40, t (8.5)	5.17, dd (9.0, 5.0) 5.10, dd (9.5, 4.5)
11	2.88, m 2.36, br s 2.25, ddd (18.0, 14.0, 5.5)	2.86, m 2.36, m	2.81, m 2.40, m 2.29, ddd (17.5, 14.0, 5.5) 2.03, m	2.75-2.57, m 2.30, br s 2.01, ddd (19.0, 10.0, 10.0)	2.36, m 2.30, m 2.11, m
12	3.10, t (10.0) 3.01, dd (15.0, 12.0) 2.91, m	3.00, m 2.88, m	2.97-2.76, m	2.94-2.77, m	2.55, ddd (15.0, 5.0, 5.0) 2.27, m 2.11, m 2.02, dd (20.0, 10.0)
13	5.96, s 5.94, s	5.90, s 5.86, s	5.98, s 5.95, s 5.92, s	6.00, s 5.96, s 5.95, s	5.58, s 5.40, s 5.26, s
R	2.43, s 2.41, s 2.00, s 1.95, s	4.04, s 4.00, s 3.68, s 3.48, s	3.03, 2.53, 2.44 2.40, sep (9.0) 1.33, 1.32, 1.29, 1.28, 1.24, 1.13, 1.12, 1.05, 1.05, 0.98, 0.95, d (9.0)	1.39, s 1.36, s 1.35, s 1.11, s 1.10, s 1.08, s	8.34, br s 8.25, 8.20, 8.08, 8.06, 7.91, d (7.5) 7.21-7.04, m

[a] 500 MHz.

Table S3. Selected NMR spectral data for **9** (CD_3CN)

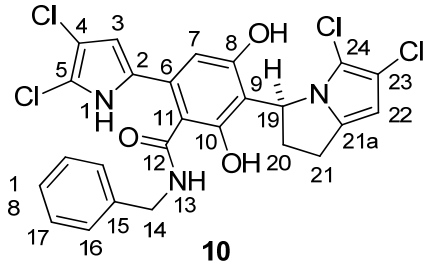


9

position	δ_C[a]	δ_H, mult. (*J*, Hz)[b]	HMBC
1		9.66, br s	3
3	107.7	6.24, s	
7	102.7	6.46, s	8,9,11,19
8	164.4		
9	114.9		
10	158.1		
11	106.0		
12	--		
14	31.5	2.55, m	15
15	39.9	3.32, m	14,17
16		6.84, br s	
17	171.8		
18	22.6	1.87, s	17
19	38.6	4.43, m	8,10,20,21
20	33.6	2.28, m 2.17, m	9,19,21,21a
21	26.5	2.51, m	19,20,21a,22
21a	131.6		
22	106.4	5.81, d (3.0)	21,21a

[a] Carbon assignments were based solely on HSQC and HMBC data collected at 600 MHz. Carbons 2, 4, 5, 6, 12, 23, and 24 could not be assigned. [b] 600 MHz.

Table S4. Selected NMR spectral data for **10** (CD_3CN)

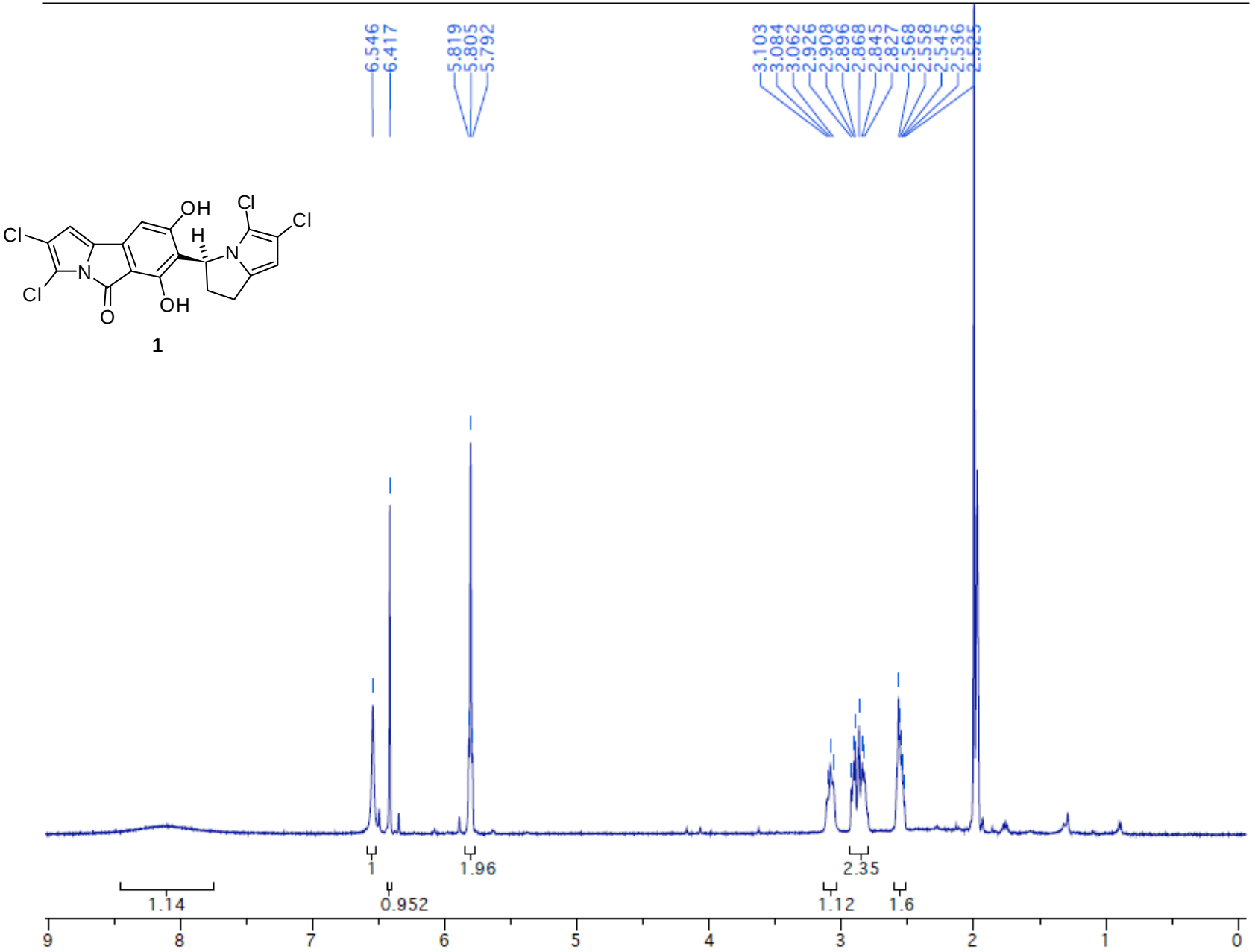


10

position	δ_C[a]	δ_H, mult. (*J*, Hz)[b]	HMBC
1		10.03, br s	
3	110.0	6.21, s	
7	110.2	6.32, br s	
9	114.3		
11	106.2		
12	170.3		
13		6.46, br s	
14	44.1	4.34, br s	12,15,16
15	138.3		
16	128.3	7.15, m	14
17	129.1	7.32, m	14,15
18	127.8	7.26, m	15,16
19	53.9	5.89, br s	
20	32.8	2.81, m 2.53, br s	9,19,21,21a
21	26.0	3.07, br s 2.87, m	19,20,21a,22
21a	137.5		
22	99.5	5.77, s	19,21,21a

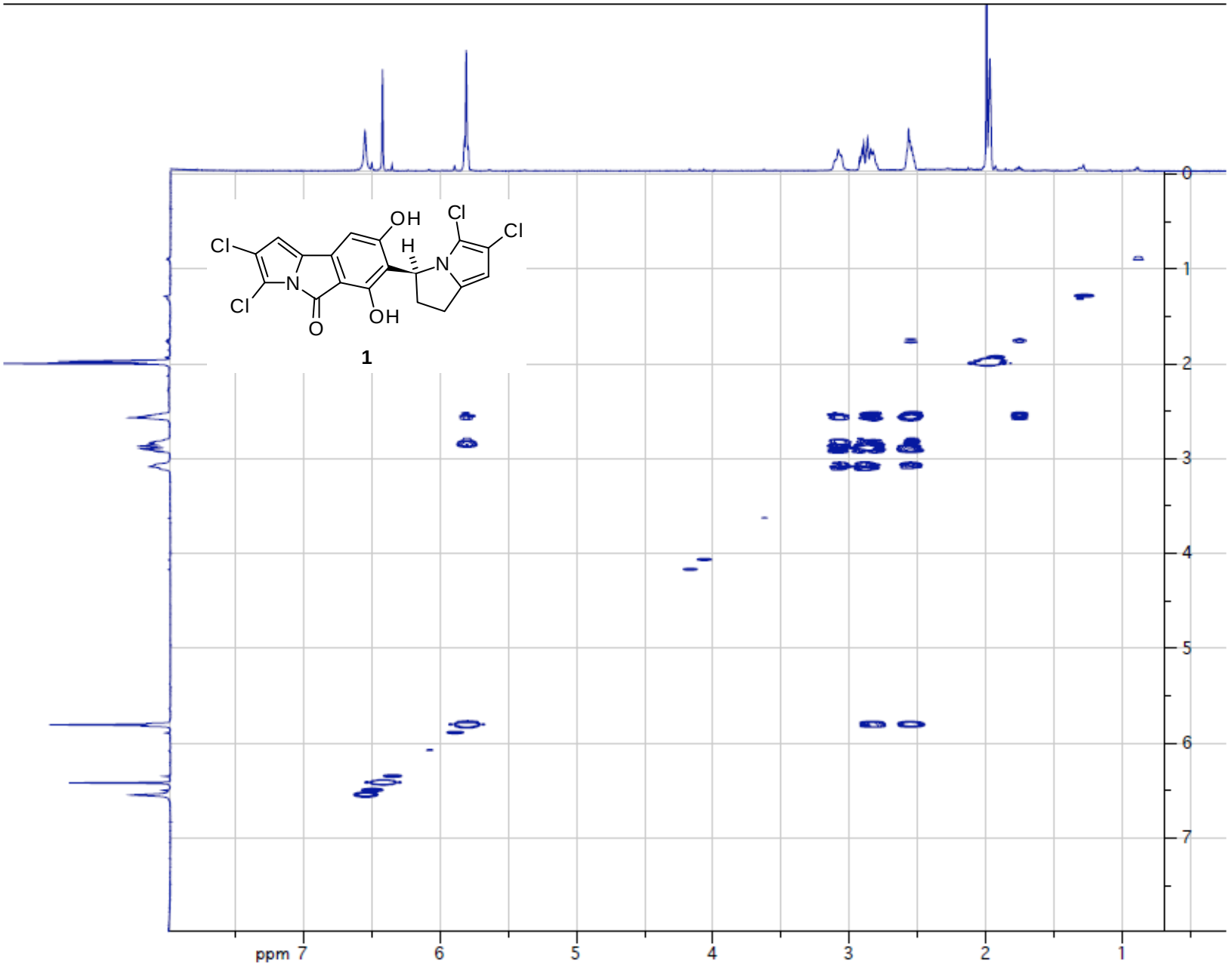
[a] Carbon assignments were based solely on HSQC and HMBC data collected at 600 MHz. Carbons 2, 4, 5, 6, 8, 10, 23, and 24 could not be assigned. [b] 600 MHz.

^{1}H NMR (500 MHz, CD_3CN) of chlorizidine A (**1**)

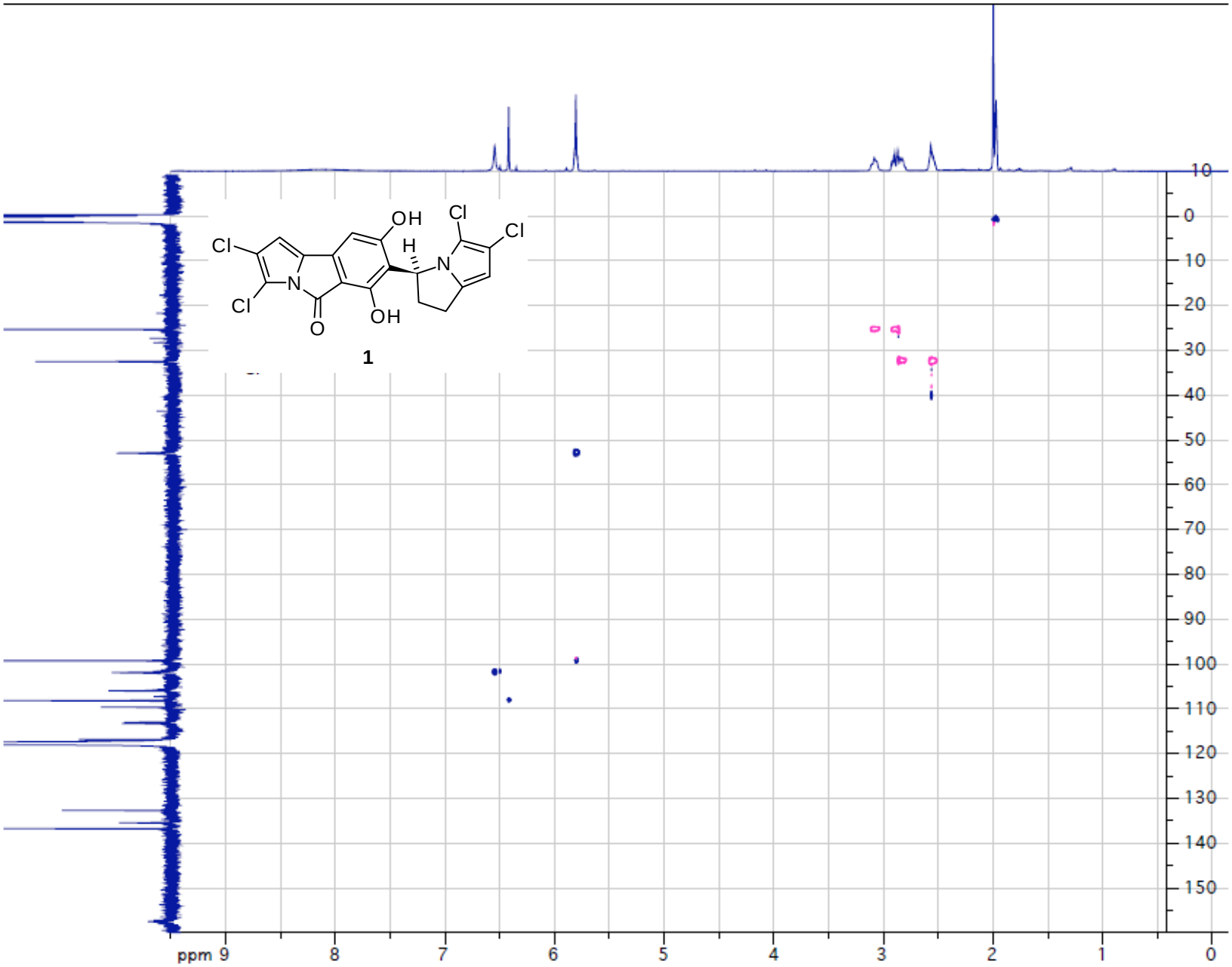


1

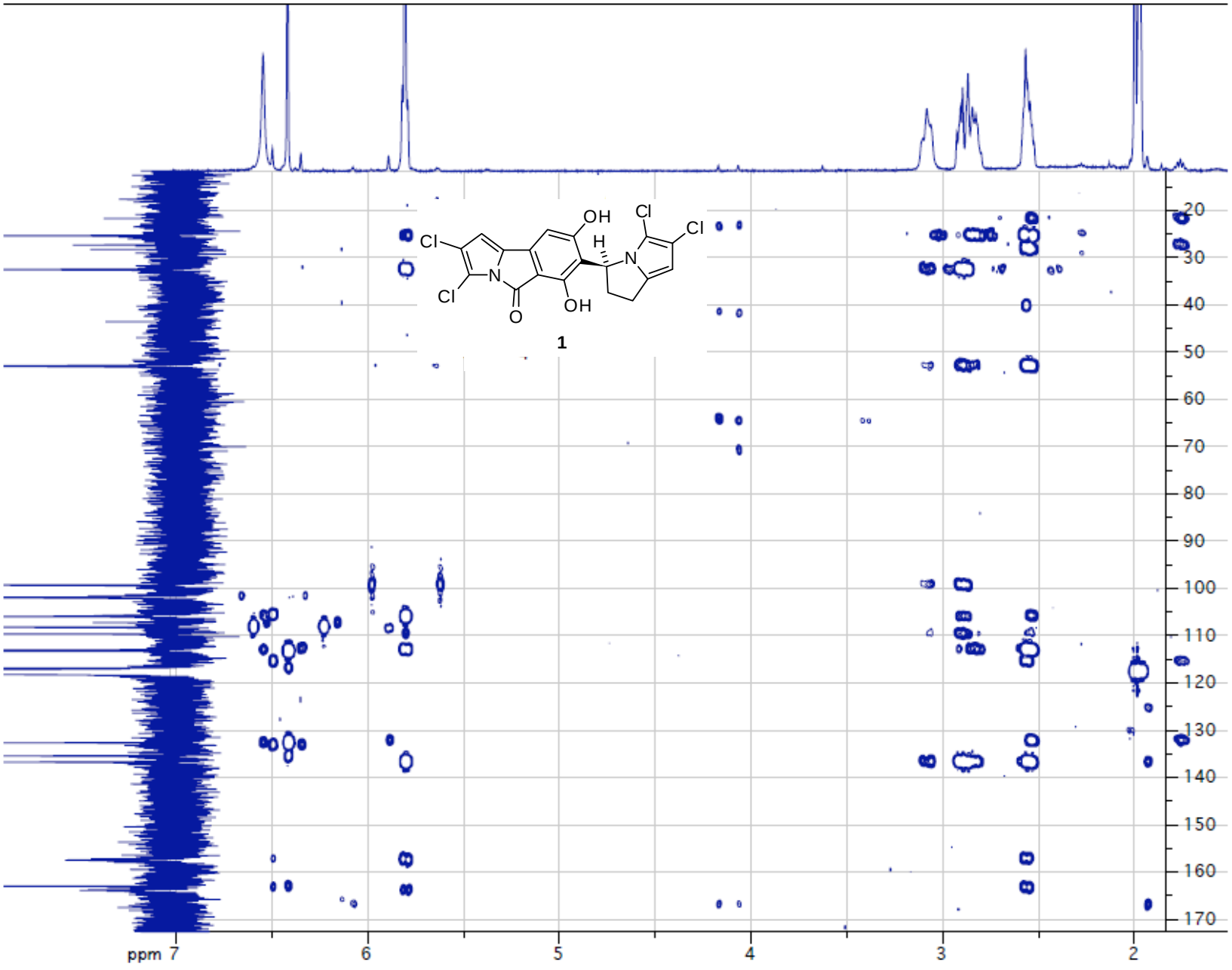
COSY (500 MHz, CD_3CN) of chlorizidine A (**1**)



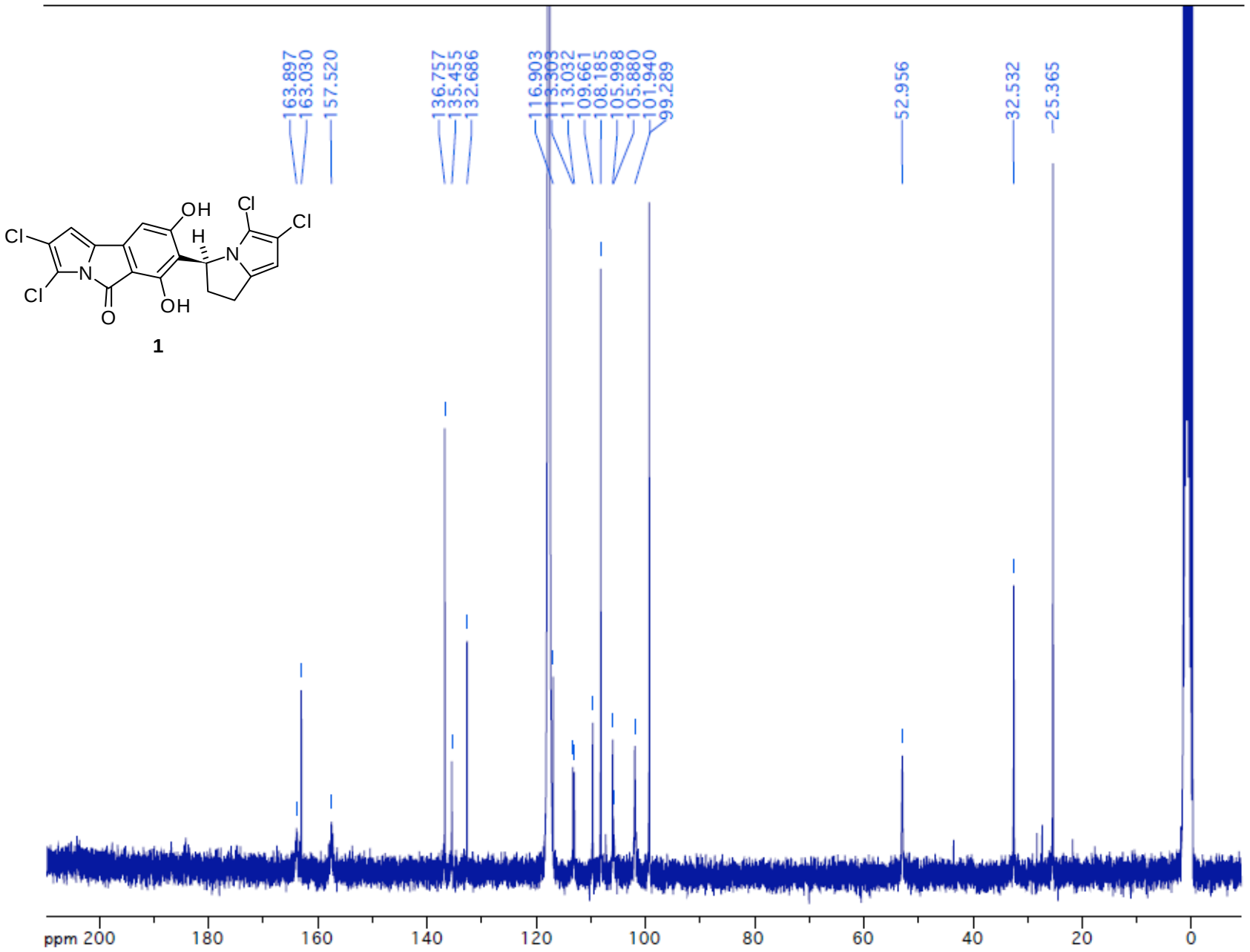
HSQC (500 MHz, CD_3CN) of chlorizidine A (**1**)



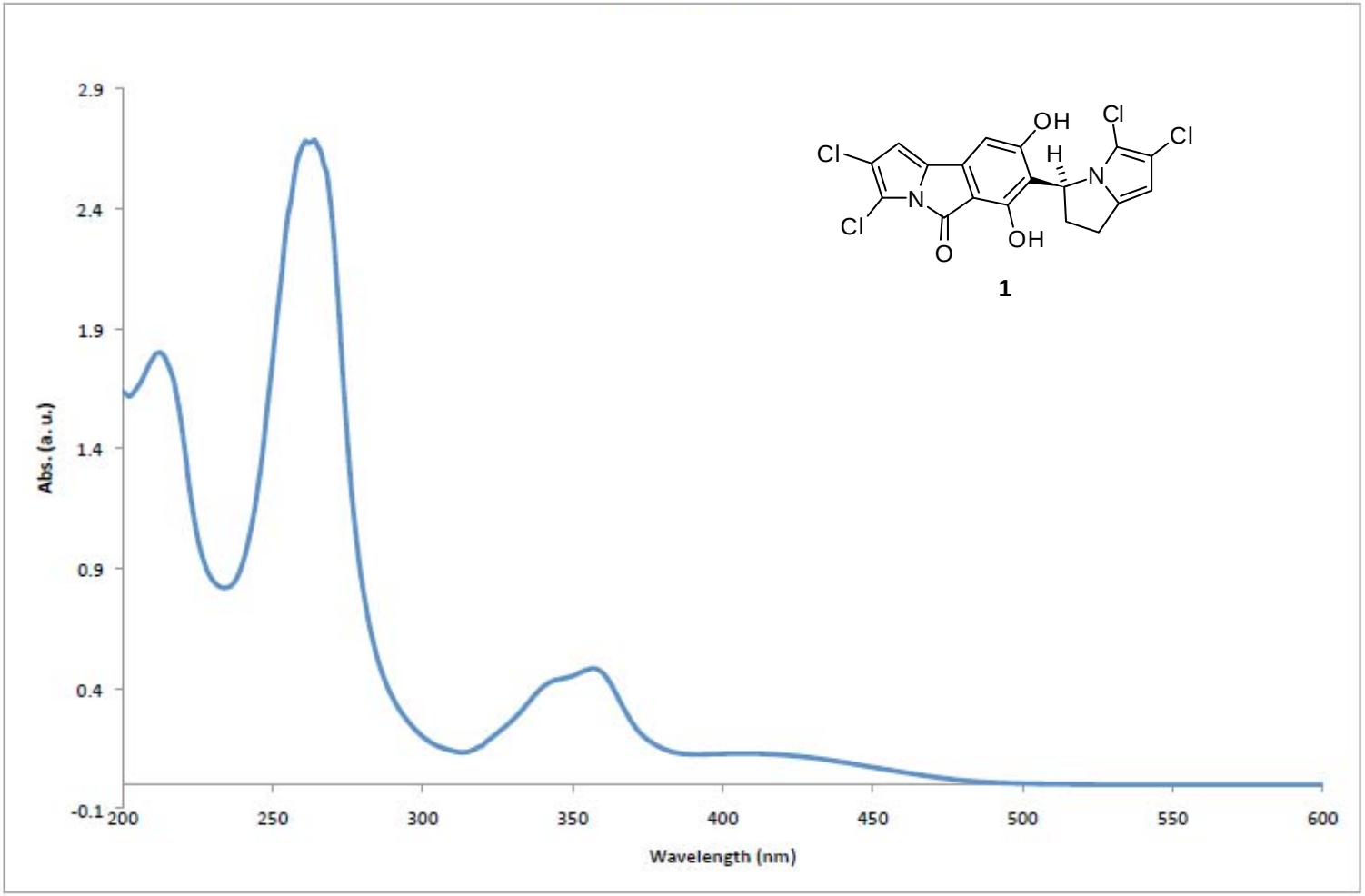
HMBC (500 MHz, CD_3CN) of chlorizidine A (**1**)



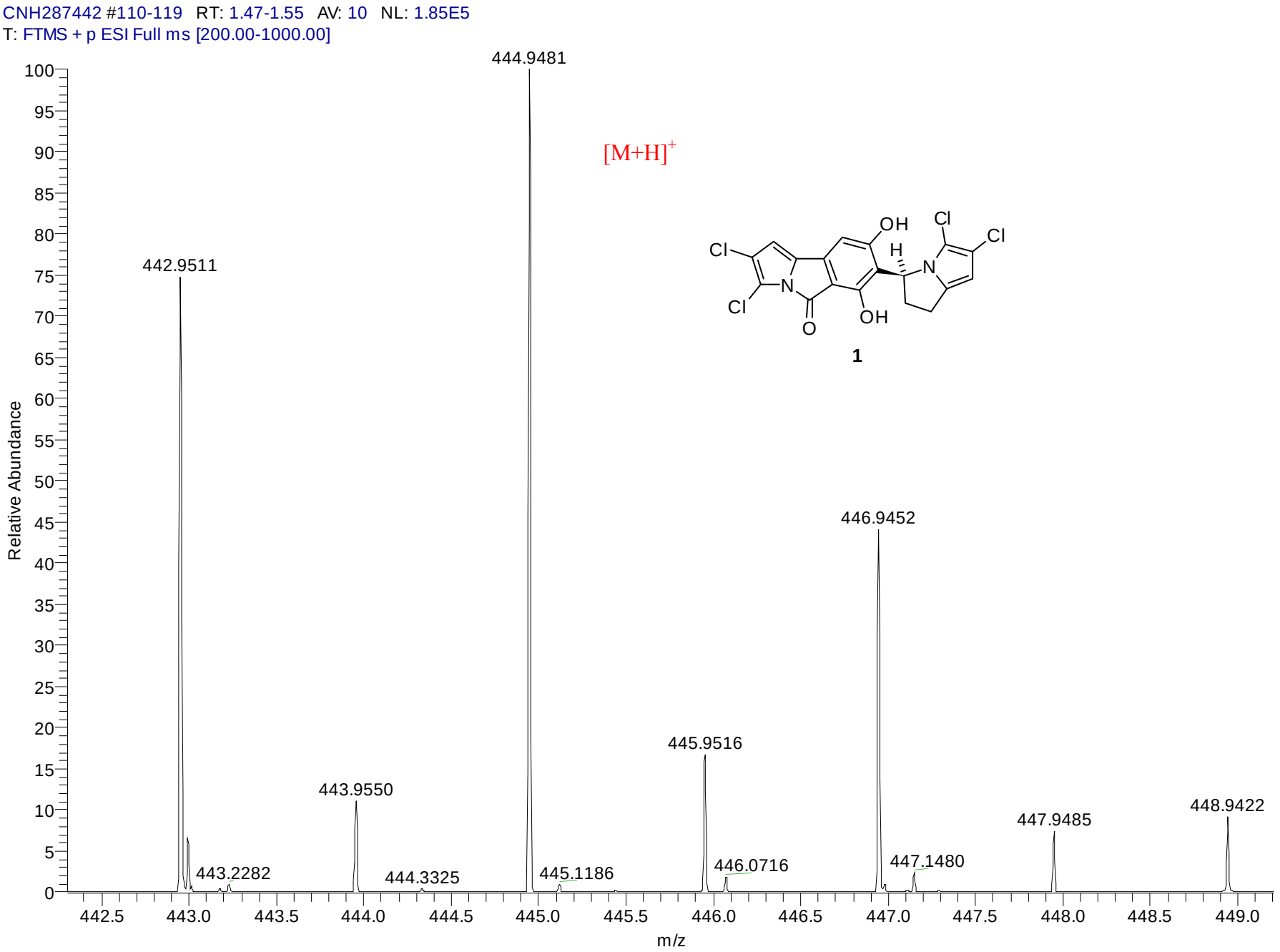
^{13}C NMR (125 MHz, CD_3CN) of chlorizidine A (**1**)



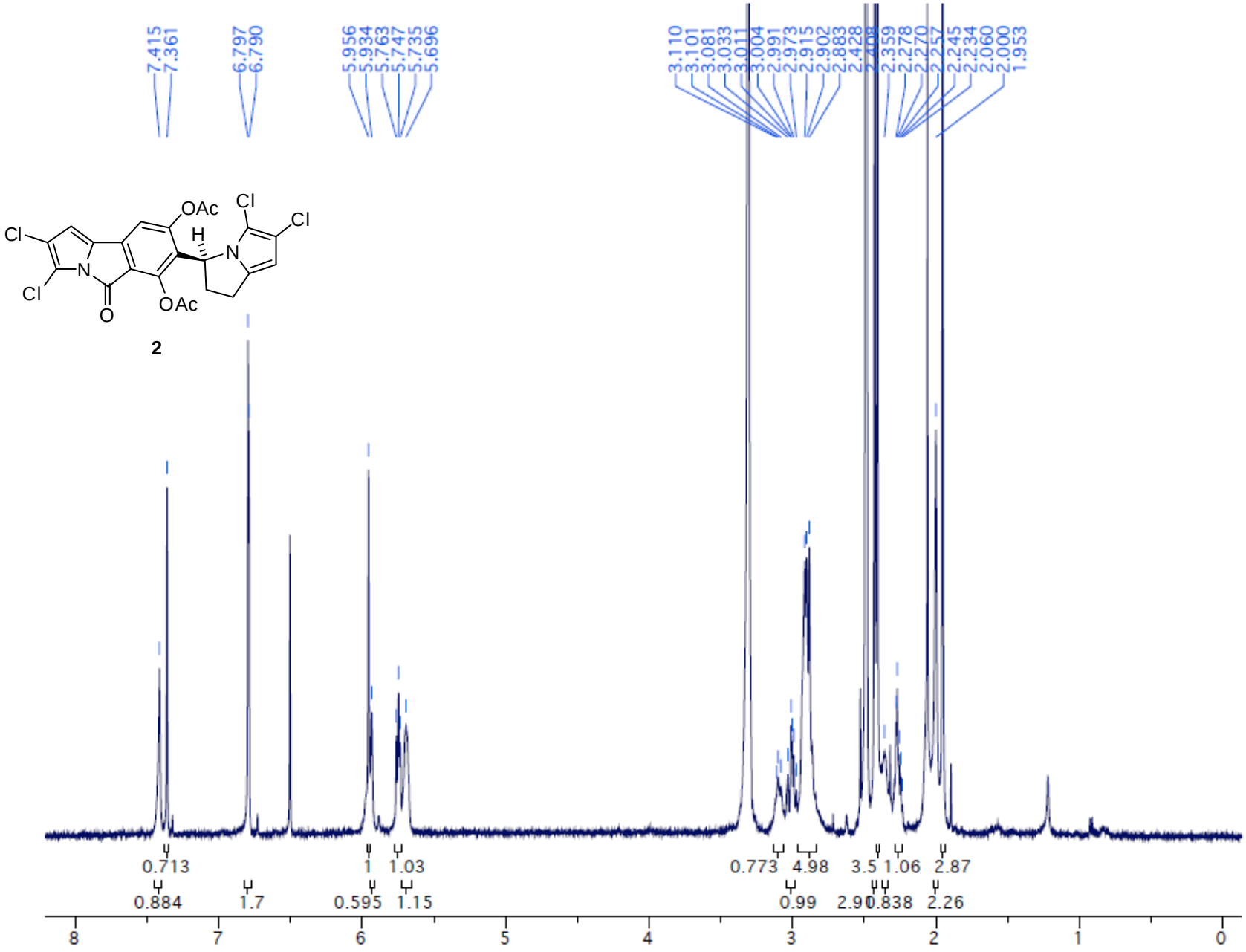
UV/vis spectrum (CH_3CN) of chlorizidine A (**1**)



1

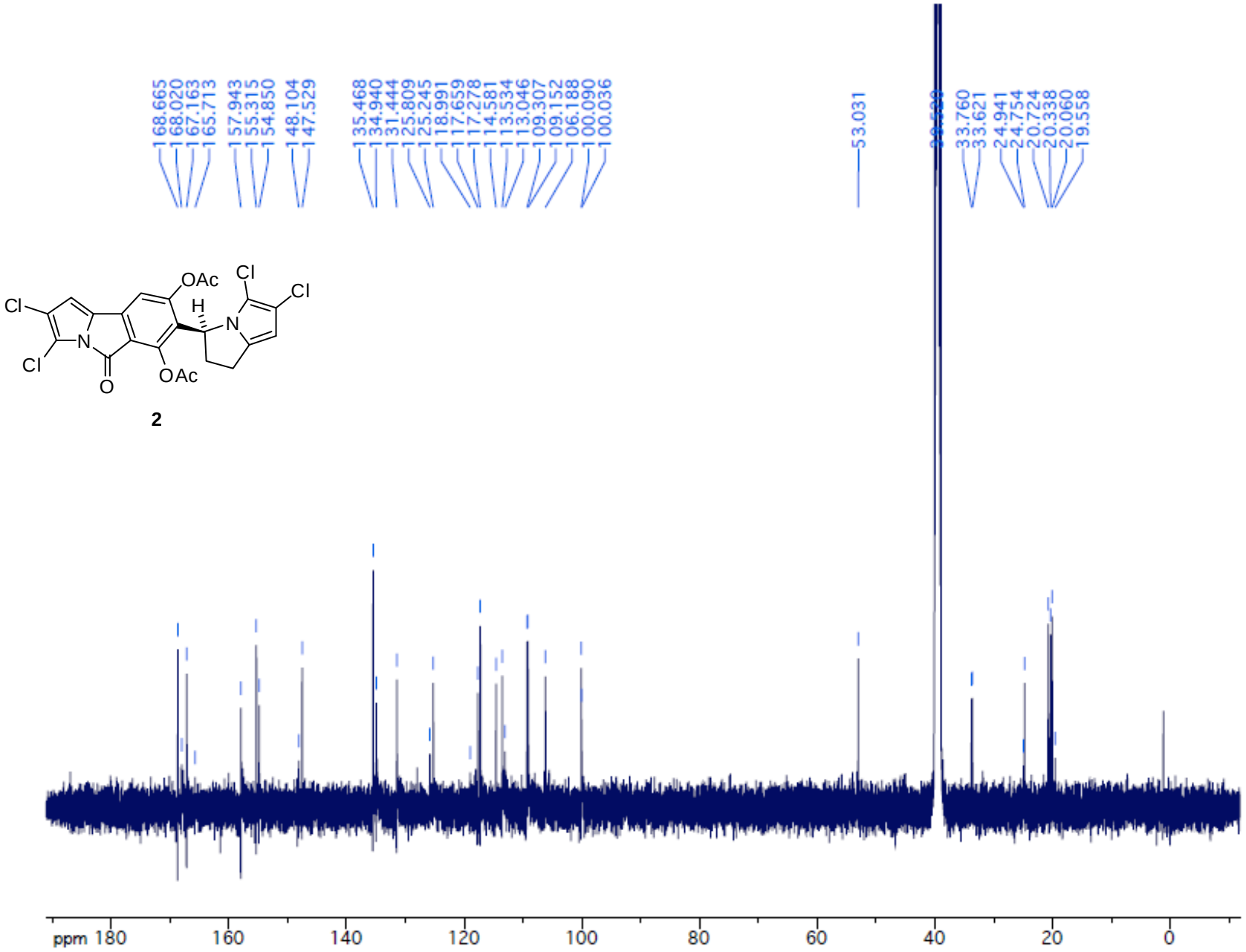

CNH287442 #110-119 RT: 1.47-1.55 AV: 10 NL: 1.85E5
T: FTMS + p ESI Full ms [200.00-1000.00]
444.9481
[M+H]+
442.9511
446.9452
445.9516
443.9550
448.9422
447.9485
443.2282
444.3325
445.1186
446.0716
447.1480
Relative Abundance
m/z
OH
Cl
Cl
H
N
Cl
N
Cl
O
OH
1

^{1}H NMR (500 MHz, DMSO-d_6) of diacetyl chlorizidine A (**2**)



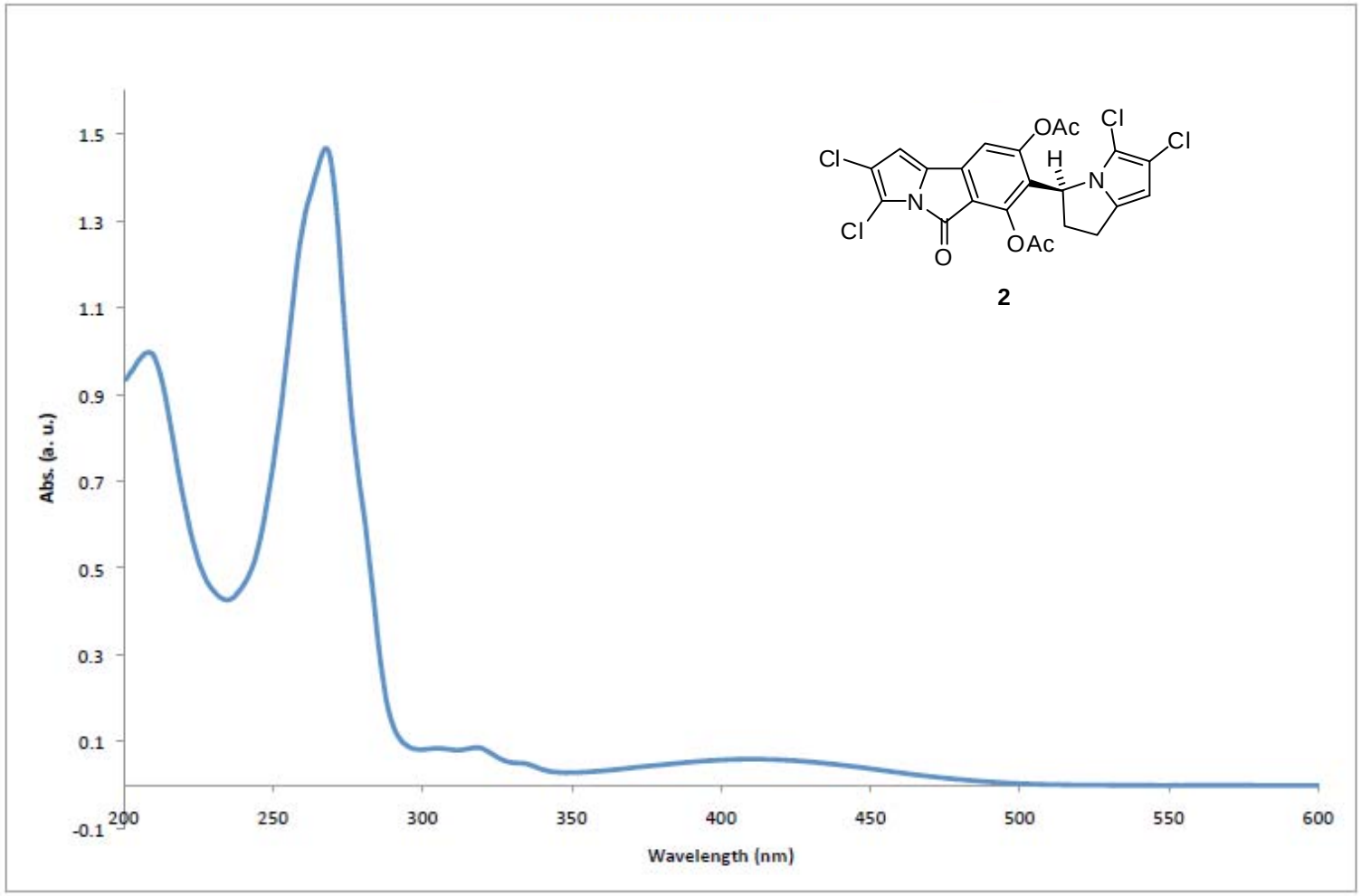
2

^{13}C NMR (125 MHz, DMSO-d_6) of diacetyl chlorizidine A (**2**)

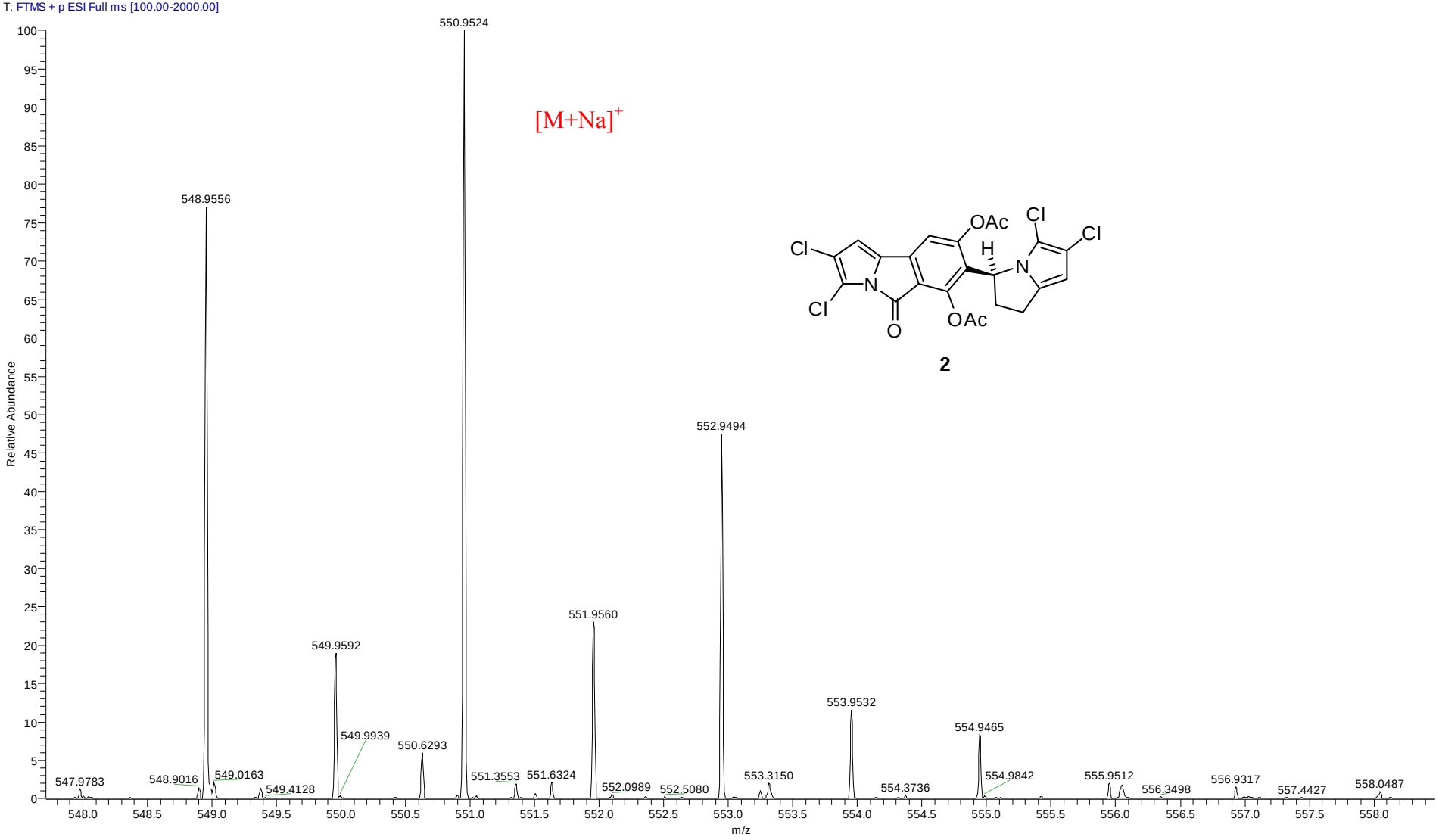


2

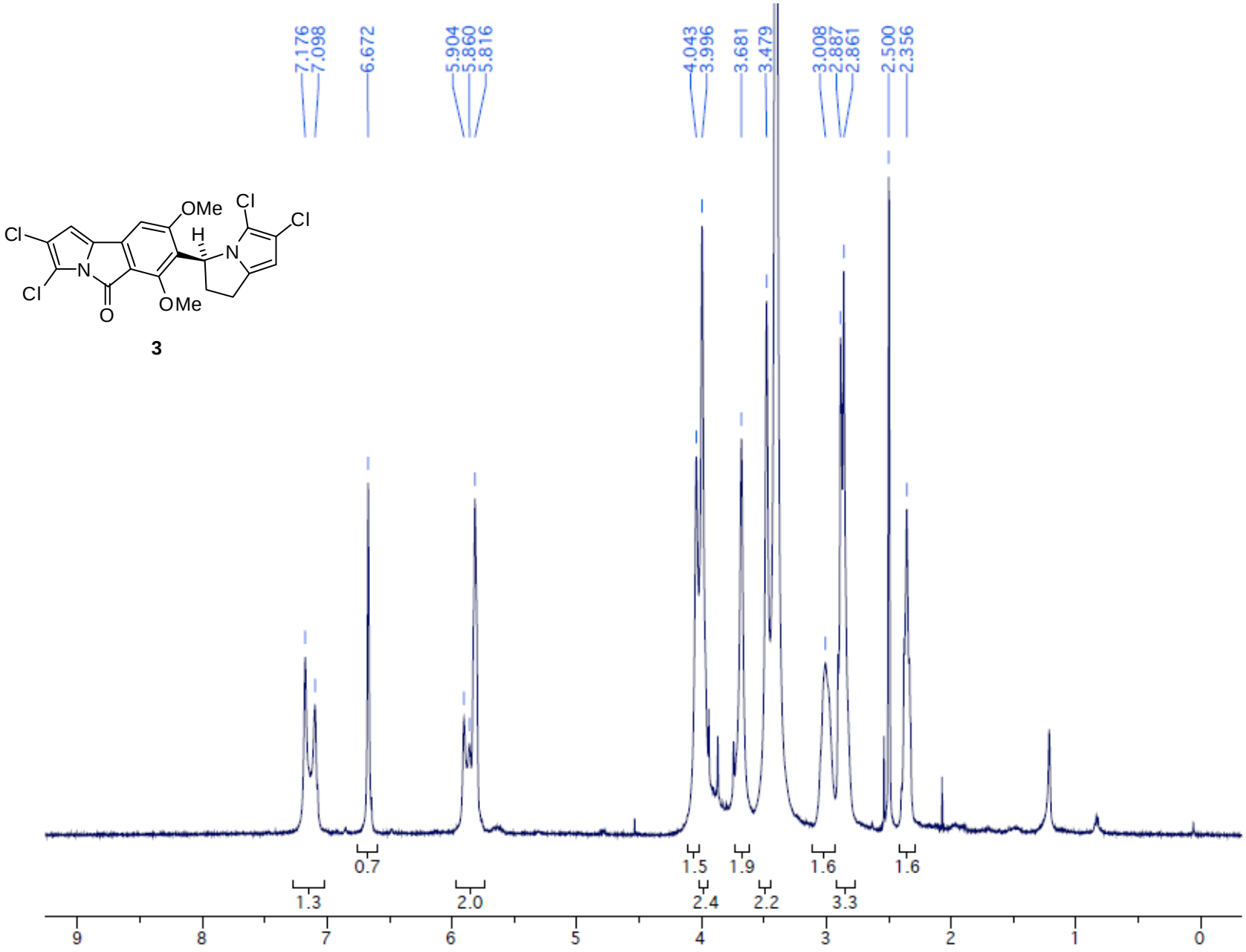
UV/vis spectrum (CH_3CN) of diacetyl chlorizidine A (**2**)



2

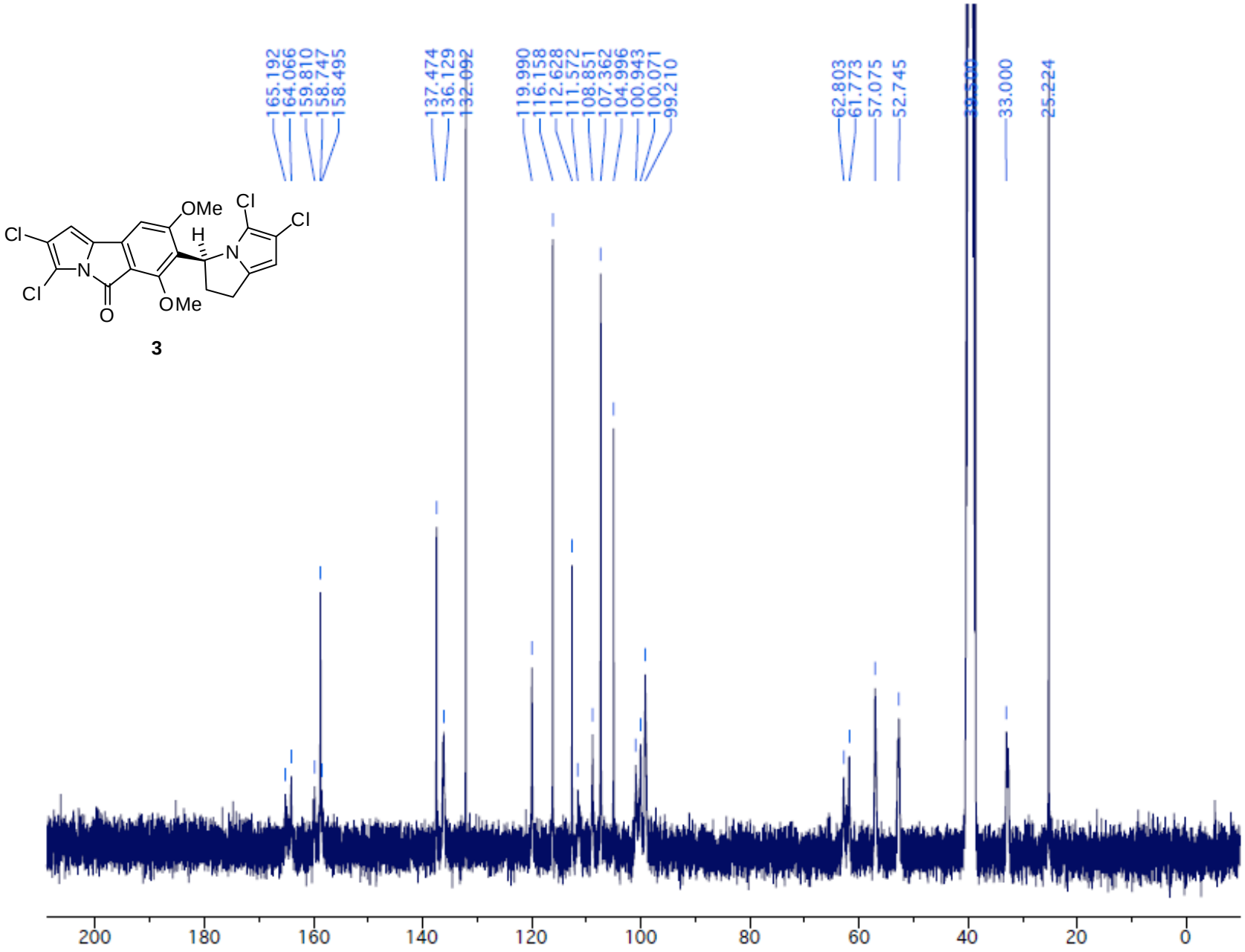

H287-Ac #114-117 RT: 0.99-1.02 AV: 4 NL: 1.31E6
T: FTMS + p ESI Full ms [100.00-2000.00]
[M+Na]+
Relative Abundance
m/z
550.9524
548.9556
552.9494
551.9560
549.9592
553.9532
554.9465
549.9939
550.6293
547.9783
548.9016
549.0163
549.4128
551.3553
551.6324
552.0989
552.5080
553.3150
554.3736
554.9842
555.9512
556.3498
556.9317
557.4427
558.0487
OAc
Cl
H
N
O
2

^{1}H NMR (500 MHz, DMSO-d_6) of dimethyl chlorizidine A (**3**)



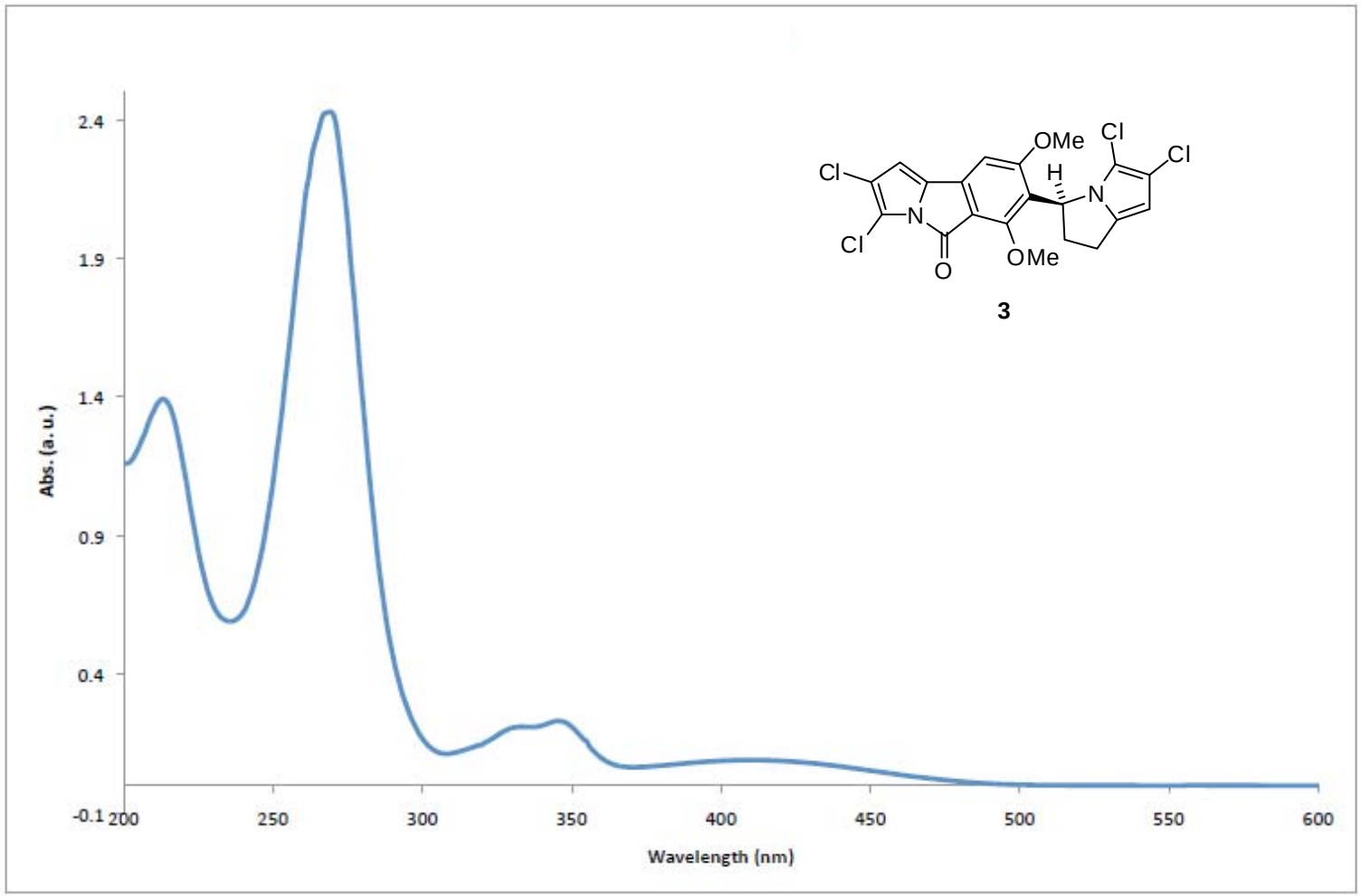
3

^{13}C NMR (125 MHz, DMSO-d_6) of dimethyl chlorizidine A (**3**)



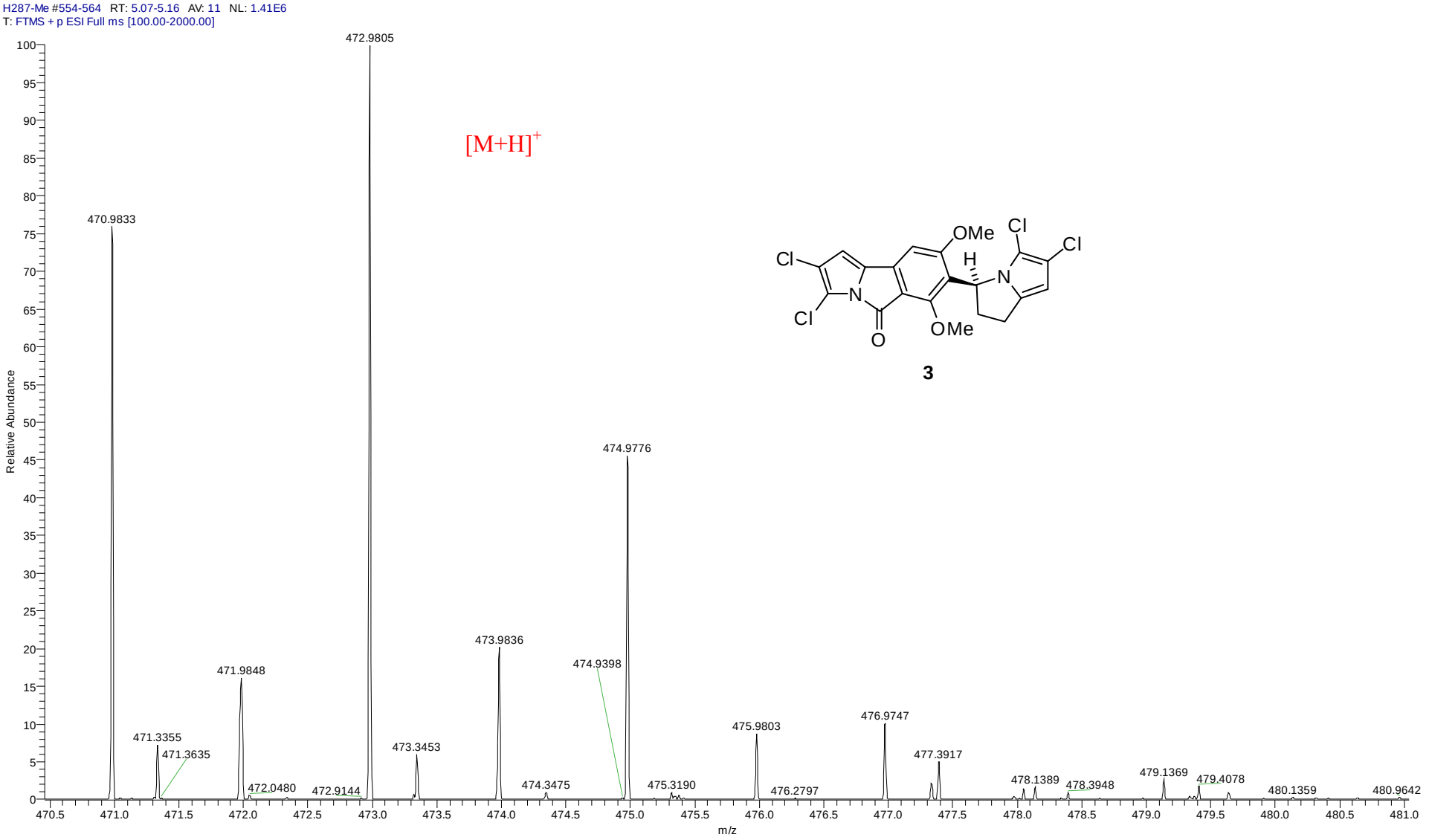
3

UV/vis spectrum (CH_3CN) of dimethyl chlorizidine A (**3**)



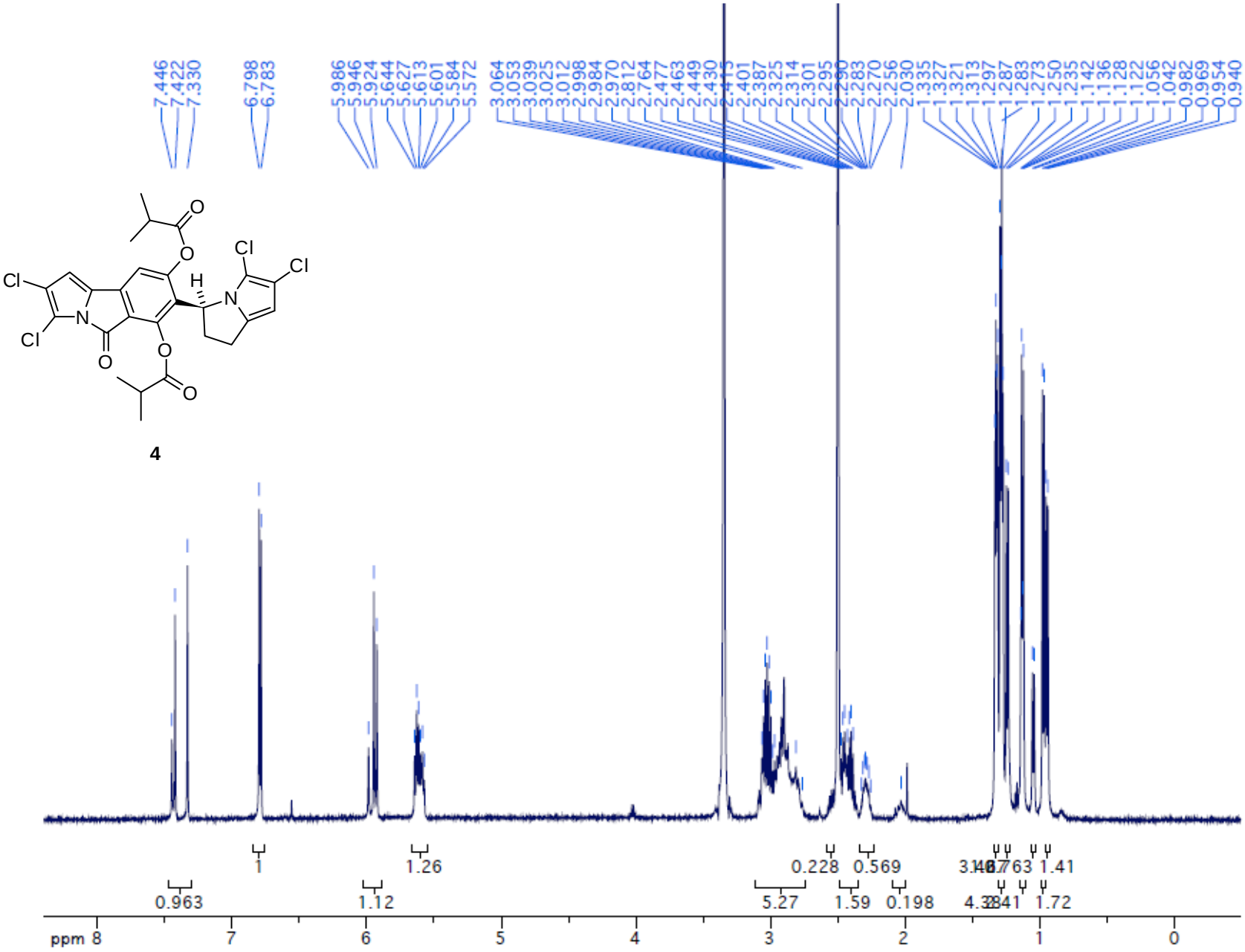
3

HRESI-FT-MS (Orbit-Trap-MS) of dimethyl chlorizidine A (**3**)



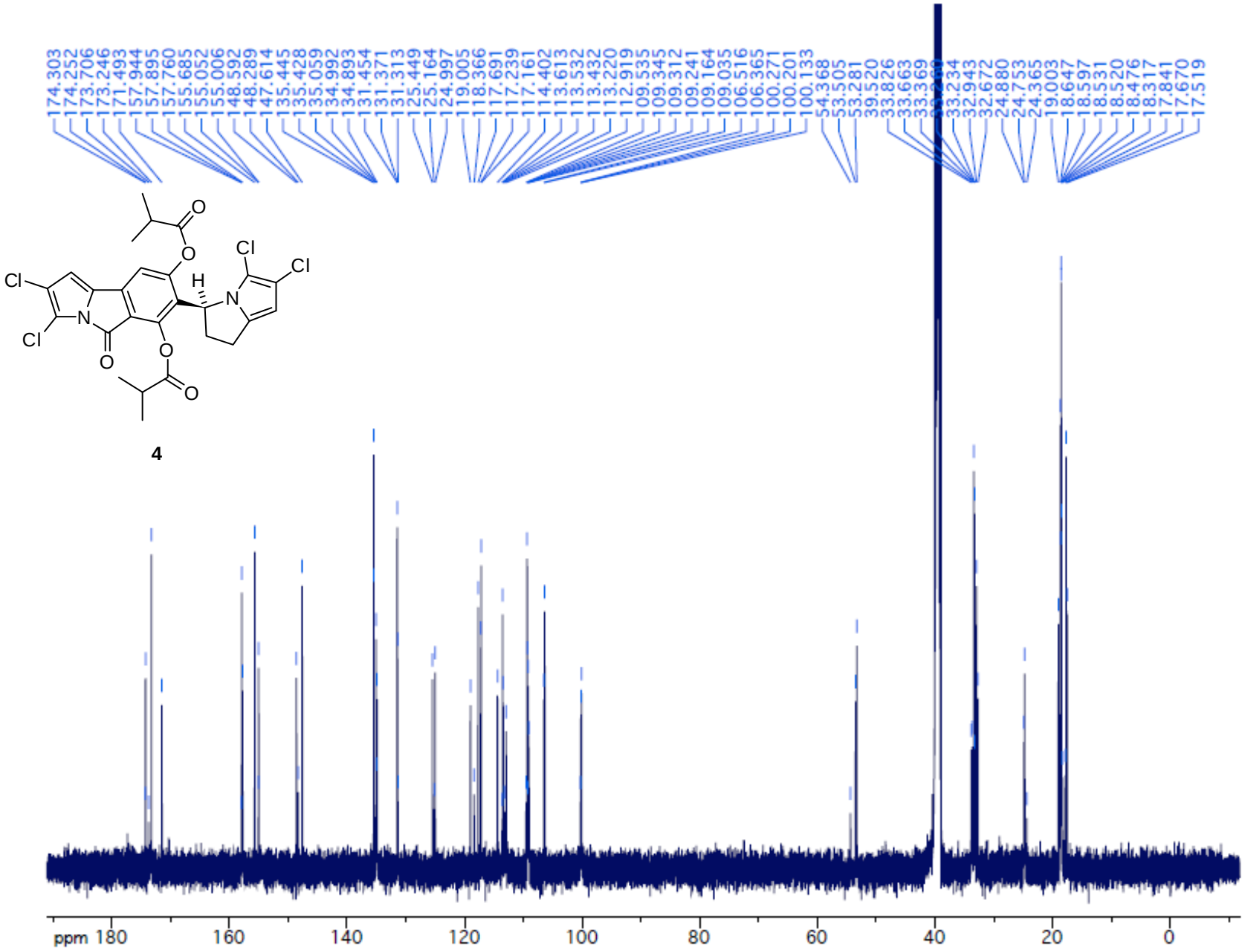
3

^{1}H NMR (500 MHz, DMSO-d_6) of diisobutoyl chlorizidine A (**4**)

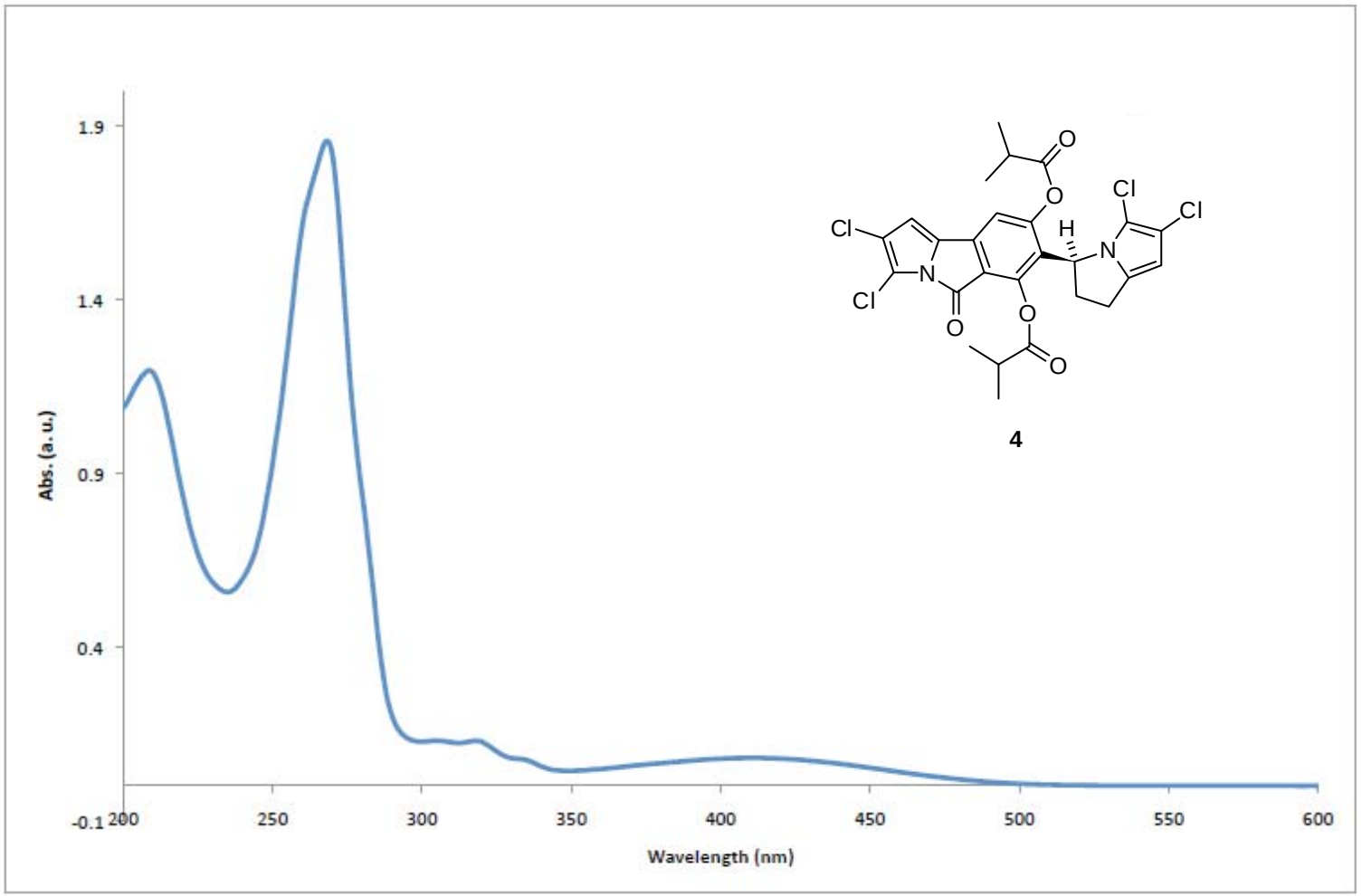


4

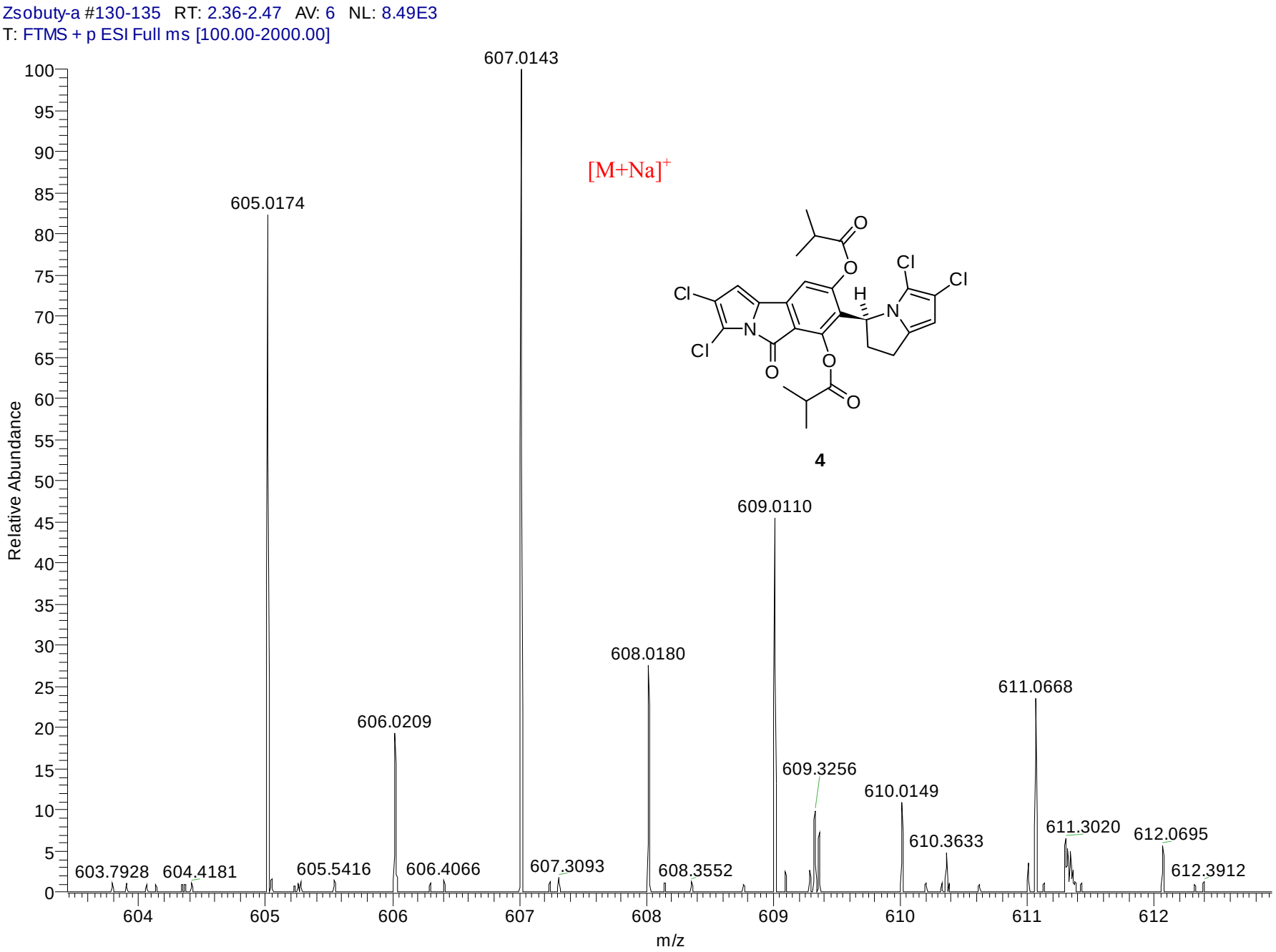
^{13}C NMR (125 MHz, DMSO-d_6) of diisobutoyl chlorizidine A (**4**)



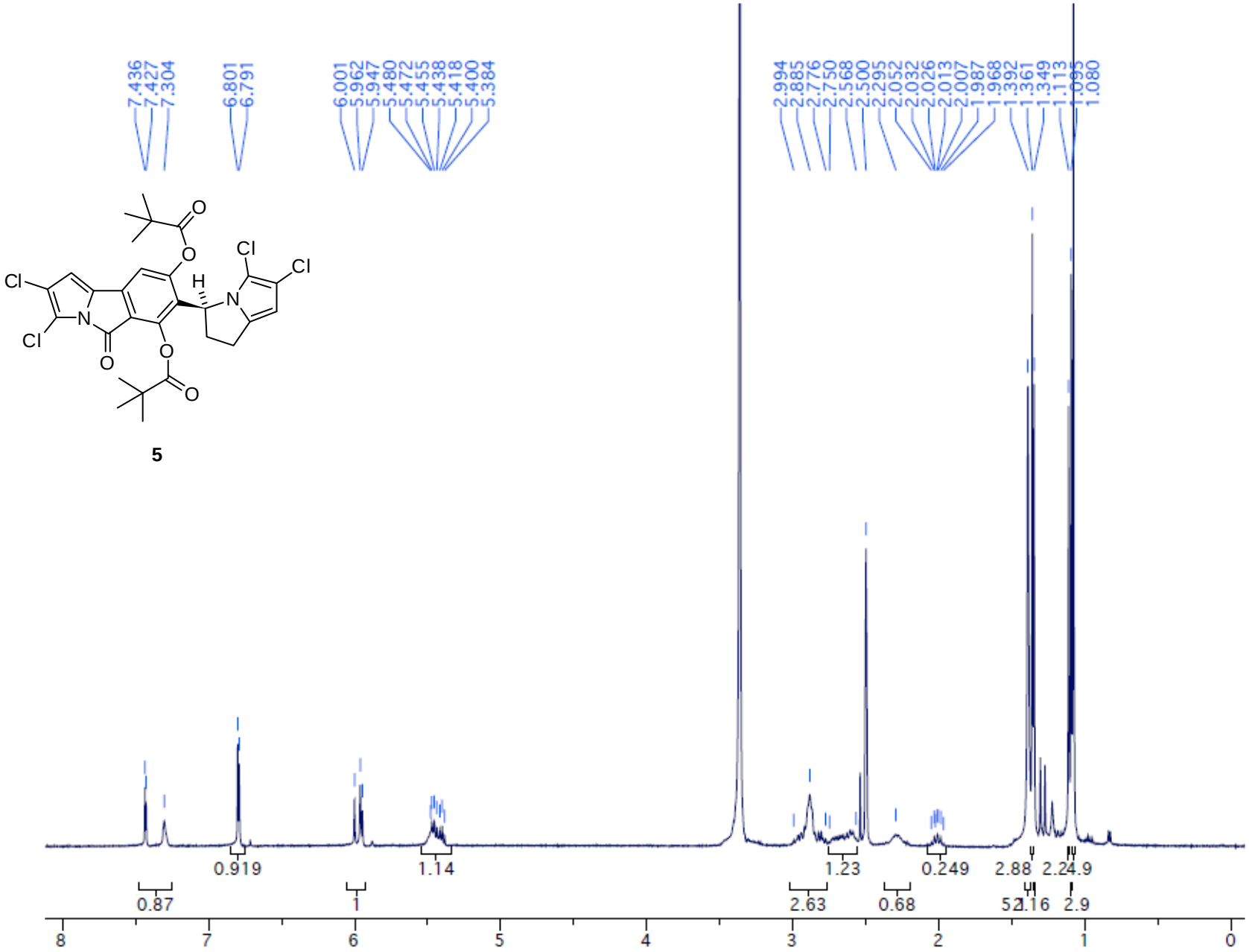
UV/vis spectrum (CH_3CN) of diisobutoyl chlorizidine A (**4**)



4

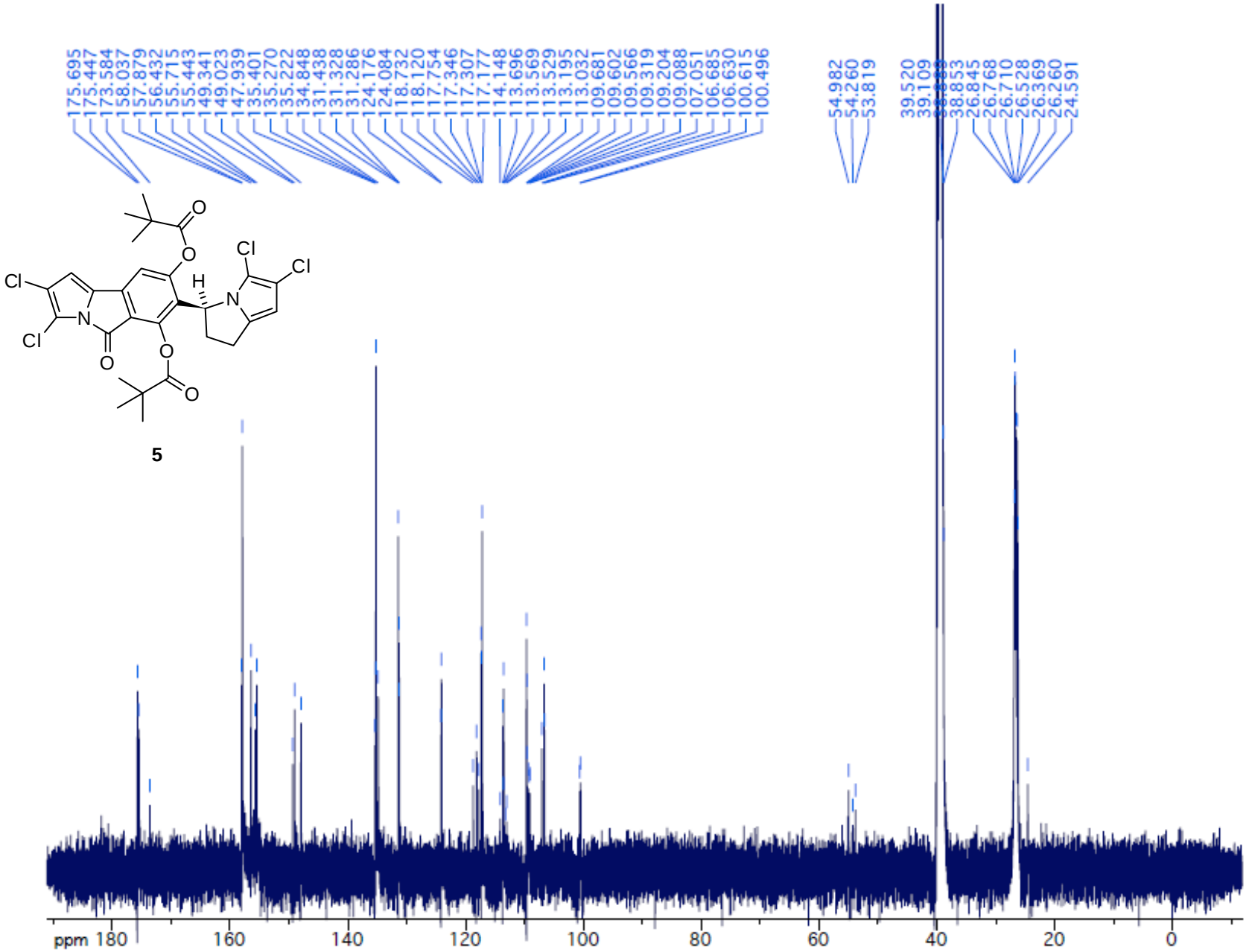

Zsobuty-a #130-135 RT: 2.36-2.47 AV: 6 NL: 8.49E3
T: FTMS + p ESI Full ms [100.00-2000.00]
607.0143
605.0174
[M+Na]+
609.0110
608.0180
606.0209
611.0668
609.3256
610.0149
610.3633
611.3020
612.0695
603.7928
604.4181
605.5416
606.4066
607.3093
608.3552
612.3912
Relative Abundance
m/z
4

^{1}H NMR (500 MHz, DMSO-d_6) of dipivaloyl chlorizidine A (**5**)



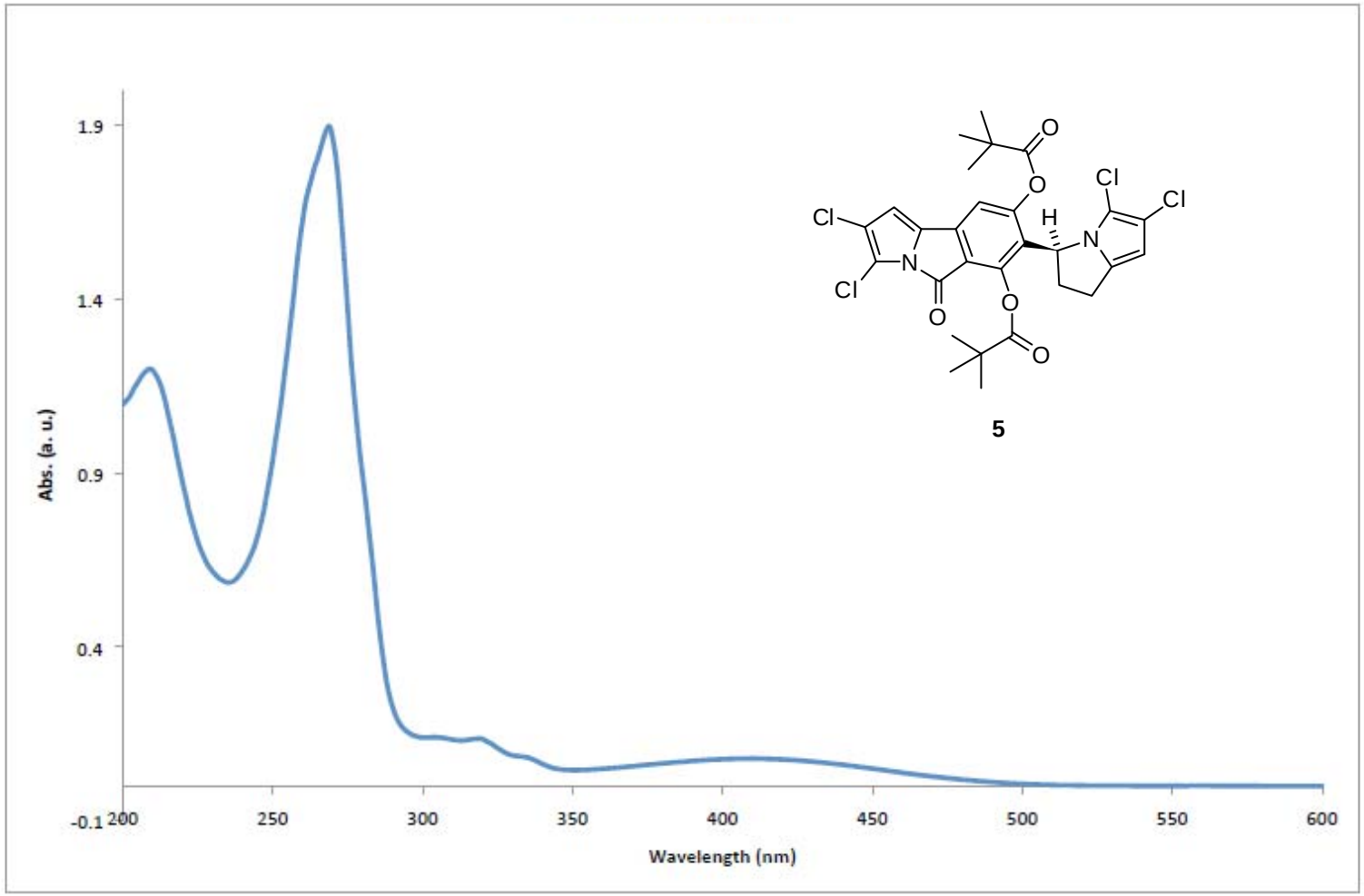
5

^{13}C NMR (125 MHz, DMSO-d_6) of dipivaloyl chlorizidine A (**5**)



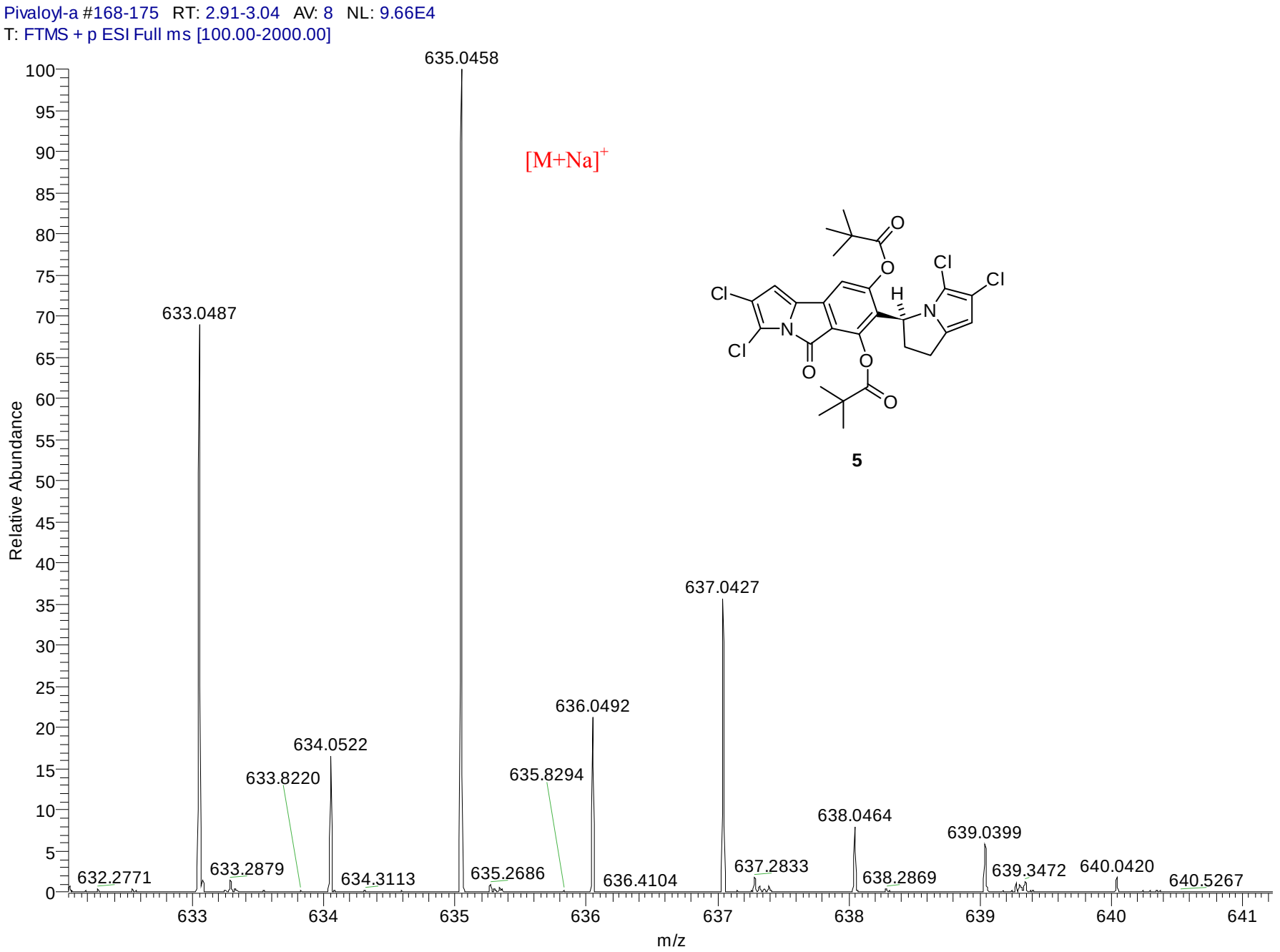
5

UV/vis spectrum (CH_3CN) of dipivaloyl chlorizidine A (**5**)



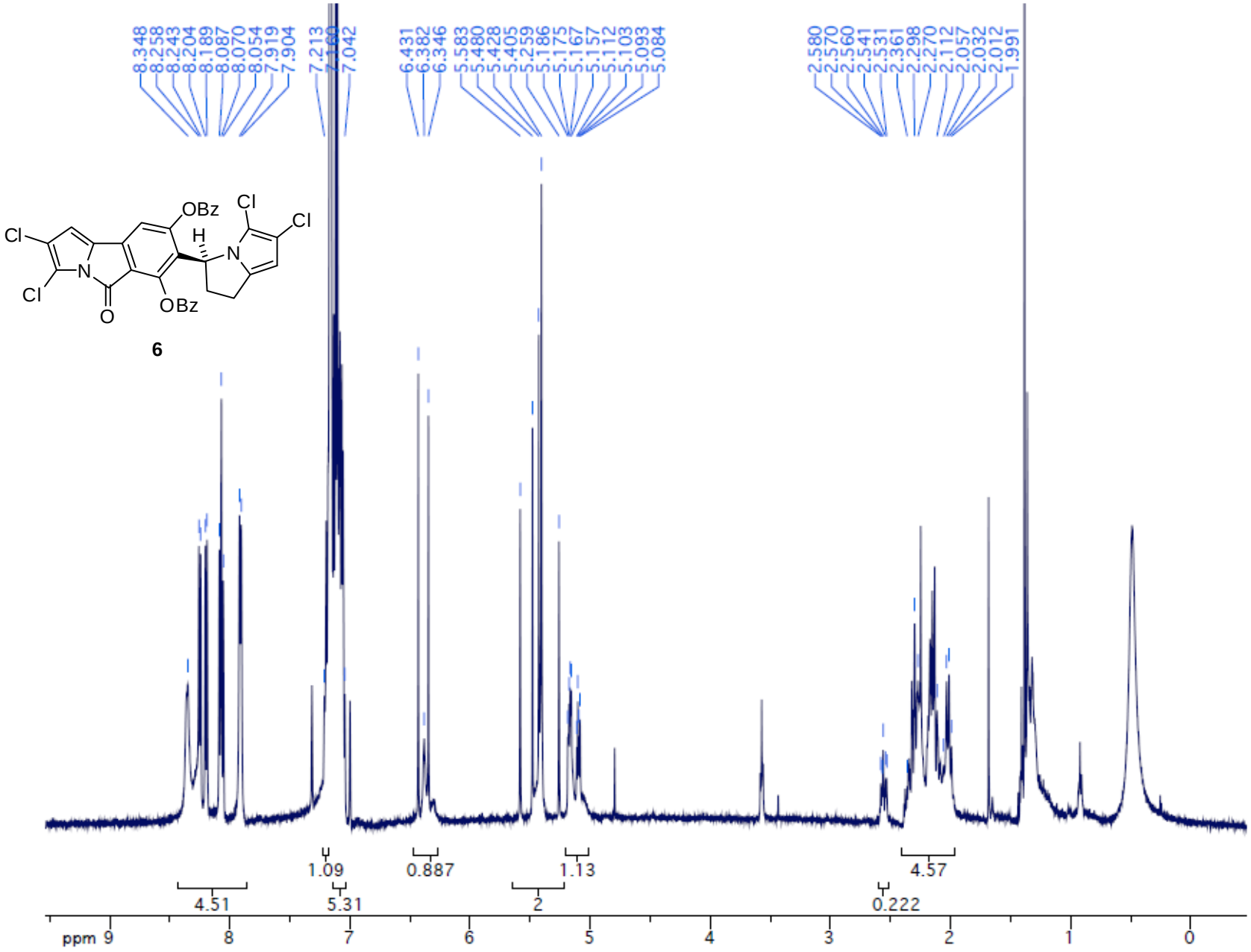
5

HRESI-FT-MS (Orbit-Trap-MS) of dipivaloyl chlorizidine A (**5**)



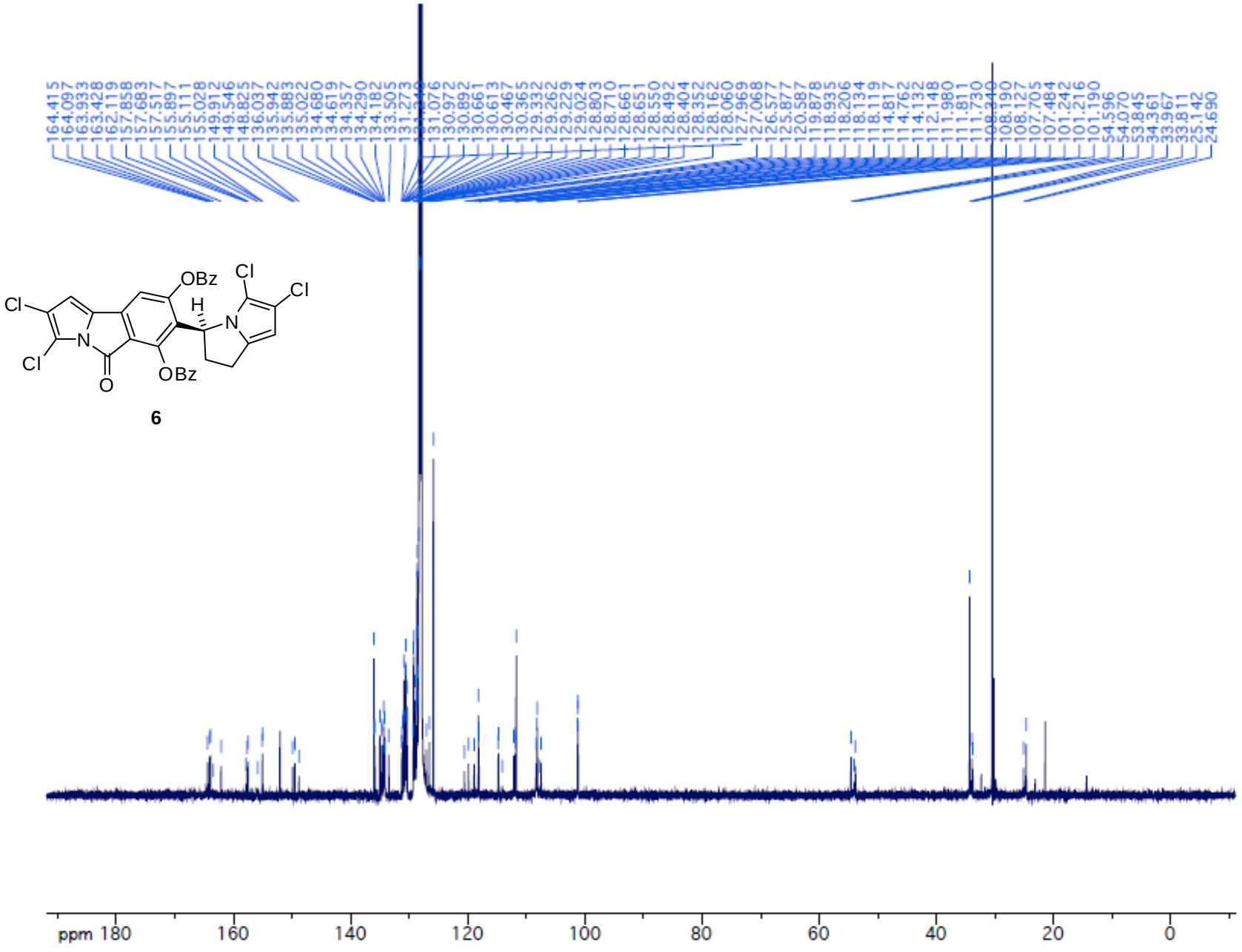
5

^{1}H NMR (500 MHz, benzene-d_6) of dibenzoyl chlorizidine A (**6**)

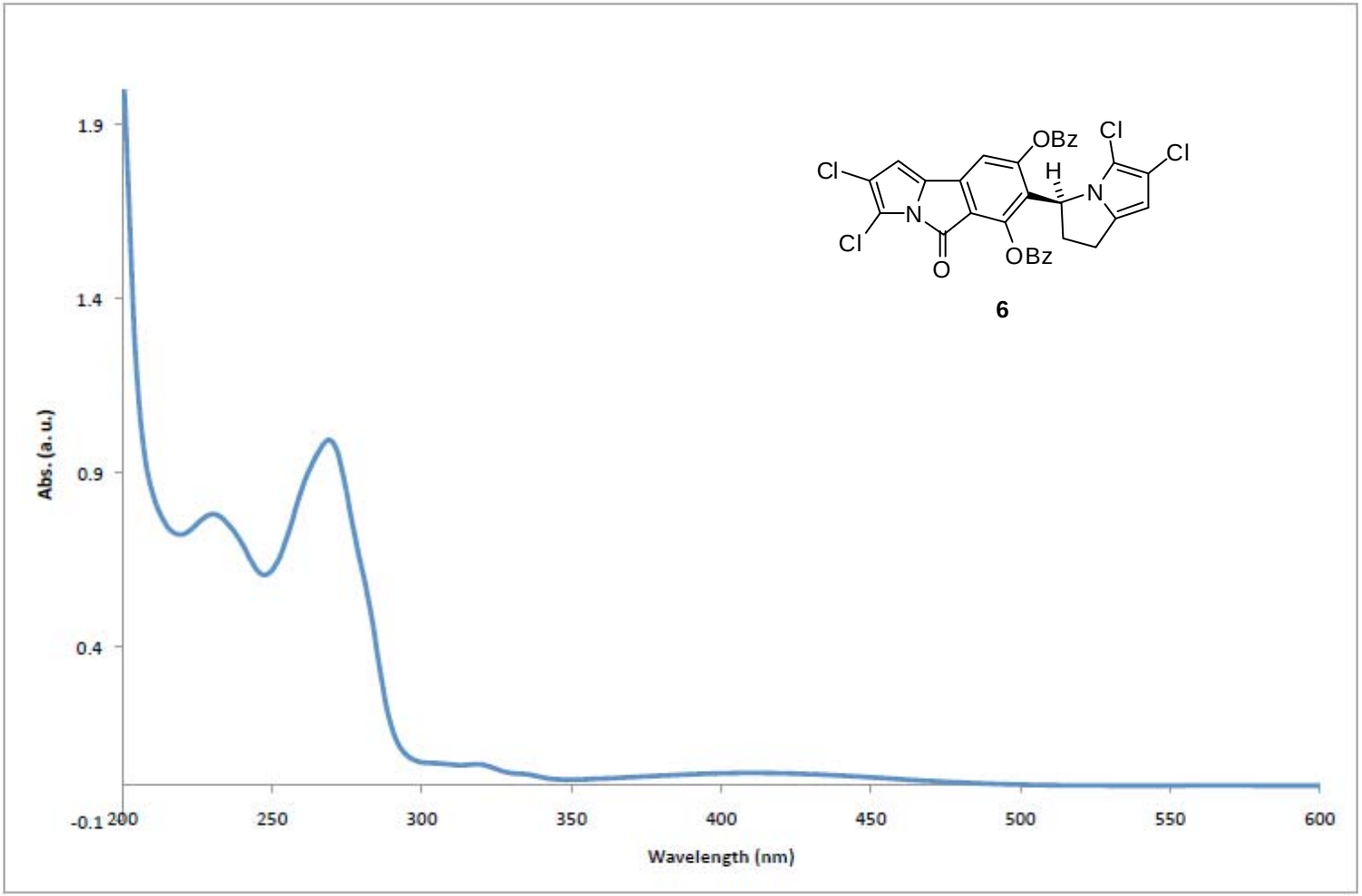


6

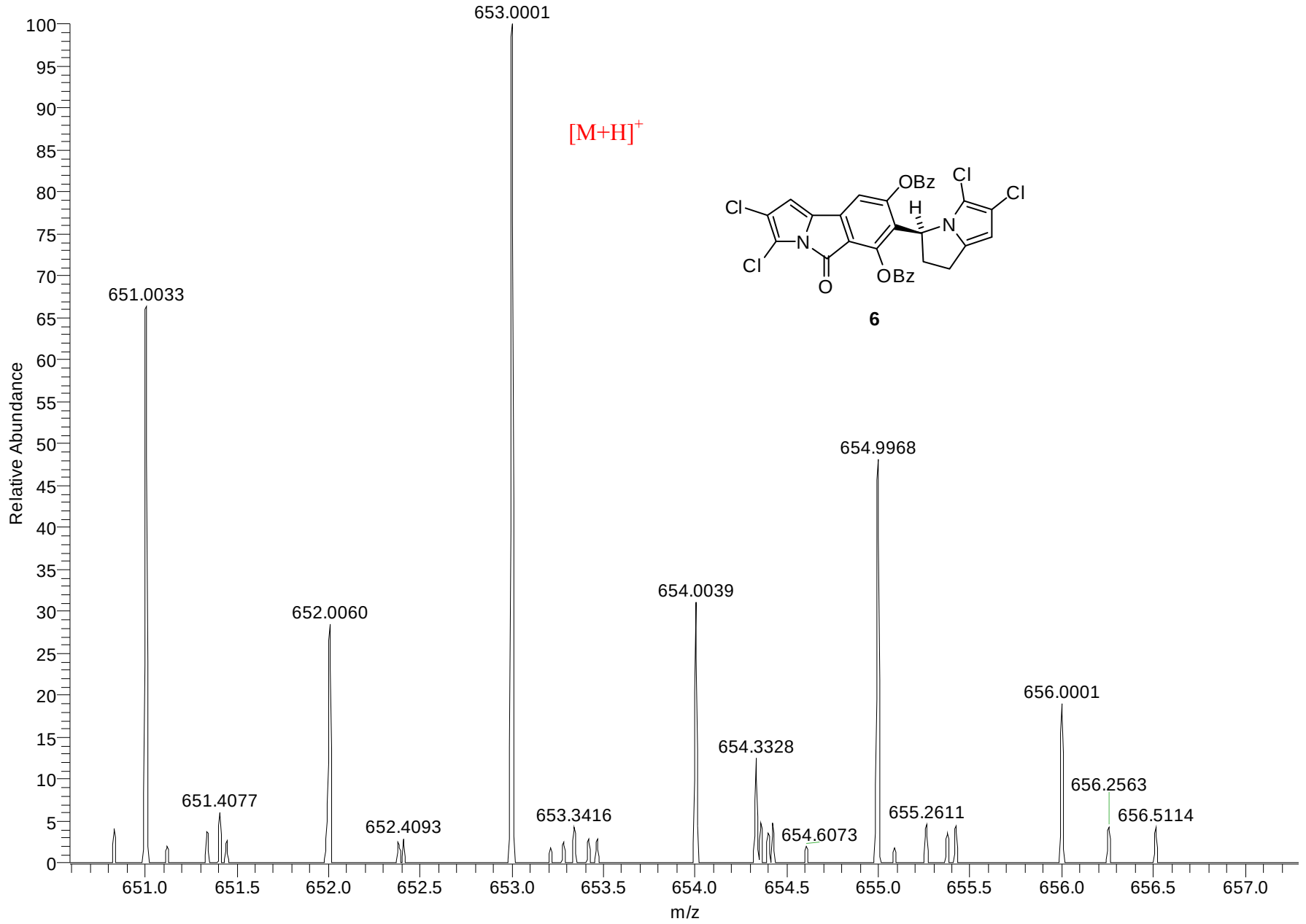
^{13}C NMR (125 MHz, benzene-d_6) of dibenzoyl chlorizidine A (**6**)



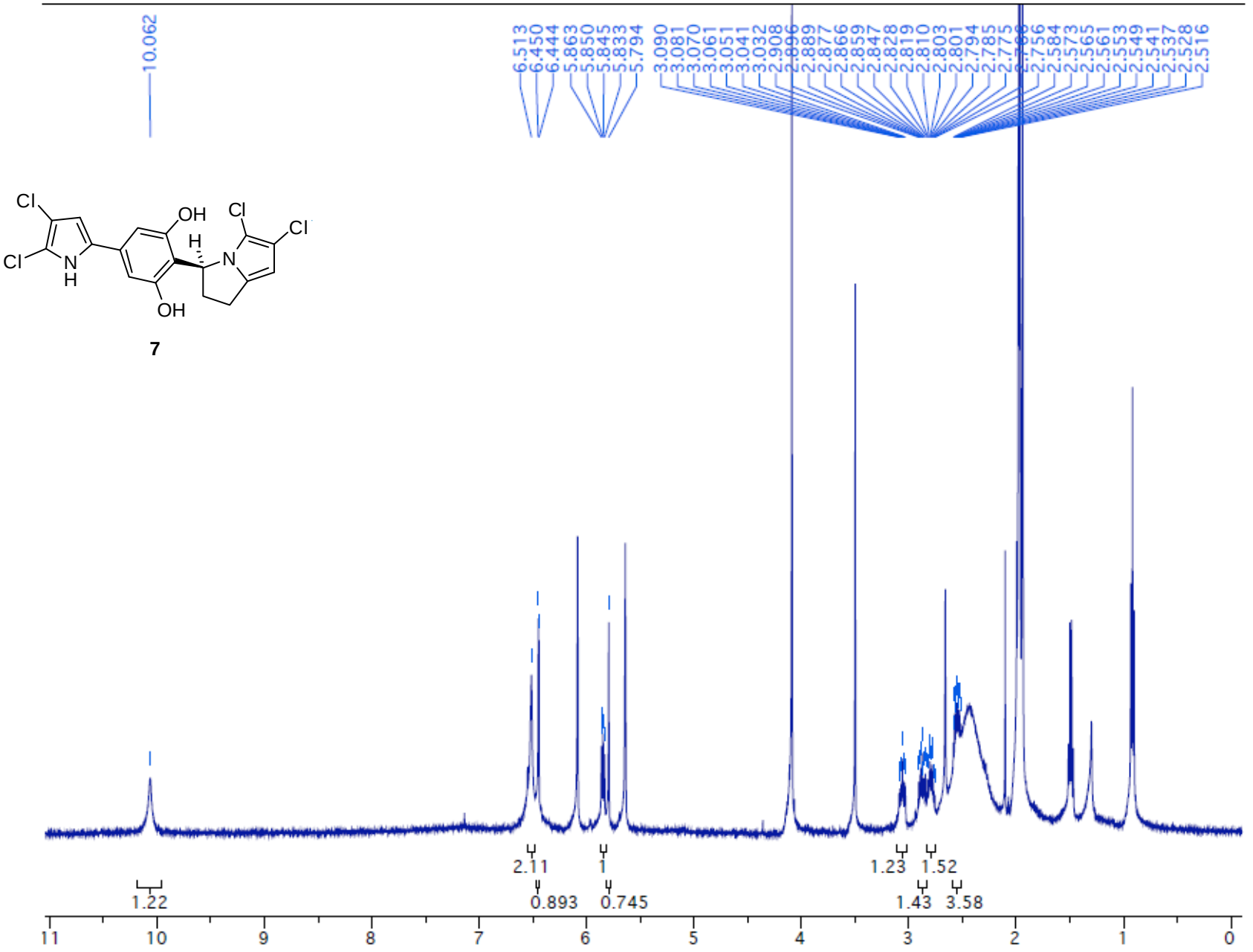
UV/vis spectrum (CH_3CN) of dibenzoyl chlorizidine A (**6**)



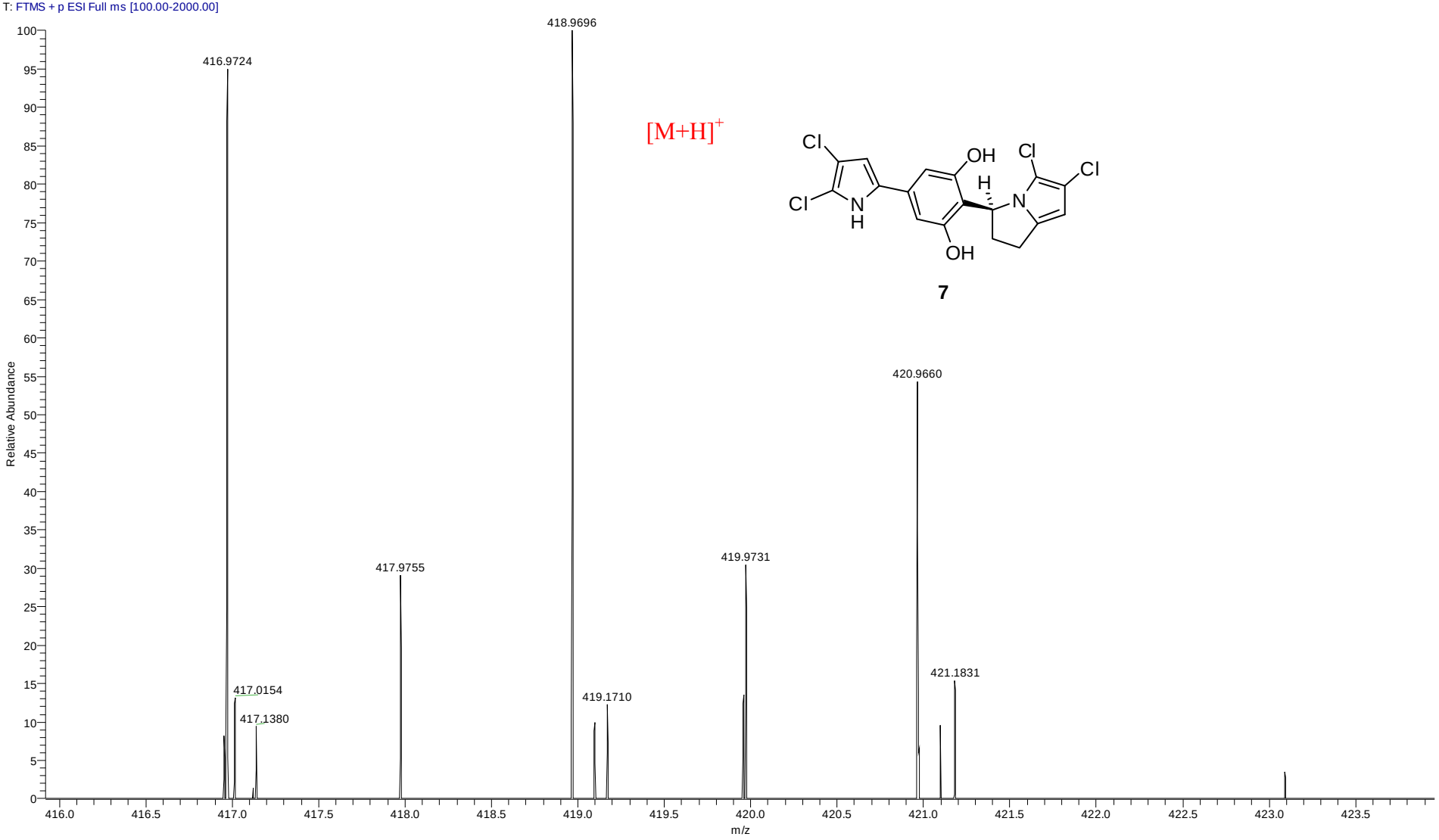
6


650-b #33-35 RT: 0.53-0.56 AV: 3 NL: 3.68E4
T: FTMS + p ESI Full ms [100.00-2000.00]
653.0001
[M+H]+
OBz
Cl
Cl
H
N
Cl
N
Cl
O
OBz
6
651.0033
654.9968
654.0039
652.0060
656.0001
654.3328
656.2563
651.4077
656.5114
653.3416
655.2611
652.4093
654.6073
Relative Abundance
m/z
100
95
90
85
80
75
70
65
60
55
50
45
40
35
30
25
20
15
10
5
0
651.0
651.5
652.0
652.5
653.0
653.5
654.0
654.5
655.0
655.5
656.0
656.5
657.0

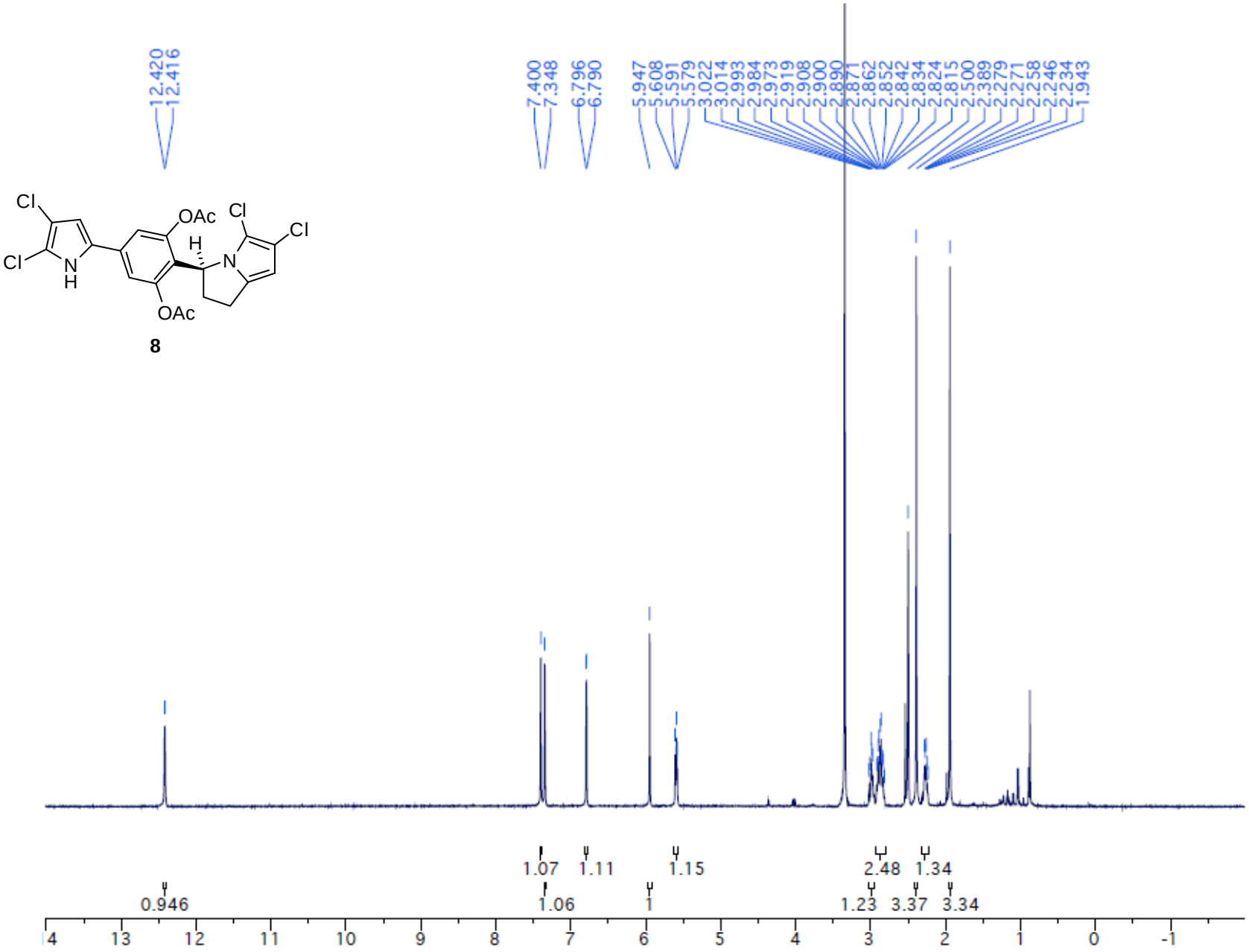
^{1}H NMR (500 MHz, CD_3CN) of chlorizidine B (**7**)



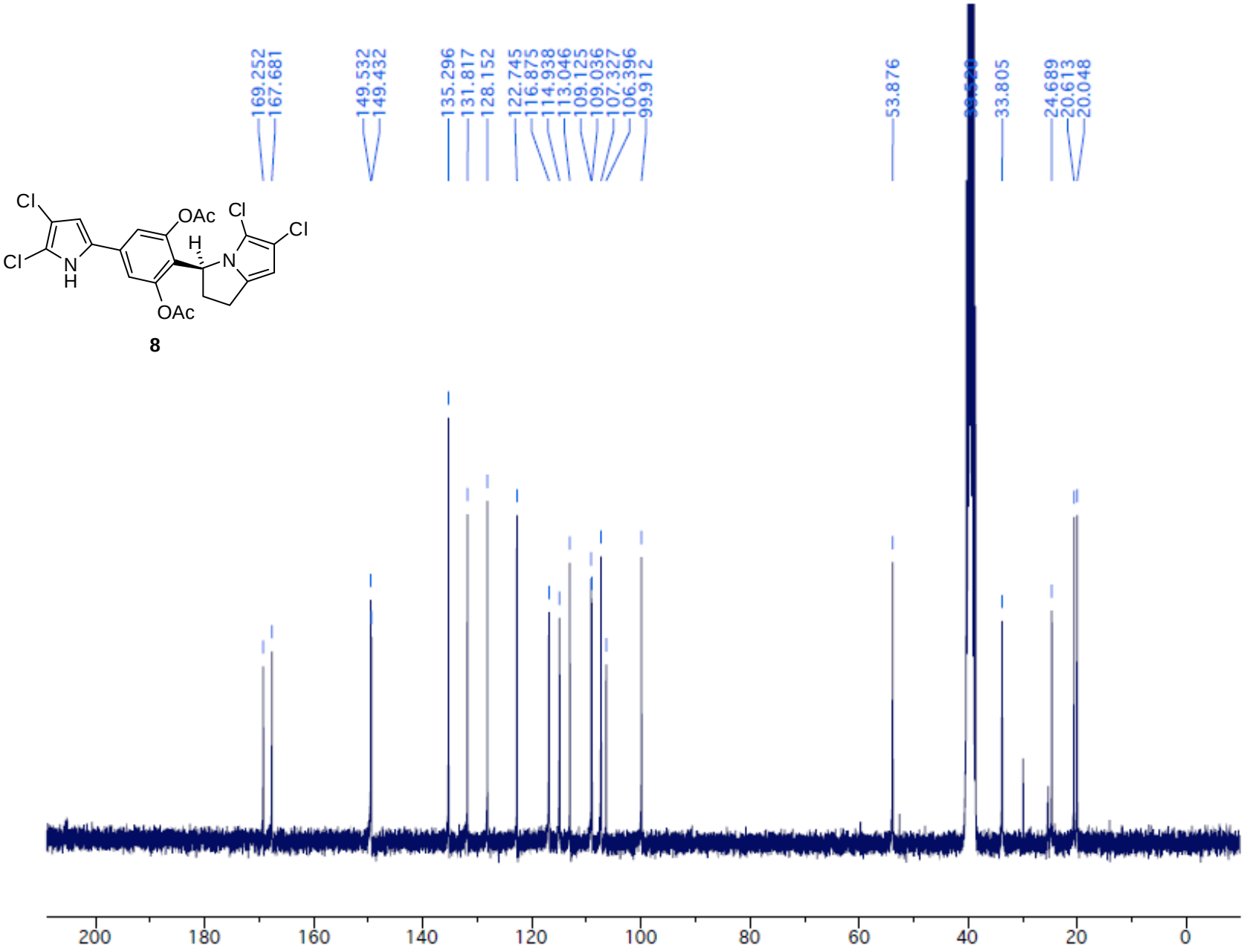
7


H287416 #316 RT: 5.61 AV: 1 SB: 4 3.37-3.41 NL: 4.08E4
T: FTMS + p ESI Full ms [100.00-2000.00]
418.9696
416.9724
[M+H]+
Cl
Cl
N
H
OH
H
OH
Cl
Cl
N
7
420.9660
419.9731
417.9755
421.1831
419.1710
417.0154
417.1380
Relative Abundance
m/z

^{1}H NMR (500 MHz, DMSO-d_6) of diacetyl chlorizidine B (**8**)

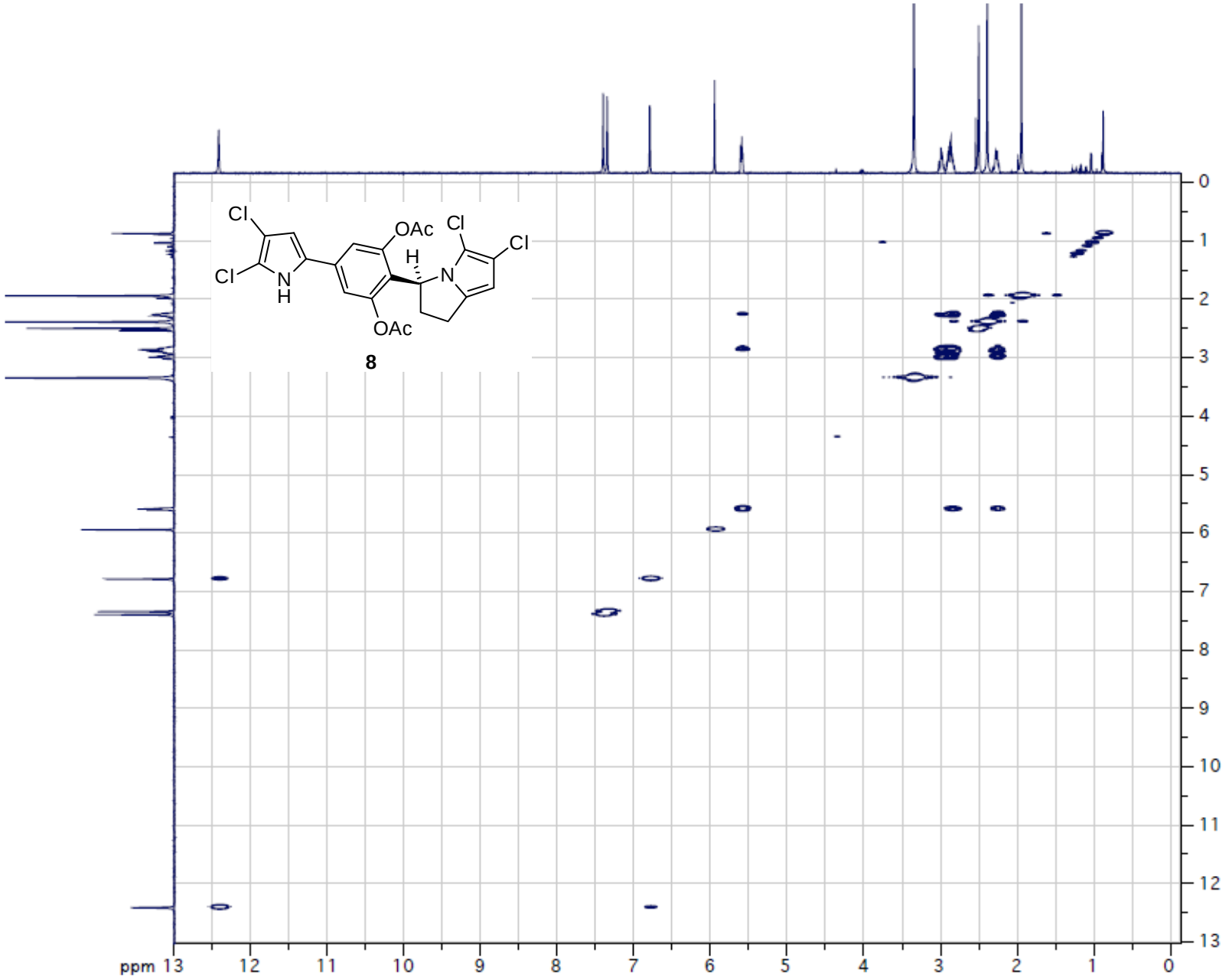


^{13}C NMR (125 MHz, DMSO-d_6) diacetyl chlorizidine B (**8**)

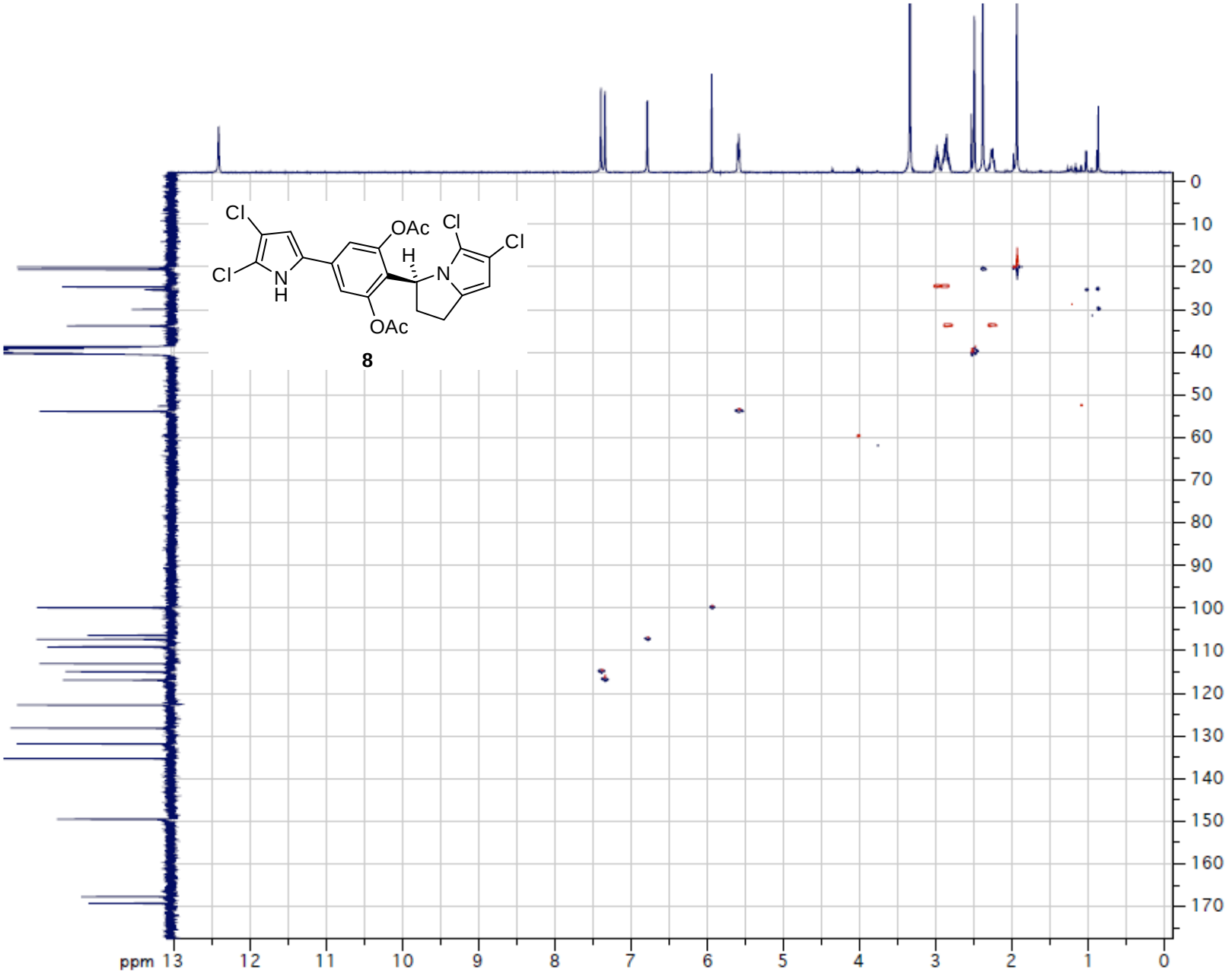


8

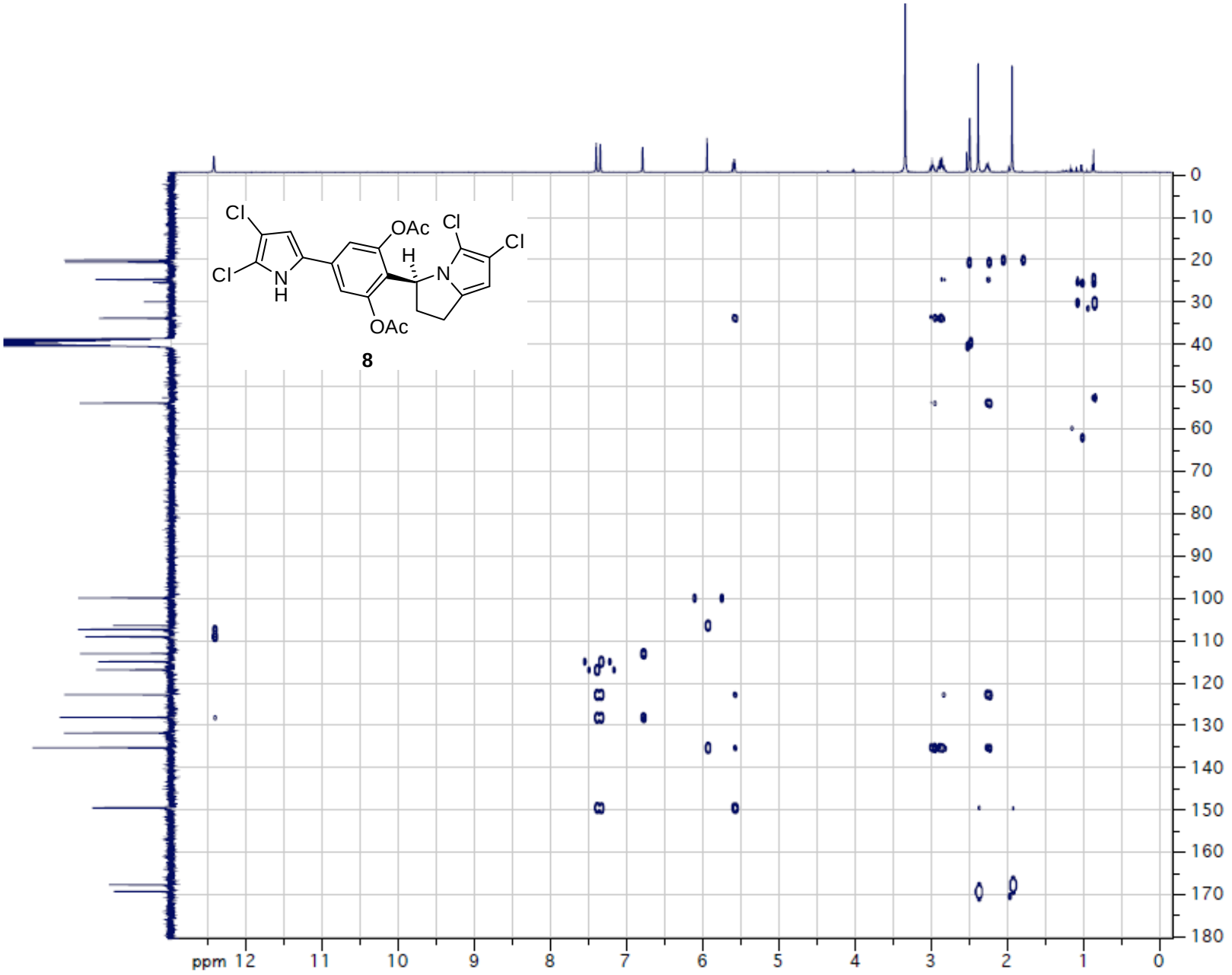
COSY (500 MHz, DMSO-d_6) of diacetyl chlorizidine B (**8**)



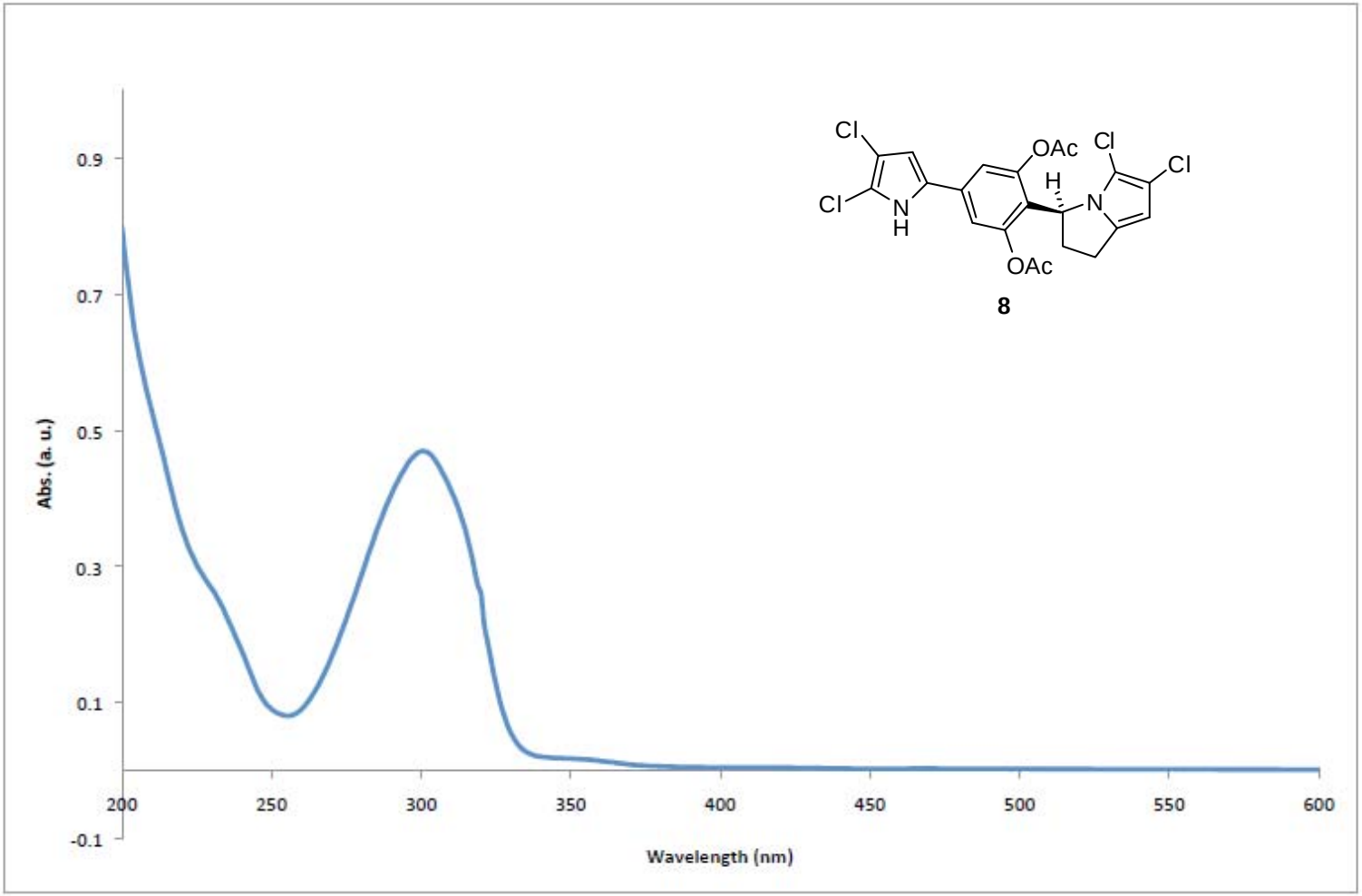
HSQC (500 MHz, DMSO-d_6) of diacetyl chlorizidine B (**8**)

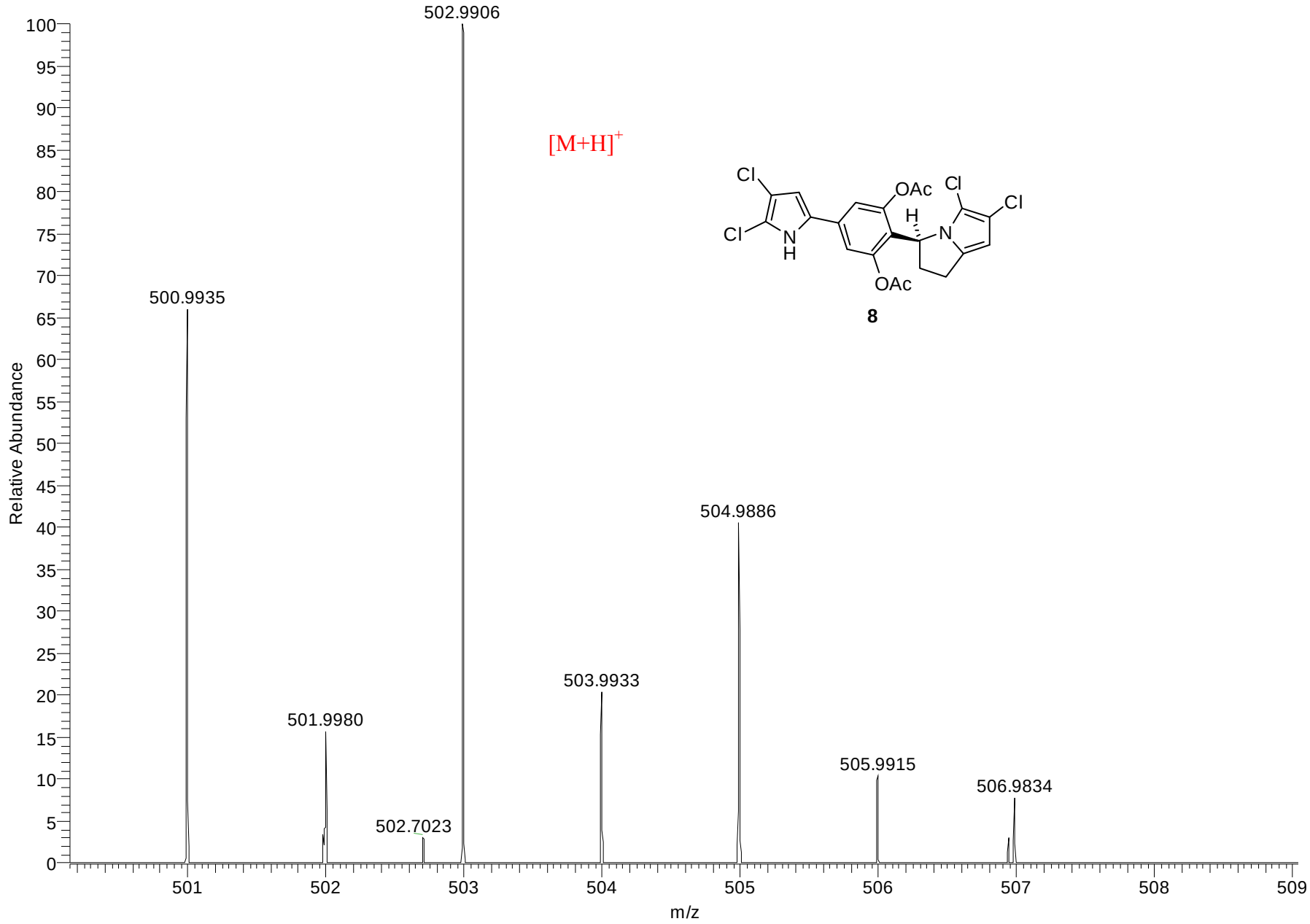


HMBC (500 MHz, DMSO-d_6) of diacetyl chlorizidine B (**8**)

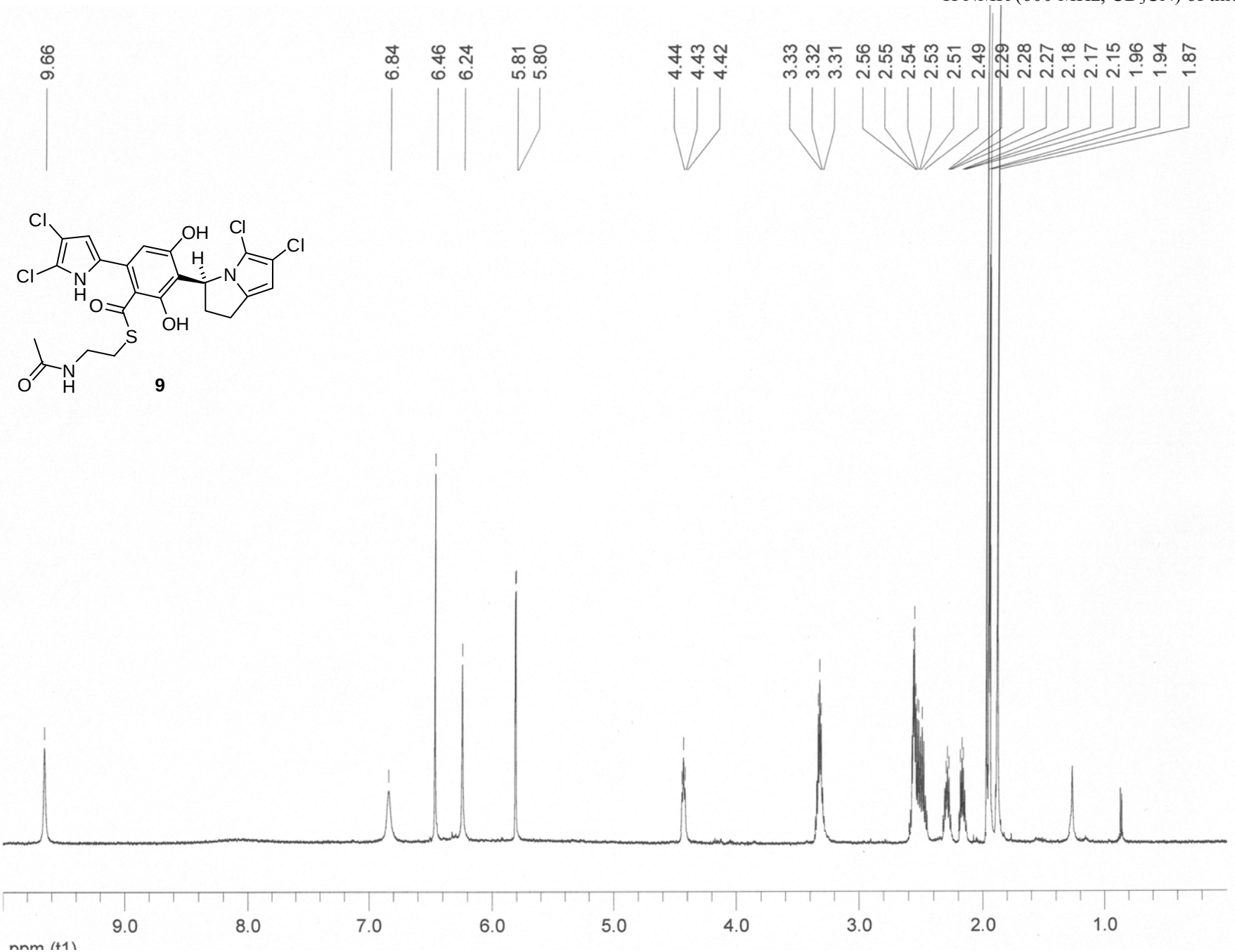


UV/vis spectrum (CH_3CN) of diacetyl chlorizidine B (**8**)

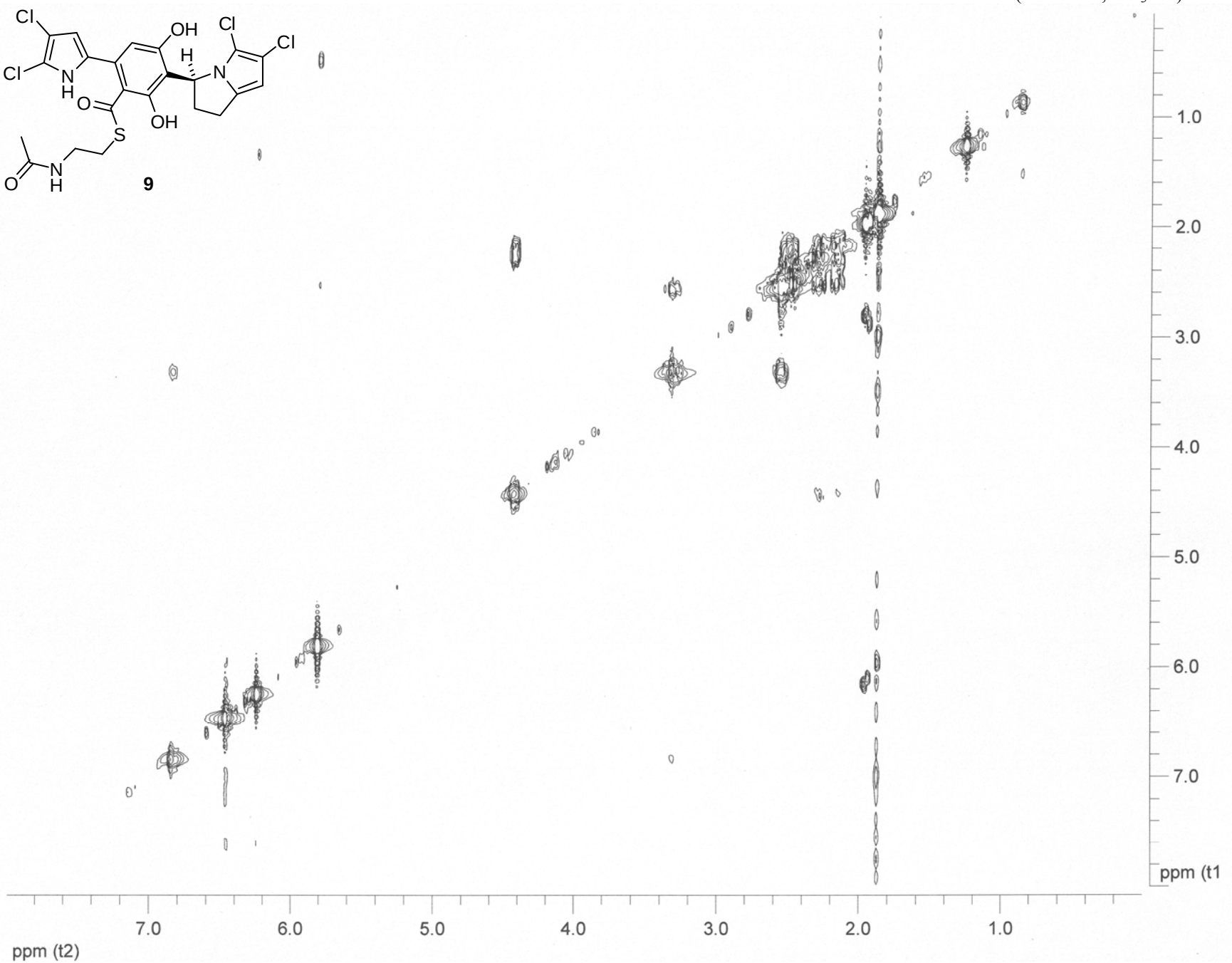



500 #98 RT: 1.57 AV: 1 NL: 3.62E5
T: FTMS + p ESI Full ms [100.00-2000.00]
502.9906
[M+H]+
500.9935
501.9980
502.7023
503.9933
504.9886
505.9915
506.9834
Relative Abundance
m/z
Cl
Cl
N
H
OAc
OAc
H
N
Cl
Cl
8

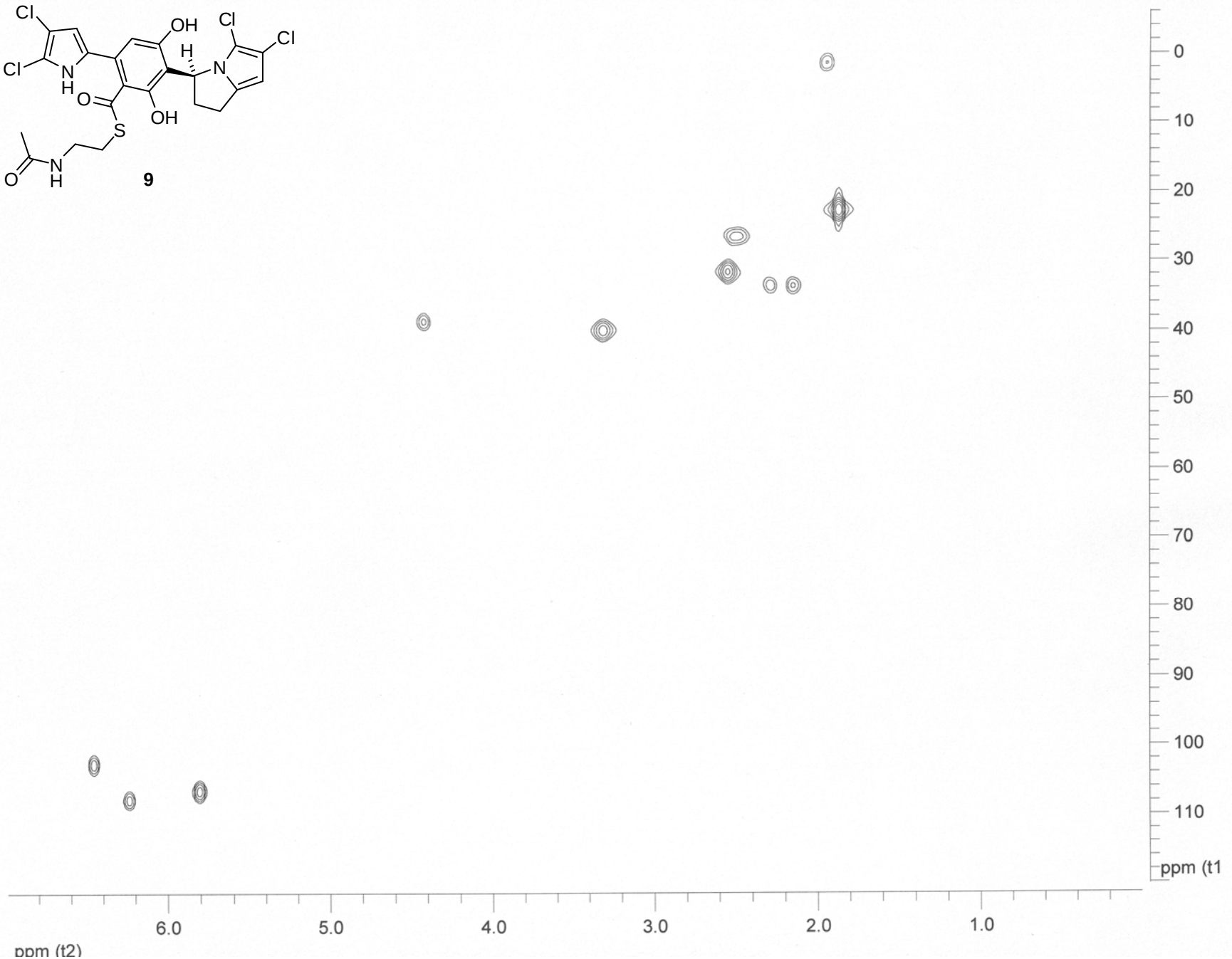
^{1}H NMR (600 MHz, CD_3CN) of thioester **9**



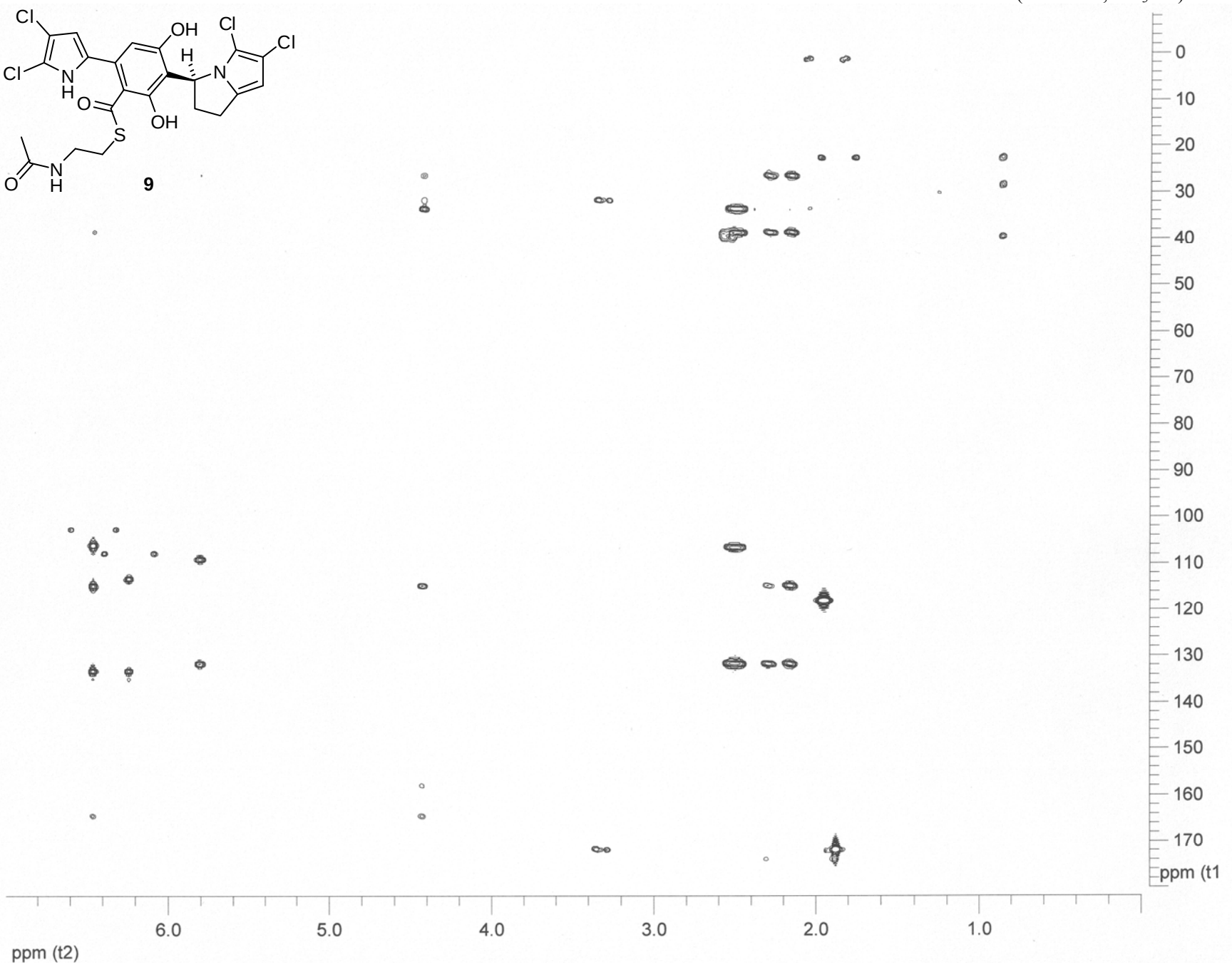
COSY (600 MHz, CD_3CN) of thioester **9**



HSQC (600 MHz, CD_3CN) of thioester **9**



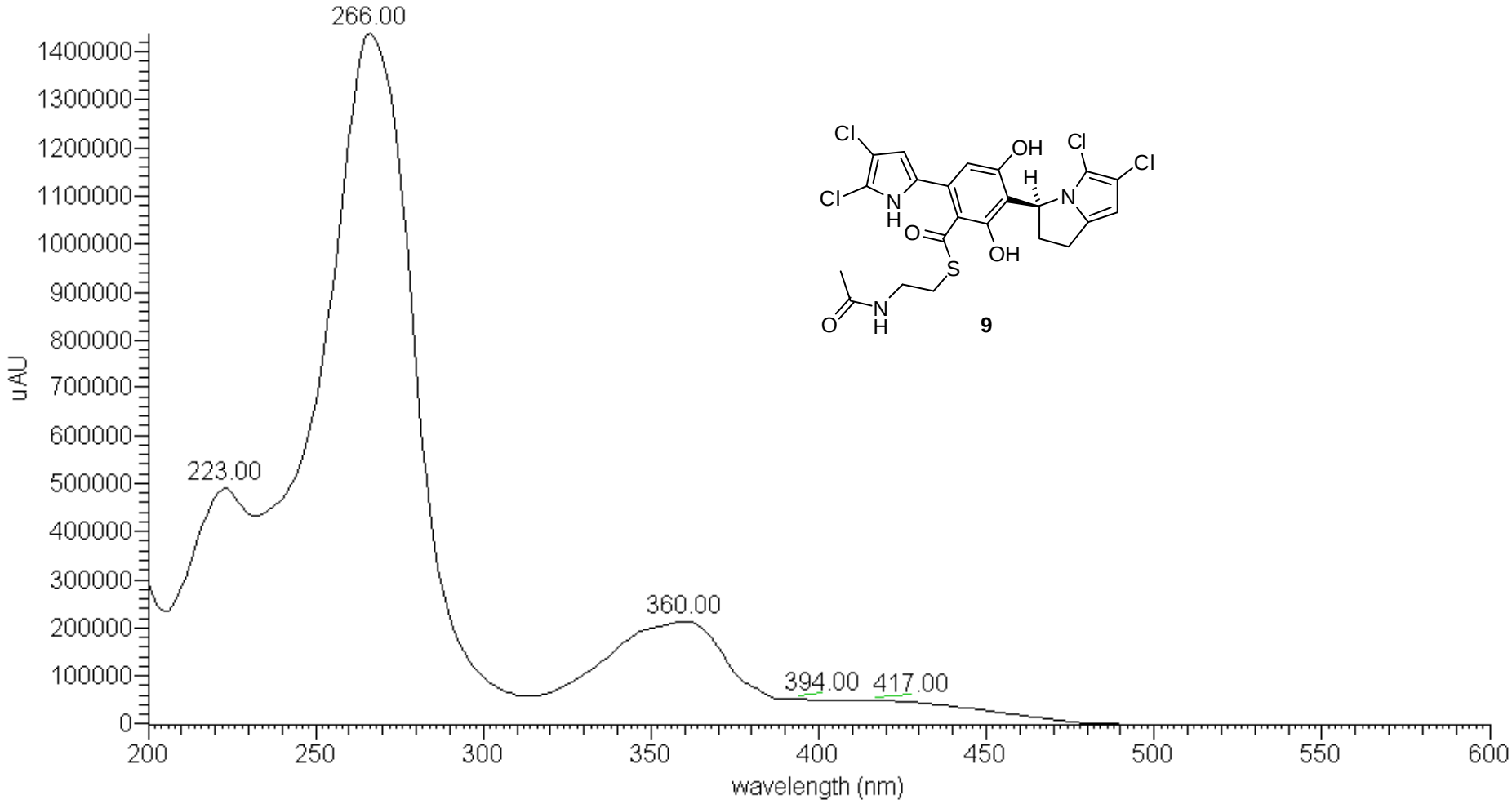
HMBC (600 MHz, CD_3CN) of thioester **9**



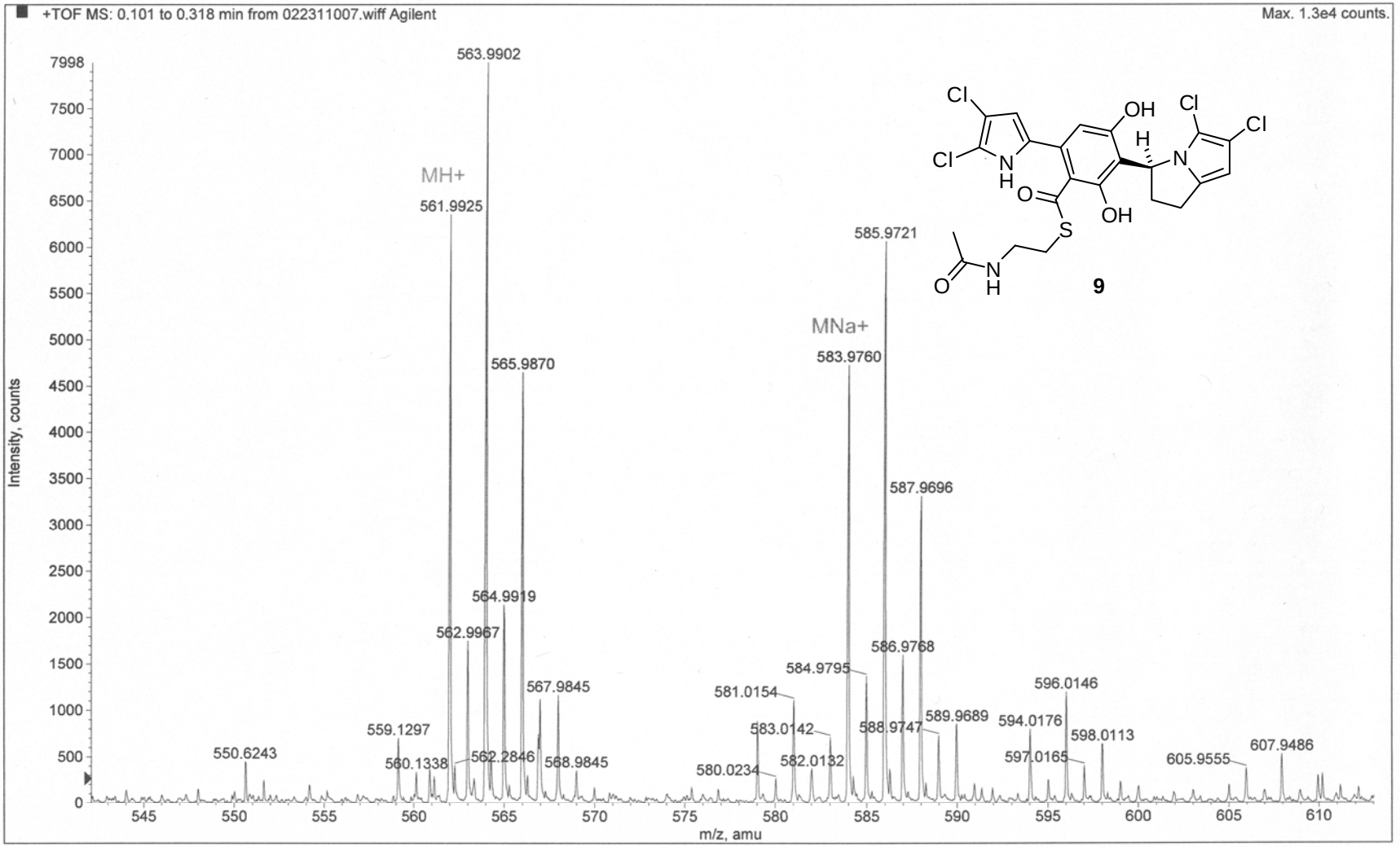
UV/vis spectrum (CH_3CN/water) of thioester **9**

C:\Xcalibur\data\Chambers\thiolsemip1 11/5/2010 11:06:35 AM

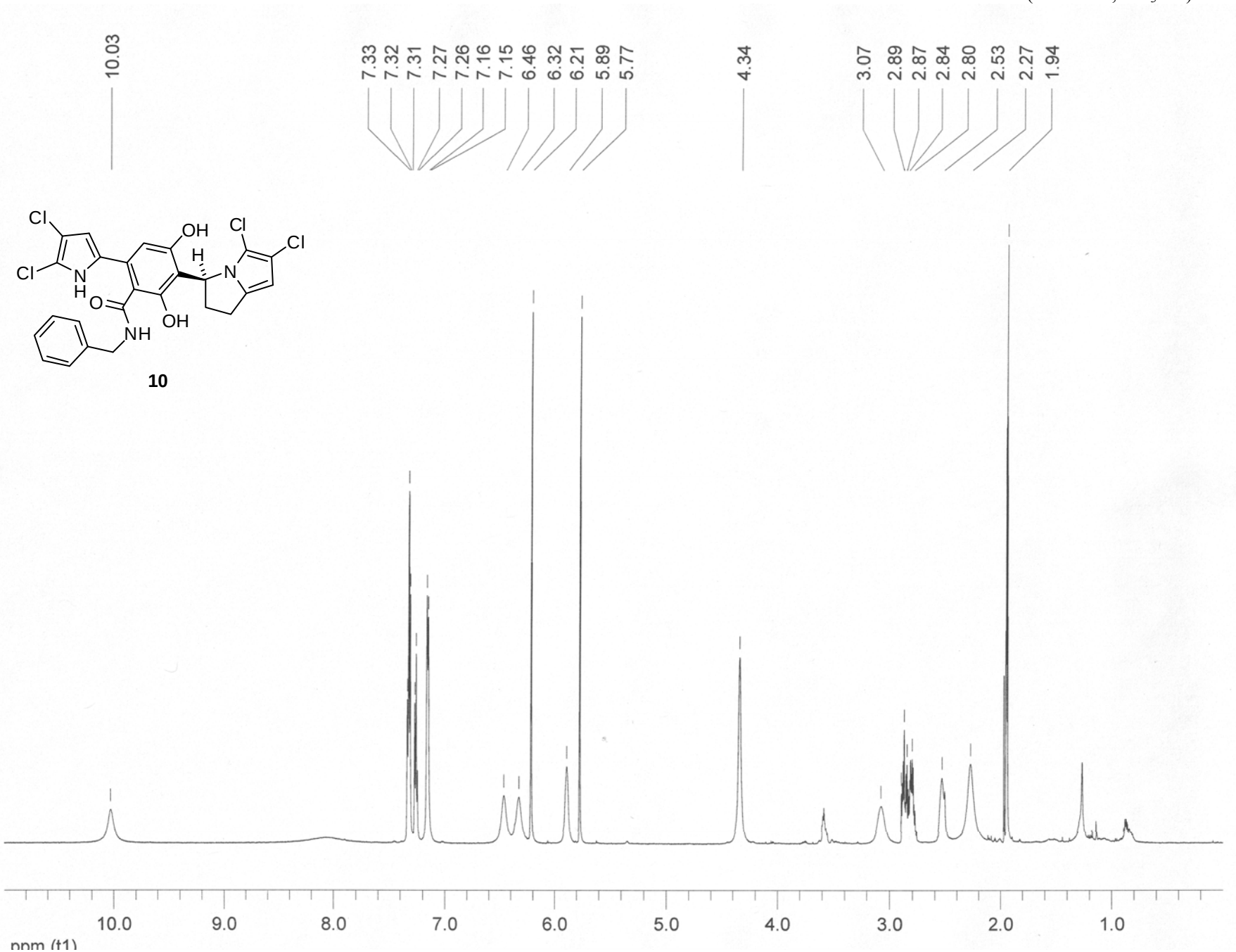
thiolsemip1 #4764 RT: 15.88 AV: 1 NL: 1.44E6 microAU



9

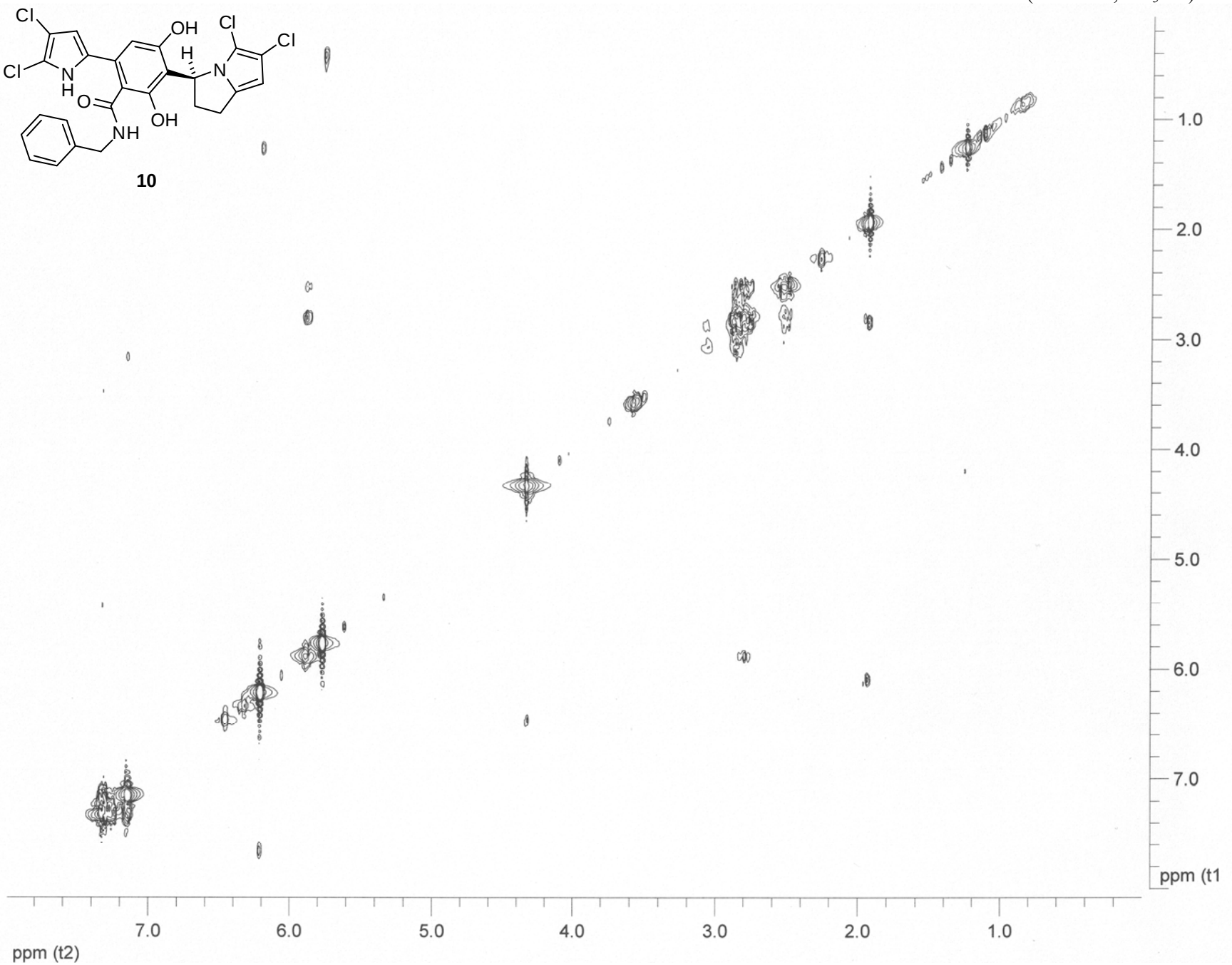

Polarity/Scan Type: Positive
Sample Name: flch8376
Acq. File: 022311007.wiff
Acq. Date: Wednesday, February 23
+TOF MS: 0.101 to 0.318 min from 022311007.wiff Agilent
Max. 1.3e4 counts.
Intensity, counts
m/z, amu
MH+
MNa+
563.9902
561.9925
565.9870
564.9919
562.9967
567.9845
559.1297
560.1338
562.2846
568.9845
550.6243
585.9721
583.9760
587.9696
586.9768
584.9795
581.0154
583.0142
588.9747
589.9689
596.0146
594.0176
598.0113
597.0165
607.9486
605.9555
580.0234
582.0132
9

^{1}H NMR (600 MHz, CD_3CN) of amide **10**

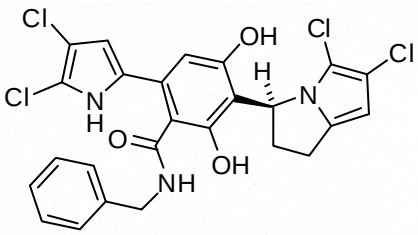


10

COSY (600 MHz, CD_3CN) of amide **10**

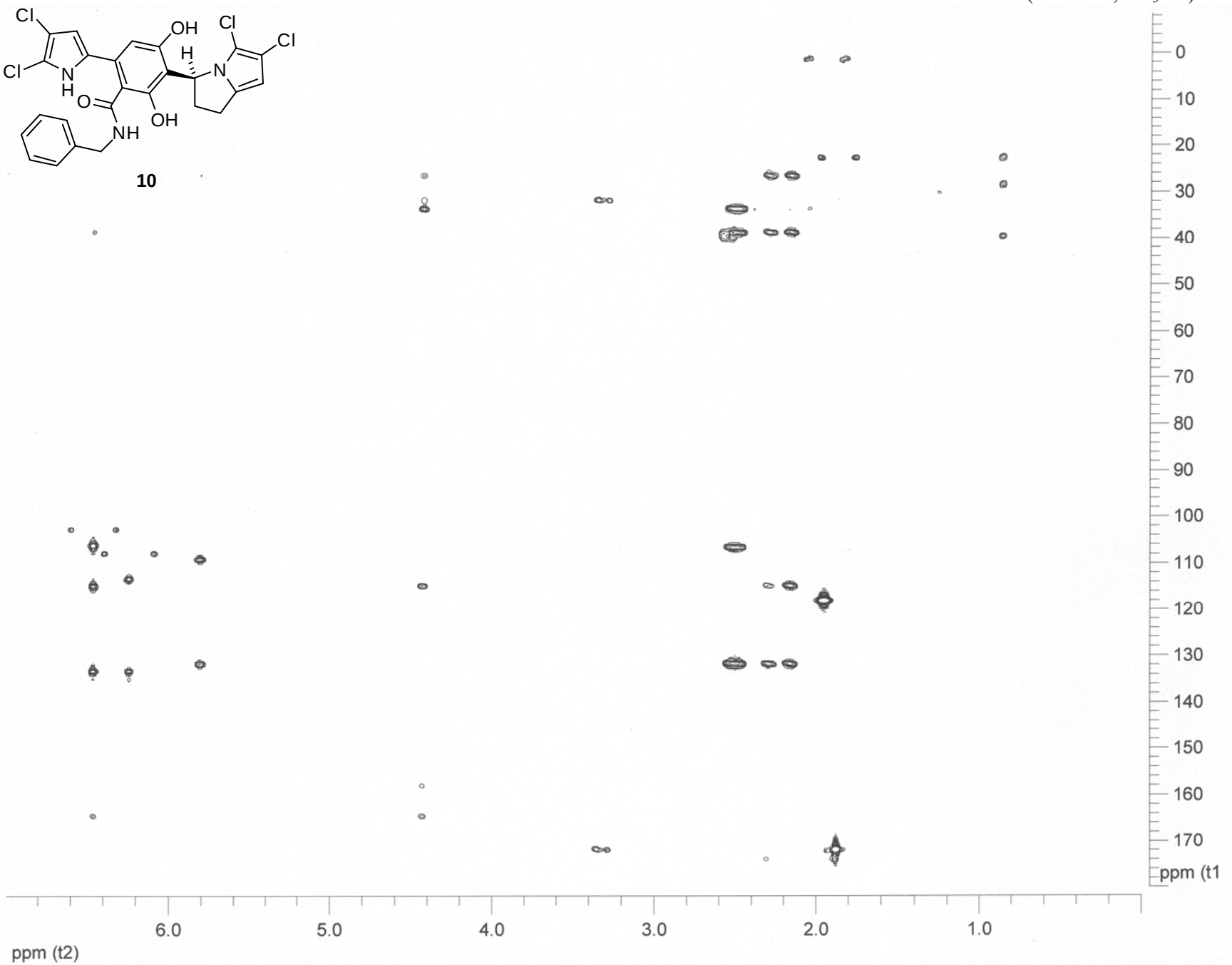


HSQC (600 MHz, CD_3CN) of amide **10**



10

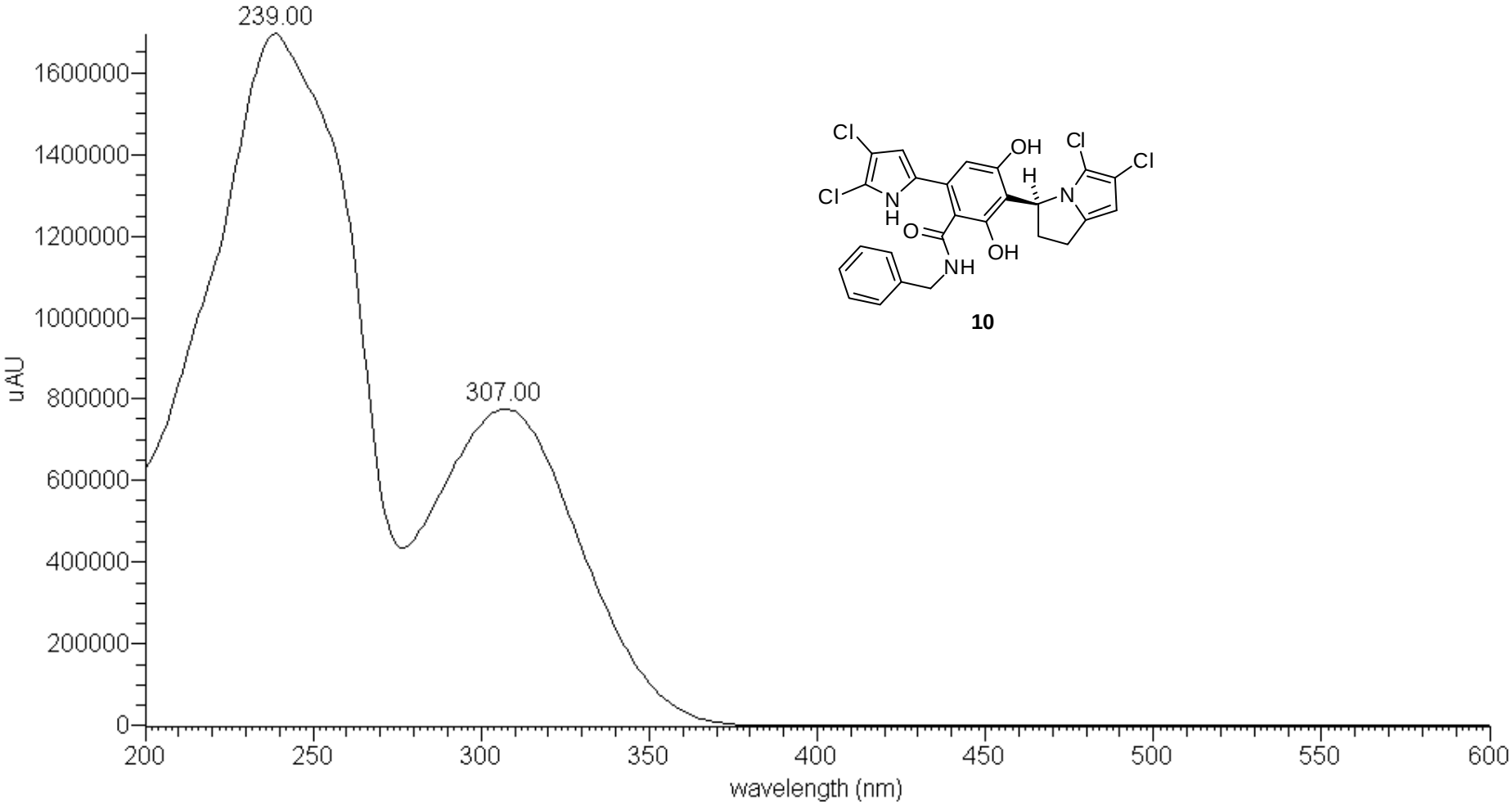
HMBC (600 MHz, CD_3CN) of amide **10**



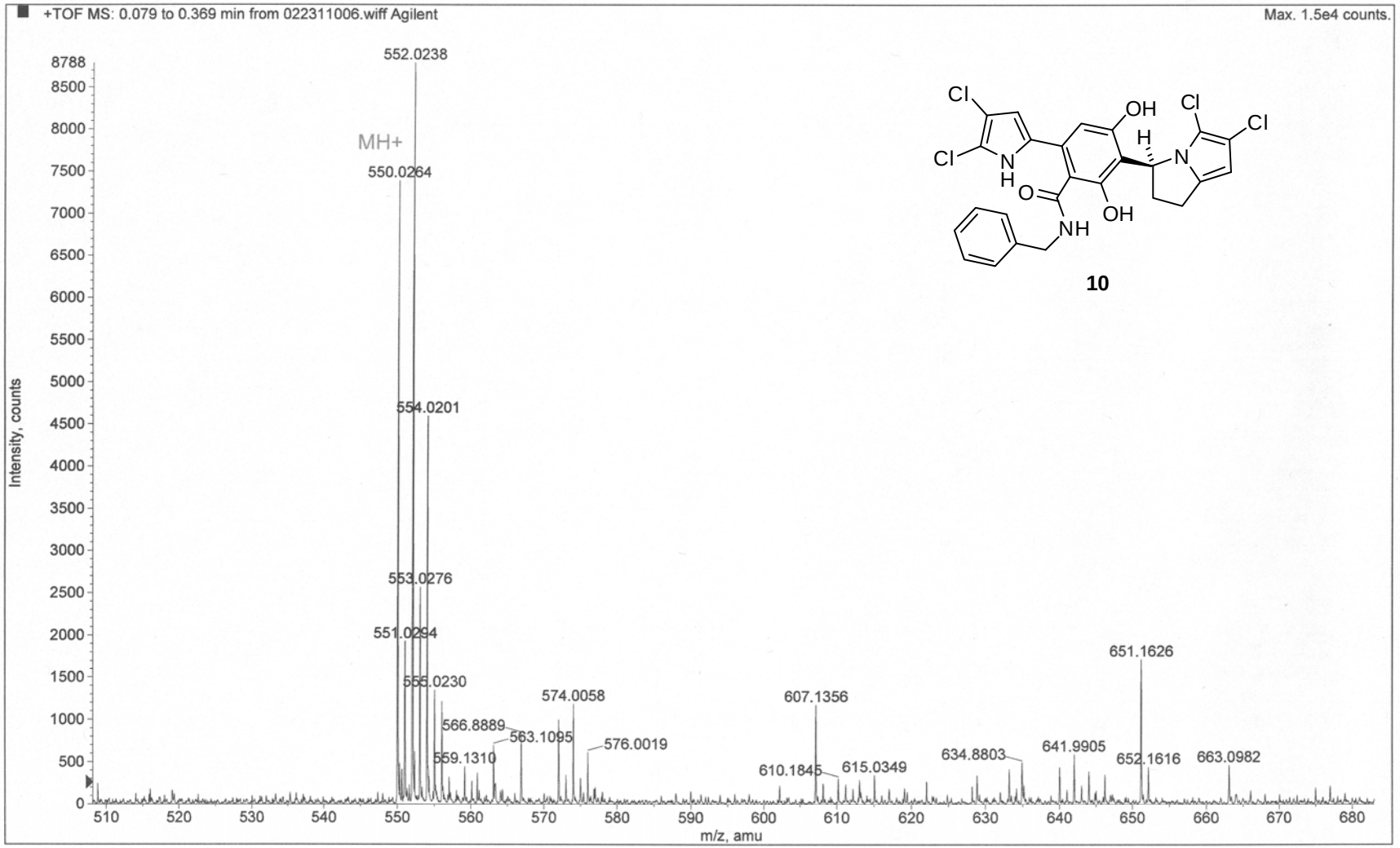
UV/vis spectrum (CH_3CN/water) of amide **10**

C:\Xcalibur\...\may3benzylaminebig1 5/3/2011 2:16:43 PM

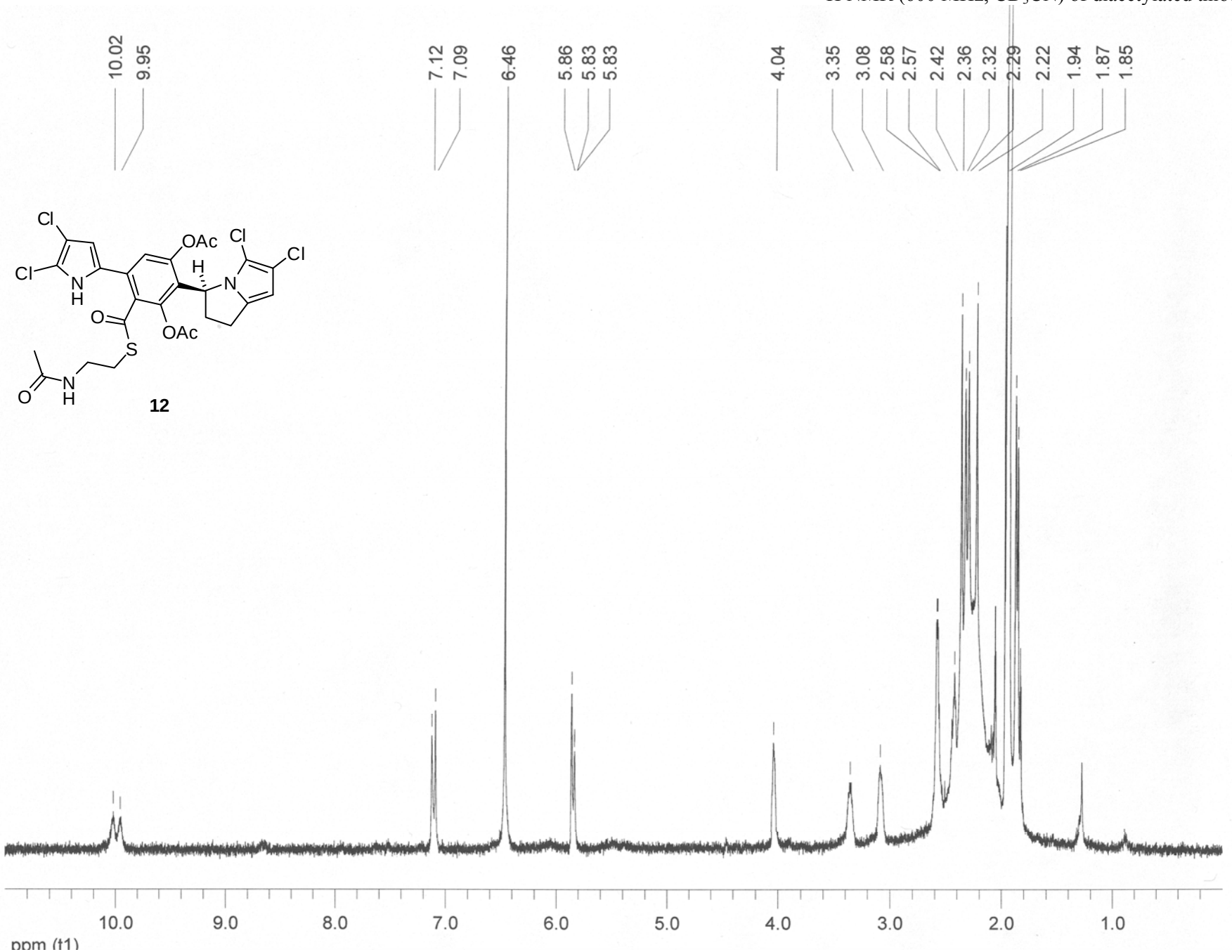
may3benzylaminebig1 #5605 RT: 18.68 AV: 1 NL: 1.69E6 microAU



10


Polarity/Scan Type: Positive
Sample Name: flch8375
Acq. File: 022311006.wiff
Acq. Date: Wednesday, February 23
+TOF MS: 0.079 to 0.369 min from 022311006.wiff Agilent
Max. 1.5e4 counts.
552.0238
MH+
550.0264
554.0201
553.0276
551.0294
556.0230
574.0058
566.8889
563.1095
576.0019
559.1310
607.1356
610.1845
615.0349
634.8803
641.9905
651.1626
652.1616
663.0982
Intensity, counts
m/z, amu
Cl
OH
Cl
H
Cl
N
H
N
O
OH
NH
10

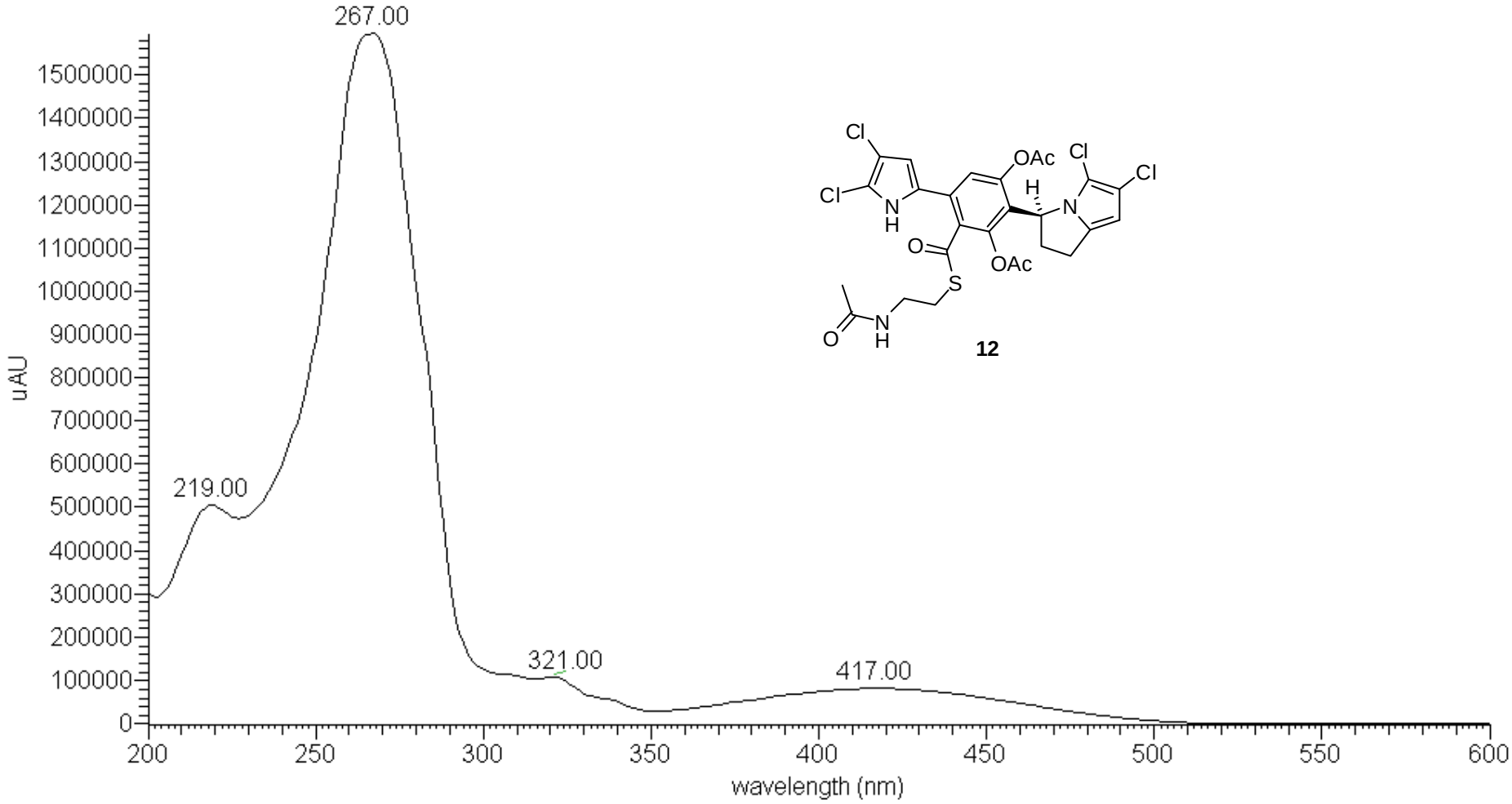
^{1}H NMR (600 MHz, CD_3CN) of diacetylated thioester **12**



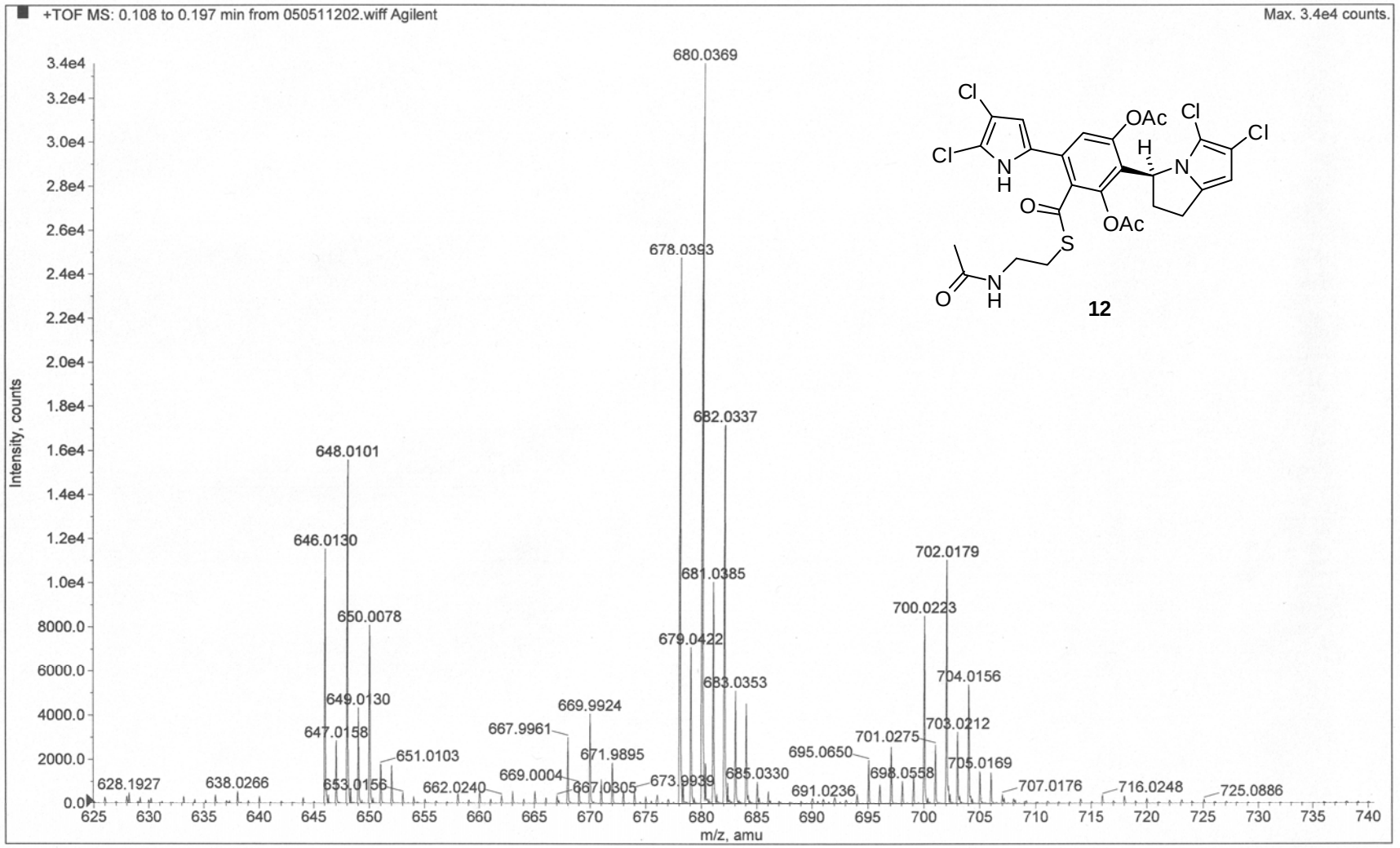
UV/vis spectrum (CH_3CN/water) of diacetylated thioester **12**

C:\Xcalibur\...\may4thioesterAc2O1P 5/4/2011 9:23:55 AM

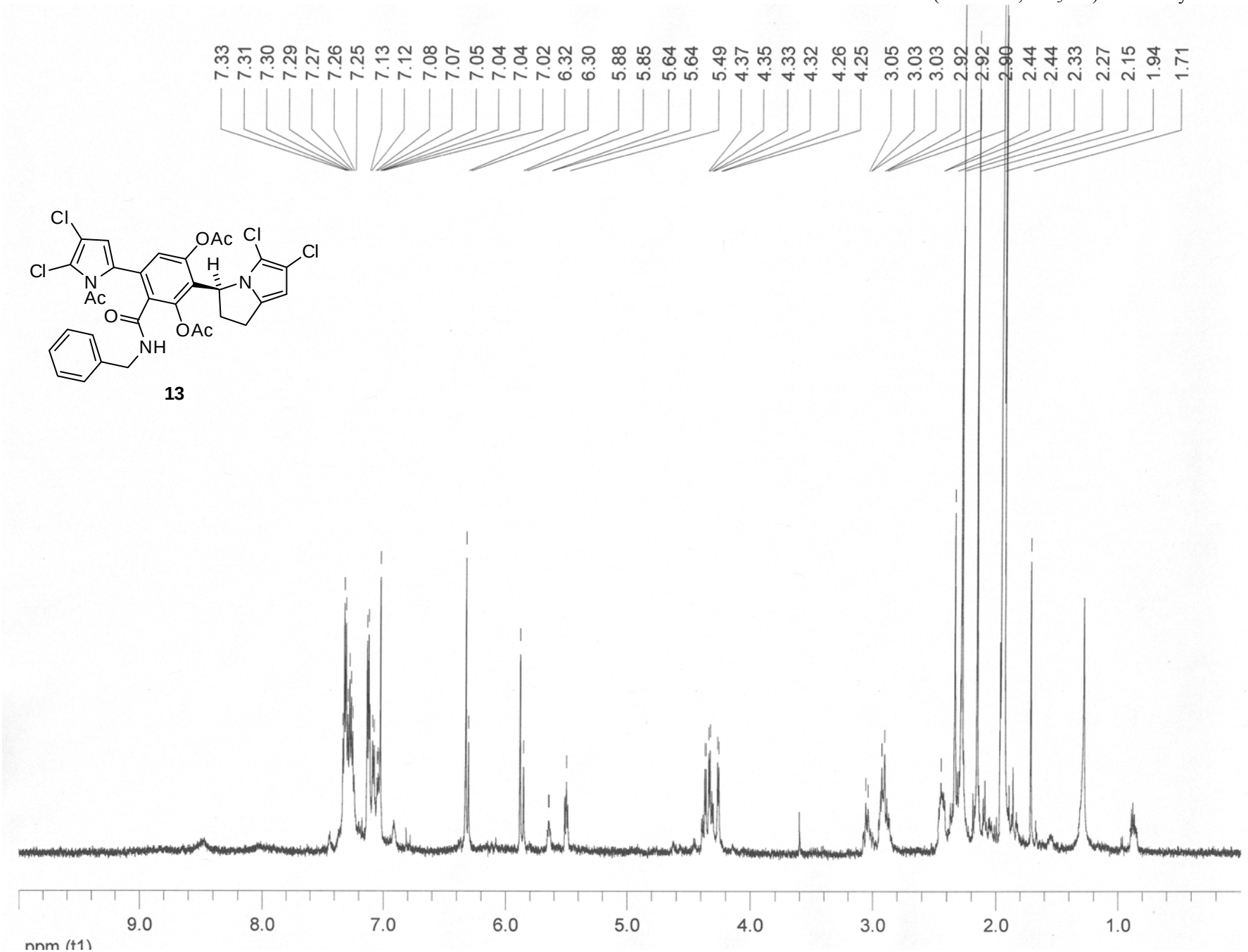
may4thioesterAc2O1P #5285 RT: 17.61 AV: 1 NL: 1.59E6 microAU



12


Polarity/Scan Type: Positive
Sample Name: flch1277
Acq. File: 050511202.wiff
Acq. Date: Thursday, May 05, 2011
+TOF MS: 0.108 to 0.197 min from 050511202.wiff Agilent
Max. 3.4e4 counts.
Intensity, counts
m/z, amu
680.0369
678.0393
682.0337
648.0101
646.0130
681.0385
702.0179
650.0078
700.0223
679.0422
704.0156
683.0353
649.0130
669.9924
703.0212
667.9961
701.0275
647.0158
695.0650
651.0103
671.9895
705.0169
628.1927
638.0266
653.0156
669.0004
662.0240
667.0305
673.9939
685.0330
691.0236
698.0558
707.0176
716.0248
725.0886
12

^{1}H NMR (600 MHz, CD_3CN) of triacetylated amide **13**

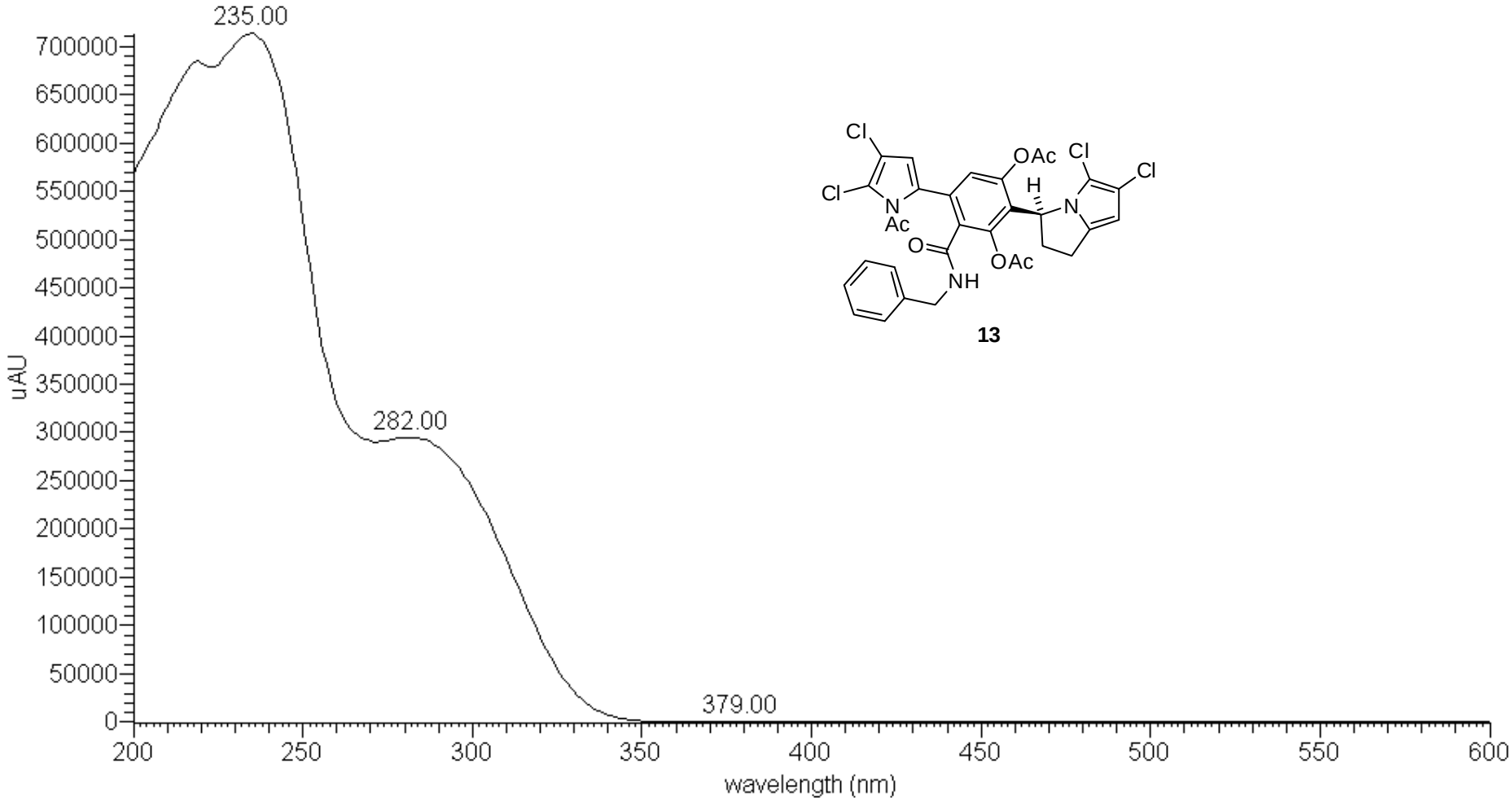


13

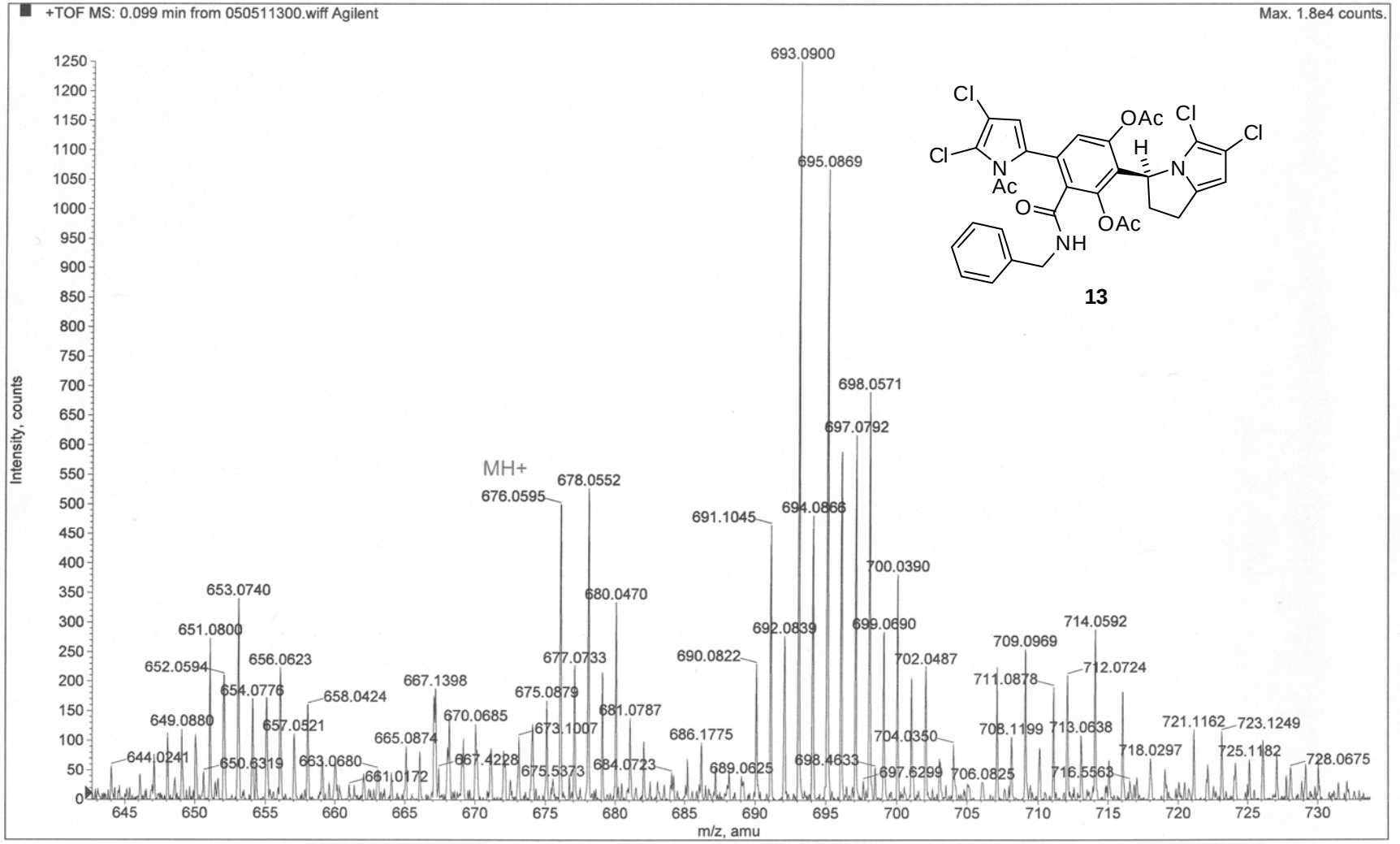
UV/vis spectrum (CH_3CN/water) of triacetylated amide **13**

C:\Xcalibur\data\Chambers\may3amideAc2O1 5/3/2011 9:03:45 AM

may3amideAc2O1 #5523 RT: 18.41 AV: 1 NL: 7.13E5 microAU

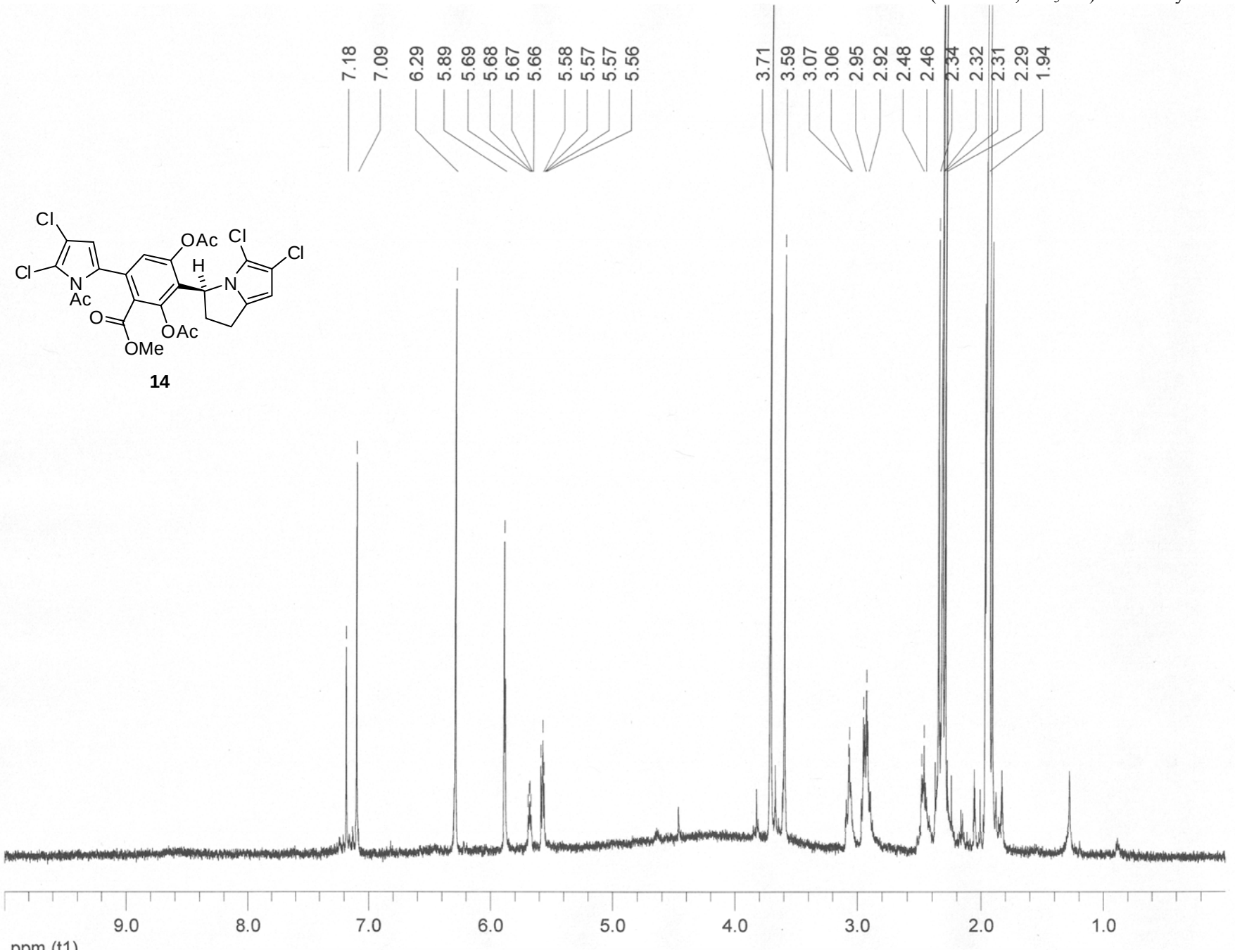


13


Polarity/Scan Type: Positive
Sample Name: flch1280
Acq. File: 050511300.wiff
Acq. Date: Thursday, May 05, 2011
+TOF MS: 0.099 min from 050511300.wiff Agilent
Max. 1.8e4 counts.
Intensity, counts
m/z, amu
MH+
693.0900
695.0869
698.0571
697.0792
678.0552
676.0595
691.1045
694.0866
700.0390
680.0470
653.0740
651.0800
692.0839
699.0690
714.0592
709.0969
656.0623
652.0594
690.0822
702.0487
712.0724
711.0878
667.1398
654.0776
658.0424
677.0733
675.0879
681.0787
649.0880
657.0521
670.0685
673.1007
686.1775
704.0350
721.1162
723.1249
665.0874
708.1199
713.0638
718.0297
725.1182
728.0675
644.0241
650.6319
663.0680
661.0172
667.4228
675.5373
684.0723
689.0625
698.4633
697.6299
706.0825
716.5563
Cl
Cl
N
Ac
OAc
Cl
Cl
H
N
O
OAc
NH

13

^{1}H NMR (600 MHz, CD_3CN) of triacetylated ester **14**

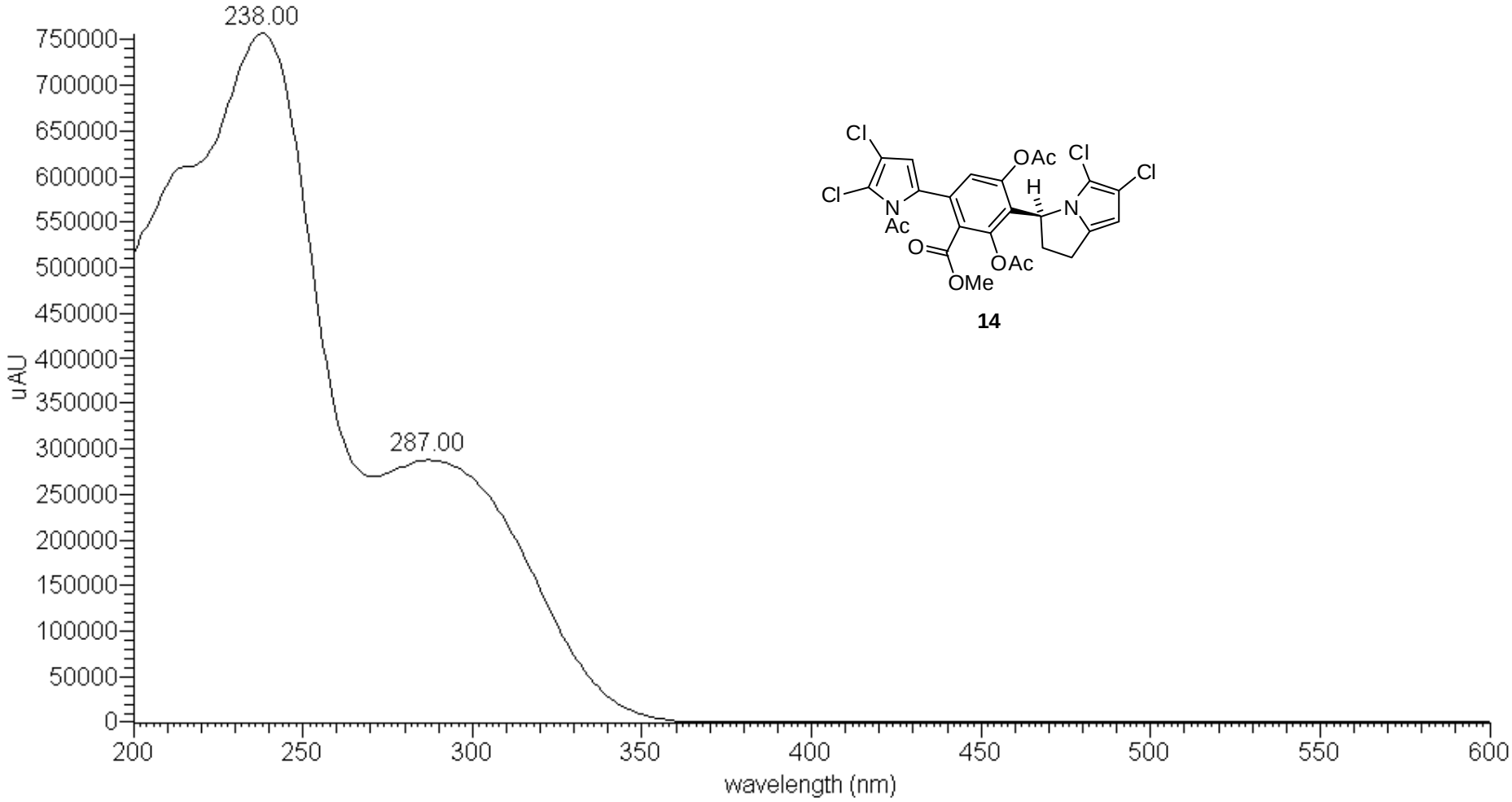


14

UV/vis spectrum (CH_3CN/water) of triacetylated ester **14**

C:\Xcalibur\data\Chambers\may3esterAc2O1 5/3/2011 9:59:34 AM

may3esterAc2O1 #5523 RT: 18.41 AV: 1 NL: 7.56E5 microAU



14

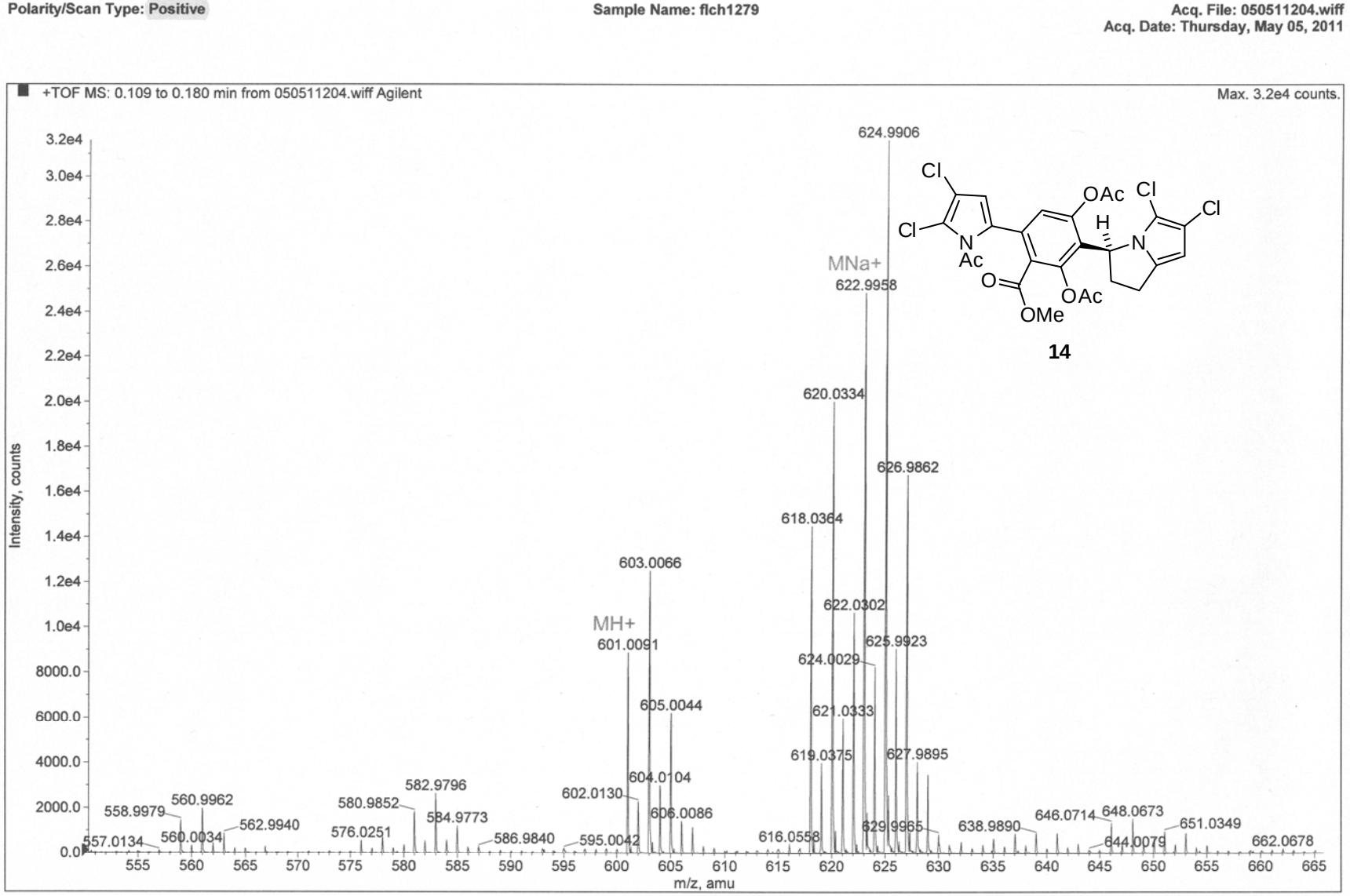

Polarity/Scan Type: Positive
Sample Name: flch1279
Acq. File: 050511204.wiff
Acq. Date: Thursday, May 05, 2011
+TOF MS: 0.109 to 0.180 min from 050511204.wiff Agilent
Max. 3.2e4 counts.
Intensity, counts
m/z, amu
MNa+
MH+
624.9906
622.9958
620.0334
626.9862
618.0364
603.0066
601.0091
622.0302
625.9923
624.0029
605.0044
621.0333
619.0375
627.9895
604.0104
602.0130
606.0086
582.9796
580.9852
584.9773
560.9962
558.9979
562.9940
560.0034
557.0134
576.0251
586.9840
595.0042
616.0558
629.9965
638.9890
646.0714
648.0673
651.0349
644.0079
662.0678
Cl
OAc
H
N
Ac
O
OMe
14

Figure S3. LC/(–)-LRESI-MS chromatogram of chlorizidine (**1**) in pH 10 buffer

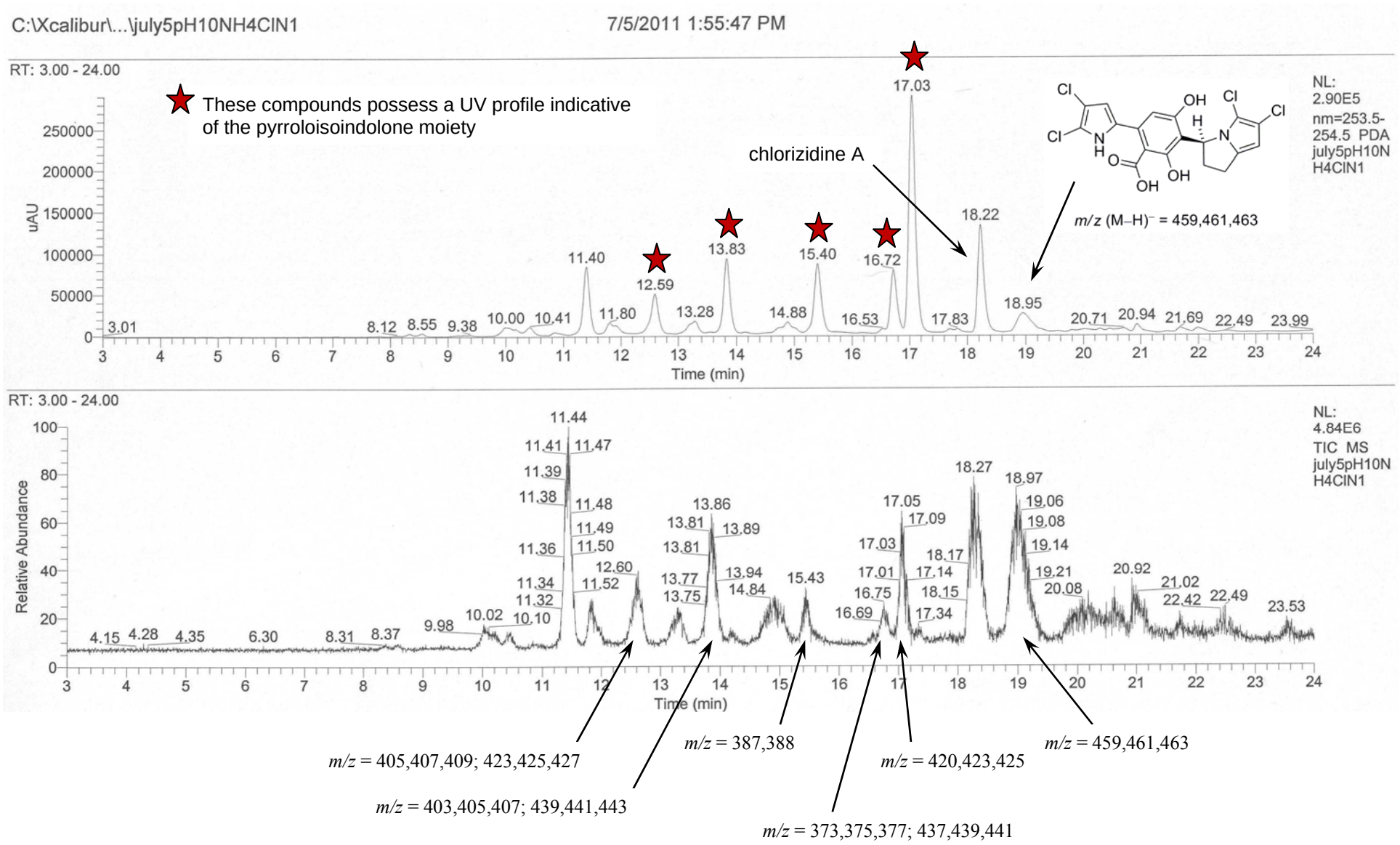


Figure S4. X-Ray structure of chlorizidine A (**1**)

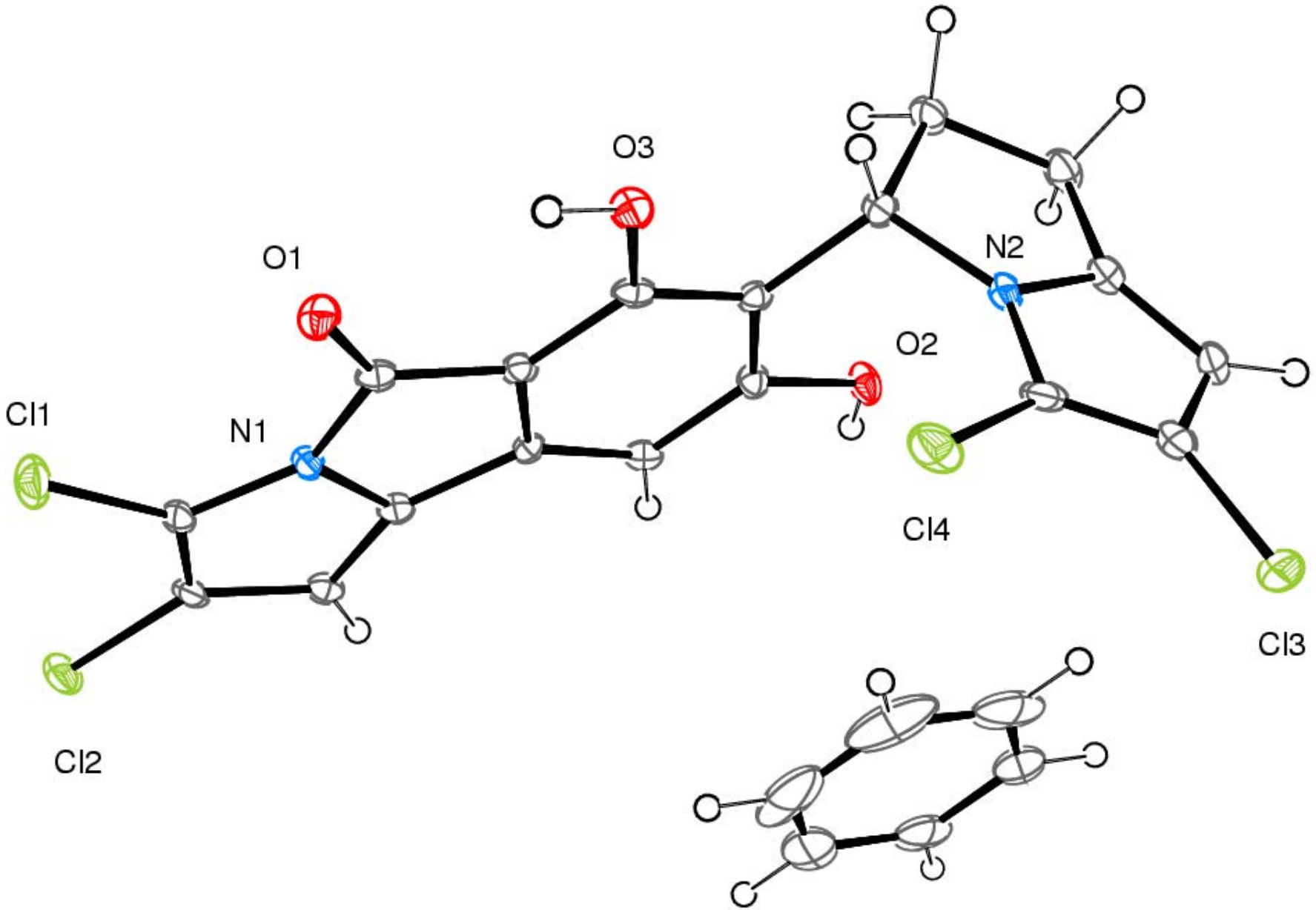
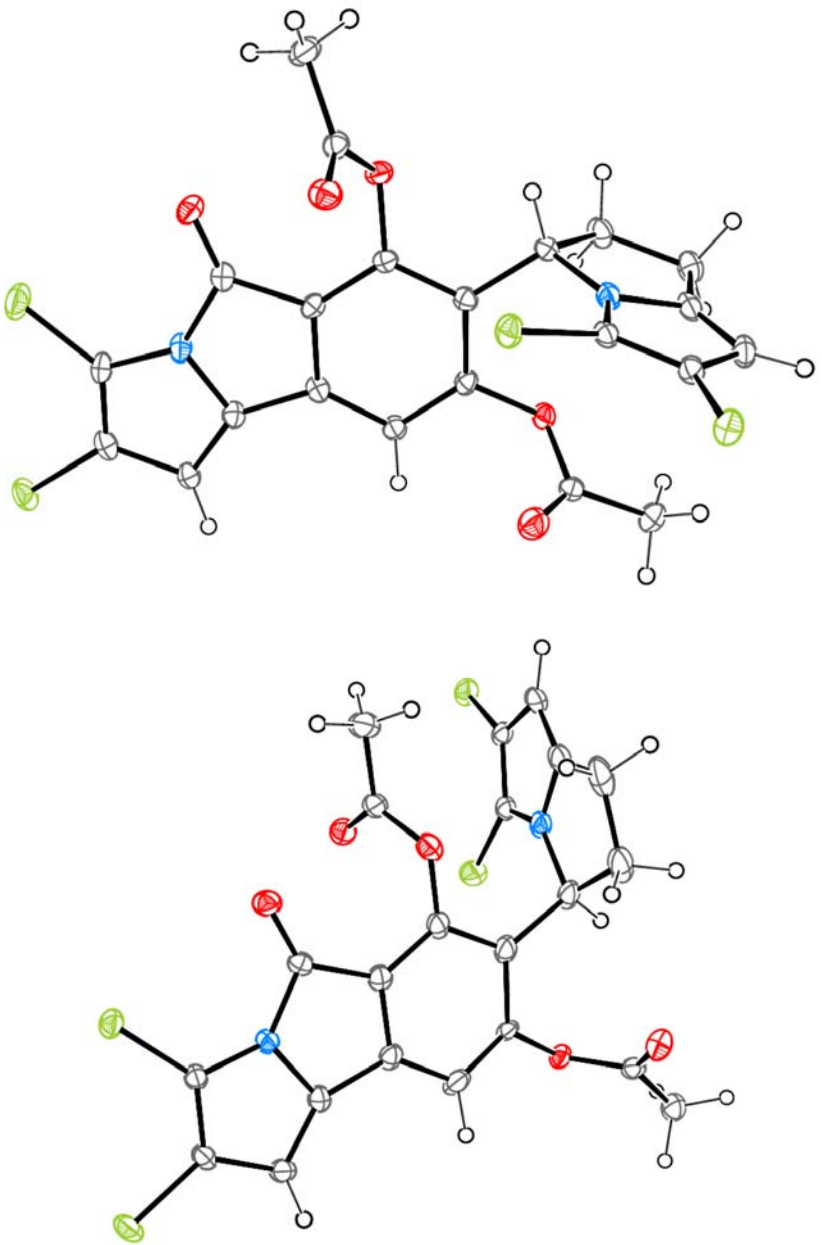


Figure S5. X-Ray structure of diacetyl chlorizidine A (**2**)



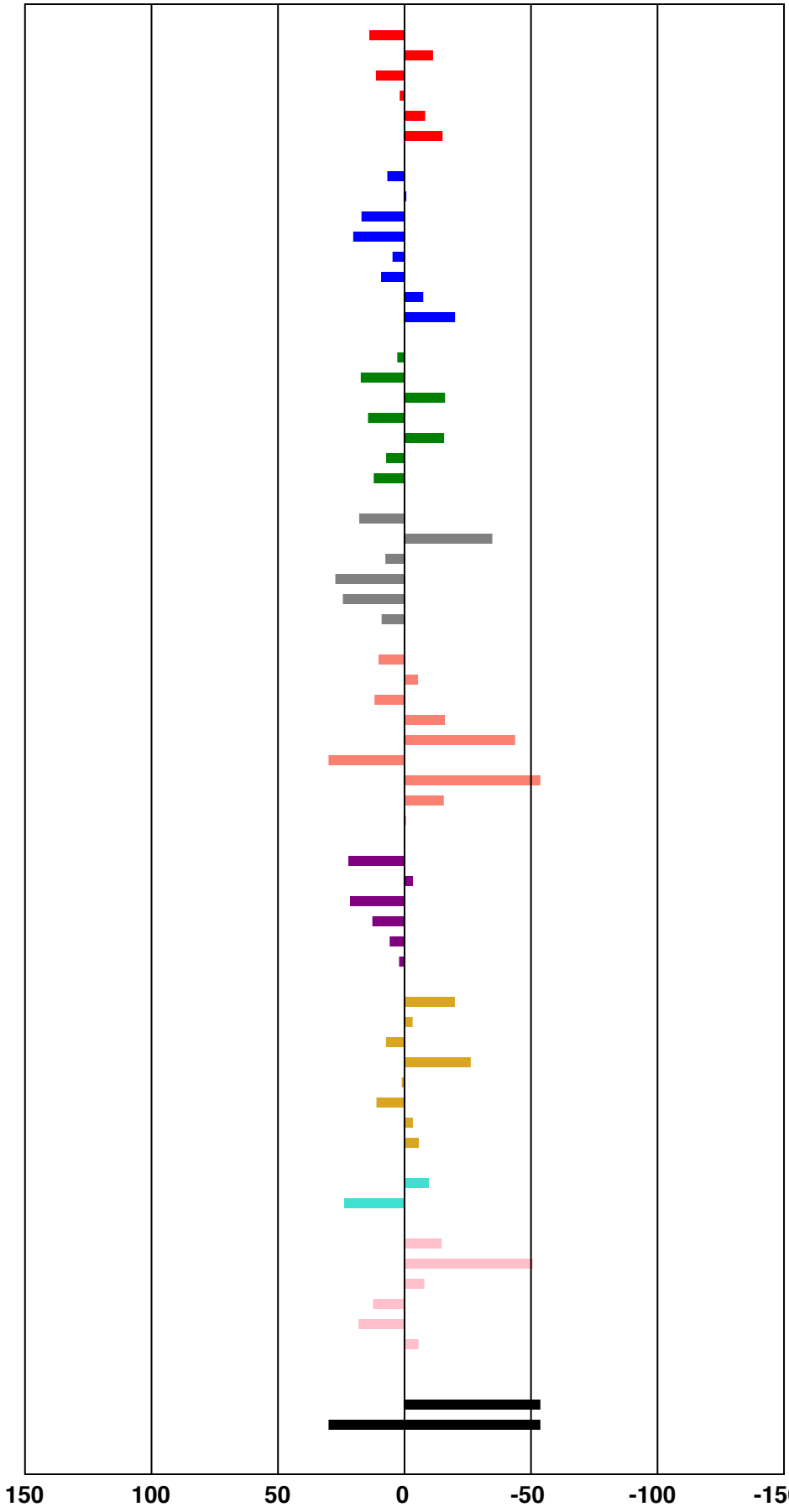
Developmental Therapeutics Program

One Dose Mean Graph

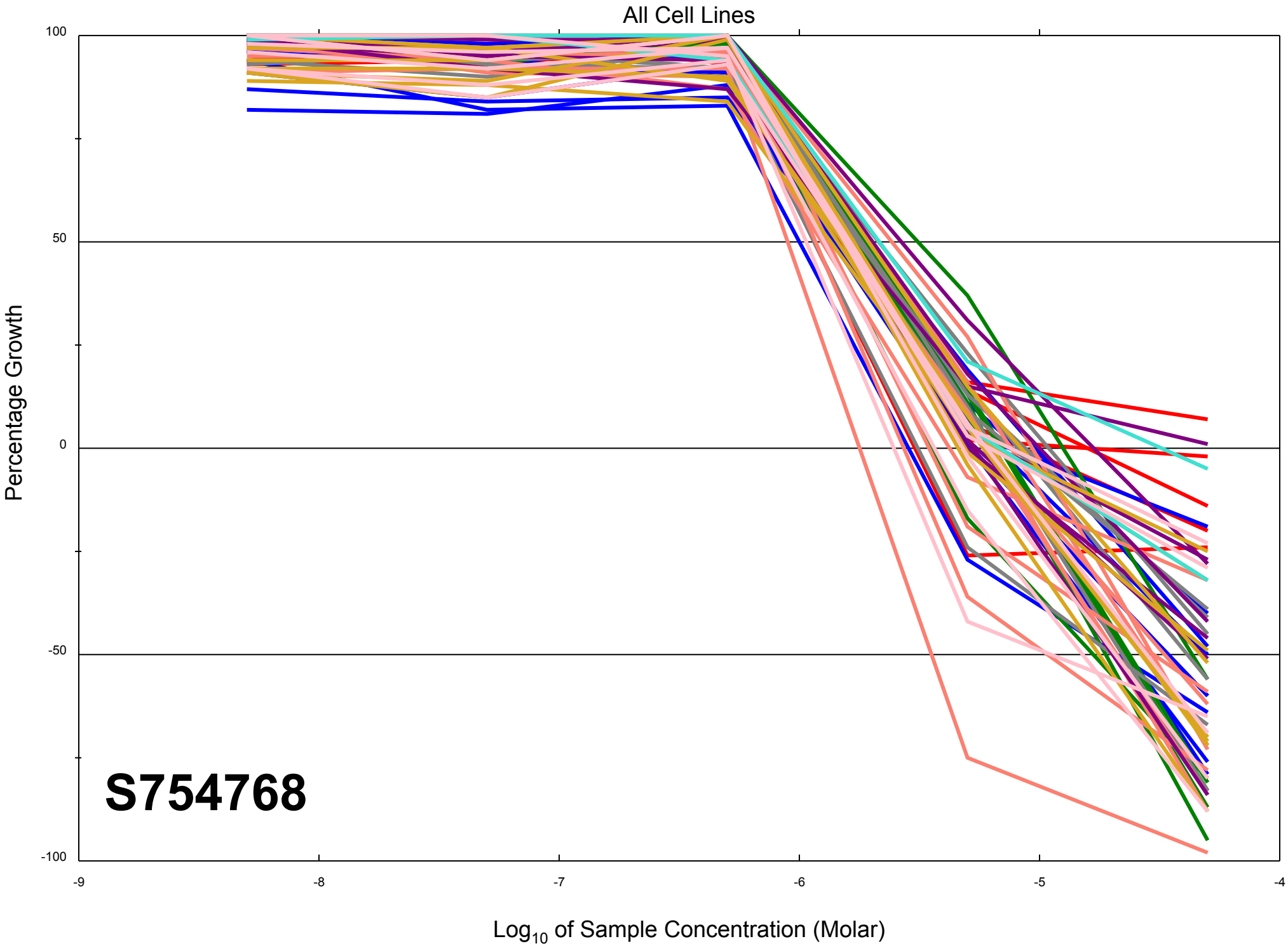
NSC: D-754768 / 1	**Conc:** 1.00E-5 Molar	**Test Date:** Nov 01, 2010
Experiment ID: 1011OS45		**Report Date:** Dec 03, 2010

Panel/Cell Line	Growth Percent
Leukemia	
CCRF-CEM	7.99
HL-60(TB)	-16.56
K-562	5.35
MOLT-4	-4.04
RPMI-8226	-13.44
SR	-20.32
Non-Small Cell Lung Cancer	
A549/ATCC	0.85
EKVX	-6.01
HOP-62	11.07
NCI-H226	14.34
NCI-H23	-1.21
NCI-H322M	3.31
NCI-H460	-12.69
NCI-H522	-25.26
Colon Cancer	
COLO 205	-3.09
HCC-2998	11.34
HCT-116	-21.31
HCT-15	8.48
HT29	-20.94
KM12	1.32
SW-620	6.23
CNS Cancer	
SF-268	11.96
SF-295	-39.98
SF-539	1.62
SNB-19	21.40
SNB-75	18.42
U251	3.11
Melanoma	
LOX IMVI	4.34
MALME-3M	-10.65
M14	5.92
MDA-MB-435	-21.27
SK-MEL-2	-48.99
SK-MEL-28	24.08
SK-MEL-5	-58.99
UACC-257	-20.81
UACC-62	-5.90
Ovarian Cancer	
IGROV1	16.27
OVCAR-3	-8.63
OVCAR-5	15.60
OVCAR-8	6.76
NCI/ADR-RES	-0.07
SK-OV-3	-3.80
Renal Cancer	
786-0	-25.20
A498	-8.48
ACHN	1.37
CAKI-1	-31.46
RXF 393	-4.85
SN12C	5.14
TK-10	-8.60
UO-31	-10.92
Prostate Cancer	
PC-3	-14.91
DU-145	17.92
Breast Cancer	
MCF7	-19.99
MDA-MB-231/ATCC	-55.99
HS 578T	-13.11
BT-549	6.52
T-47D	12.23
MDA-MB-468	-10.81
Mean	-5.61
Delta	53.38
Range	83.07

Mean Growth Percent - Growth Percent



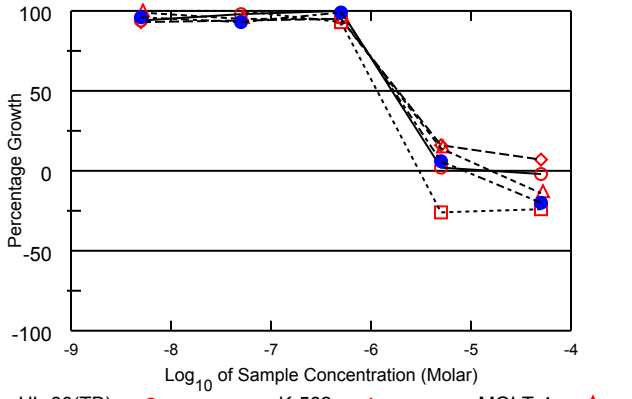
National Cancer Institute Developmental Therapeutics Program Dose Response Curves	NSC : D - 754768/1	SSPL:075T	EXP. ID:1012NS49
	Report Date:February 01, 2011	Test Date:December 06, 2010	



National Cancer Institute Developmental Therapeutics Program
Dose Response Curves

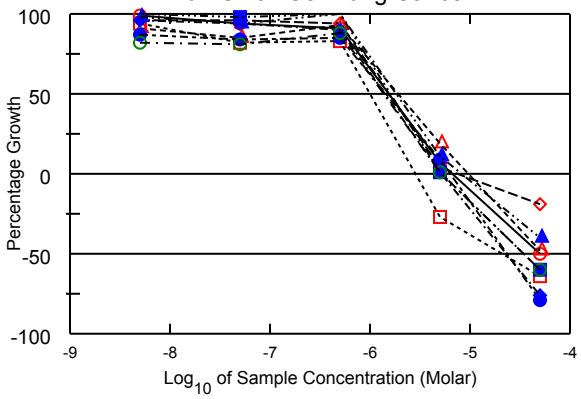
NSC: D - 754768 / 1	SSPL: 075T	EXP. ID: 1012NS49
Report Date: February 01, 2011	Test Date: December 06, 2010	

Leukemia



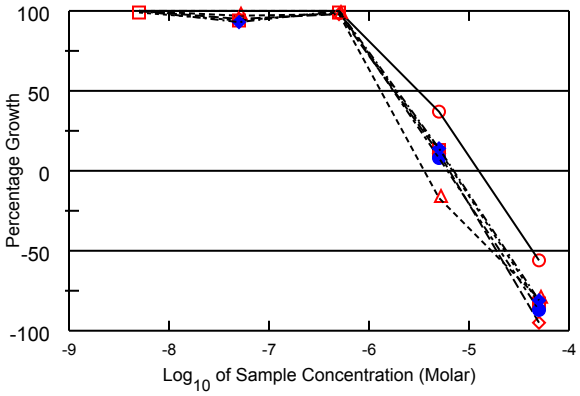
HL-60(TB) K-562 MOLT-4
RPMI-8226 SR

Non-Small Cell Lung Cancer



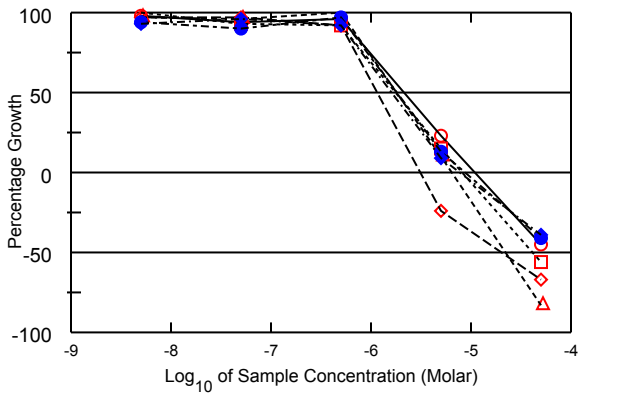
A549/ATCC EKVX HOP-62
HOP-92 NCI-H226 NCI-H23
NCI-H322M NCI-H460 NCI-H522

Colon Cancer



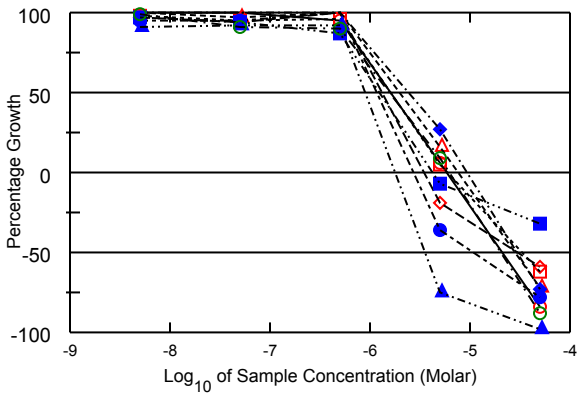
COLO 205 HCC-2998 HCT-116
HCT-15 KM12 SW-620

CNS Cancer



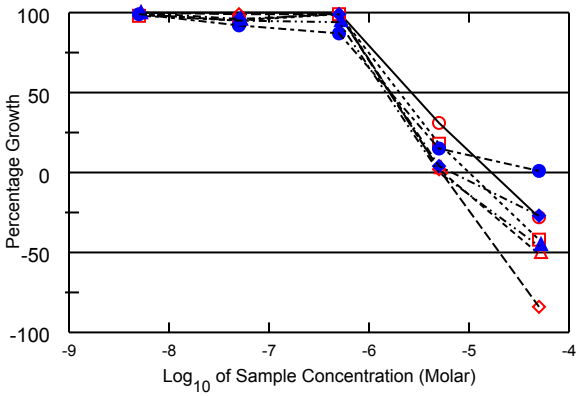
SF-268 SF-295 SF-539
SNB-19 SNB-75 U251

Melanoma



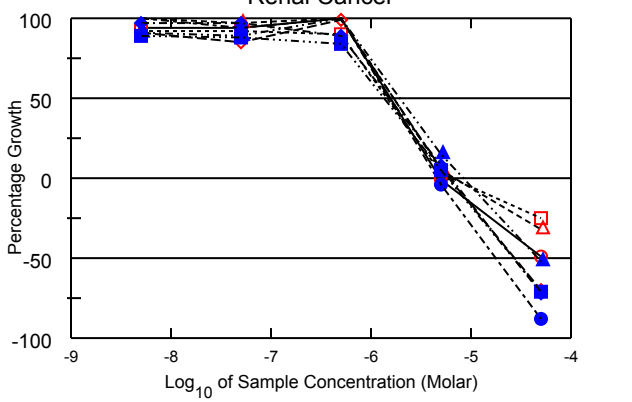
LOX IMVI MALME-3M M14
MDA-MB-435 SK-MEL-2 SK-MEL-28
SK-MEL-5 UACC-257 UACC-62

Ovarian Cancer



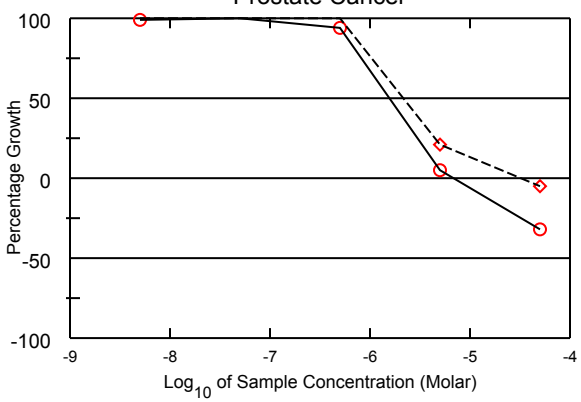
IGROV1 OVCAR-3 OVCAR-4
OVCAR-5 OVCAR-8 NCI/ADR-RES
SK-OV-3

Renal Cancer



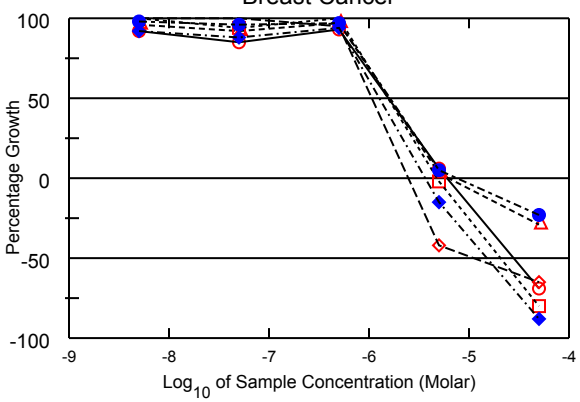
786-0 A498 ACHN
CAKI-1 RXF 393 SN12C
TK-10 UO-31

Prostate Cancer



PC-3 DU-145

Breast Cancer



MCF7 MDA-MB-231/ HS 578T
BT-549 T-47D MDA-MB-468

National Cancer Institute Developmental Therapeutics Program
In-Vitro Testing Results

NSC : D - 754768 / 1	Experiment ID : 1012NS49	Test Type : 08	Units : Molar
Report Date : February 01, 2011	Test Date : December 06, 2010	QNS :	MC :
COMI : CNH287.528_SS_XM (100117)	Stain Reagent : SRB Dual-Pass Related	SSPL : 075T	

			Log10 Concentration												
	Time		Mean Optical Densities					Percent Growth							
Panel/Cell Line	Zero	Ctrl	-8.3	-7.3	-6.3	-5.3	-4.3	-8.3	-7.3	-6.3	-5.3	-4.3	GI50	TGI	LC50
Leukemia															
HL-60(TB)	0.986	2.925	2.818	2.883	2.971	1.031	0.965	94	98	102	2	-2	1.67E-6	1.65E-5	> 5.00E-5
K-562	0.187	1.718	1.603	1.620	1.648	0.437	0.291	93	94	95	16	7	1.88E-6	> 5.00E-5	> 5.00E-5
MOLT-4	0.707	2.625	2.613	2.528	2.534	0.972	0.607	99	95	95	14	-14	1.80E-6	1.56E-5	> 5.00E-5
RPMI-8226	0.773	2.231	2.265	2.225	2.125	0.571	0.590	102	100	93	-26	-24	1.14E-6	3.01E-6	> 5.00E-5
SR	0.593	2.320	2.252	2.200	2.296	0.693	0.474	96	93	99	6	-20	1.67E-6	8.36E-6	> 5.00E-5
Non-Small Cell Lung Cancer															
A549/ATCC	0.415	2.088	2.068	1.982	1.931	0.538	0.210	99	94	91	7	-50	1.54E-6	6.73E-6	> 5.00E-5
EKVX	0.717	1.904	1.862	1.856	1.838	0.769	0.583	96	96	94	4	-19	1.56E-6	7.72E-6	> 5.00E-5
HOP-62	0.386	0.898	0.852	0.819	0.863	0.485	0.200	91	85	93	19	-48	1.92E-6	9.64E-6	> 5.00E-5
HOP-92	1.057	1.512	1.485	1.431	1.433	0.768	0.376	94	82	83	-27	-64	9.92E-7	2.82E-6	2.04E-5
NCI-H226	0.508	1.261	1.160	1.138	1.152	0.573	0.109	87	84	85	9	-79	1.45E-6	6.28E-6	2.35E-5
NCI-H23	0.483	1.631	1.590	1.557	1.521	0.498	0.117	96	94	90	1	-76	1.42E-6	5.20E-6	2.31E-5
NCI-H322M	0.767	1.697	1.679	1.641	1.718	0.870	0.463	98	94	102	11	-40	1.87E-6	8.26E-6	> 5.00E-5
NCI-H460	0.228	2.072	2.071	2.041	2.118	0.253	0.091	100	98	103	1	-60	1.65E-6	5.26E-6	3.43E-5
NCI-H522	0.614	1.548	1.385	1.373	1.434	0.620	0.247	82	81	88	1	-60	1.36E-6	5.12E-6	3.44E-5
Colon Cancer															
COLO 205	0.257	0.965	1.003	0.963	0.967	0.517	0.113	105	100	100	37	-56	3.09E-6	1.24E-5	4.29E-5
HCC-2998	0.948	2.808	2.845	2.722	2.763	1.152	0.049	102	95	98	11	-95	1.77E-6	6.34E-6	1.88E-5
HCT-116	0.267	1.841	1.879	1.796	1.810	0.222	0.054	102	97	98	-17	-80	1.31E-6	3.57E-6	1.68E-5
HCT-15	0.265	1.639	1.629	1.556	1.623	0.446	0.046	99	94	99	13	-83	1.86E-6	6.86E-6	2.28E-5
KM12	0.302	1.529	1.549	1.541	1.544	0.402	0.040	102	101	101	8	-87	1.77E-6	6.09E-6	2.05E-5
SW-620	0.186	1.193	1.197	1.128	1.194	0.327	0.036	100	93	100	14	-81	1.91E-6	7.02E-6	2.36E-5
CNS Cancer															
SF-268	0.379	1.389	1.366	1.327	1.352	0.609	0.210	98	94	96	23	-45	2.13E-6	1.09E-5	> 5.00E-5
SF-295	0.884	2.778	2.728	2.727	2.631	0.673	0.288	97	97	92	-24	-67	1.16E-6	3.11E-6	1.99E-5
SF-539	0.634	1.706	1.669	1.658	1.718	0.737	0.107	97	96	101	10	-83	1.81E-6	6.35E-6	2.20E-5
SNB-19	0.342	1.292	1.317	1.226	1.215	0.480	0.149	103	93	92	15	-56	1.74E-6	8.01E-6	4.06E-5
SNB-75	0.585	1.133	1.099	1.076	1.118	0.657	0.346	94	90	97	13	-41	1.83E-6	8.76E-6	> 5.00E-5
U251	0.264	1.416	1.341	1.373	1.325	0.366	0.160	93	96	92	9	-39	1.60E-6	7.62E-6	> 5.00E-5
Melanoma															
LOX IMVI	0.172	1.225	1.236	1.223	1.168	0.235	0.027	101	100	95	6	-84	1.59E-6	5.82E-6	2.08E-5
MALME-3M	0.593	1.019	1.005	0.992	1.021	0.478	0.244	97	94	100	-19	-59	1.32E-6	3.44E-6	2.98E-5
M14	0.304	1.075	1.105	1.050	1.113	0.426	0.085	104	97	105	16	-72	2.06E-6	7.55E-6	2.80E-5
MDA-MB-435	0.401	1.850	1.819	1.776	1.793	0.474	0.153	98	95	96	5	-62	1.60E-6	5.95E-6	3.33E-5
SK-MEL-2	0.896	1.739	1.694	1.700	1.790	0.573	0.193	95	95	106	-36	-78	1.24E-6	2.79E-6	1.06E-5
SK-MEL-28	0.383	0.891	0.914	0.896	0.948	0.519	0.102	104	101	111	27	-73	2.65E-6	9.25E-6	2.91E-5
SK-MEL-5	0.464	2.591	2.402	2.424	2.420	0.117	0.010	91	92	92	-75	-98	8.93E-7	1.78E-6	3.55E-6
UACC-257	0.669	1.647	1.619	1.585	1.523	0.621	0.456	97	94	87	-7	-32	1.24E-6	4.20E-6	> 5.00E-5
UACC-62	0.591	2.068	2.058	1.935	1.921	0.728	0.074	99	91	90	9	-88	1.57E-6	6.23E-6	2.05E-5
Ovarian Cancer															
IGROV1	0.575	1.727	1.778	1.746	1.782	0.935	0.412	104	102	105	31	-28	2.78E-6	1.67E-5	> 5.00E-5
OVCAR-3	0.341	1.026	1.038	1.021	1.016	0.357	0.056	102	99	99	2	-84	1.60E-6	5.31E-6	2.03E-5
OVCAR-4	0.589	1.033	1.029	1.009	1.102	0.600	0.288	99	95	116	2	-51	1.90E-6	5.56E-6	4.75E-5
OVCAR-5	0.463	1.188	1.171	1.149	1.178	0.593	0.271	98	95	99	18	-42	2.00E-6	1.00E-5	> 5.00E-5
OVCAR-8	0.122	0.590	0.583	0.554	0.528	0.190	0.126	99	92	87	15	1	1.62E-6	> 5.00E-5	> 5.00E-5
NCI/ADR-RES	0.406	1.497	1.504	1.455	1.486	0.446	0.298	101	96	99	4	-27	1.63E-6	6.59E-6	> 5.00E-5
SK-OV-3	0.639	1.561	1.548	1.515	1.503	0.643	0.348	99	95	94	.	-46	1.47E-6	5.11E-6	> 5.00E-5
Renal Cancer															
786-0	0.661	2.073	1.990	1.989	2.111	0.656	0.337	94	94	103	-1	-49	1.62E-6	4.92E-6	> 5.00E-5
A498	0.498	0.987	0.944	0.916	0.981	0.527	0.148	91	85	99	6	-70	1.67E-6	5.96E-6	2.71E-5
ACHN	0.497	1.852	1.907	1.817	1.881	0.560	0.337	104	97	102	5	-32	1.71E-6	6.67E-6	> 5.00E-5
CAKI-1	0.716	2.470	2.329	2.333	2.293	0.763	0.541	92	92	90	3	-25	1.43E-6	6.26E-6	> 5.00E-5
RXF 393	0.655	1.164	1.164	1.132	1.161	0.632	0.082	100	94	100	-4	-88	1.51E-6	4.62E-6	1.79E-5
SN12C	0.398	1.599	1.567	1.569	1.467	0.491	0.112	97	97	89	8	-72	1.51E-6	6.25E-6	2.65E-5
TK-10	0.548	1.229	1.166	1.154	1.314	0.654	0.266	91	89	112	15	-52	2.20E-6	8.51E-6	4.74E-5
UO-31	0.615	1.501	1.406	1.392	1.361	0.657	0.177	89	88	84	5	-71	1.35E-6	5.76E-6	2.62E-5
Prostate Cancer															
PC-3	0.451	1.716	1.698	1.722	1.637	0.515	0.305	99	100	94	5	-32	1.56E-6	6.80E-6	> 5.00E-5
DU-145	0.241	0.936	0.934	0.957	0.960	0.386	0.230	100	103	103	21	-5	2.22E-6	3.26E-5	> 5.00E-5
Breast Cancer															
MCF7	0.284	1.200	1.127	1.066	1.136	0.338	0.087	92	85	93	6	-69	1.56E-6	5.99E-6	2.76E-5
MDA-MB-231/ATCC	0.430	1.041	1.045	1.055	1.011	0.250	0.152	101	102	95	-42	-65	1.07E-6	2.47E-6	1.13E-5
HS 578T	0.812	1.671	1.639	1.602	1.642	0.834	0.574	96	92	97	3	-29	1.56E-6	5.99E-6	> 5.00E-5
BT-549	0.726	1.283	1.299	1.249	1.323	0.709	0.149	103	94	107	-2	-80	1.66E-6	4.76E-6	2.07E-5
T-47D	0.647	1.347	1.336	1.321	1.327	0.679	0.501	98	96	97	5	-23	1.62E-6	7.36E-6	> 5.00E-5
MDA-MB-468	0.575	1.135	1.088	1.069	1.100	0.491	0.069	92	88	94	-15	-88	1.27E-6	3.66E-6	1.51E-5

National Cancer Institute Developmental Therapeutics Program

Mean Graphs

NSC : D - 754768/1 | Units :Molar | SSPL :075T | EXP. ID :1012NS49

Report Date :February 01, 2011 | Test Date :December 06, 2010

Panel/Cell Line	Log_{10}GI50	Log_{10}TGI	Log_{10}LC50
Leukemia			
HL-60(TB)	-5.78	-4.78	> -4.30
K-562	-5.73	> -4.30	> -4.30
MOLT-4	-5.75	-4.81	> -4.30
RPMI-8226	-5.94	-5.52	> -4.30
SR	-5.78	-5.08	> -4.30
Non-Small Cell Lung Cancer			
A549/ATCC	-5.81	-5.17	> -4.30
EKVX	-5.81	-5.11	> -4.30
HOP-62	-5.72	-5.02	> -4.30
HOP-92	-6.00	-5.55	-4.69
NCI-H226	-5.84	-5.20	-4.63
NCI-H23	-5.85	-5.28	-4.64
NCI-H322M	-5.73	-5.08	> -4.30
NCI-H460	-5.78	-5.28	-4.47
NCI-H522	-5.87	-5.29	-4.46
Colon Cancer			
COLO 205	-5.51	-4.91	-4.37
HCC-2998	-5.75	-5.20	-4.72
HCT-116	-5.88	-5.45	-4.78
HCT-15	-5.73	-5.16	-4.64
KM12	-5.75	-5.22	-4.69
SW-620	-5.72	-5.15	-4.63
CNS Cancer			
SF-268	-5.67	-4.96	> -4.30
SF-295	-5.94	-5.51	-4.70
SF-539	-5.74	-5.20	-4.66
SNB-19	-5.76	-5.10	-4.39
SNB-75	-5.74	-5.06	> -4.30
U251	-5.80	-5.12	> -4.30
Melanoma			
LOX IMVI	-5.80	-5.23	-4.68
MALME-3M	-5.88	-5.46	-4.53
M14	-5.69	-5.12	-4.55
MDA-MB-435	-5.80	-5.23	-4.48
SK-MEL-2	-5.91	-5.56	-4.97
SK-MEL-28	-5.58	-5.03	-4.54
SK-MEL-5	-6.05	-5.75	-5.45
UACC-257	-5.91	-5.38	> -4.30
UACC-62	-5.81	-5.21	-4.69
Ovarian Cancer			
IGROV1	-5.56	-4.78	> -4.30
OVCAR-3	-5.80	-5.27	-4.69
OVCAR-4	-5.72	-5.25	-4.32
OVCAR-5	-5.70	-5.00	> -4.30
OVCAR-8	-5.79	> -4.30	> -4.30
NCI/ADR-RES	-5.79	-5.18	> -4.30
SK-OV-3	-5.83	-5.29	> -4.30
Renal Cancer			
786-0	-5.79	-5.31	> -4.30
A498	-5.78	-5.22	-4.57
ACHN	-5.77	-5.18	> -4.30
CAKI-1	-5.84	-5.20	> -4.30
RXF 393	-5.82	-5.34	-4.75
SN12C	-5.82	-5.20	-4.58
TK-10	-5.66	-5.07	-4.32
UO-31	-5.87	-5.24	-4.58
Prostate Cancer			
PC-3	-5.81	-5.17	> -4.30
DU-145	-5.65	-4.49	> -4.30
Breast Cancer			
MCF7	-5.81	-5.22	-4.56
MDA-MB-231/ATCC	-5.97	-5.61	-4.95
HS 578T	-5.81	-5.22	> -4.30
BT-549	-5.78	-5.32	-4.68
T-47D	-5.79	-5.13	> -4.30
MDA-MB-468	-5.90	-5.44	-4.82
_MID	-5.79	-5.17	-4.49
Delta	0.26	0.58	0.96
Range	0.54	1.45	1.15