Supplementary Information

Synthesis and Structure-activity Relationship Studies of Benzimidazole-4,7-dione-based P2X3 Receptor Antagonists as Novel Anti-nociceptive Agents

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** Previously reported compounds, 10a-10b[18], 11a-11b[17] were reported by mass spectroscopy data according to the Instructions for Authors of MDPI.

6-chloro-5-((4-fluorophenyl)amino)-2-methyl-1H-benzo[d]imidazole-4,7-dione (10a)[18] Following the general procedure for the synthesis of **10a-b**, the substitution reaction of **9a** with 4-fluoroaniline afforded **10a**. red purple powder. Yield 67%; ^{1}H NMR (400 MHz, DMSO-d6) δ ppm 2.32 (s, 4 H) 7.03 - 7.16 (m, 4 H); LC/MS (ESI, m/z) 303.7 $[M - H]^-$ 305.8 $[M + H]^+$.

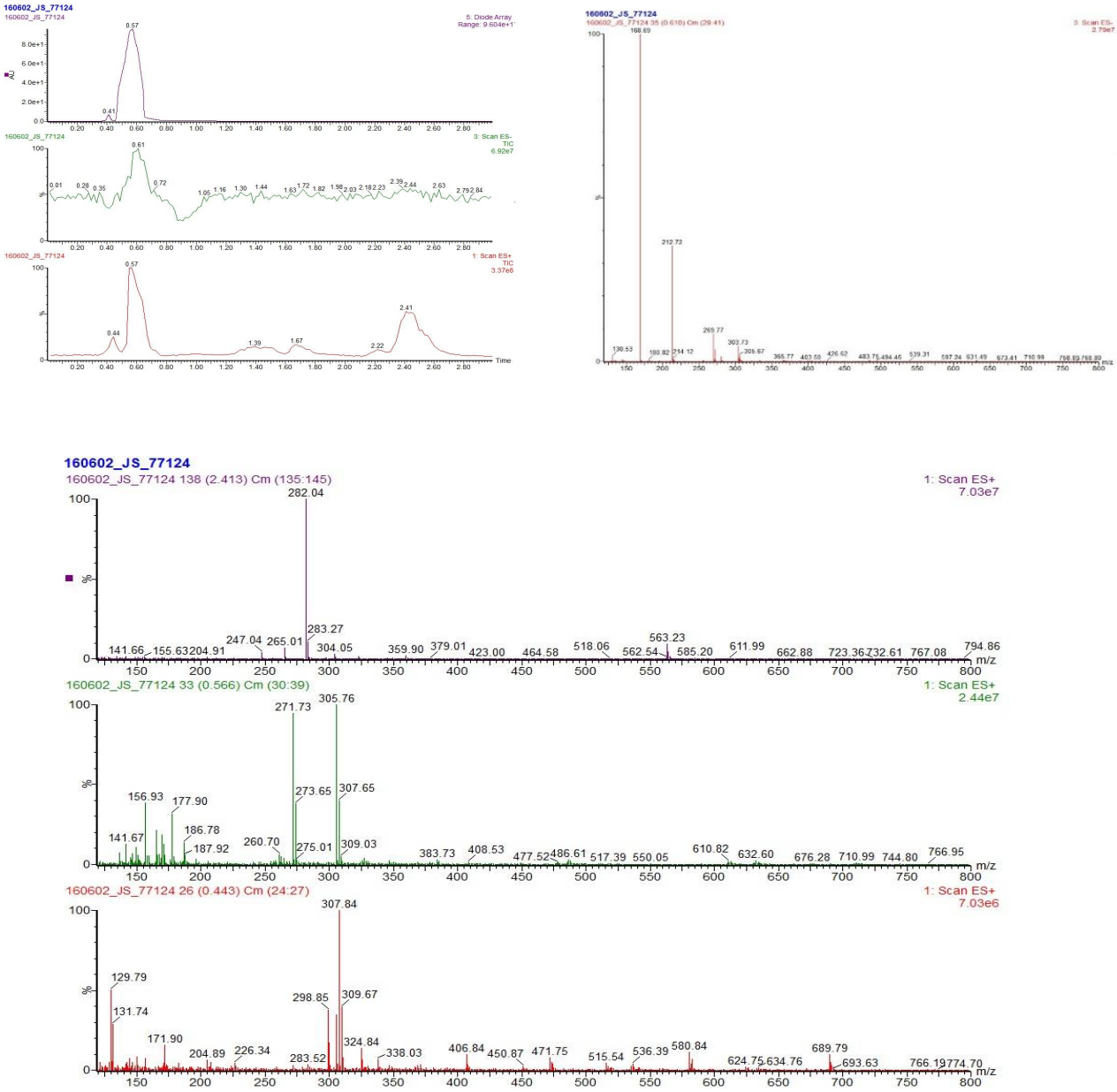


Figure S1. LC/MS spectrum of compound 10a

6-chloro-5-((4-chlorophenyl)amino)-2-methyl-1H-benzo[d]imidazole-4,7-dione (10b)[18] Following the general procedure for the synthesis of **10a-b**, the substitution reaction of **9a** with 4-chloroaniline afforded **10b**. red purple powder. Yield 71%; ^{1}H NMR (400 MHz, DMSO-d6) δ ppm 13.6 (s, 1H, NH), 9.0 (s, 1H, NH), 7.0–7.3 (dd, J = 2.4, 8.4 Hz, 4H, Ph-H), 2.2 (s, 3H, CH3); LC/MS (ESI, m/z) 319.8 $[M - H]^-$ 321.8 $[M + H]^+$.

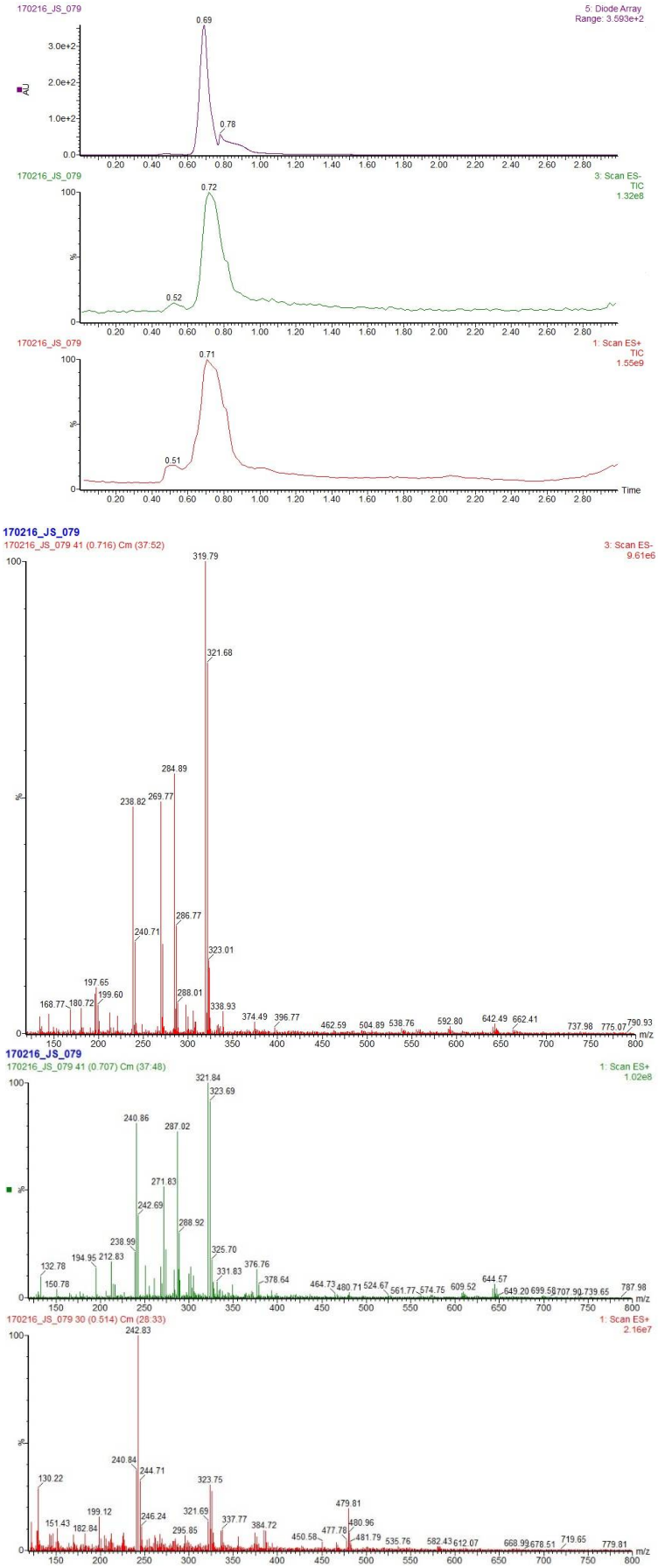


Figure S2. LC/MS spectrum of compound 10b

6-chloro-5-((4-fluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11a)[17] Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-fluoroaniline afforded **11a**. purple powder. Yield 41%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 6.98 - 7.13 (m, 4 H); LC/MS (ESI, m/z) 358.3 $[M - H]^-$ 360.0 $[M + H]^+$.

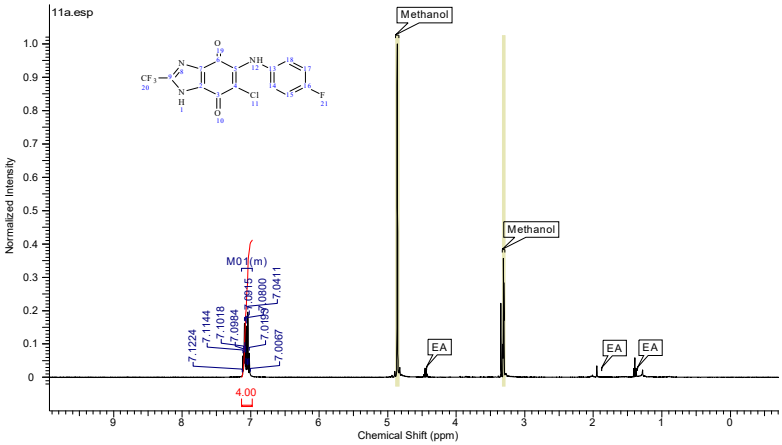


Figure S3. 1D 1H NMR spectrum of compound 11a

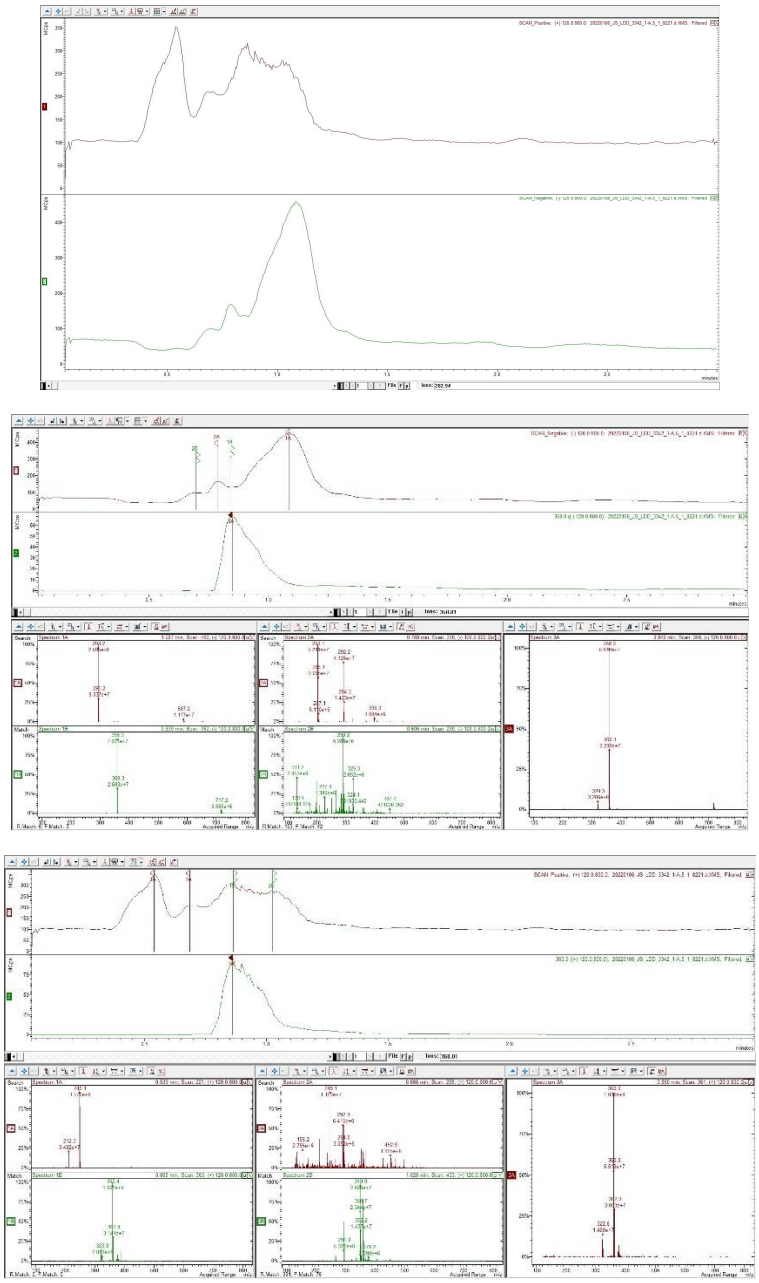


Figure S4. LC/MS spectrum of compound 11a

6-chloro-5-((4-chlorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11b)[17] Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-chloroaniline afforded **11b**. purple powder. Yield 56%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.01 - 7.10 (m, 2 H) 7.23 - 7.31 (m, 2 H); LC/MS (ESI, m/z) 374.3 $[M - H]^-$ 375.8 $[M + H]^+$.

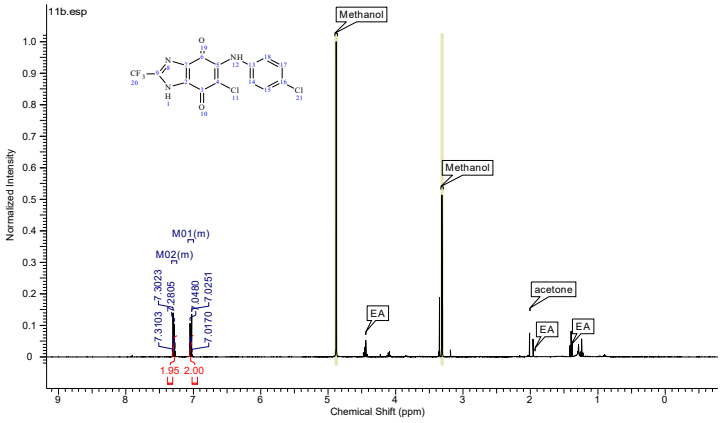


Figure S5. 1D 1H NMR spectrum of compound 11b

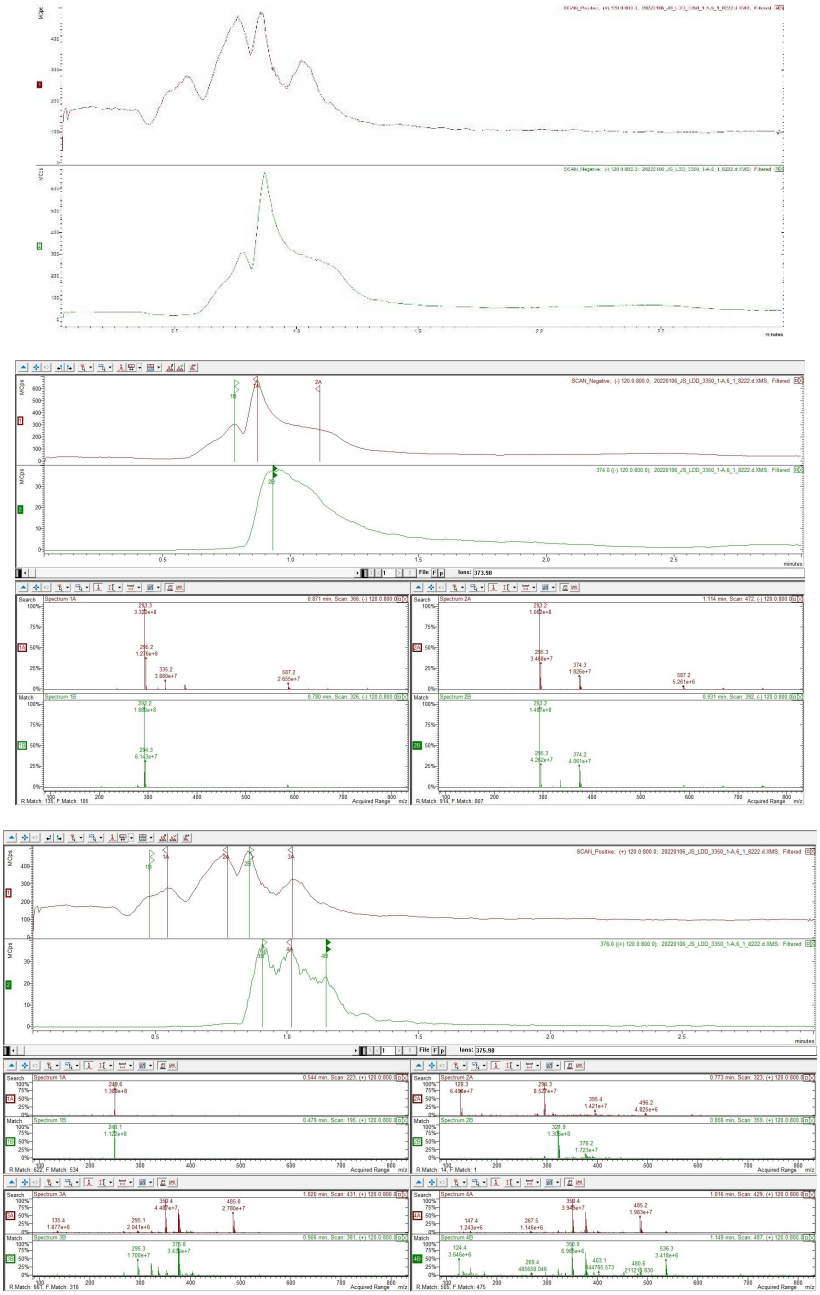


Figure S6. LC/MS spectrum of compound 11b

6-chloro-5-((3,4-difluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11c) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 3,4-difluoroaniline afforded **11c**. dark red powder. **11c** fully dissolved in 600 μL of the acetone-d6 and few drops of methanol. Yield 31%; ^{1}H NMR (400 MHz, ACETONE-d6) δ ppm 7.00 - 7.07 (m, 1 H) 7.20 (ddd, J=11.91, 7.33, 2.75 Hz, 1 H) 7.24 - 7.34 (m, 1 H) 8.51 (s, 1 H); ^{13}C NMR (100 MHz, ACETONE-d6) δ 174.18, 173.99, 142.43, 141.55, 137.99, 135.97, 135.86, 121.28, 120.73, 118.60, 116.57, 116.39, 113.53, 113.33, 111.59, 145~155 ppm peaks are considered as impurities; LC/MS (ESI, m/z) 376.3 $[M - H]^-$ 377.1 $[M + H]^+$.

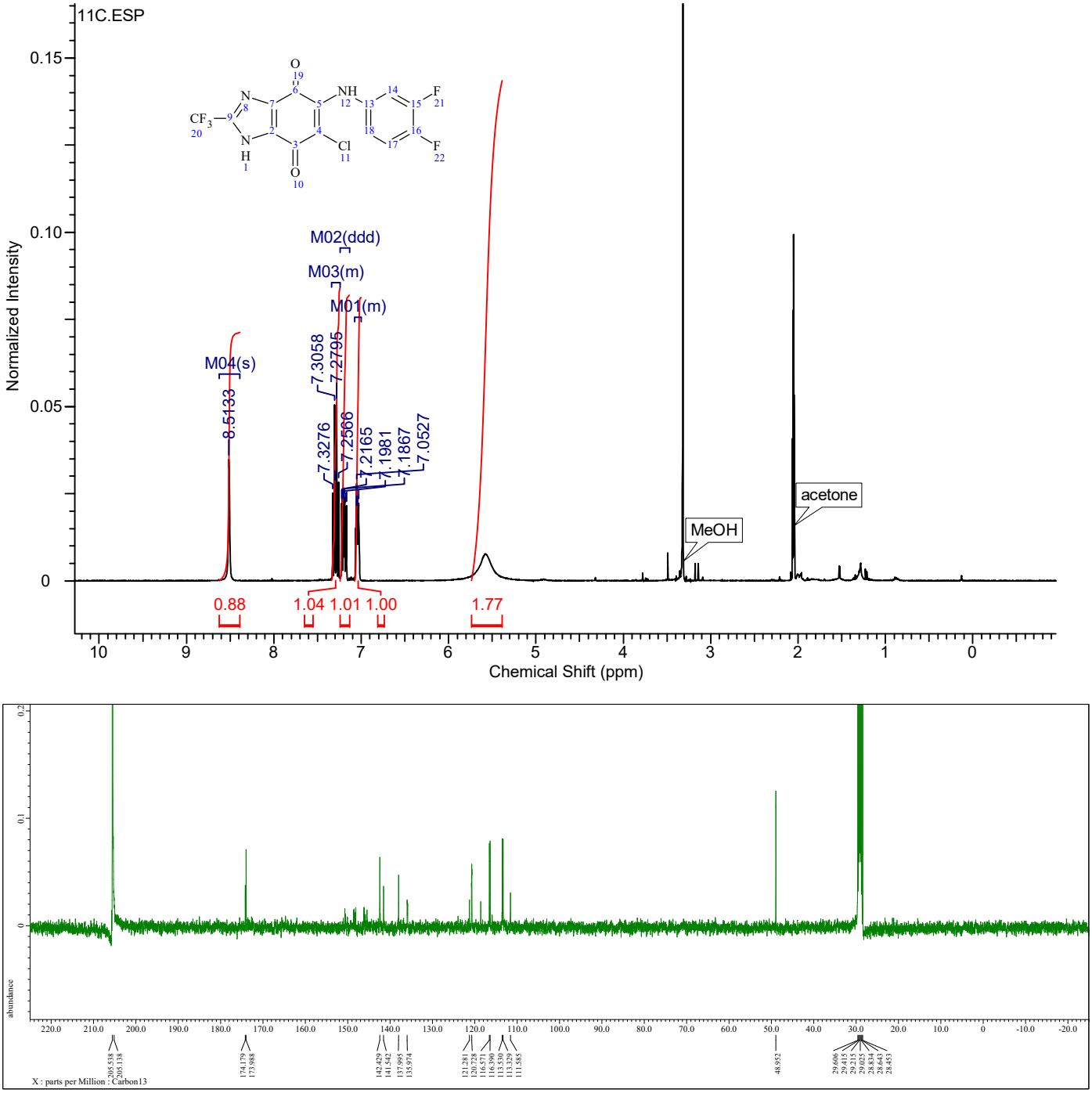


Figure S7. 1D 1H and 13C NMR spectrum of compound 11c

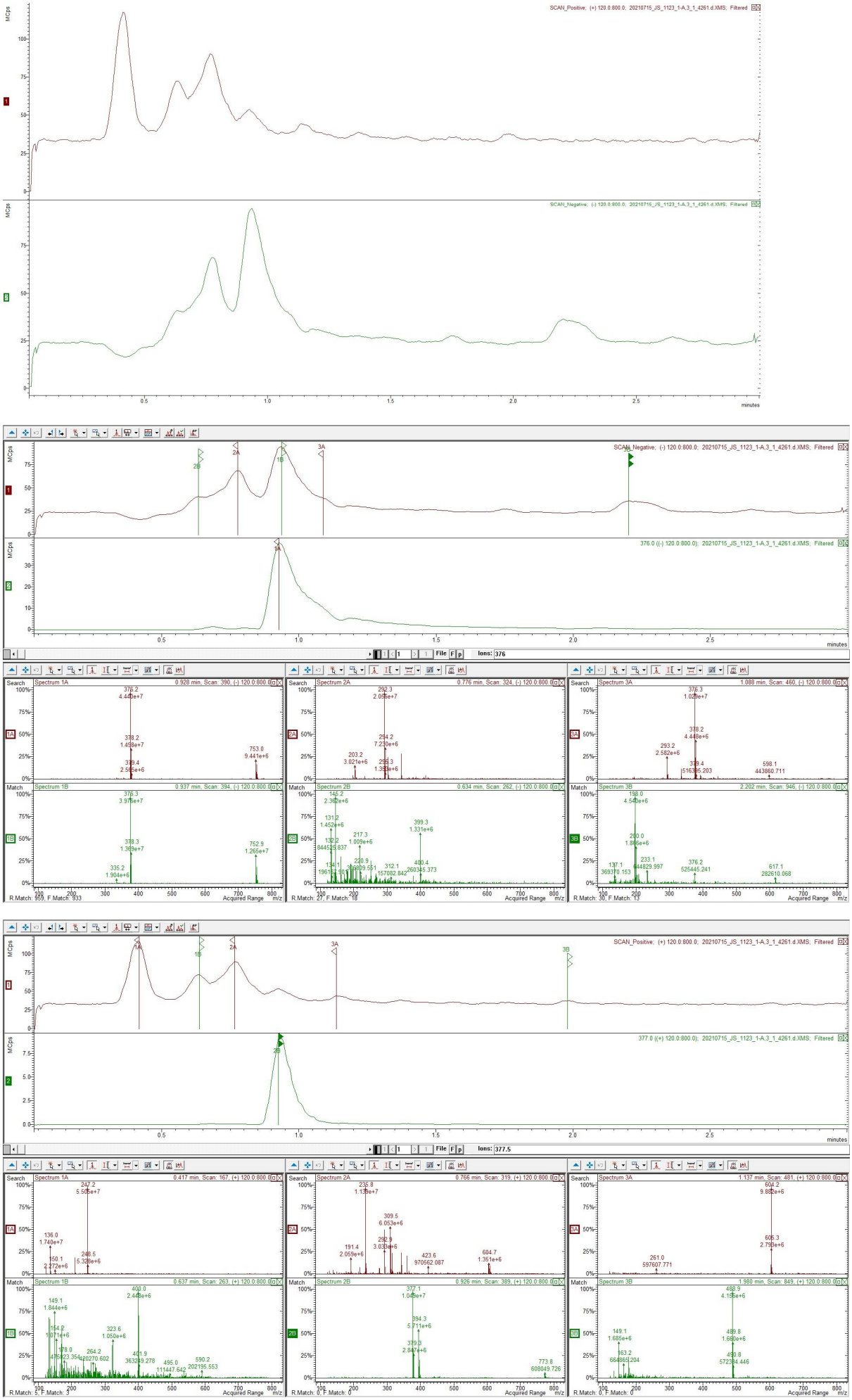
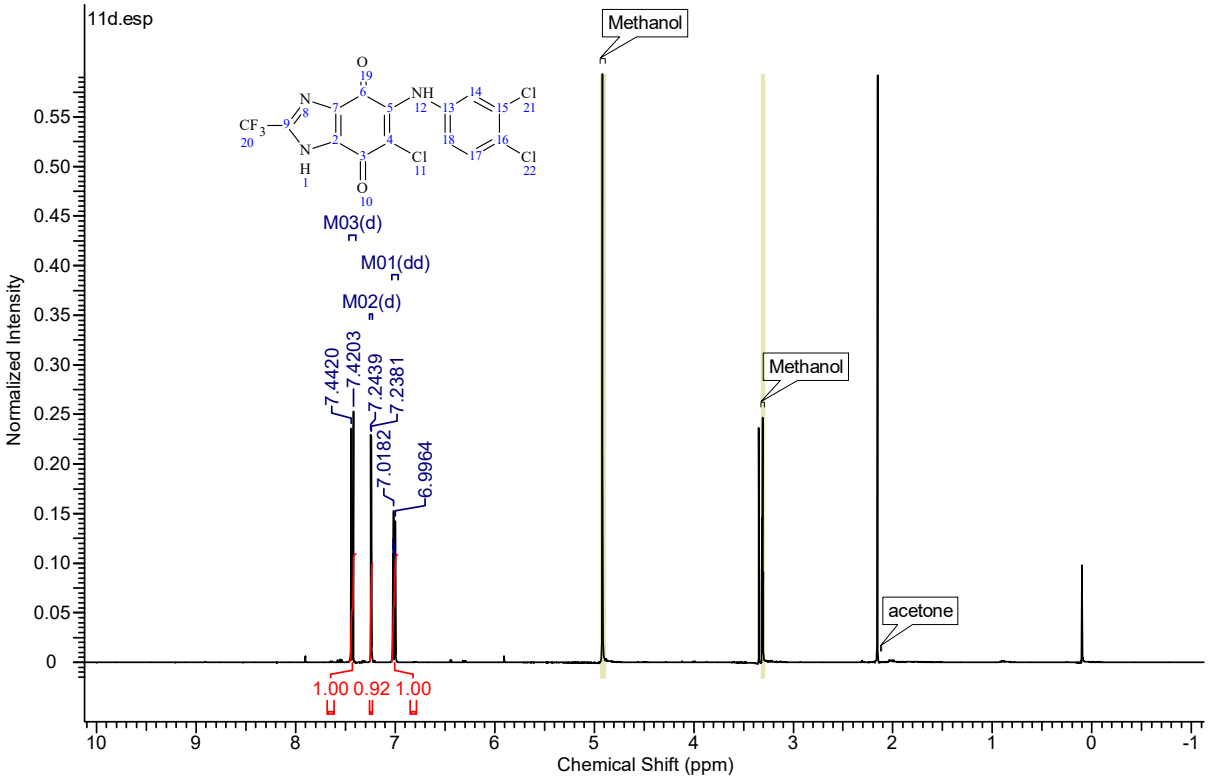


Figure S8. LC/MS spectrum of compound 11c

6-chloro-5-((3,4-dichlorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11d) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 3,4-dichloroaniline afforded **11d**. red purple powder. **11d** fully dissolved in 600 μL of the methanol-d4 and few drops of acetone. Yield 38%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.01 (dd, J=8.47, 2.52 Hz, 1 H) 7.24 (d, J=2.29 Hz, 1 H) 7.43 (d, J=8.70 Hz, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 174.16, 173.42, 142.22, 139.94, 139.24, 137.54, 131.91, 129.96, 127.88, 125.59, 123.75, 120.98, 113.37, 2 carbon peaks in 125.59 ppm overlapped; LC/MS (ESI, m/z) 408.0 [M - H]$^{-}$ 410.2 [M + H]$^{+}$.



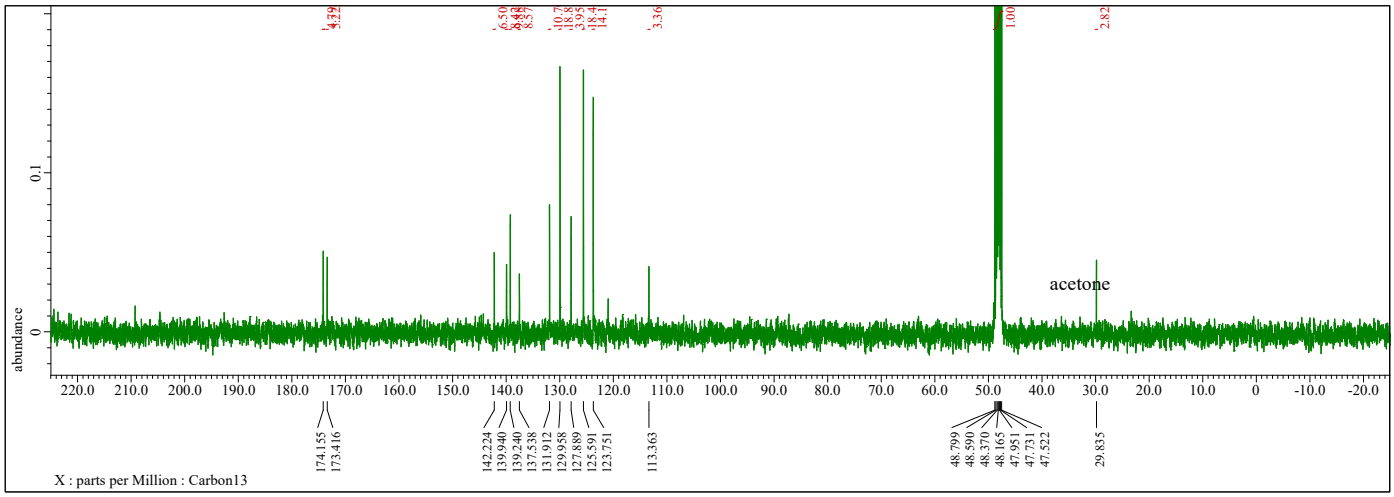


Figure S9. 1D 1H and 13C NMR spectrum of compound 11d

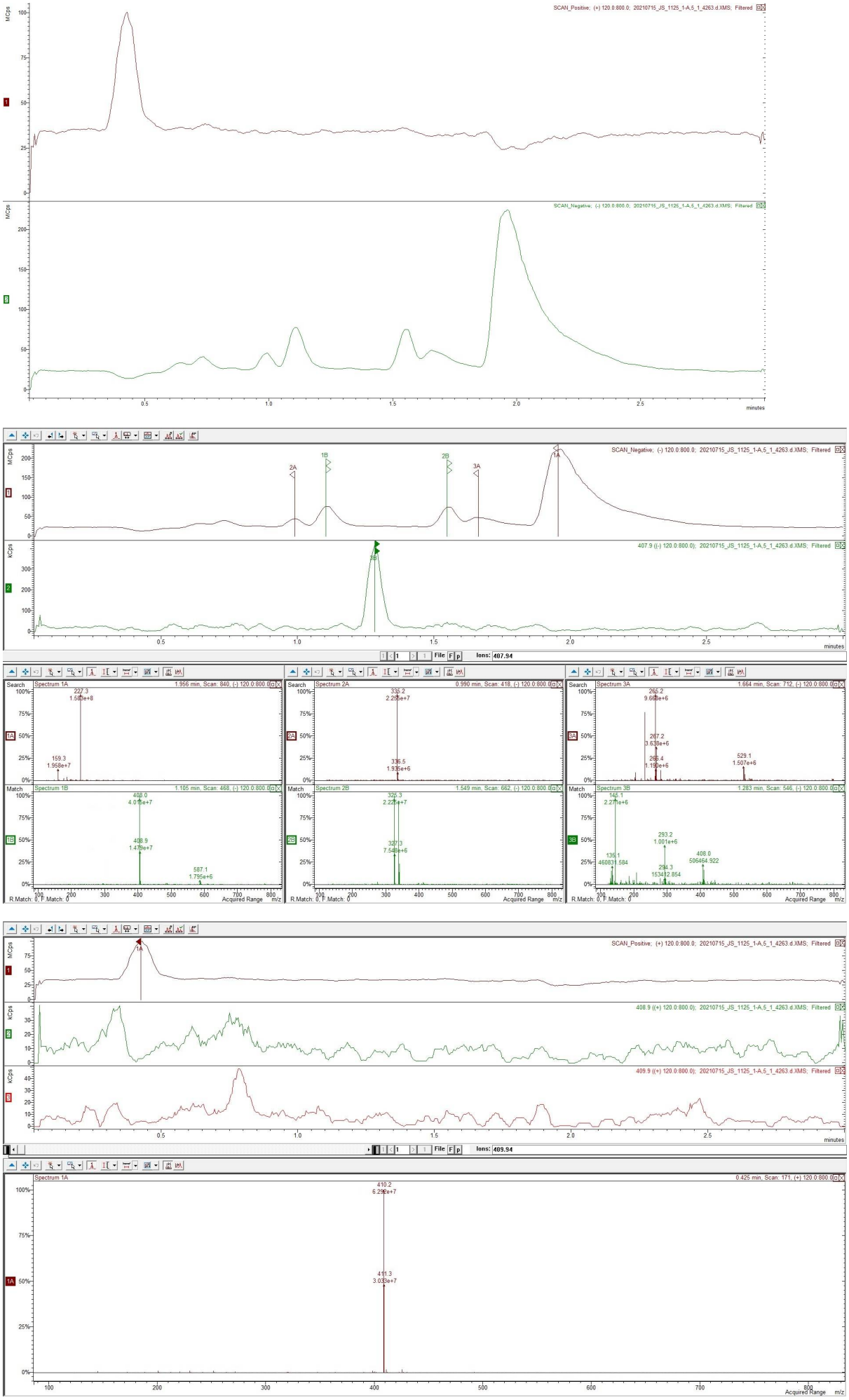
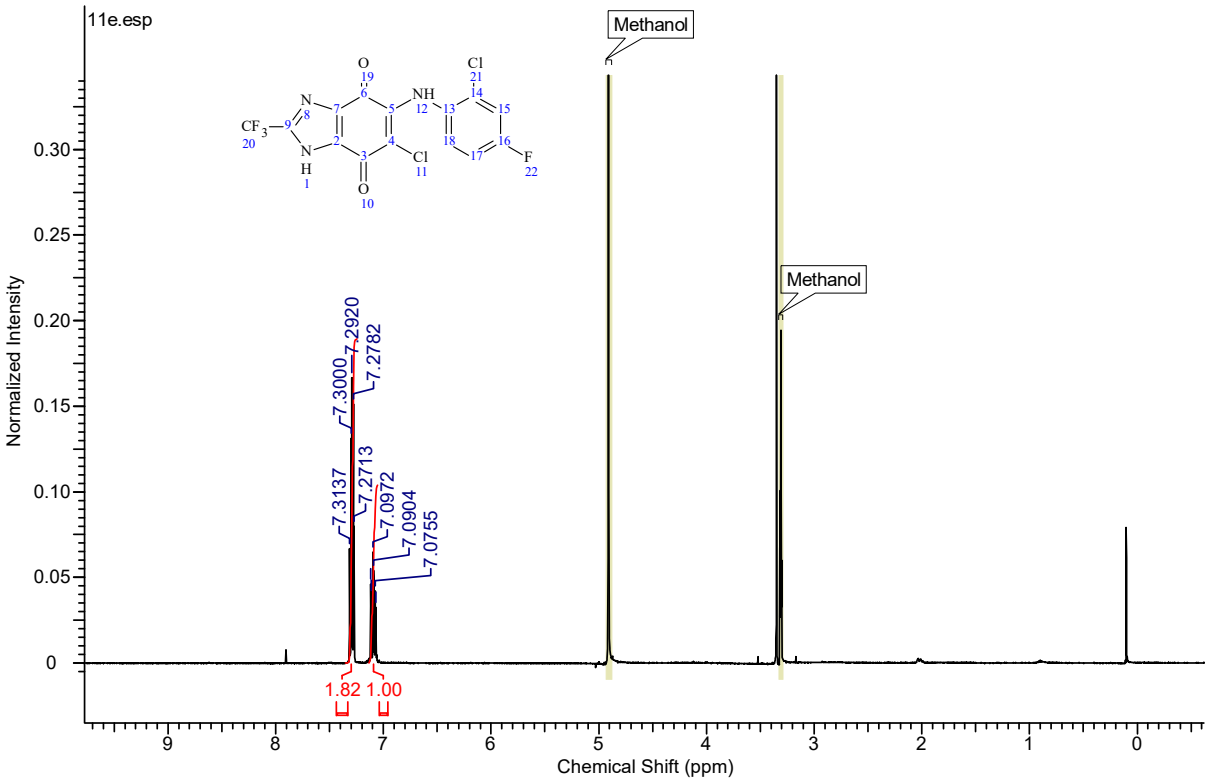


Figure S10. LC/MS spectrum of compound 11d

6-chloro-5-((2-chloro-4-fluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11e) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 2-chloro-4-fluoroaniline afforded **11e**. red purple powder. Yield 41%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.09 (td, J=8.47, 2.75 Hz, 1 H) 7.24 - 7.35 (m, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.34, 173.26, 142.69, 140.09, 136.91, 132.87, 131.87, 131.76, 129.34, 120.64, 117.95, 116.14, 115.89, 113.73, 113.50, 109.94; LC/MS (ESI, m/z) 392.2 $[M - H]^-$ 394.1 $[M + H]^+$.



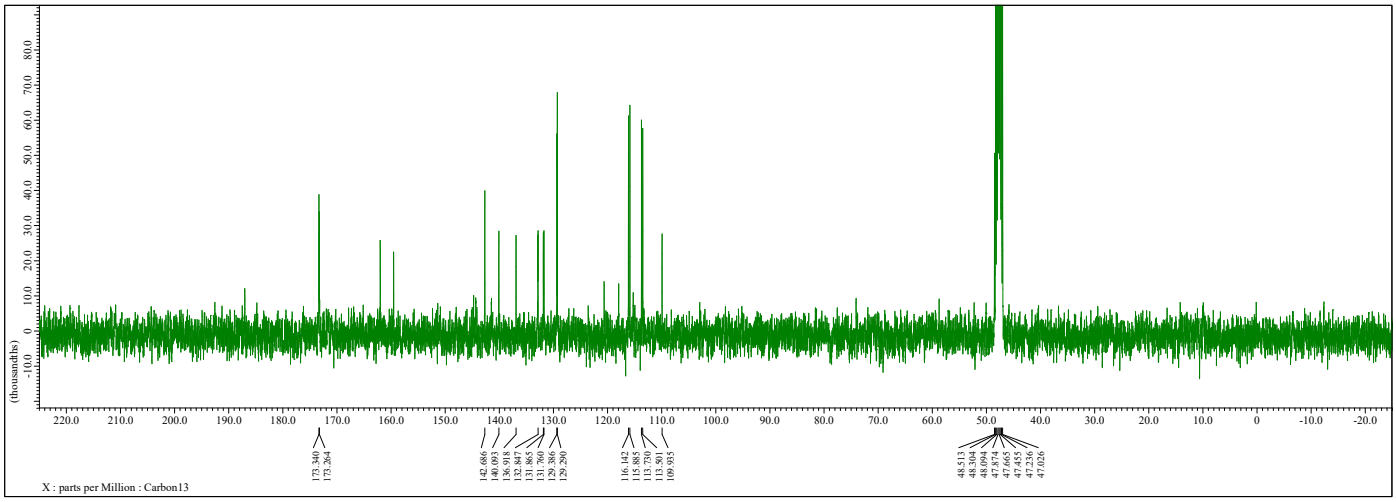


Figure S11. 1D 1H and 13C NMR spectrum of compound 11e

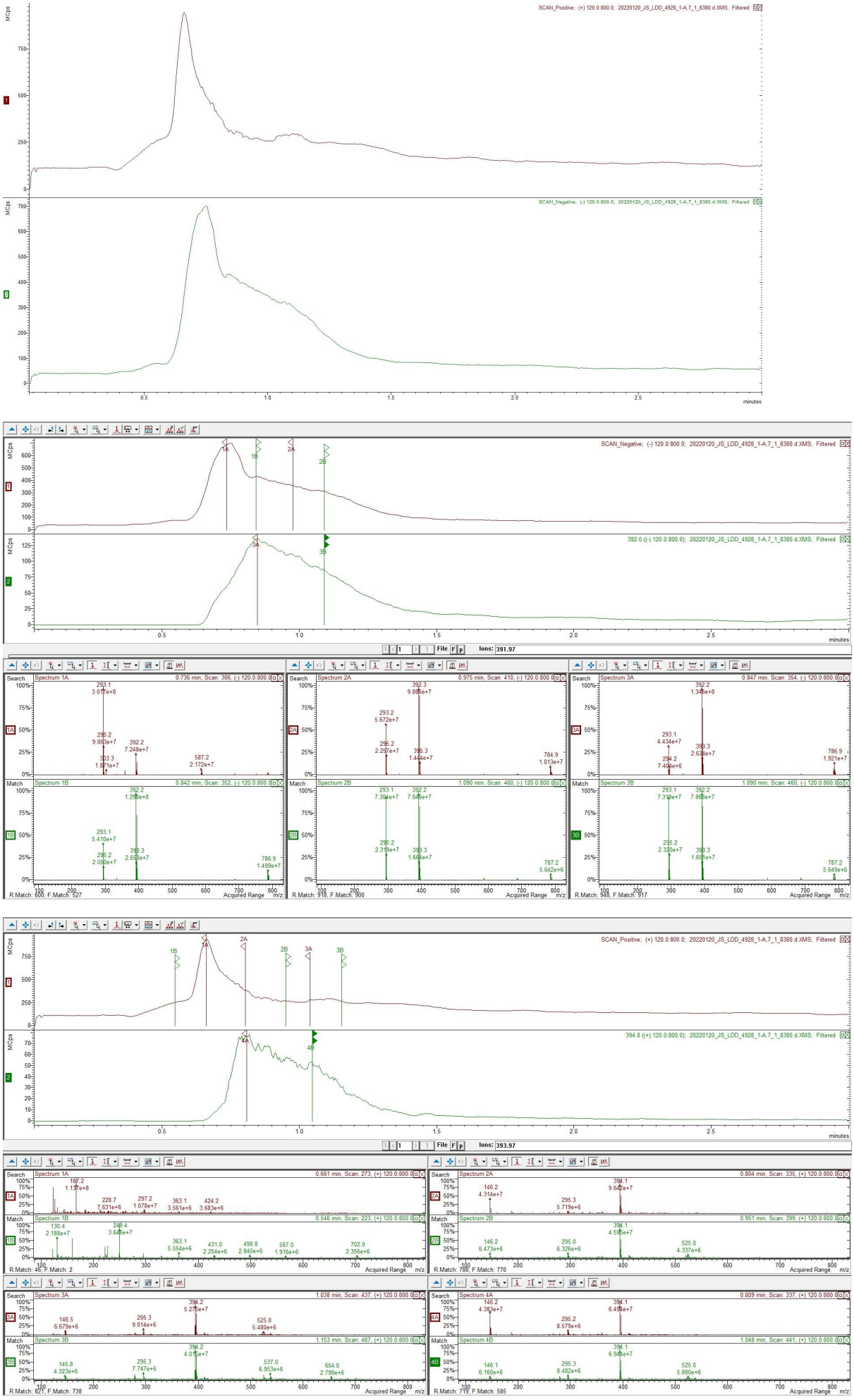
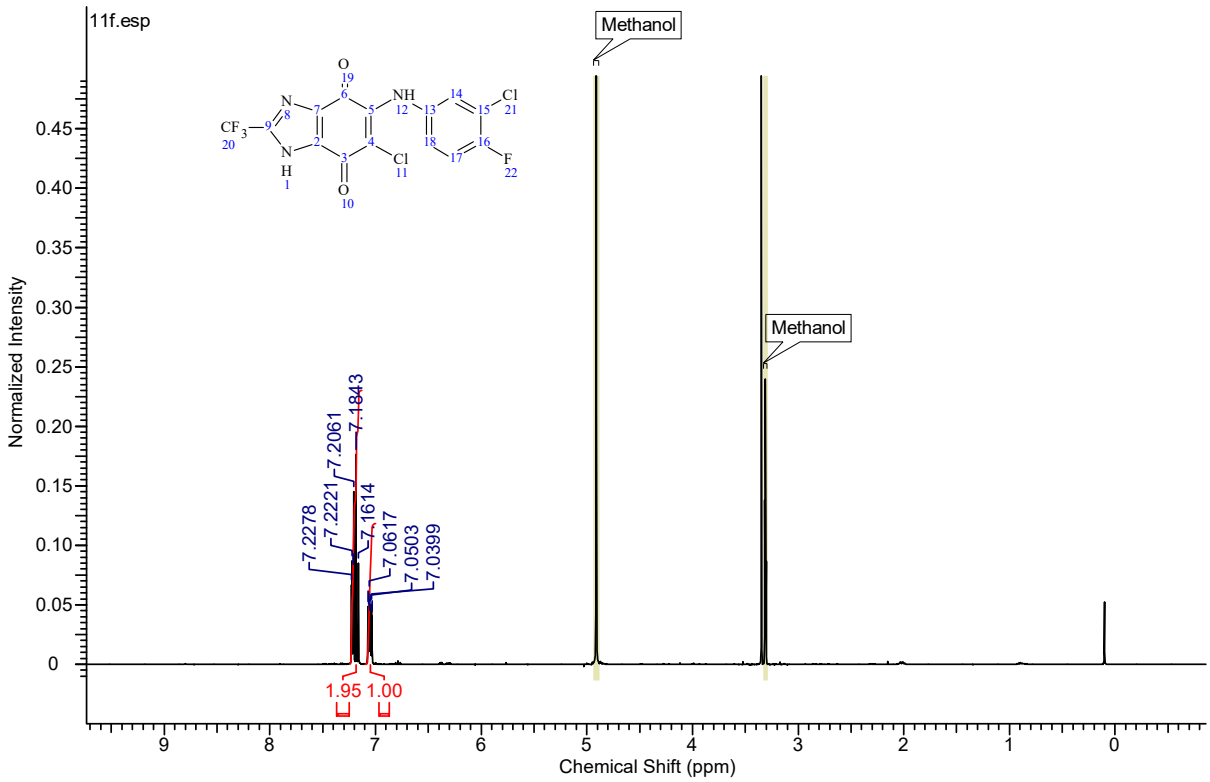


Figure S12. LC/MS spectrum of compound 11e

6-chloro-5-((3-chloro-4-fluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11f) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 3-chloro-4-fluoroaniline afforded **11f**. red purple powder. Yield 46%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.05 (ddd, J=8.93, 4.12, 2.52 Hz, 1 H) 7.13 - 7.24 (m, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 178.81, 142.11, 140.72, 137.51, 135.81, 126.00, 124.28, 124.21, 119.86, 119.67, 115.65, 115.42, 111.30, 2 carbon peaks in 178.81 ppm overlapped, other peaks are impurities; LC/MS (ESI, m/z) 392.2 [M - H]$^{-}$ 393.7 [M + H]$^{+}$.



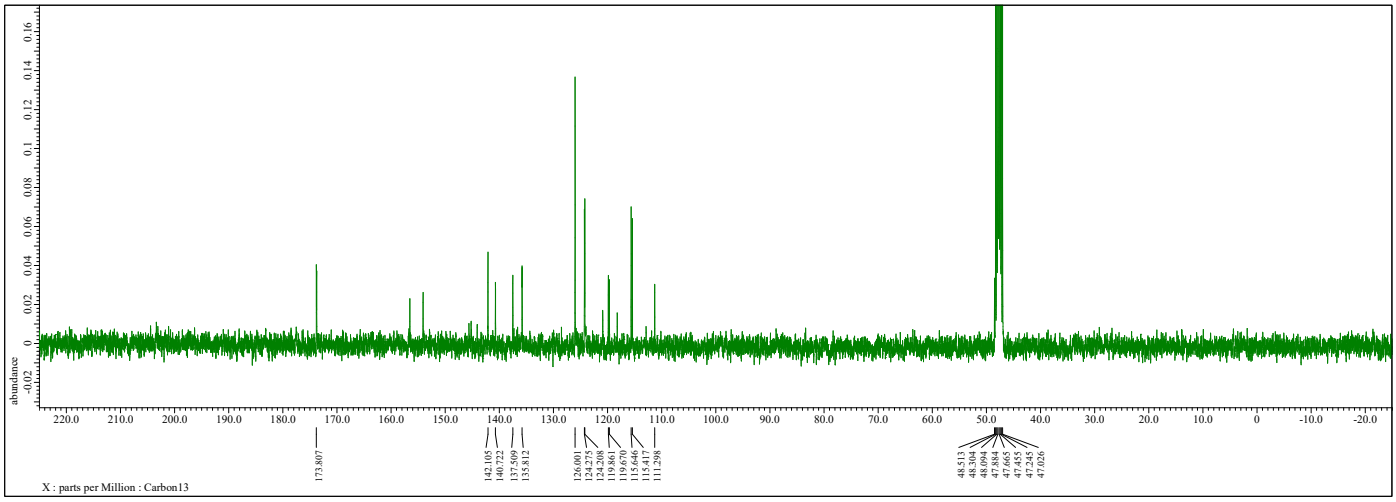


Figure S13. 1D 1H and 13C NMR spectrum of compound 11f

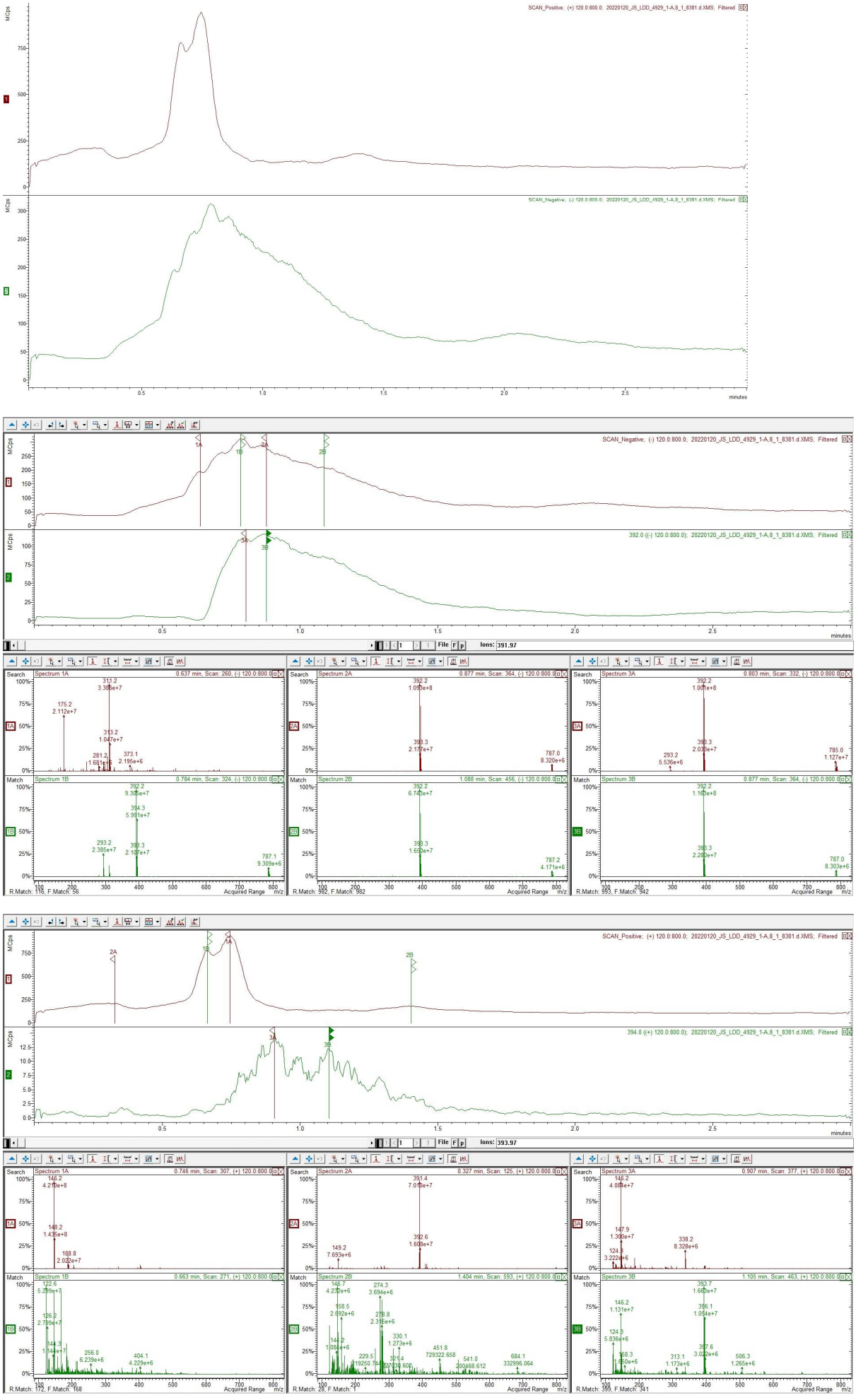
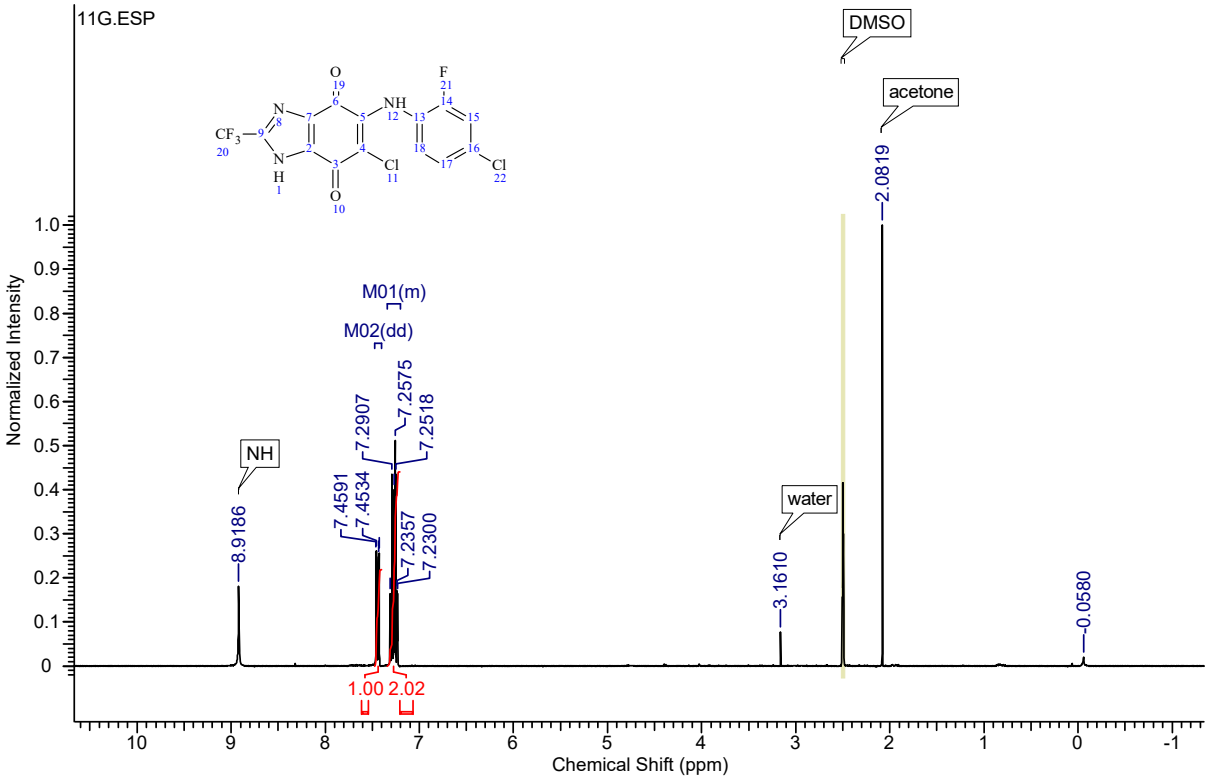


Figure S14. LC/MS spectrum of compound 11f

6-chloro-5-((4-chloro-2-fluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11g) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-chloro-2-fluoroaniline afforded **11g**. red purple powder. **11g** fully dissolved in 600 µL of the DMSO-d6 and few drops of acetone. Yield 52%; ^{1}H NMR (400 MHz, DMSO-d6) δ ppm 7.20 - 7.34 (m, 2 H) 7.44 (dd, J=10.30, 2.06 Hz, 1 H); ^{13}C NMR (100 MHz, DMSO-d6) δ 174.06, 173.44, 142.84, 142.73, 138.43, 130.20, 130.09, 128.65, 127.65, 127.53, 124.67, 116.48, 116.25, 111.93, peaks in 155 and 157 ppm are impurities; LC/MS (ESI, m/z) 392.2 $[M - H]^-$ 394.1 $[M + H]^+$.



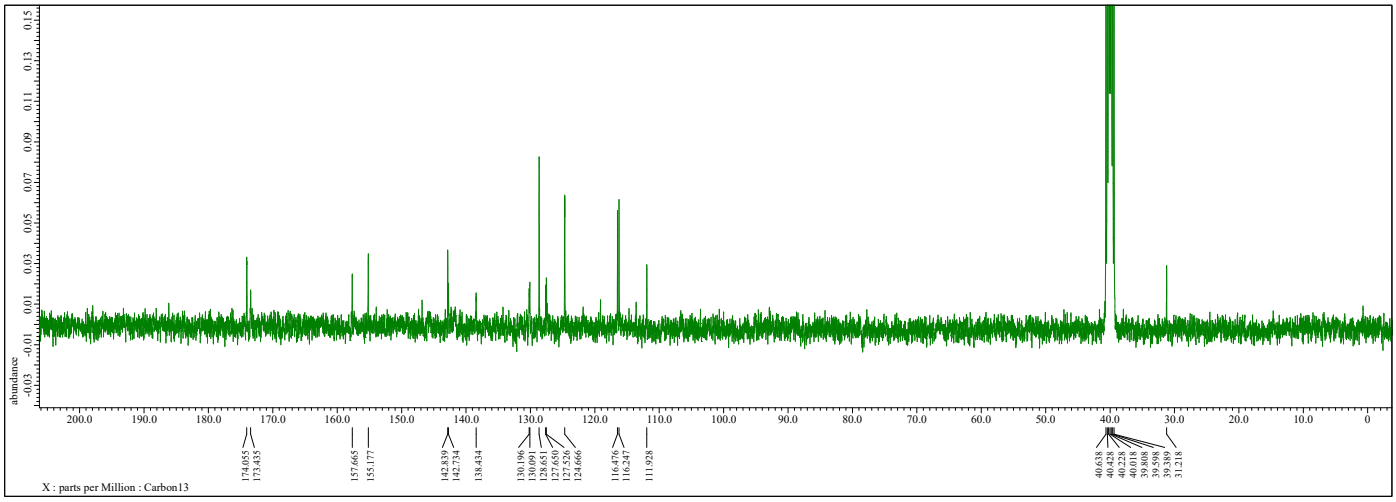


Figure S15. 1D 1H and 13C NMR spectrum of compound 11g

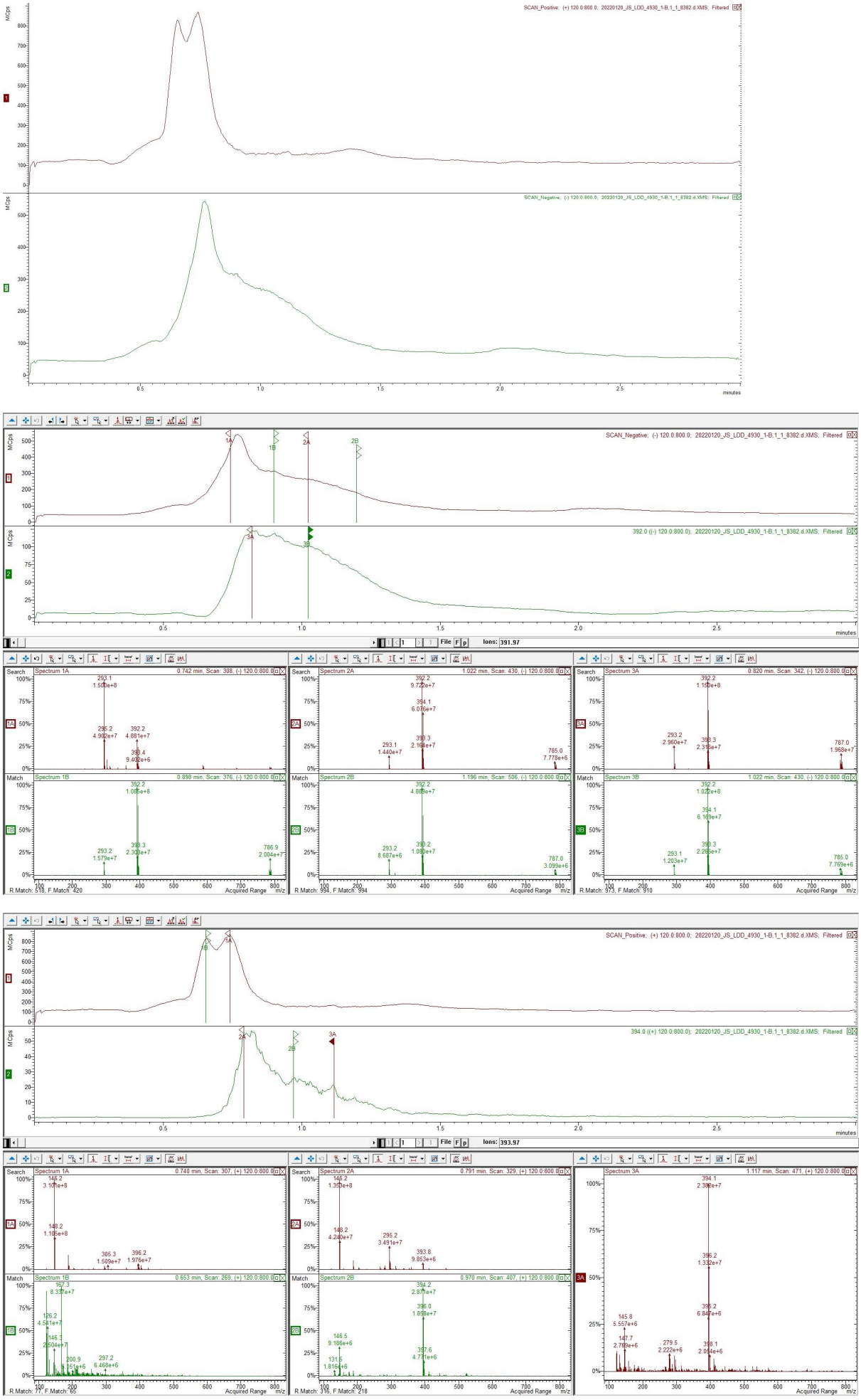
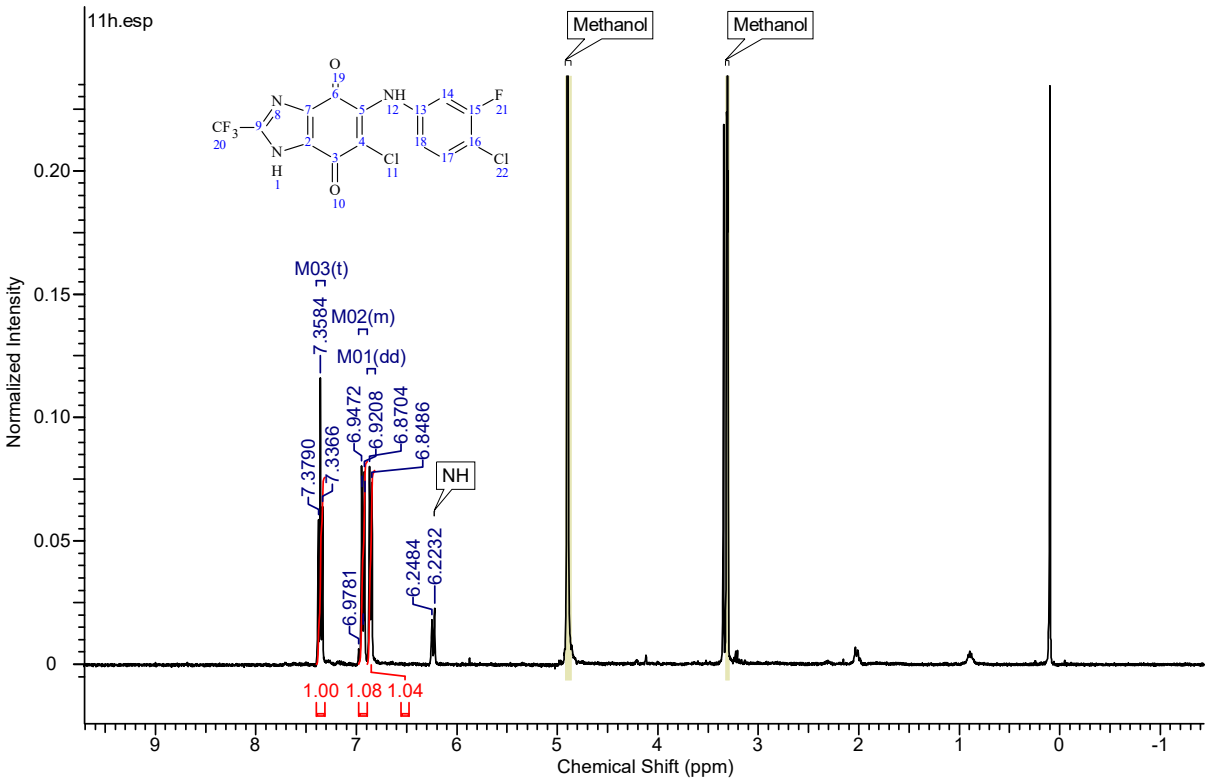


Figure S16. LC/MS spectrum of compound 11g

6-chloro-5-((4-chloro-3-fluorophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11h) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-chlro-3-fluoroaniline afforded **11h**. red purple powder. Yield 39%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 6.86 (dd, J=8.70, 0.92 Hz, 1 H) 6.89 - 6.98 (m, 1 H) 7.36 (t, J=8.47 Hz, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 175.09, 174.29, 143.04, 141.47, 139.63, 139.05, 129.36, 119.91, 119.87, 115.26, 115.07, 113.25, 111.45, 111.21, peaks in 158.67 and 156.22 ppm are impurities; LC/MS (ESI, m/z) 392.2 [M - H]$^{-}$ 394.1 [M + H]$^{+}$.



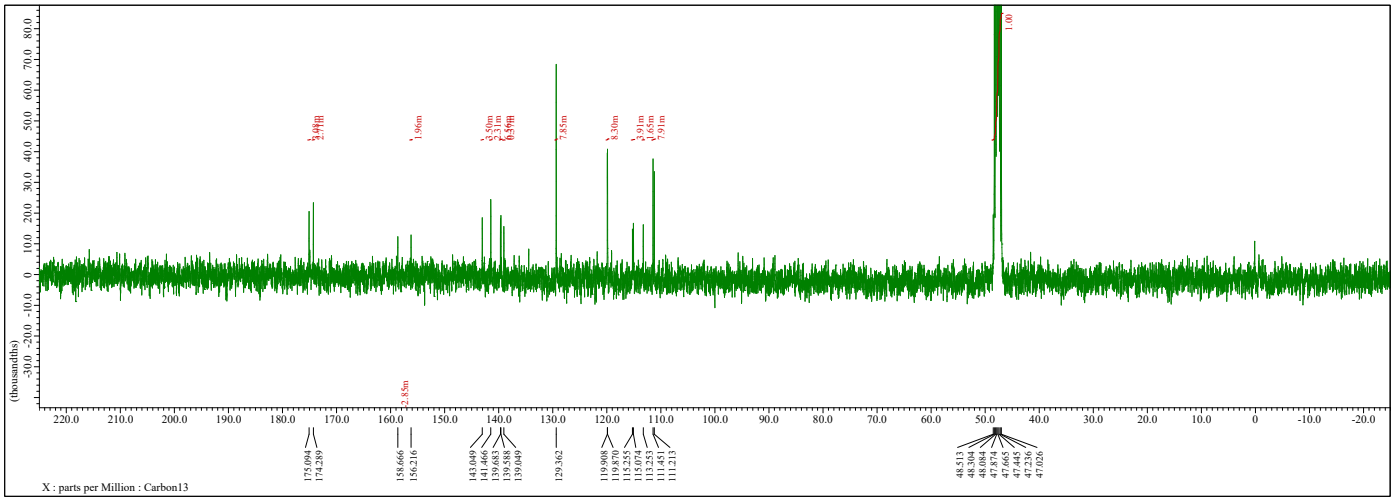


Figure S17. 1D 1H and 13C NMR spectrum of compound 11h

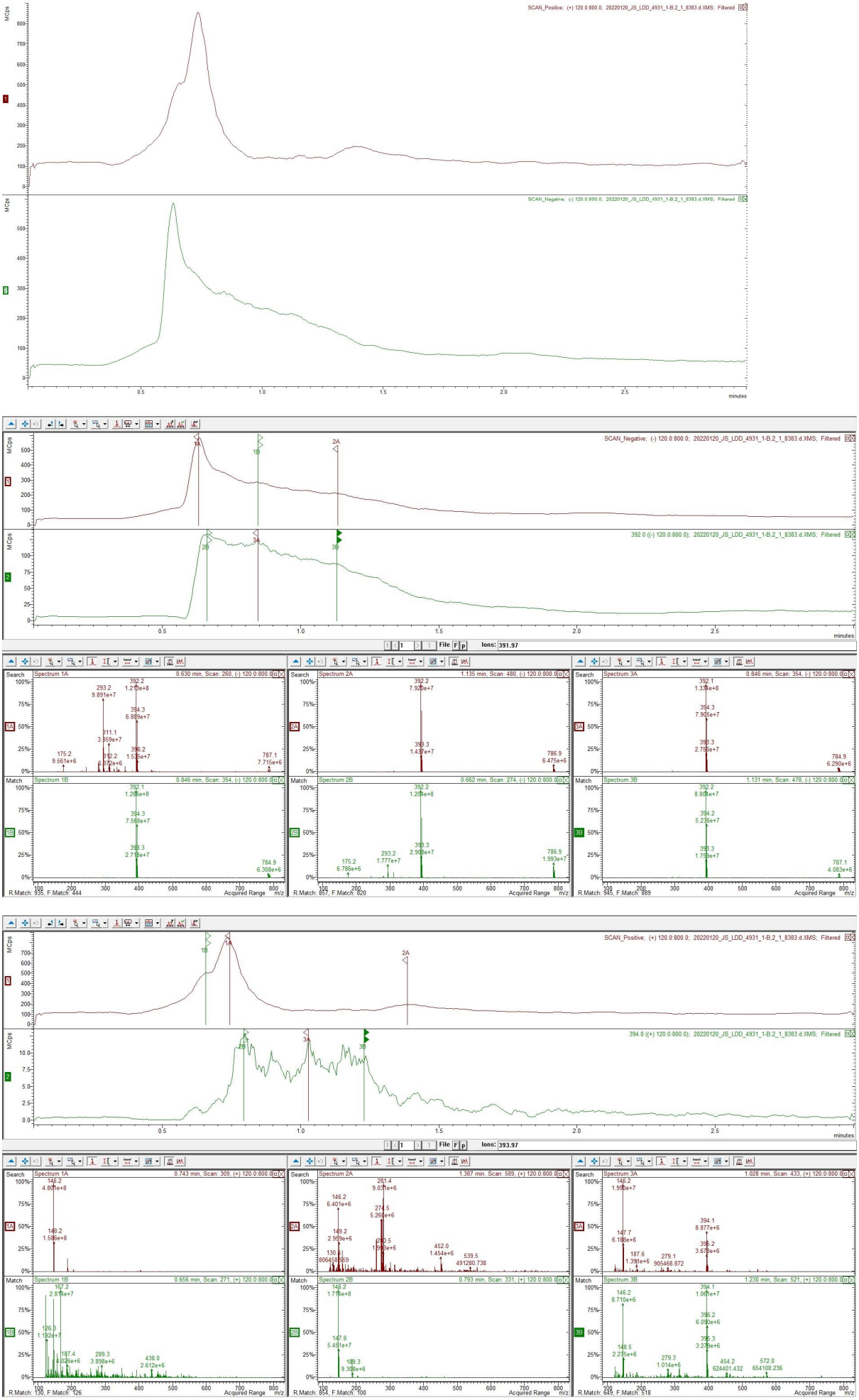
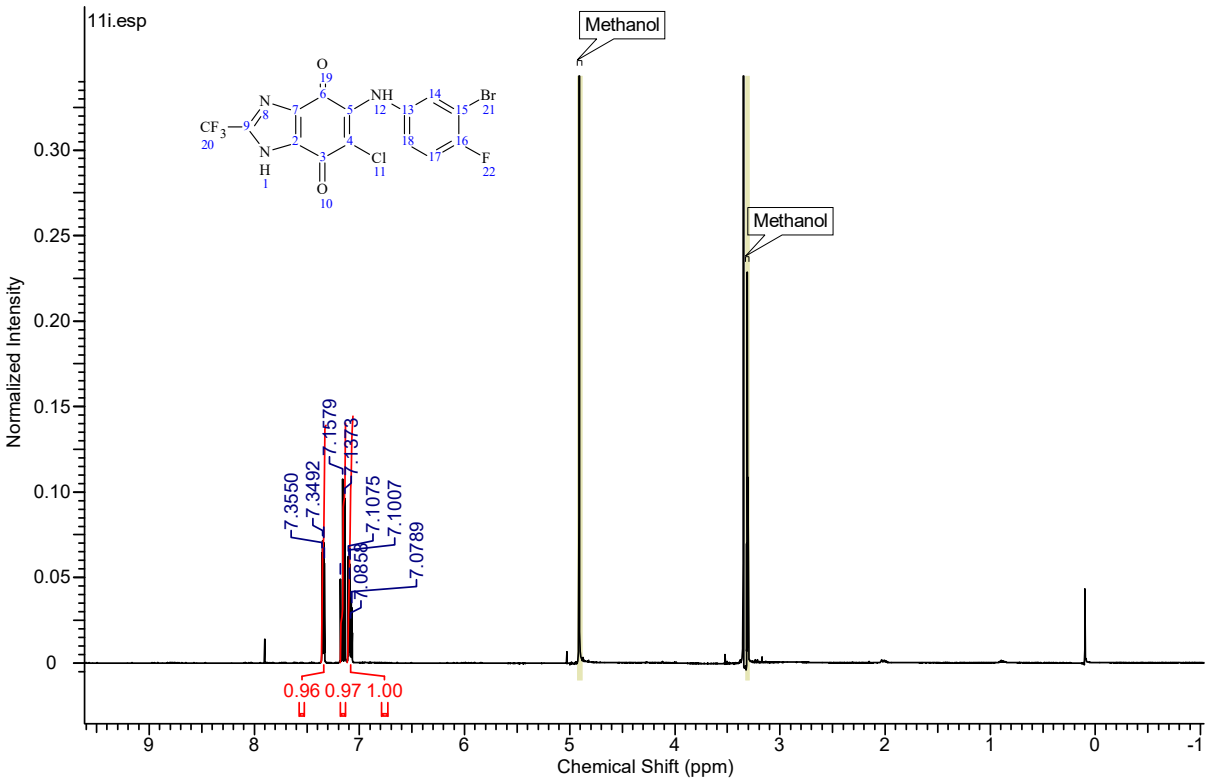


Figure S18. LC/MS spectrum of compound 11h

5-((3-bromo-4-fluorophenyl)amino)-6-chloro-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11i) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 3-bromo-4-fluoroaniline afforded **11i**. purple powder. Yield 63%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.06 - 7.11 (m, 1 H) 7.13 - 7.18 (m, 1 H) 7.34 (dd, J=5.95, 2.29 Hz, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 174.43, 173.89, 142.12, 141.68, 137.99, 136.05, 128.83, 124.98, 124.90, 115.41, 115.17, 111.07, 107.54, 107.32, peaks in 157.57 and 155.14 ppm are impurities; LC/MS (ESI, m/z) 436.1 [M - H]$^{-}$ 437.8 [M + H]$^{+}$.



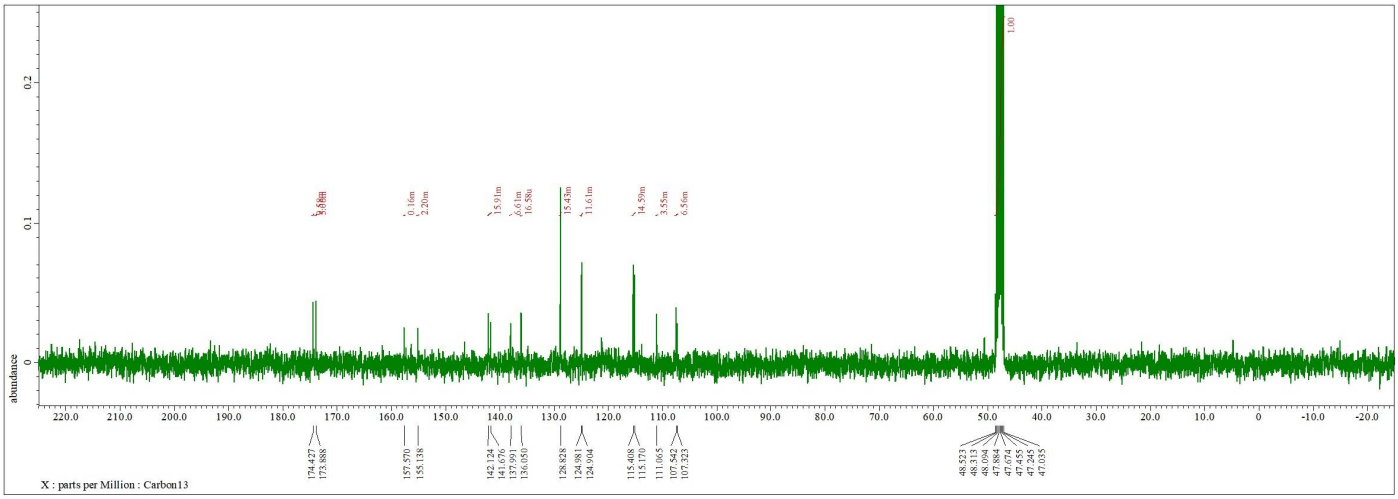


Figure S19. 1D 1H and 13C NMR spectrum of compound 11i

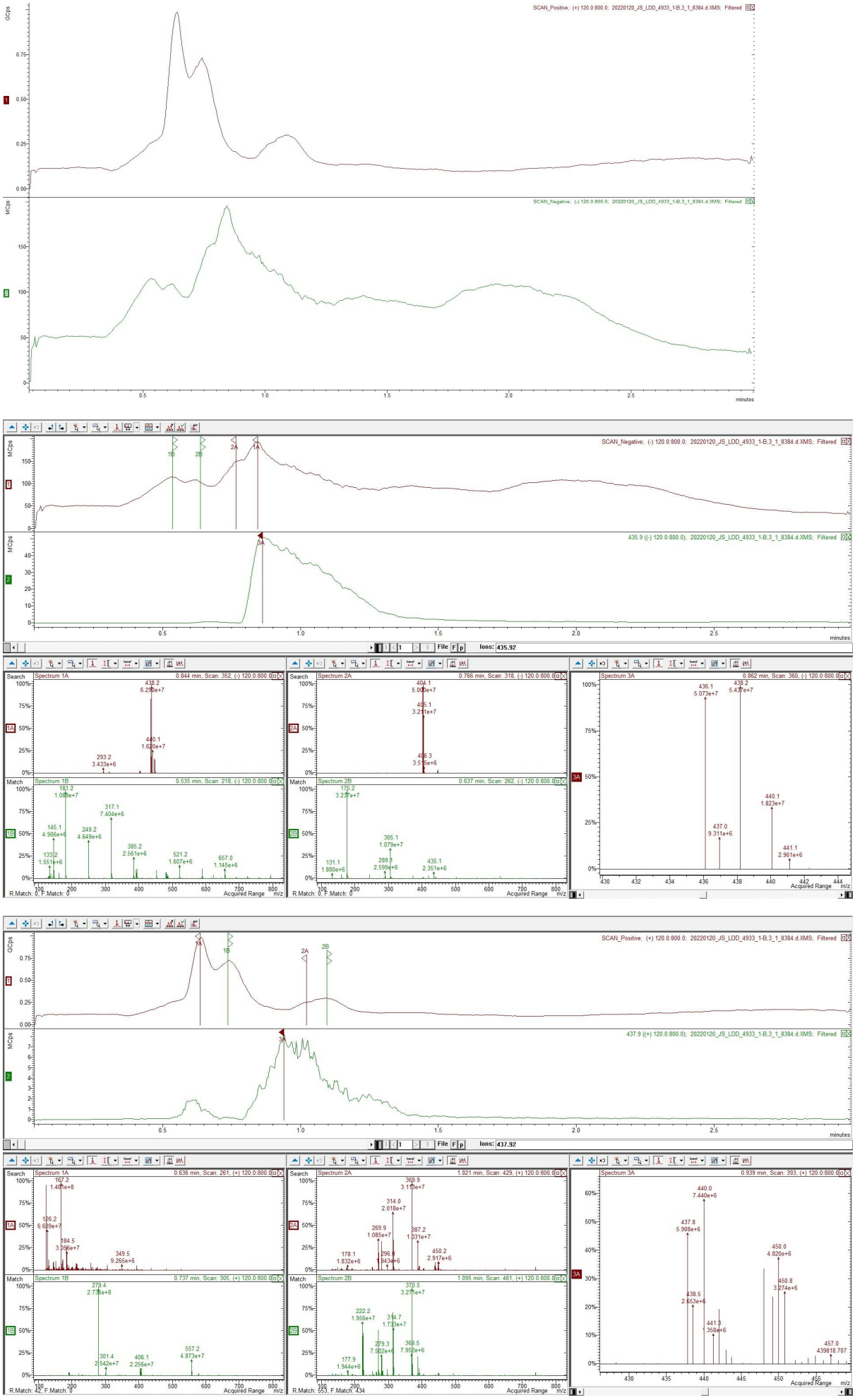
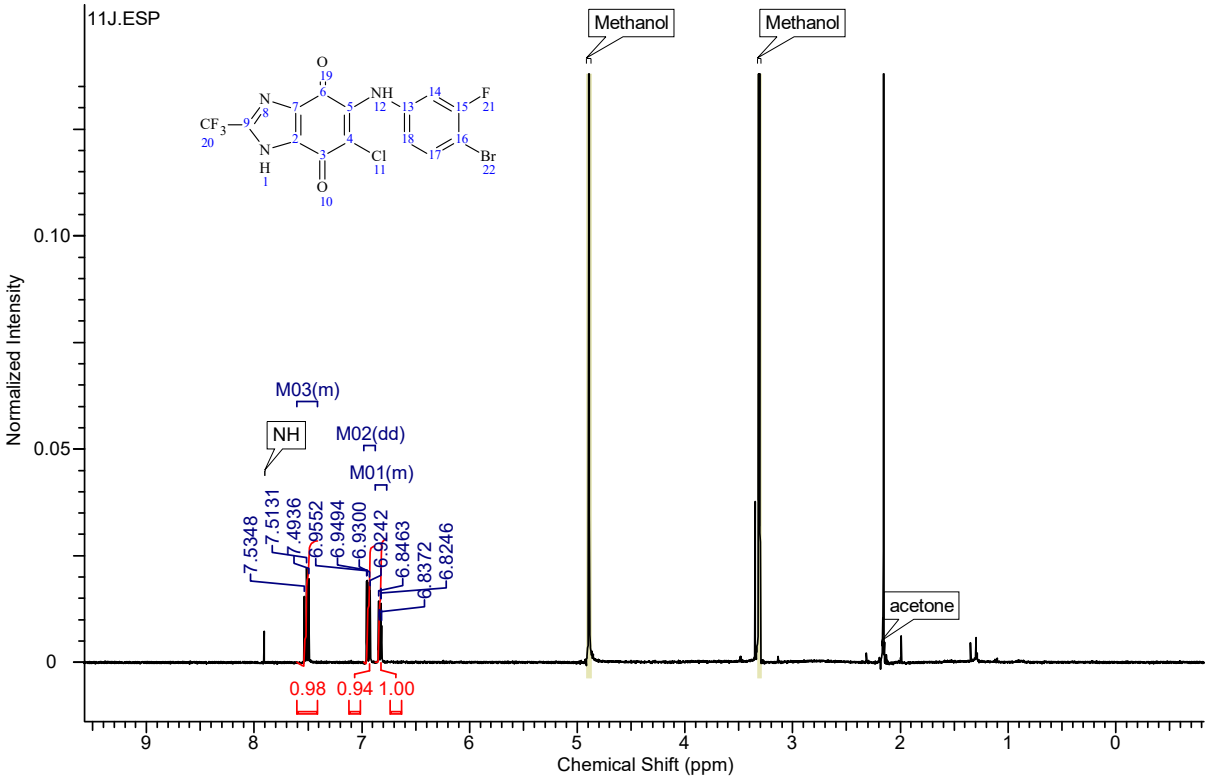


Figure S20. LC/MS spectrum of compound 11i

5-((4-bromo-3-fluorophenyl)amino)-6-chloro-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11j) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-bromo-3-fluoroaniline afforded **11j**. red purple powder. **11j** fully dissolved in 600 μL of the methanol-d4 and few drops of acetone. Yield 51%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 6.77 - 6.88 (m, 1 H) 6.94 (dd, J=10.08, 2.29 Hz, 1 H) 7.41 - 7.60 (m, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 174.81, 174.27, 148.44, 148.06, 142.67, 141.38, 140.45, 140.35, 138.87, 132.27, 120.21, 113.56, 111.23, 110.98, peaks in 159.75, 157.32, and 102.54 ppm are impurities; LC/MS (ESI, m/z) 436.1 [M - H]$^{-}$ 438.0 [M + H]$^{+}$.



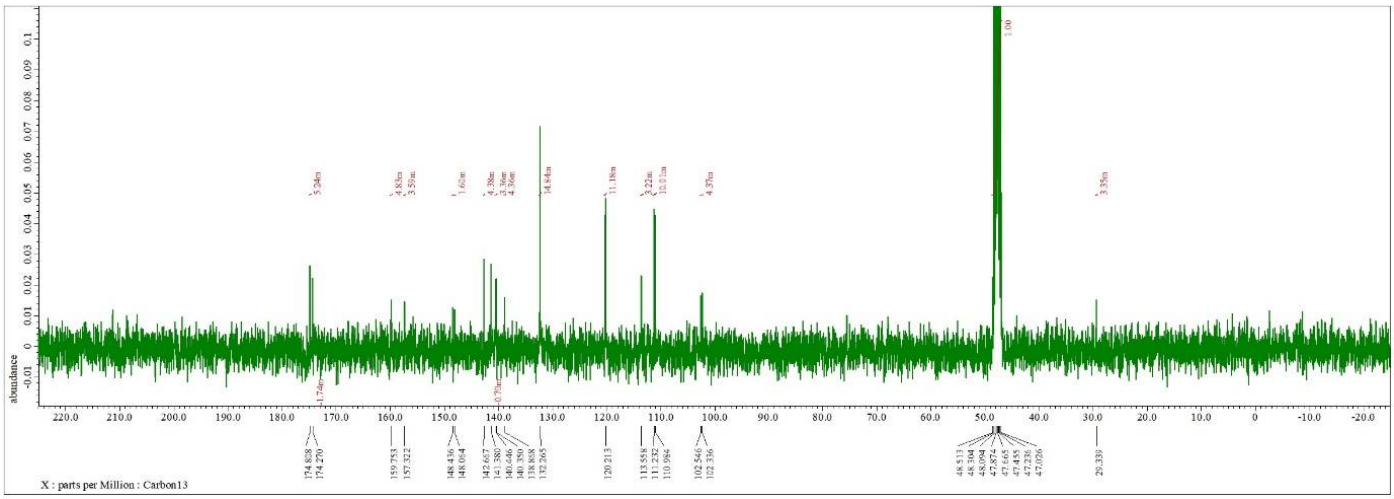


Figure S21. 1D 1H and 13C NMR spectrum of compound 11j

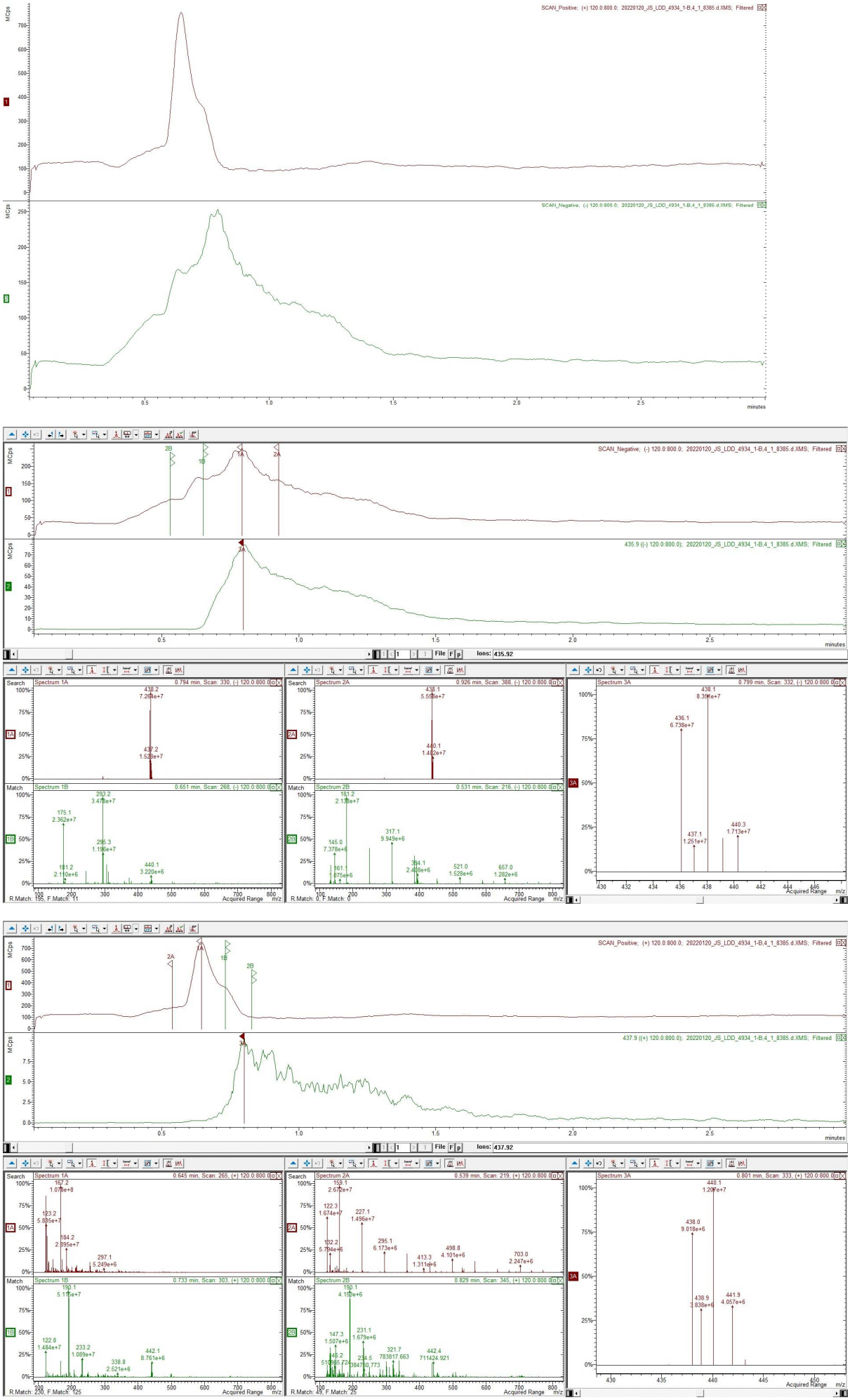
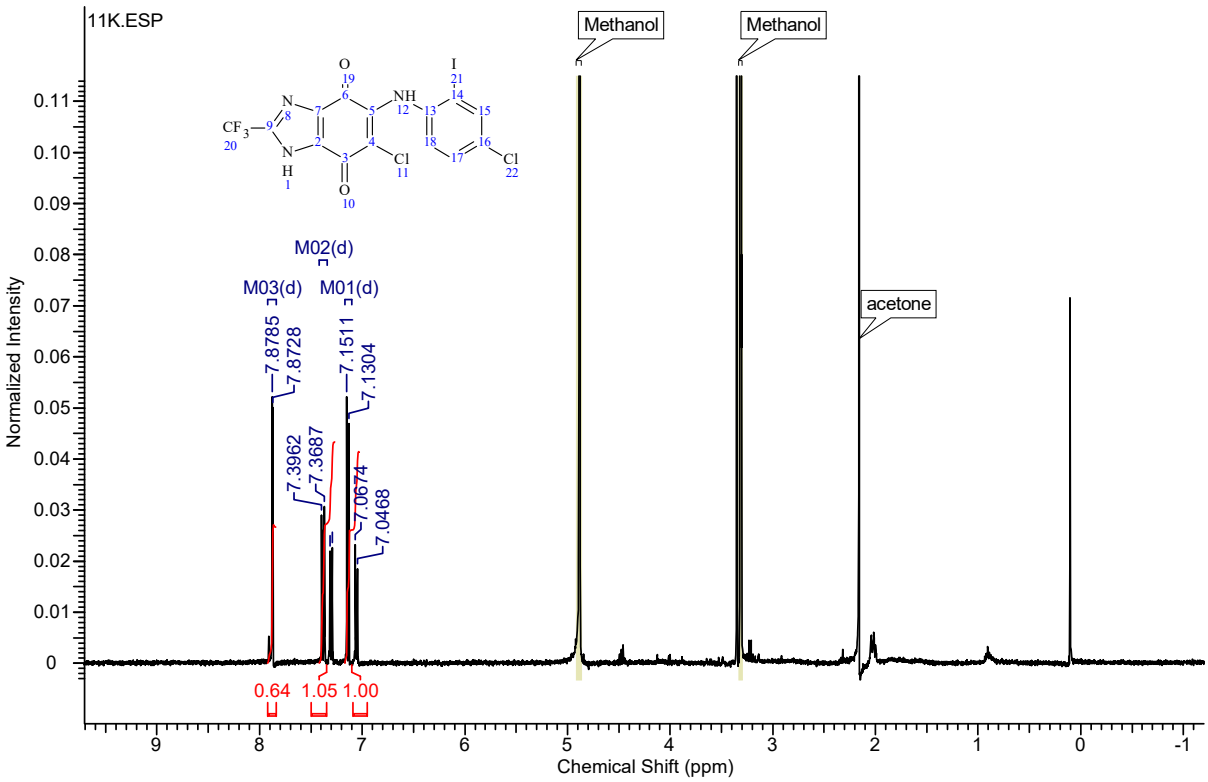


Figure S22. LC/MS spectrum of compound 11j

6-chloro-5-((4-chloro-2-iodophenyl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (11k) Following the general procedure for the synthesis of **11a-k**, the substitution reaction of **9b** with 4-chloro-2-iodoaniline afforded **11k**. purple powder. **11k** fully dissolved in 600 μL of the methanol-d4 and few drops of acetone. Yield 24%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 7.14 (d, J=8.24 Hz, 1 H) 7.38 (d, J=10.99 Hz, 1 H) 7.88 (d, J=2.29 Hz, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 175.75, 174.94, 141.80, 140.10, 138.43, 137.95, 131.35, 127.92, 127.78, 127.54, 125.09, 117.77, 110.53, 96.60, other peaks are impurities; LC/MS (ESI, m/z) 499.8 [M - H]$^{-}$ 501.9 [M + H]$^{+}$.



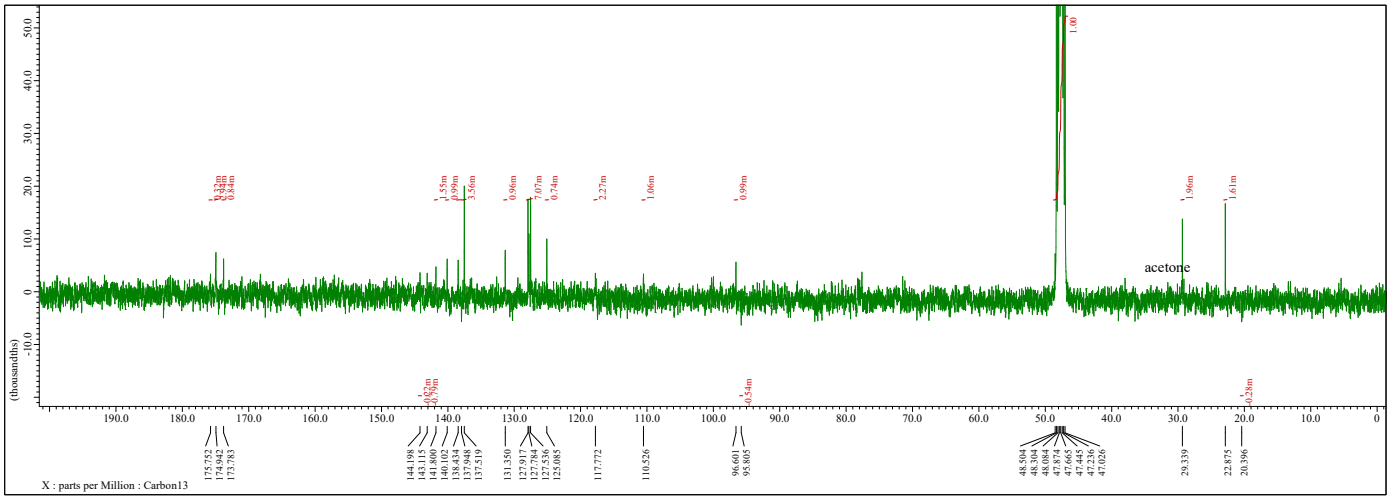


Figure S23. 1D 1H and 13C NMR spectrum of compound 11k

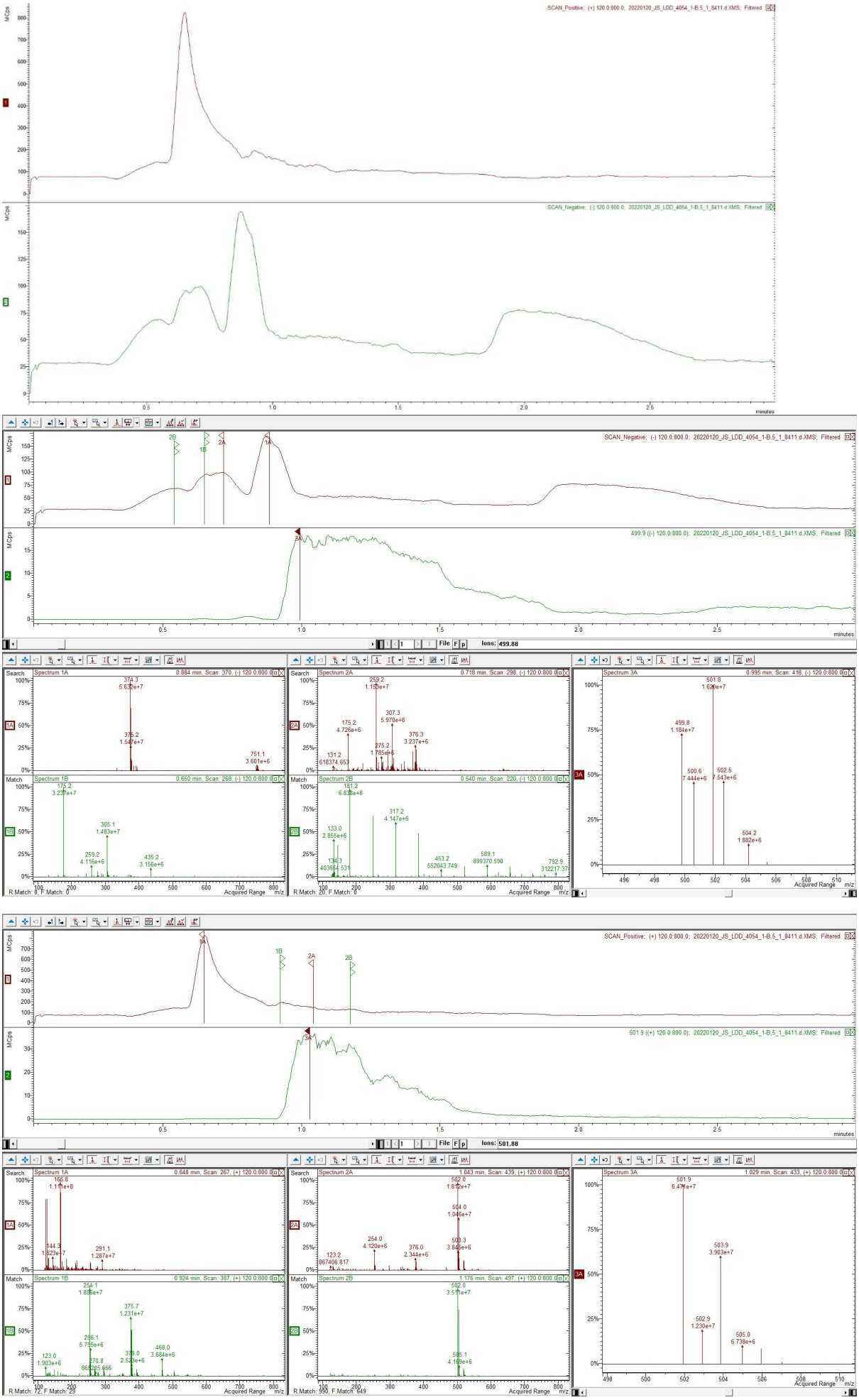
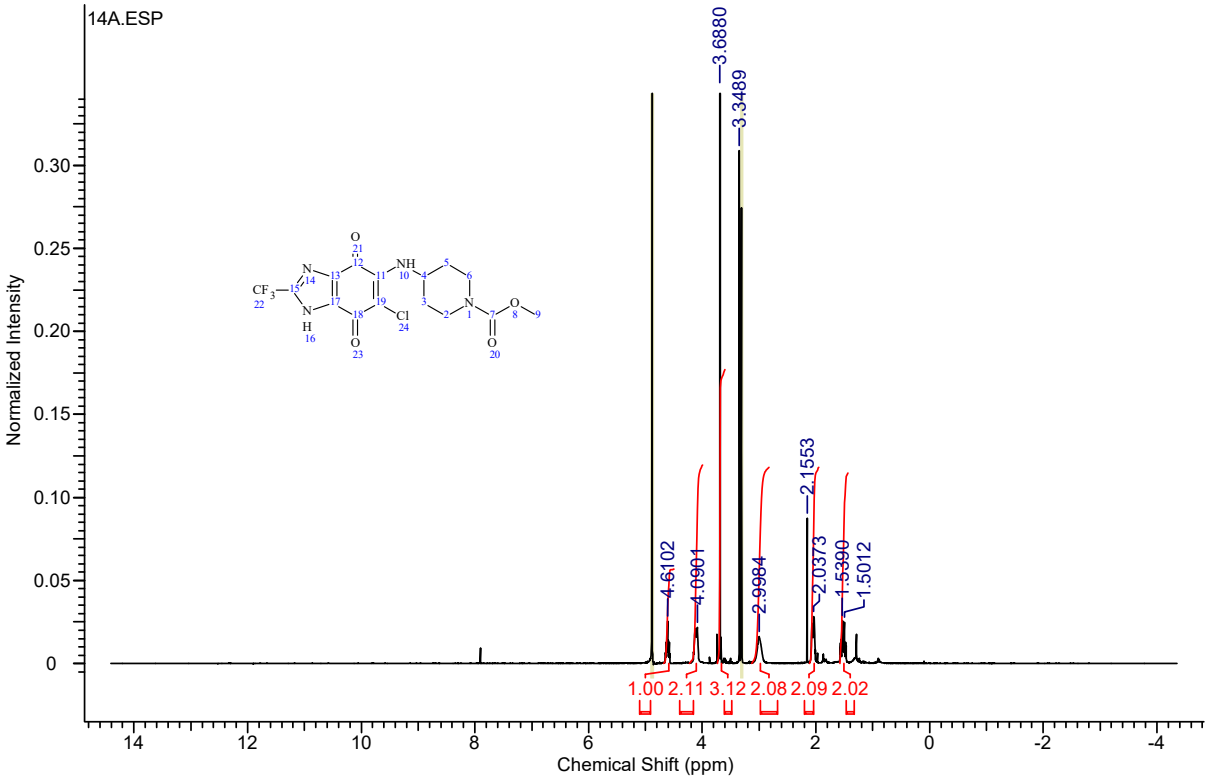


Figure S24. LC/MS spectrum of compound 11k

methyl-4-((6-chloro-4,7-dioxo-2-(trifluoromethyl)-4,7-dihydro-1H-benzo[d]imidazol-5-yl)amino)piperidine-1-carboxylate (14a) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13a** afforded **14a**. purple powder. **14a** fully dissolved in 600 µL of the methanol-d4 and a drop of acetone. Yield 37%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.52 (d, J=15.11 Hz, 2 H) 2.04 (br. s., 2 H) 3.00 (br. s., 2 H) 3.69 (s, 3 H) 4.09 (br. s., 2 H) 4.61 (s, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 175.76, 173.83, 169.11, 156.24, 144.98, 143.53, 138.29, 122.12, 119.44, 51.97, 50.52, 42.29, 32.84, other peaks are impurities; LC/MS (ESI, m/z) 404.9 [M - H]$^{-}$ 406.8 [M + H]$^{+}$.



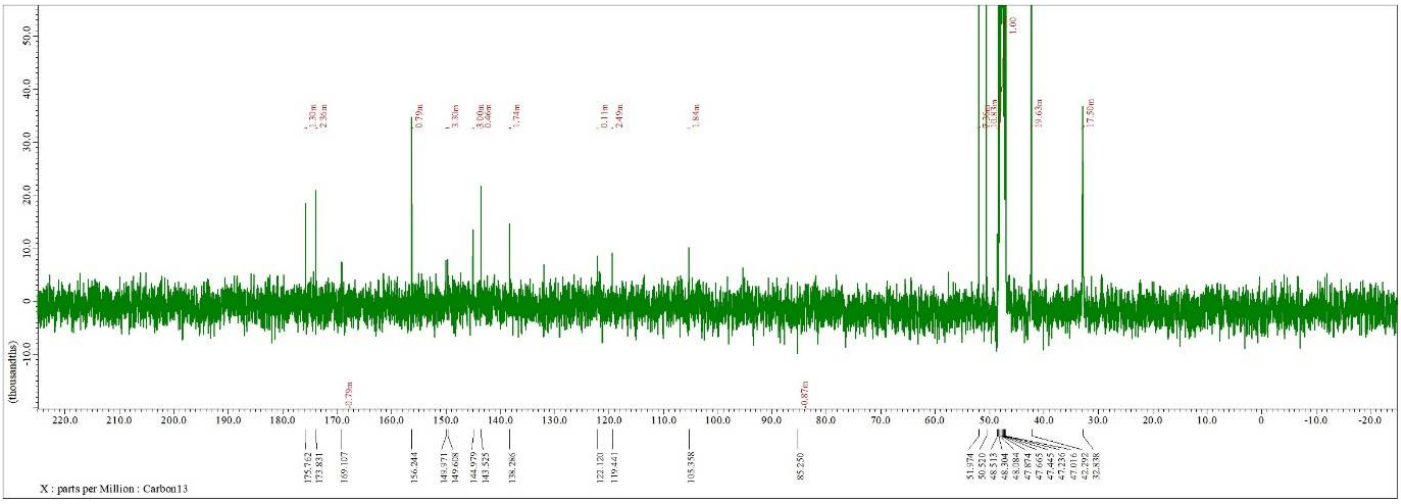


Figure S25. 1D 1H and 13C NMR spectrum of compound 14a

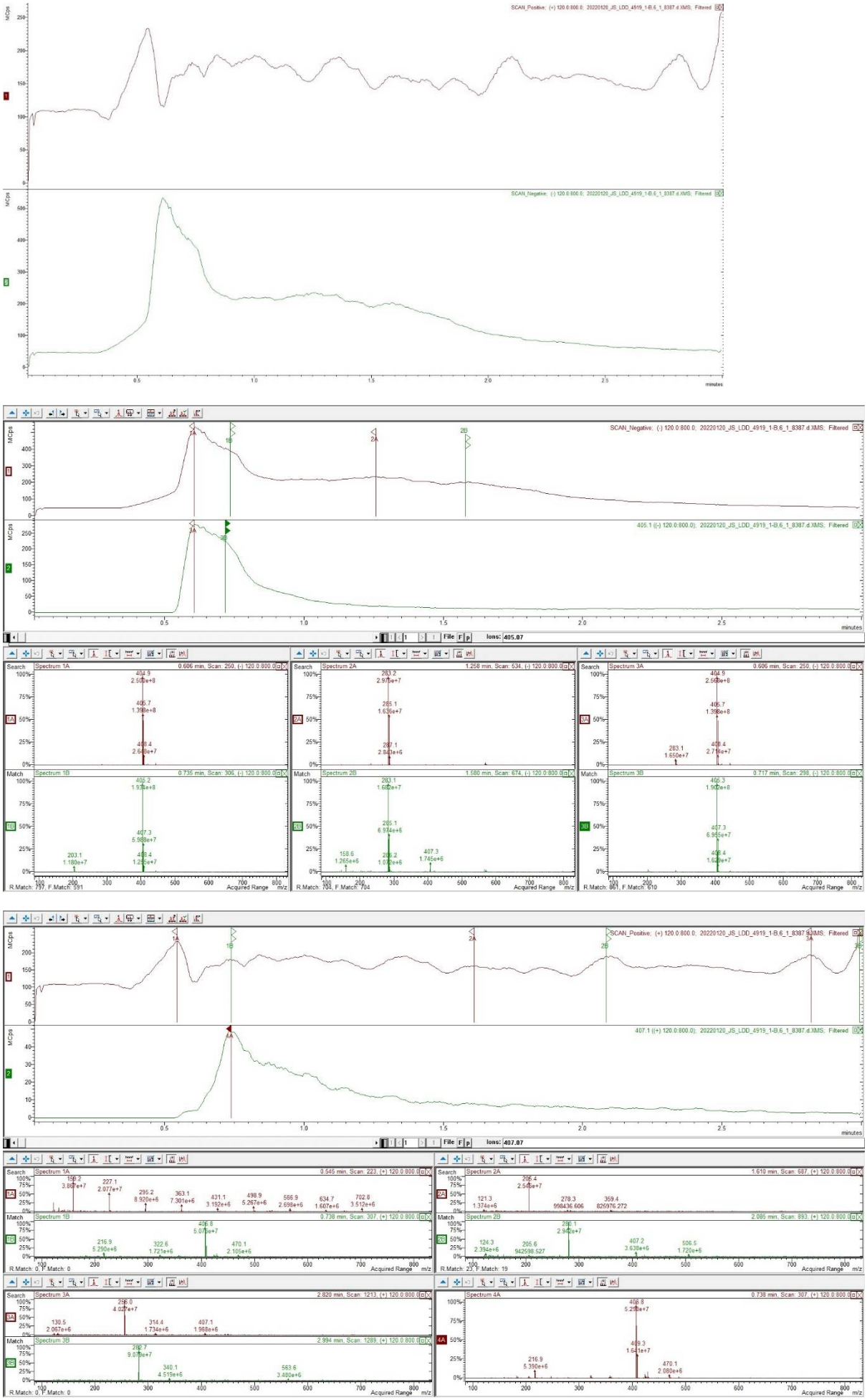


Figure S26. LC/MS spectrum of compound 14a

6-chloro-5-((1-propionylpiperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14b) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13b** afforded **14b**. red purple powder. **14b** fully dissolved in 600 μL of the methanol-d4 and a drop of acetone. Yield 30%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.06 - 1.16 (m, 3 H) 1.42 - 1.56 (m, 1 H) 1.56 - 1.64 (m, 1 H) 2.00 - 2.15 (m, 2 H) 2.44 (q, J=7.48 Hz, 2 H) 2.82 (t, J=14.20 Hz, 1 H) 3.22 (t, J=11.68 Hz, 1 H) 3.98 (d, J=15.11 Hz, 1 H) 4.51 (d, J=14.66 Hz, 1 H) 4.67 (s, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 175.44, 173.79, 173.41, 149.41, 144.47, 143.58, 138.12, 121.97, 119.29, 50.62, 43.92, 40.20, 25.95, 8.64, other peaks are impurities and acetone; LC/MS (ESI, m/z) 403.3 $[M - H]^-$ 405.4 $[M + H]^+$.

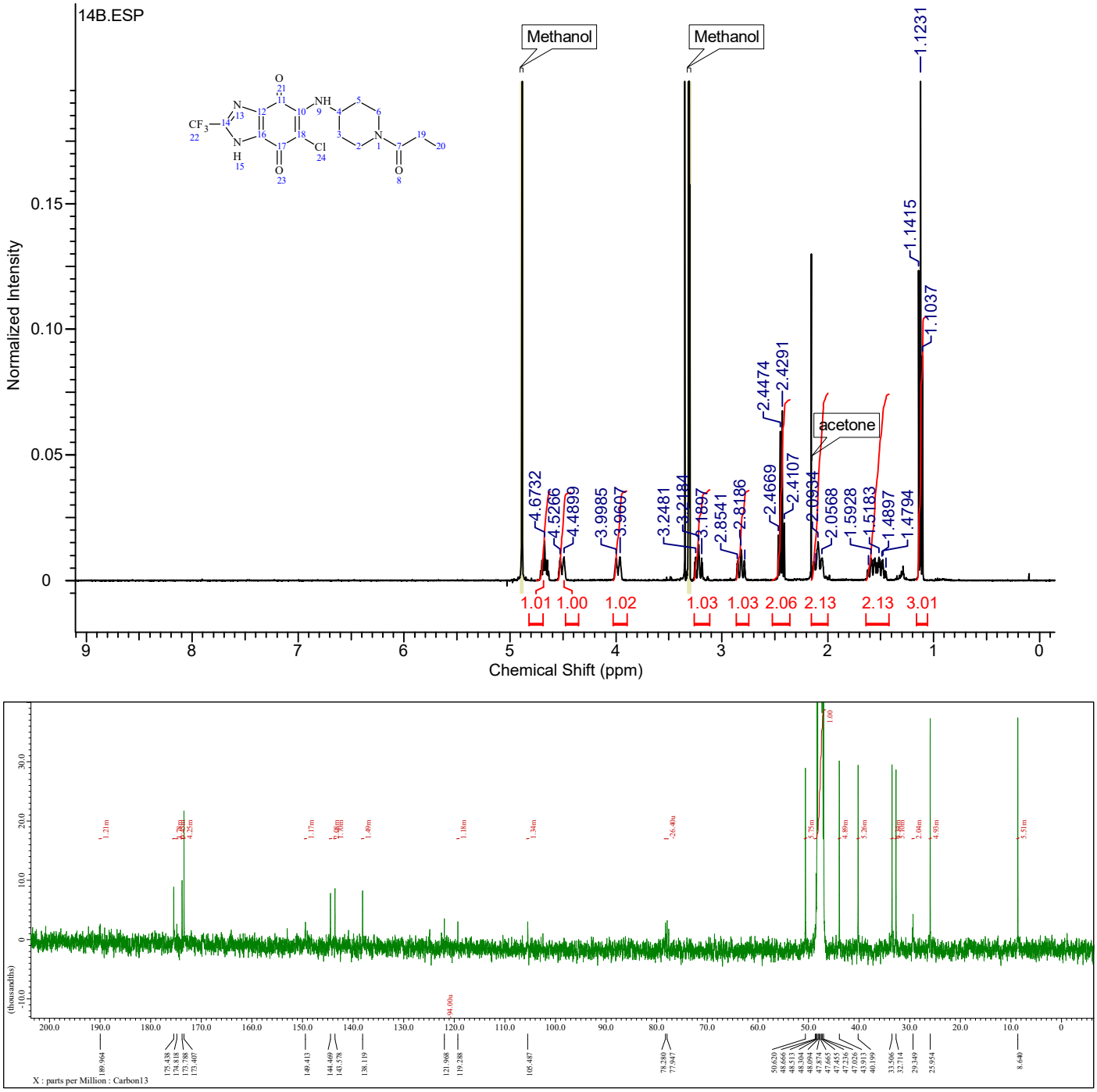


Figure S27. 1D 1H and 13C NMR spectrum of compound 14b

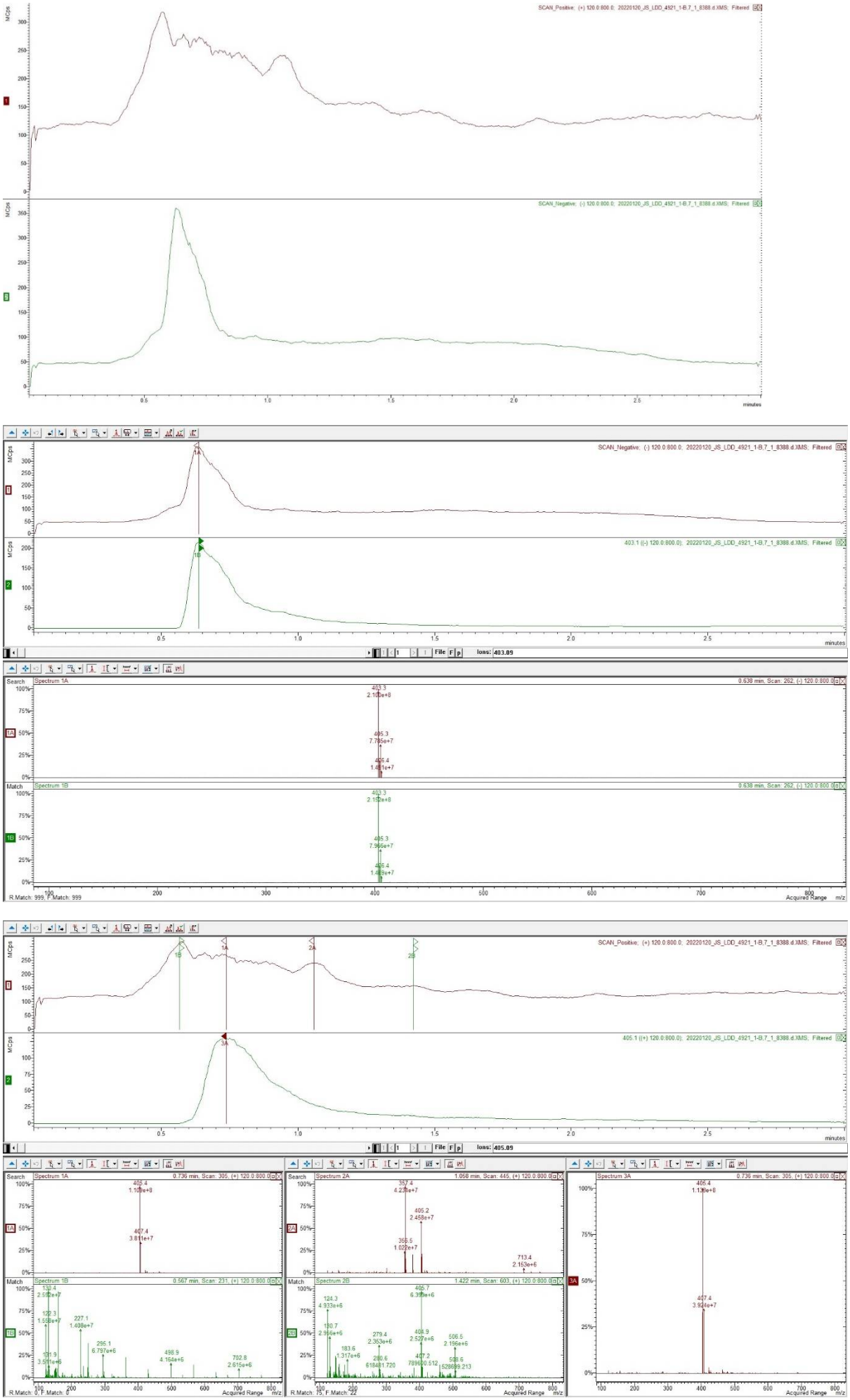
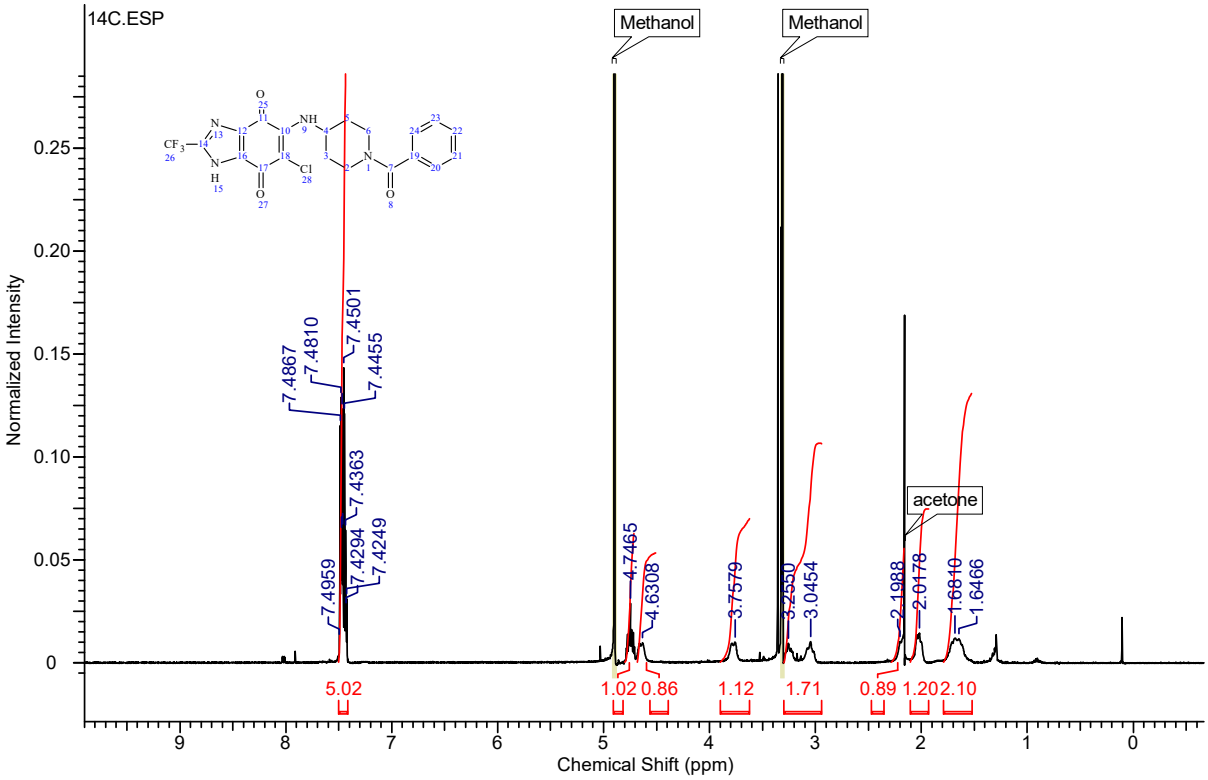


Figure S28. LC/MS spectrum of compound 14b

5-((1-benzoylpiperidin-4-yl)amino)-6-chloro-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14c) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13c** afforded **14c**. purple powder. Yield 43%; **14c** fully dissolved in 600 μL of the methanol-d4 and a drop of acetone. ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.66 (d, J=13.74 Hz, 2 H) 2.02 (br. s., 1 H) 2.20 (br. s., 1 H) 3.05 (br. s., 1 H) 3.26 (br. s., 1 H) 3.76 (br. s., 1 H) 4.63 (br. s., 1 H) 4.75 (s, 1 H) 7.42 - 7.50 (m, 5 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.56, 171.21, 146.20, 143.85, 141.35, 136.89, 135.64, 129.76, 129.37, 128.41, 126.51, 50.88, 46.23, 40.68, 2 carbon peaks in 173.56 ppm overlapped and peaks in 33 and 32 ppm are acetone; LC/MS (ESI, m/z) 451.5 $[M - H]^-$ 453.1 $[M + H]^+$.



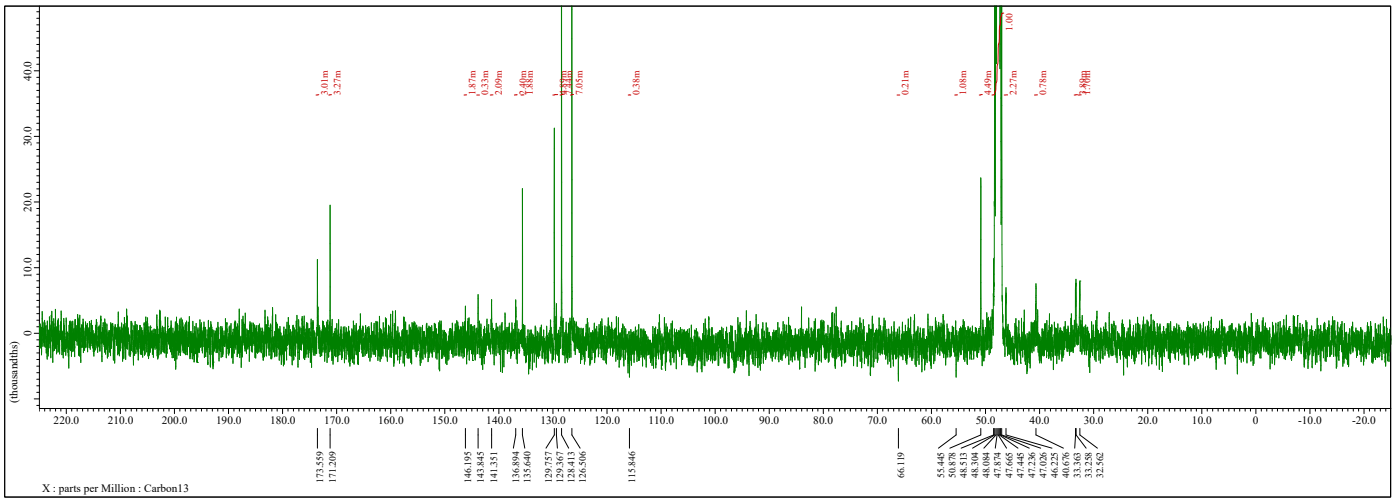


Figure S29. 1D 1H and 13C NMR spectrum of compound 14c

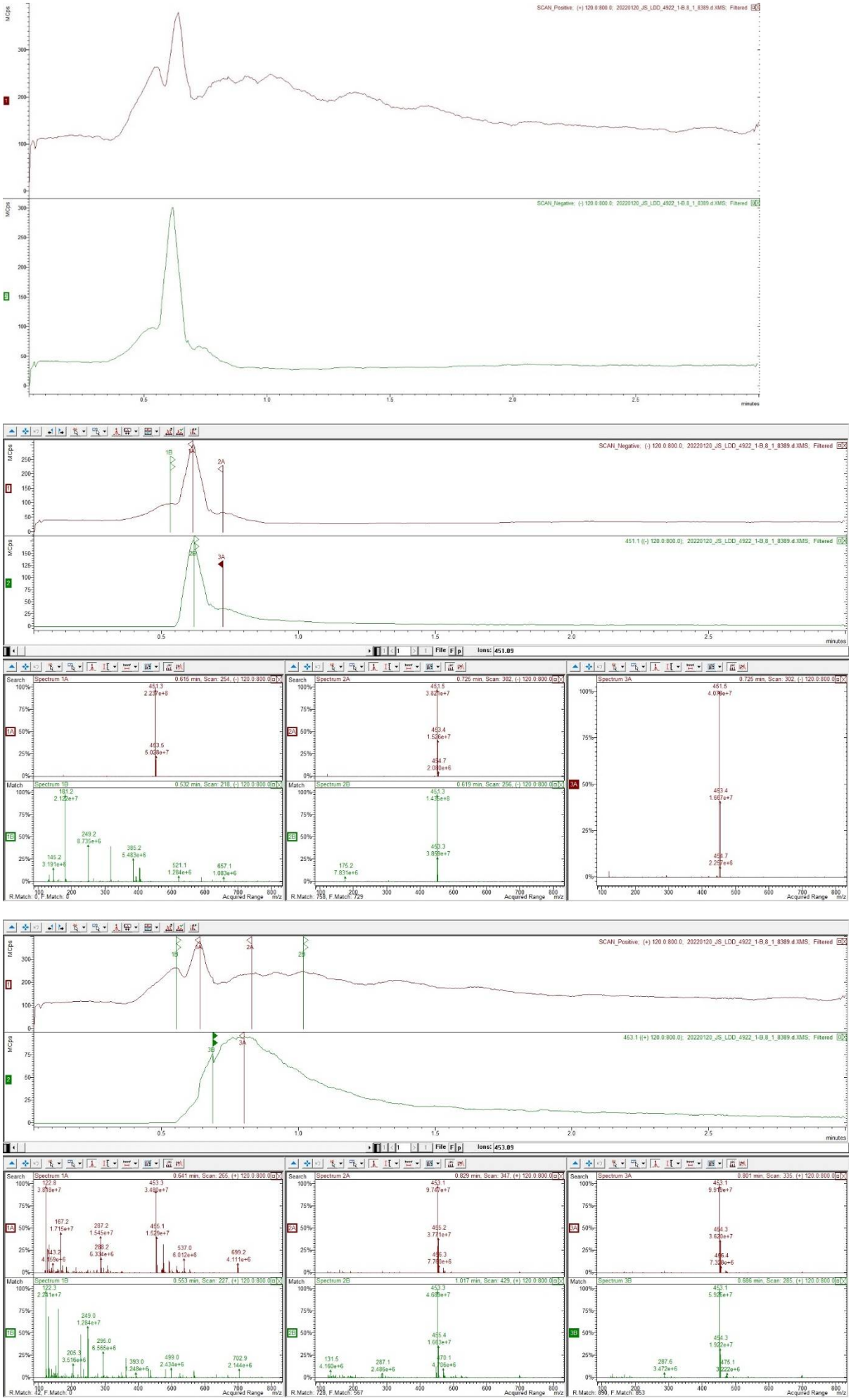
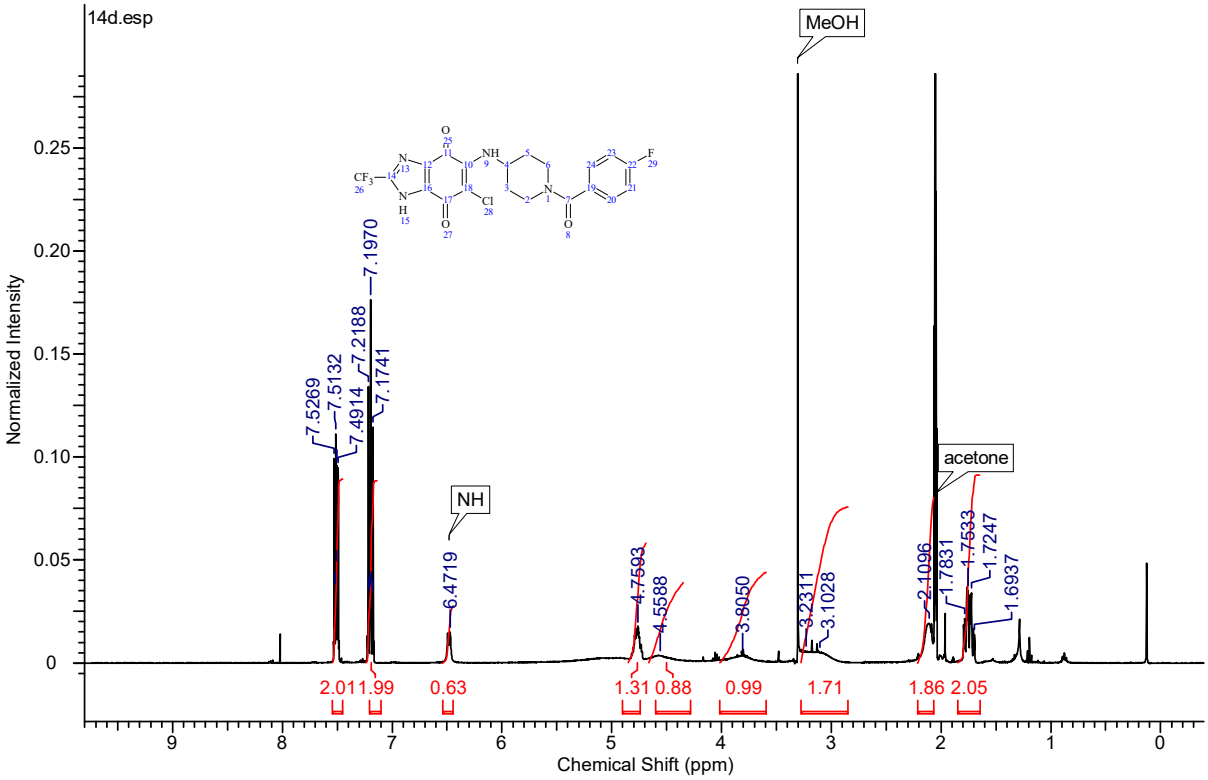


Figure S30. LC/MS spectrum of compound 14c

6-chloro-5-((1-(4-fluorobenzoyl)piperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14d)

Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13d** afforded **14d**. red purple powder. **14d** fully dissolved in 600 μL of the acetone-d6 and a drop of methanol. Yield 67%; m.p. 205-206 ℃; ^{1}H NMR (400 MHz, ACETONE-d6) δ ppm 1.65 - 1.85 (m, 2 H) 2.11 (br. s., 2 H) 2.98 - 3.28 (m, 2 H) 3.80 (br. s., 1 H) 4.56 (br. s., 1 H) 4.76 (br. s., 1 H) 6.47 (br. s., 1 H) 7.14 - 7.24 (m, 2 H) 7.45 - 7.54 (m, 2 H); ^{13}C NMR (100 MHz, DMSO-d6) δ 174.06, 173.14, 168.60, 144.11, 137.82, 137.76, 133.13, 133.10, 129.79, 129.70, 116.05, 115.83, 105.68, 51.15, 49.12, 46.49, peak in 33 ppm is acetone; LC/MS (ESI, m/z) 469.4 [M - H]$^{-}$ 471.1 [M + H]$^{+}$.



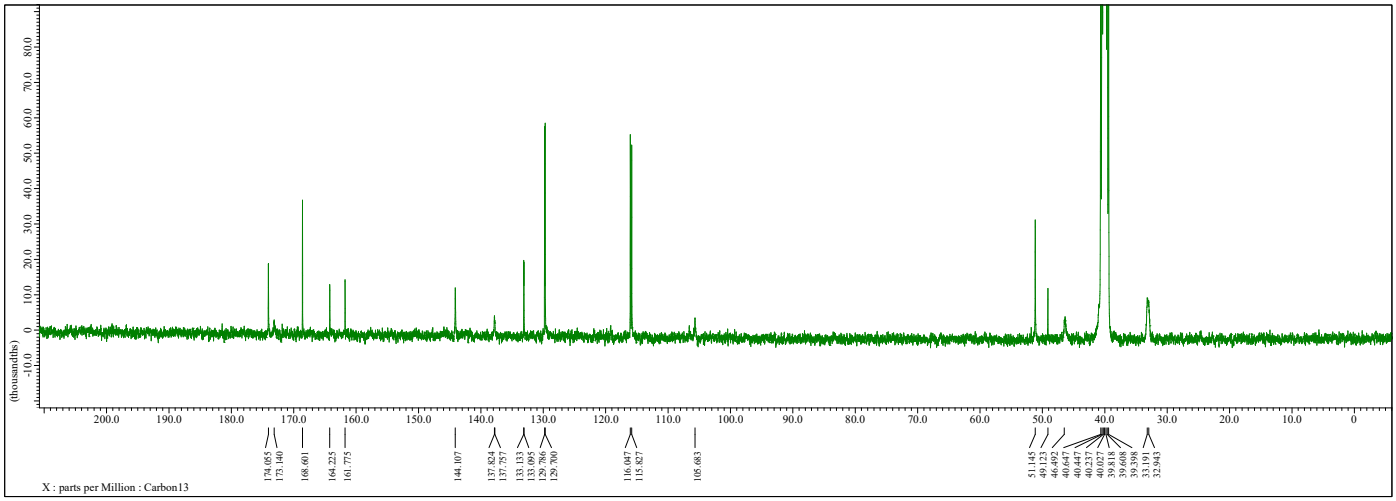


Figure S31. 1D 1H and 13C NMR spectrum of compound 14d

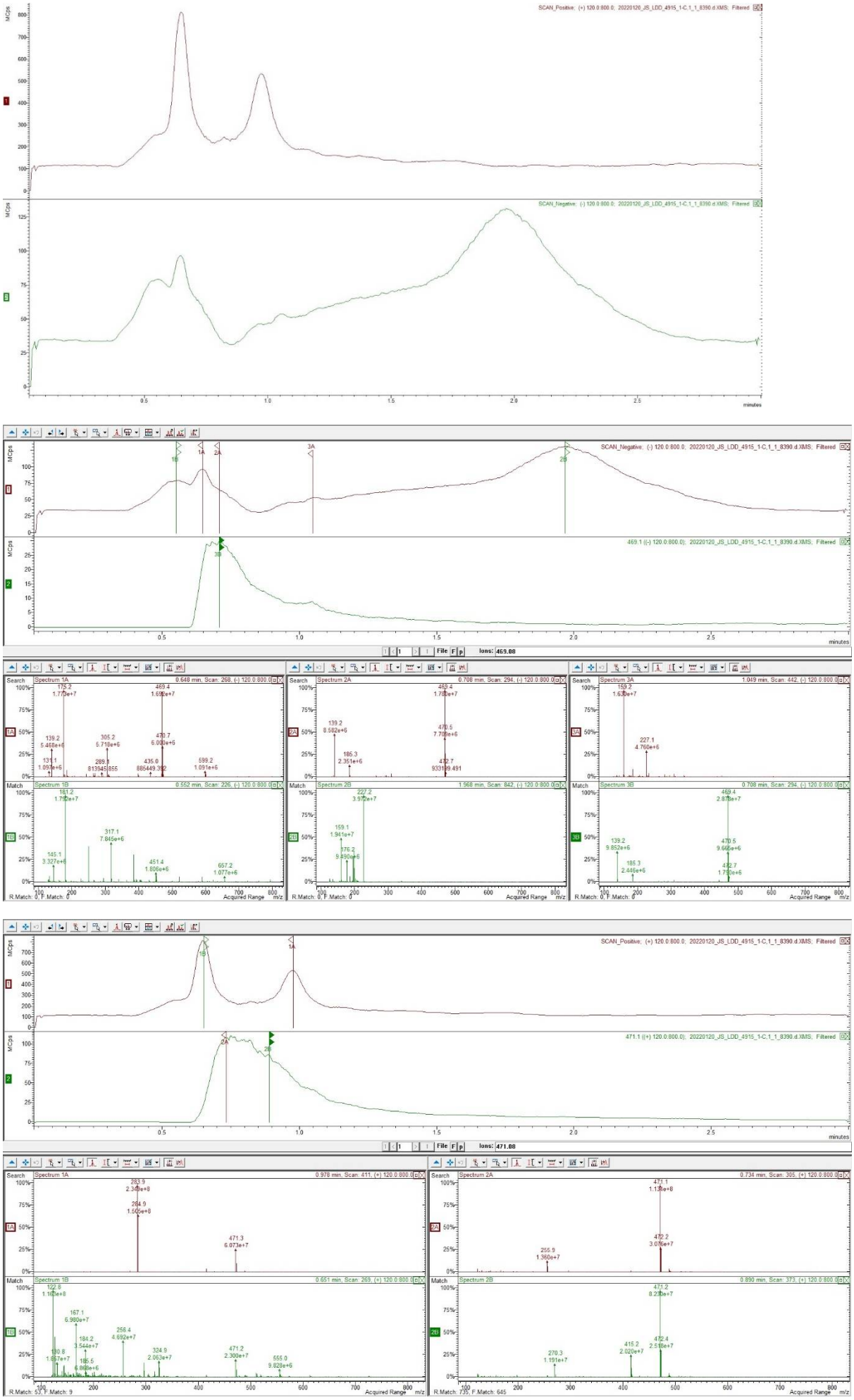
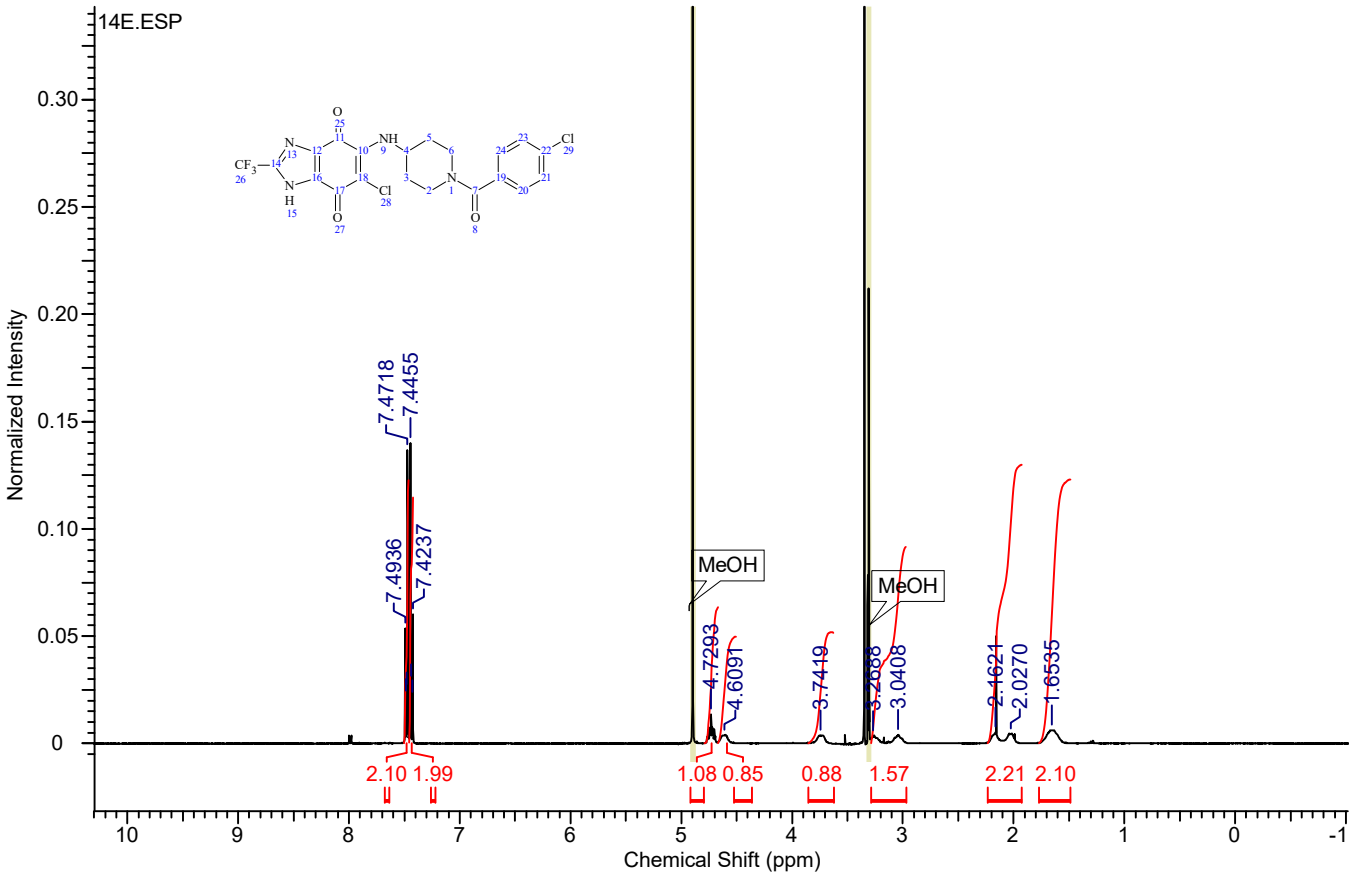


Figure S32. LC/MS spectrum of compound 14d

6-chloro-5-((1-(4-chlorobenzoyl)piperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14e)

Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13e** afforded **14e**. purple powder. Yield 77%; m.p. 249-251 ℃; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.65 (br. s., 2 H) 2.03 (br. s., 1 H) 2.16 (br. s., 1 H) 3.04 (br. s., 1 H) 3.27 (br. s., 1 H) 3.74 (br. s., 1 H) 4.61 (br. s., 1 H) 4.73 (s, 1 H) 7.42 - 7.45 (m, 2 H) 7.46 - 7.50 (m, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.72, 173.46, 170.02, 143.95, 141.67, 136.97, 135.70, 134.19, 128.61, 128.37, 50.84, 46.27, 40.77, 2 carbon peaks in 141.67 ppm are overlapped; LC/MS (ESI, m/z) 485.3 [M - H]$^{-}$ 487.1 [M + H]$^{+}$.



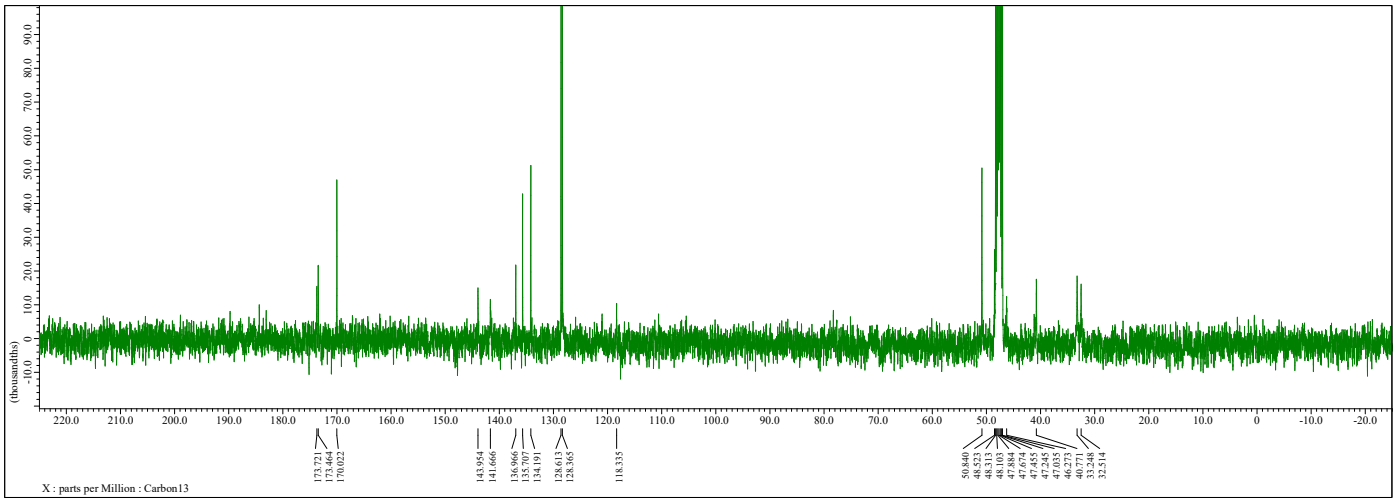


Figure S33. 1D 1H and 13C NMR spectrum of compound 14e

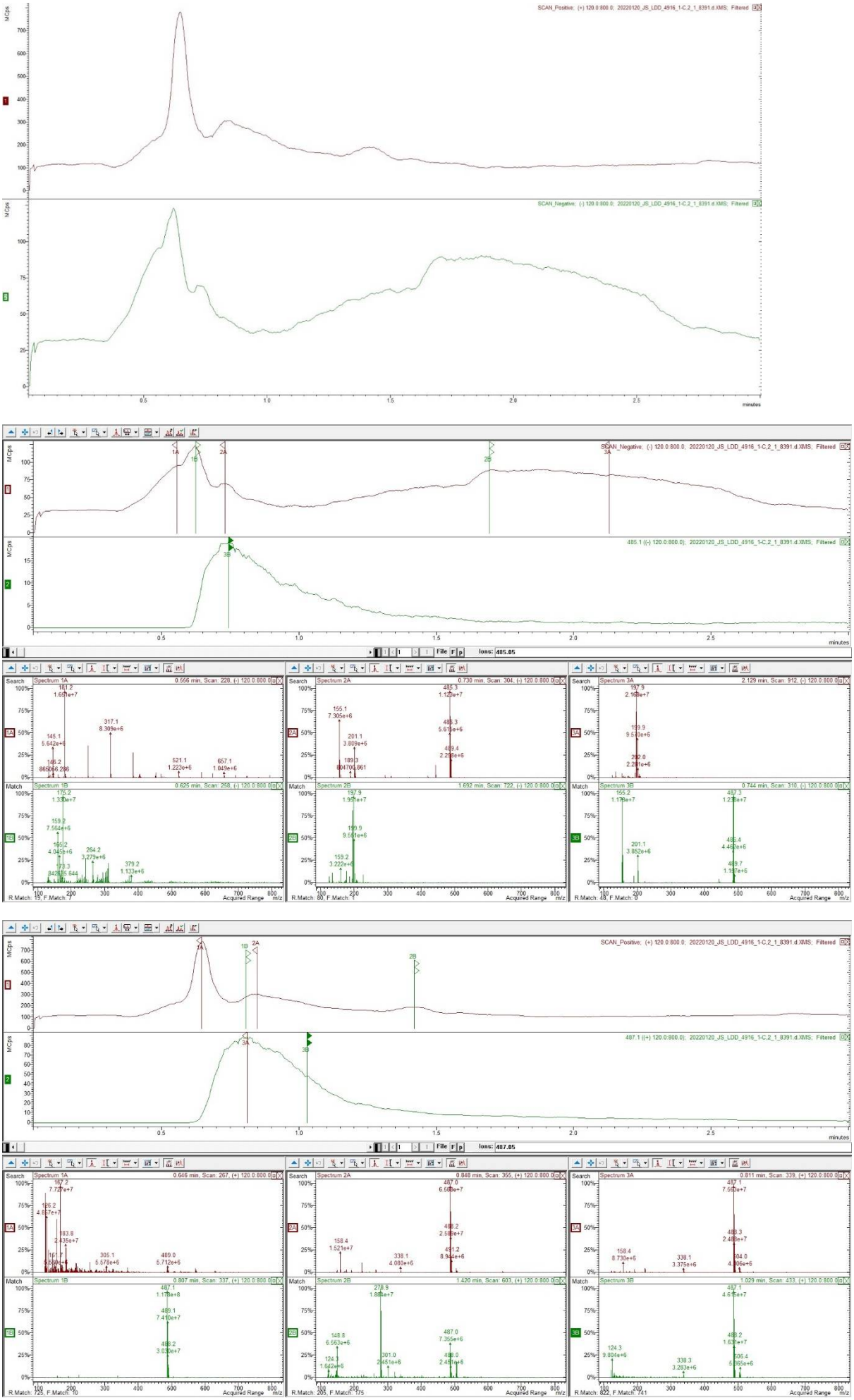
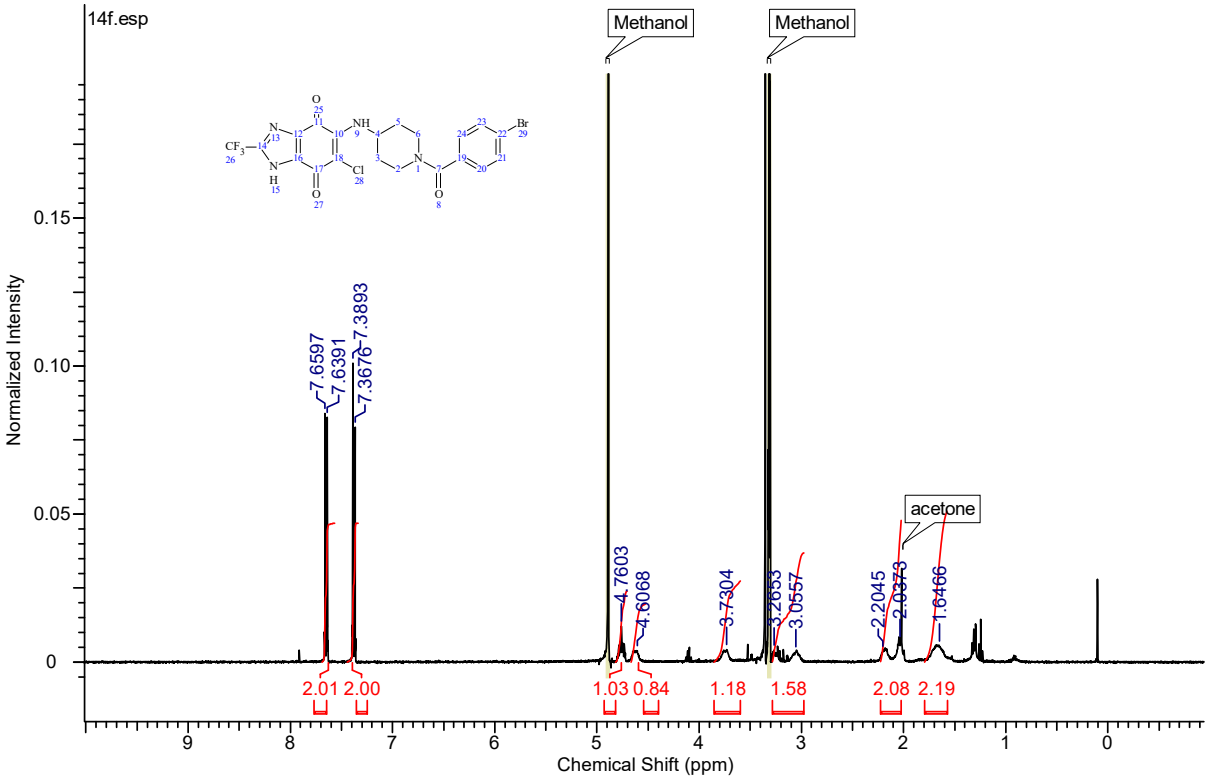


Figure S34. LC/MS spectrum of compound 14e

5-((1-(4-bromobenzoyl)piperidin-4-yl)amino)-6-chloro-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14f)

Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13f** afforded **14f**. purple powder. **14f** fully dissolved in 600 µL of the methanol-d4 and a drop of acetone. Yield 68%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.65 (br. s., 2 H) 2.04 (br. s., 1 H) 2.20 (br. s., 1 H) 3.06 (br. s., 1 H) 3.27 (br. s., 1 H) 3.73 (br. s., 1 H) 4.61 (br. s., 1 H) 4.76 (s, 1 H) 7.38 (d, J=8.70 Hz, 2 H) 7.65 (d, J=8.24 Hz, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.46, 173.36, 170.07, 143.99, 141.08, 138.59, 136.80, 134.63, 131.63, 128.51, 123.81, 120.86, 118.17, 115.484, 50.88, 46.25, 40.72, other peaks are impurities; LC/MS (ESI, m/z) 529.2 [M - H]$^{-}$ 532.9 [M + H]$^{+}$.



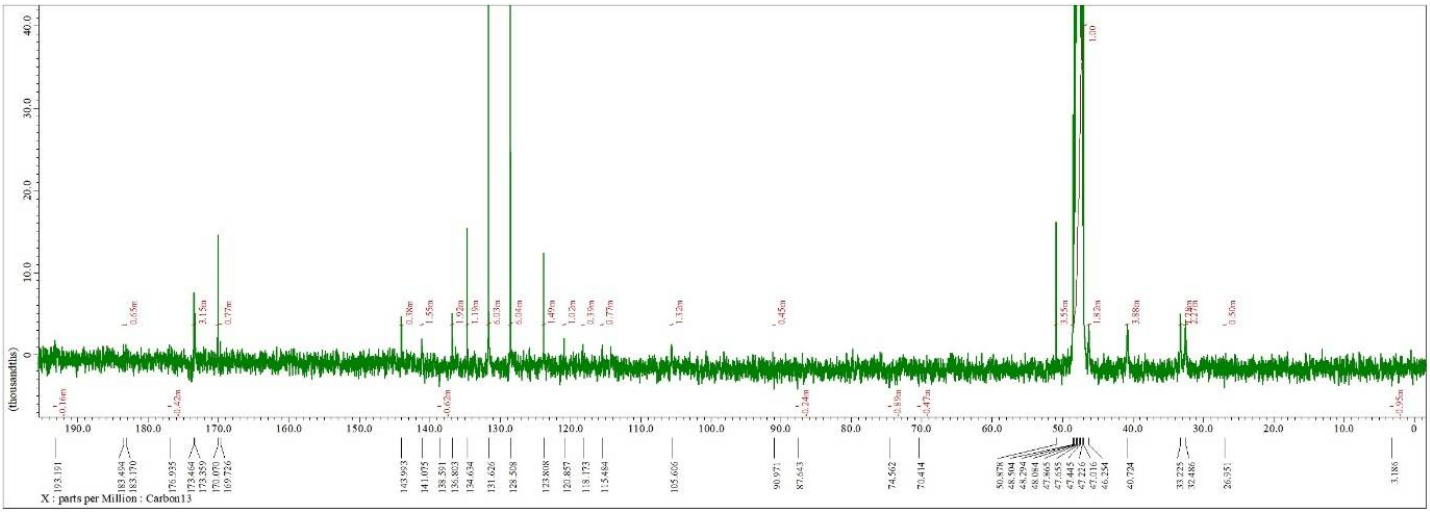


Figure S35. 1D 1H and 13C NMR spectrum of compound 14f

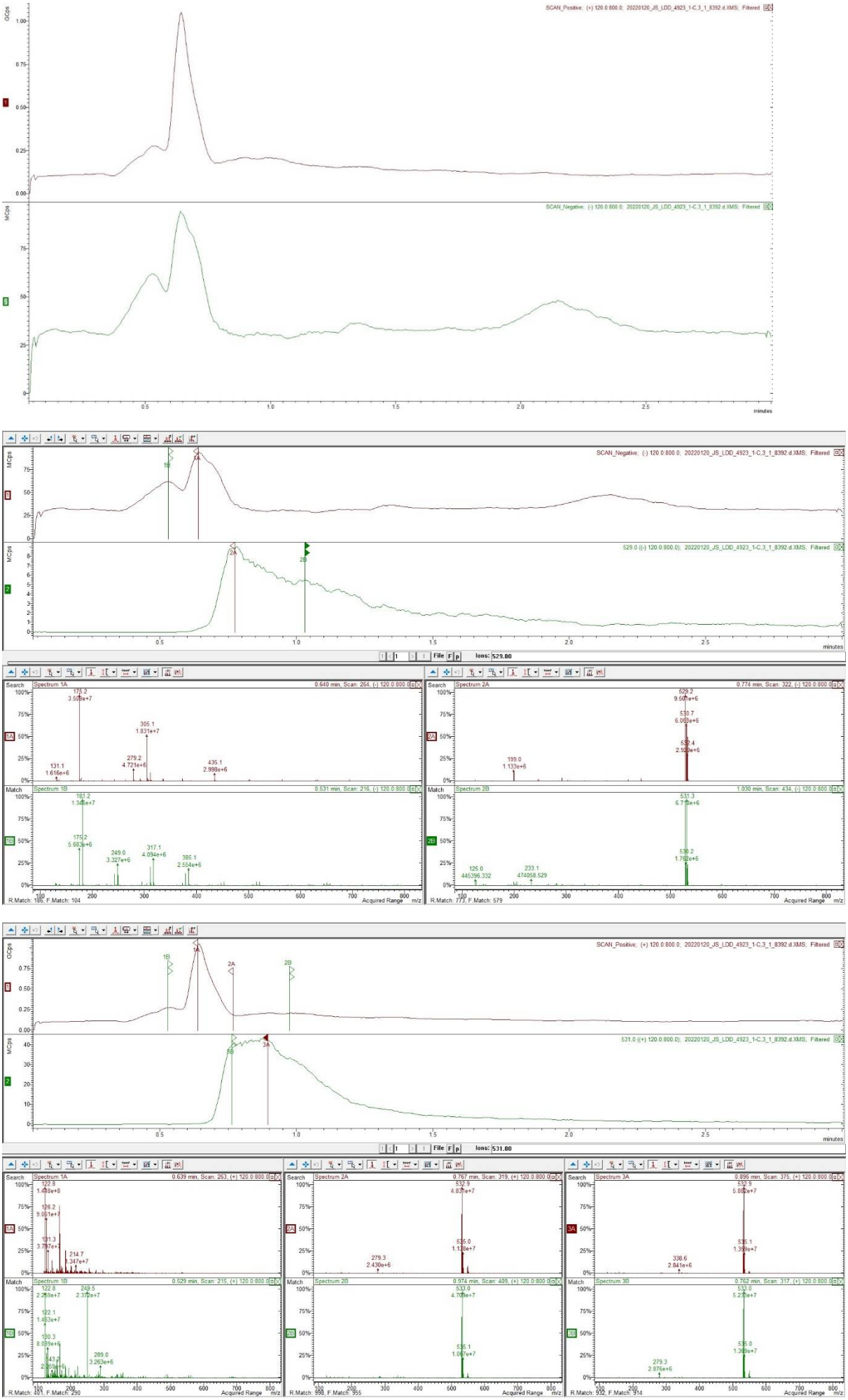


Figure S36. LC/MS spectrum of compound 14f

6-chloro-5-((1-(3,4-dichlorobenzoyl)piperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14g)
Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13g** afforded **14g**. purple powder. Yield 66%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.68 (br. s., 2 H) 2.03 (br. s., 1 H) 2.18 (br. s., 1 H) 3.05 (br. s., 1 H) 3.28 (br. s., 1 H) 3.72 (br. s., 1 H) 4.60 (br. s., 1 H) 4.76 (s, 1 H) 7.38 (dd, J=8.24, 1.83 Hz, 1 H) 7.58 - 7.68 (m, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 174.24, 173.44, 168.49, 144.06, 142.34, 137.26, 135.90, 133.71, 132.55, 130.69, 128.84, 126.41, 123.50, 122.12, 118.55, 50.77, 46.21, 40.87, 40.72, other peaks are impurities; LC/MS (ESI, m/z) 519.2 [M - H]$^{-}$ 521.0 [M + H]$^{+}$.

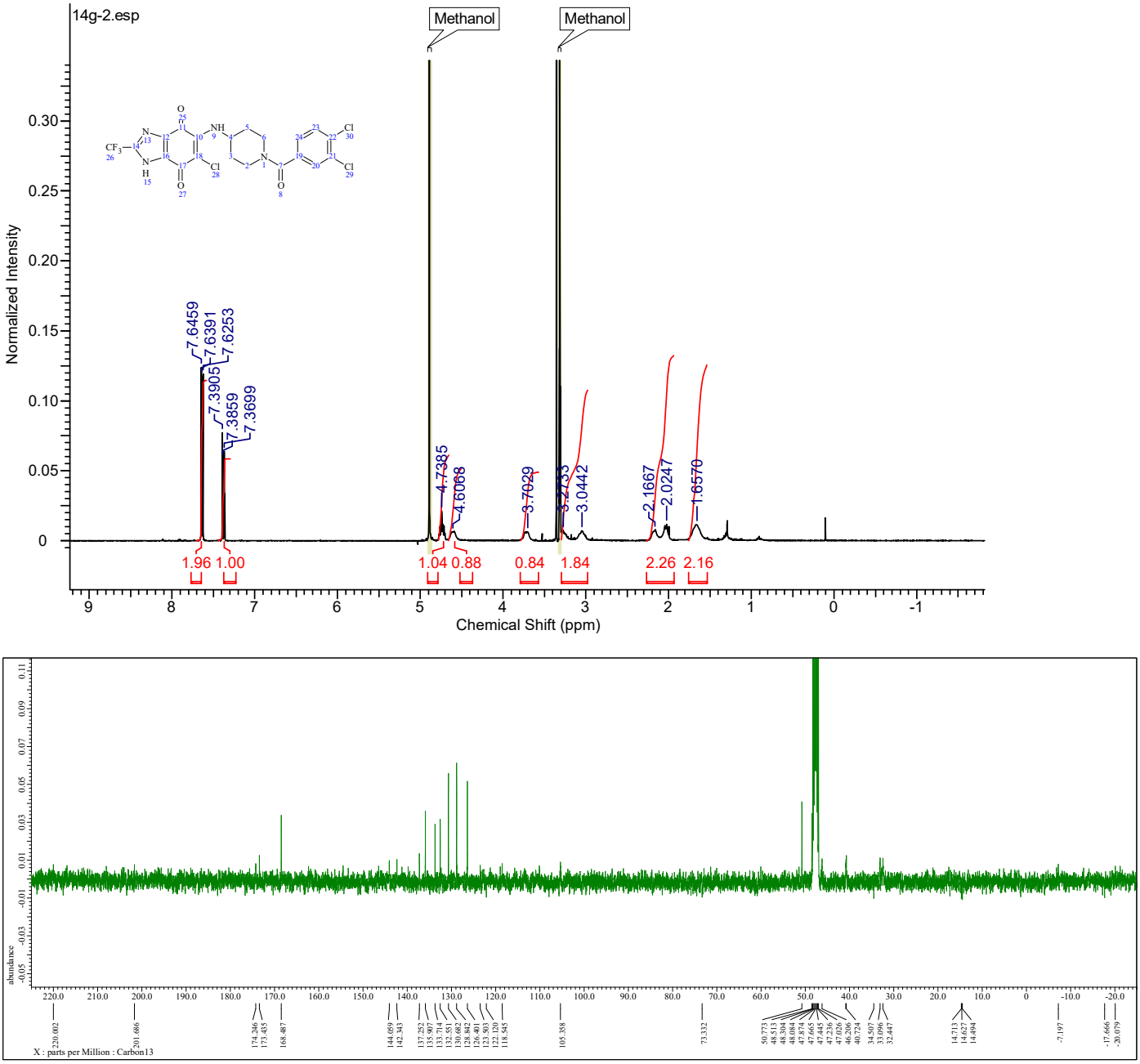


Figure S37. 1D 1H and 13C NMR spectrum of compound 14g

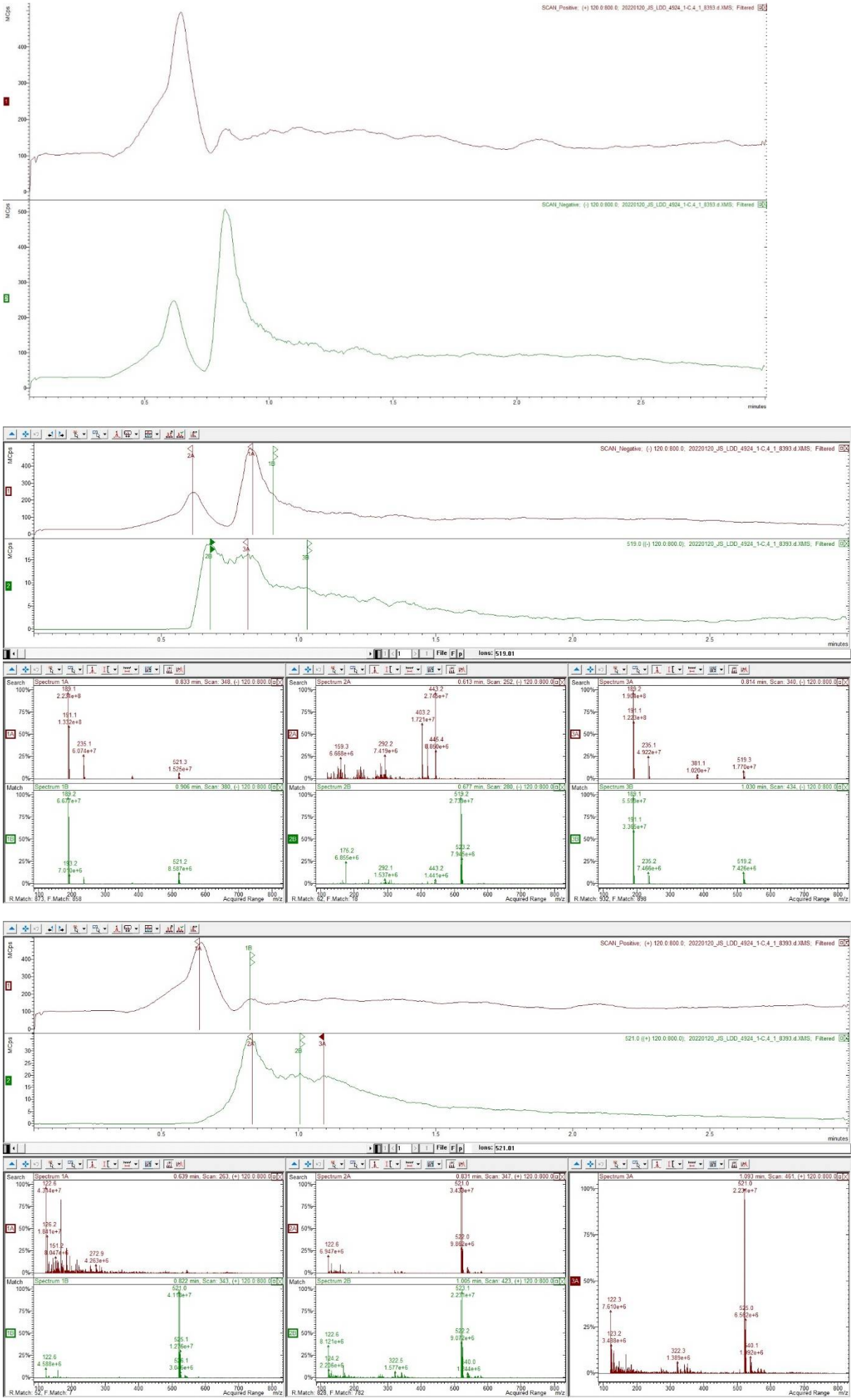
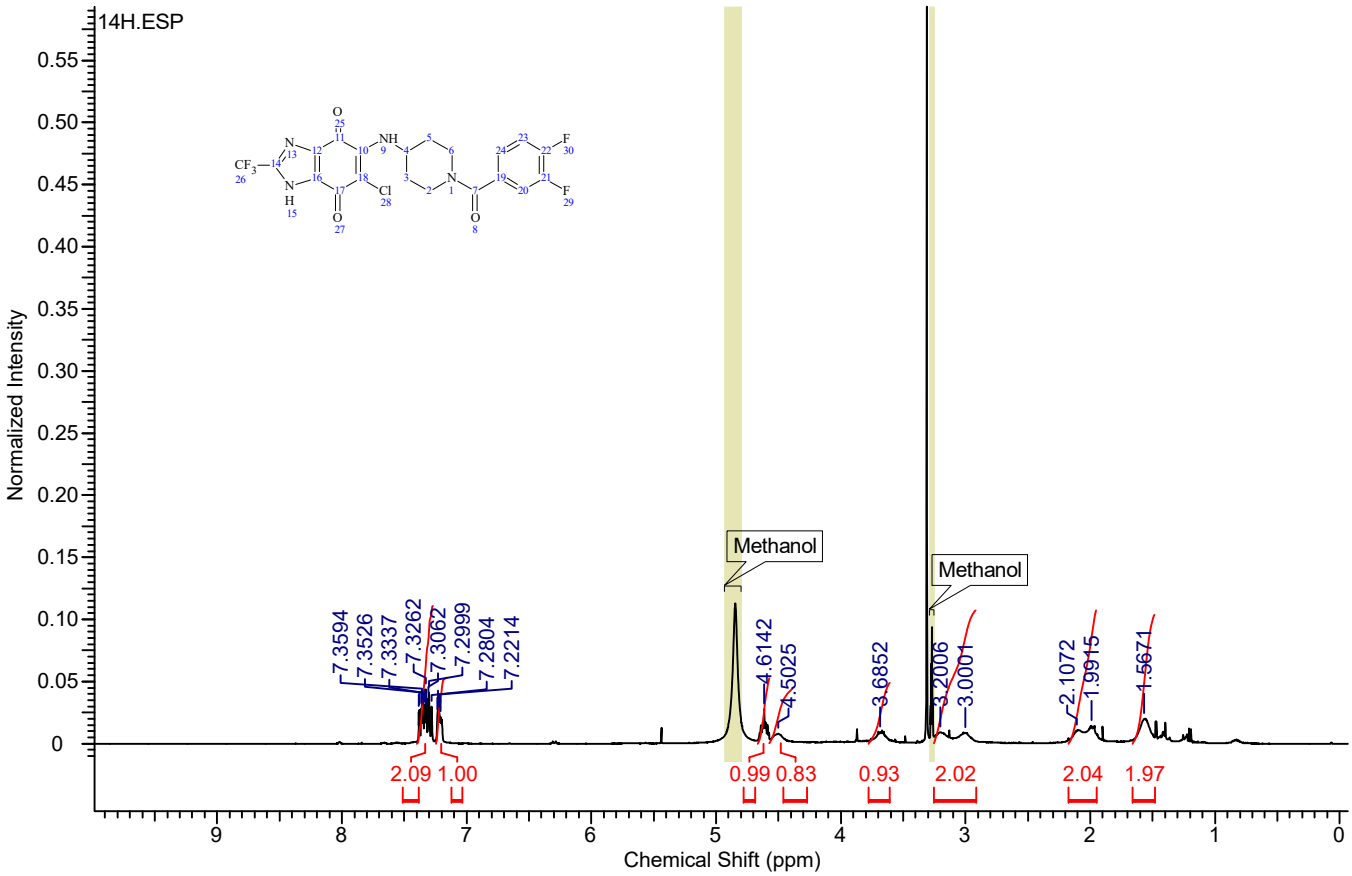


Figure S38. LC/MS spectrum of compound 14g

6-chloro-5-((1-(3,4-difluorobenzoyl)piperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14h)

Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13h** afforded **14h**. red purple powder. Yield 67%; m.p. 204-205 ℃; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.57 (br. s., 2 H) 1.99 (br. s., 1 H) 2.11 (br. s., 1 H) 3.00 (br. s., 1 H) 3.20 (br. s., 1 H) 3.69 (br. s., 1 H) 4.50 (br. s., 1 H) 4.61 (s, 1 H) 7.18 - 7.24 (m, 1 H) 7.26 - 7.41 (m, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.33, 173.02, 168.73, 143.97, 140.64, 136.48, 132.83, 132.78, 123.74, 123.70, 120.67, 117.99, 117.63, 117.46, 116.47, 116.29, 50.91, 46.24, 40.87, peaks in 152~148 ppm are impurities; LC/MS (ESI, m/z) 487.4 [M - H]$^{-}$ 489.1 [M + H]$^{+}$.



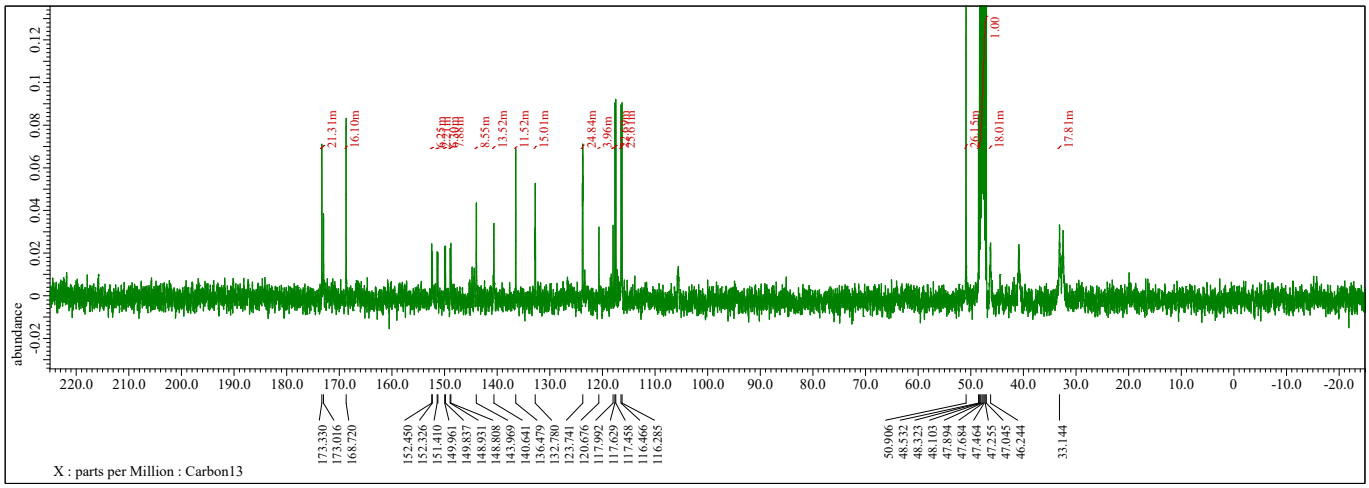


Figure S39. 1D 1H and 13C NMR spectrum of compound 14h

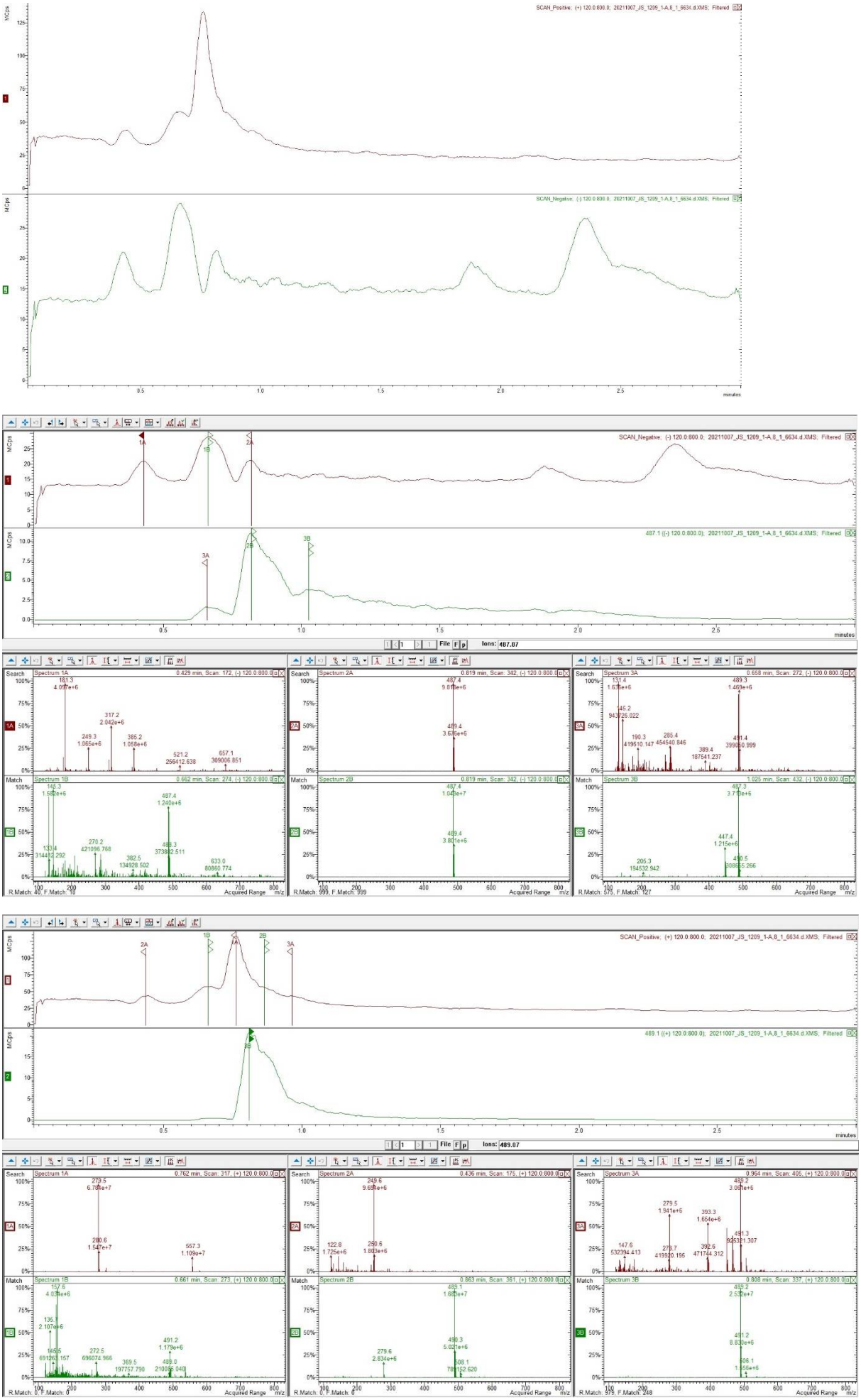


Figure S40. LC/MS spectrum of compound 14h

6-chloro-5-((1-(3-chloro-4-fluorobenzoyl)piperidin-4-yl)amino)-2-(trifluoromethyl)-1H-benzo[d]imidazole-4,7-dione (14i) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13i** afforded **14i**. red purple powder. **14i** fully dissolved in 600 µL of the acetone-d6 and a drop of methanol. Yield 75%; ^{1}H NMR (400 MHz, ACETONE-d6) δ ppm 1.68 - 1.84 (m, 2 H) 2.13 (br. s., 1 H) 3.4 (br. s., 2 H) 3.82 (br. s., 1 H) 4.51 (br. s., 1 H) 4.76 (br. s., 1 H) 7.35 - 7.41 (m, 1 H) 7.47 (ddd, J=8.47, 4.58, 2.06 Hz, 1 H) 7.58 - 7.62 (m, 1 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.49, 168.67, 136.98, 133.14, 129.42, 127.38, 127.31, 127.10, 121.10, 121.01, 120.92, 116.84, 116.73, 116.62, 50.79, 46.33, 41.43, 2 carbon peaks in 173.49 ppm overlapped and other peaks are impurities; LC/MS (ESI, m/z) 505.1 $[M - H]^-$ 503.3 $[M + H]^+$.

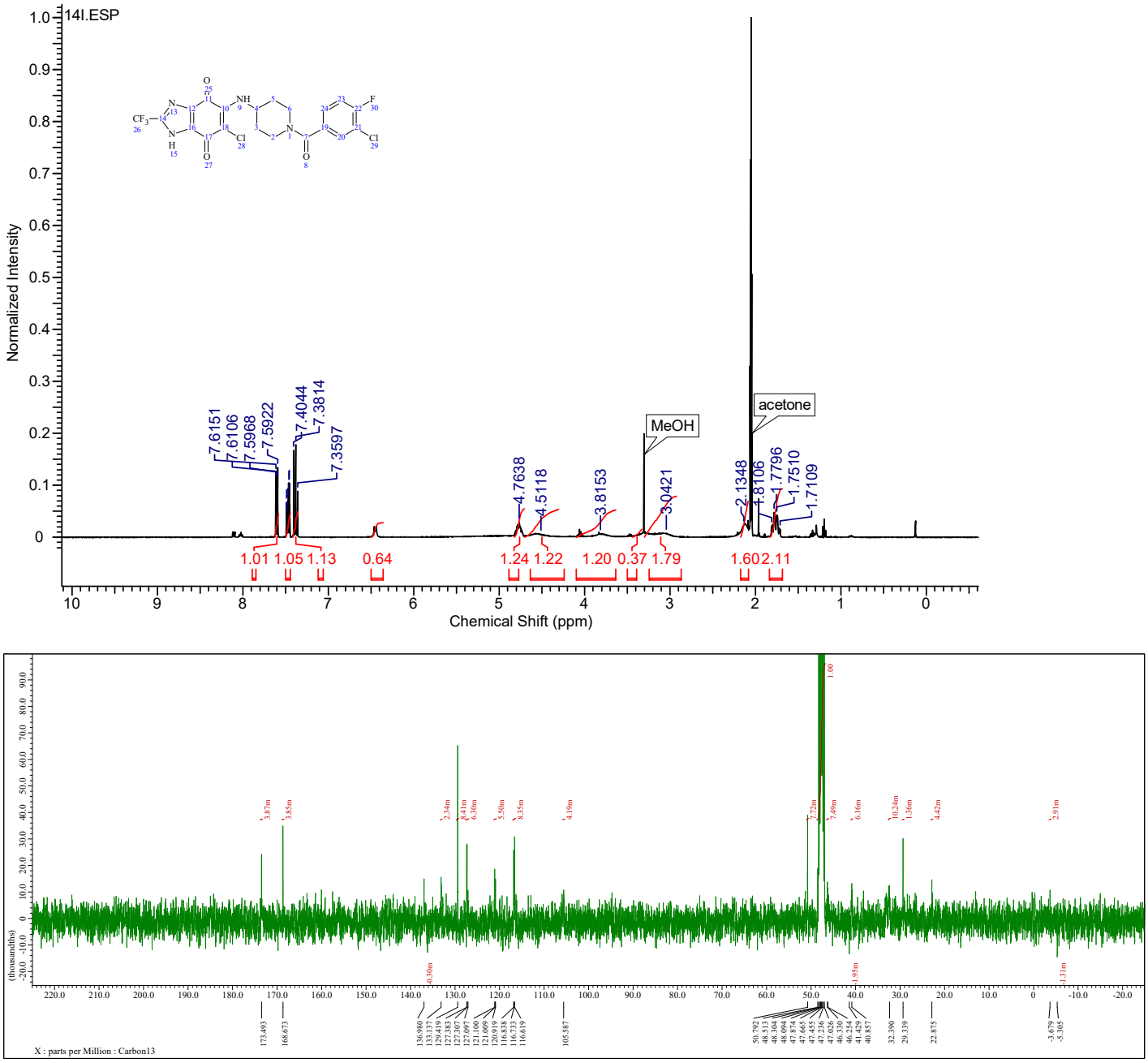


Figure S41. 1D 1H and 13C NMR spectrum of compound 14i

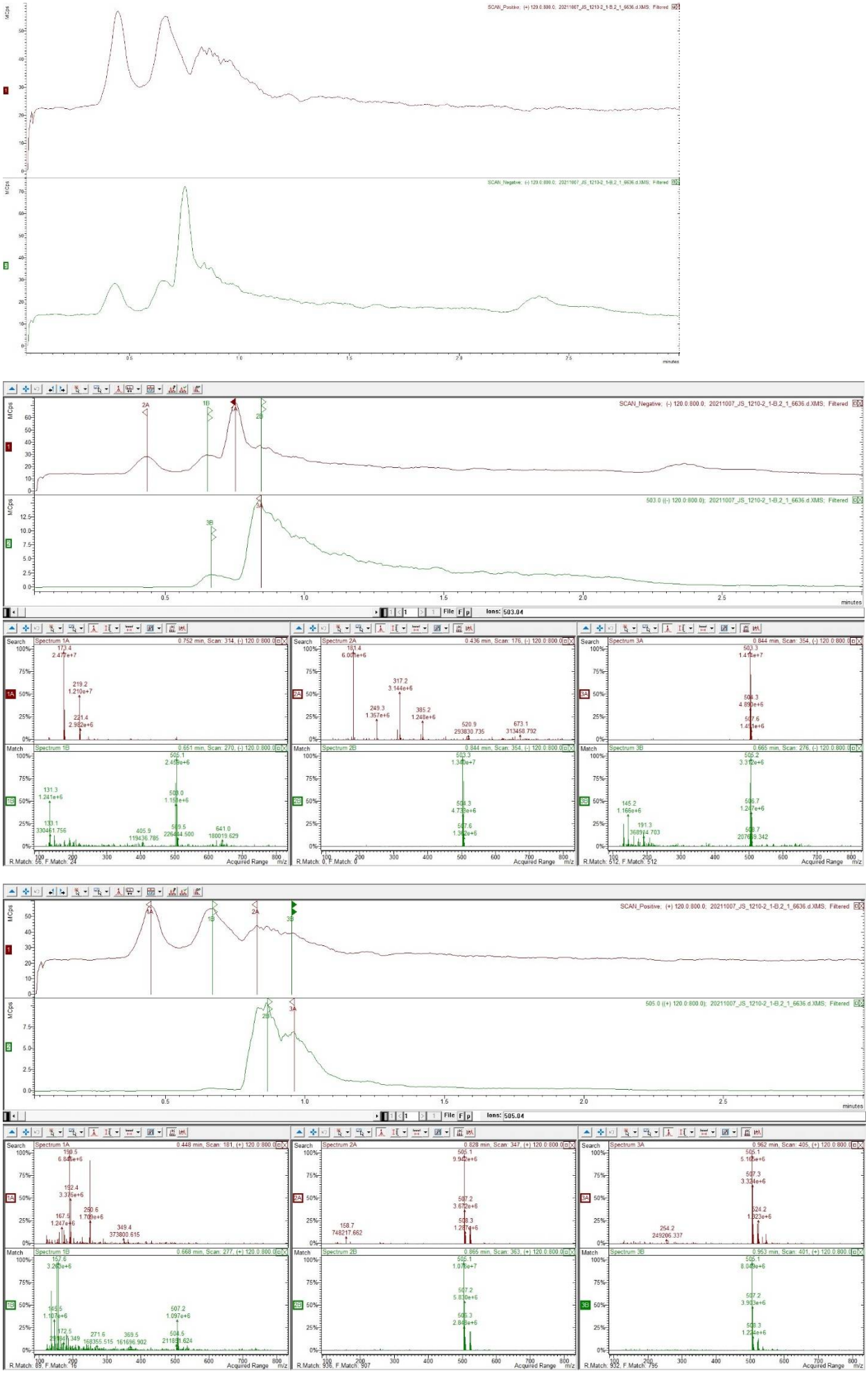
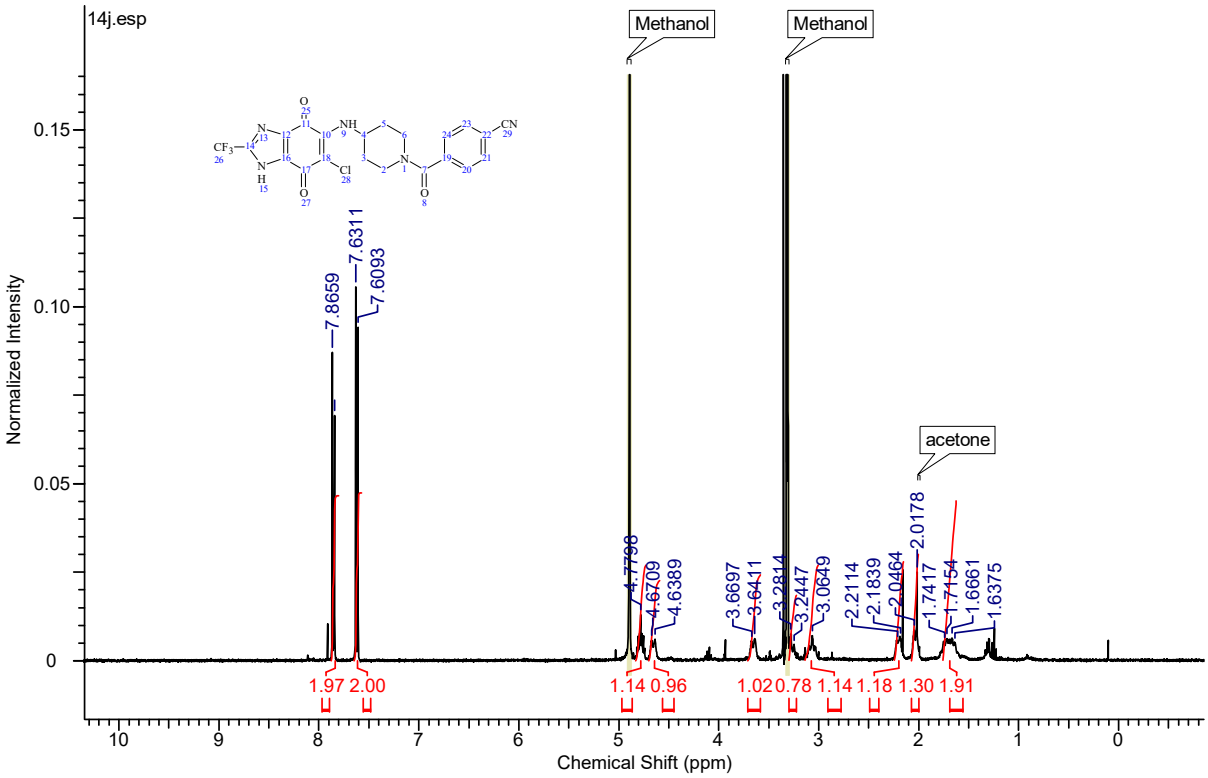


Figure S42. LC/MS spectrum of compound 14i

4-(4-((6-chloro-4,7-dioxo-2-(trifluoromethyl)-4,7-dihydro-1H-benzo[d]imidazol-5-yl)amino)piperidine-1-carbonyl)benzonitrile (14j) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13j** afforded **14j**. purple powder. **14j** fully dissolved in 600 μL of the methanol-d4 and a drop of acetone. Yield 70%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.62 - 1.76 (m, 2 H) 2.00 - 2.07 (m, 1 H) 2.20 (d, J=10.99 Hz, 1 H) 3.06 (br. s., 1 H) 3.22 - 3.30 (m, 1 H) 3.66 (d, J=11.45 Hz, 1 H) 4.65 (d, J=12.82 Hz, 1 H) 4.78 (s, 1 H) 7.62 (d, J=8.70 Hz, 2 H) 7.86 (d, J=8.24 Hz, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.55, 169.09, 143.89, 143.00, 142.10, 140.21, 137.57, 132.43, 129.39, 127.46, 117.72, 113.36, 50.66, 46.07, 40.65, 2 carbon peaks in 173.55 ppm overlapped and low peak signal in 142.10 ppm; LC/MS (ESI, m/z) 476.6 [M - H]$^{-}$ 478.3 [M + H]$^{+}$.



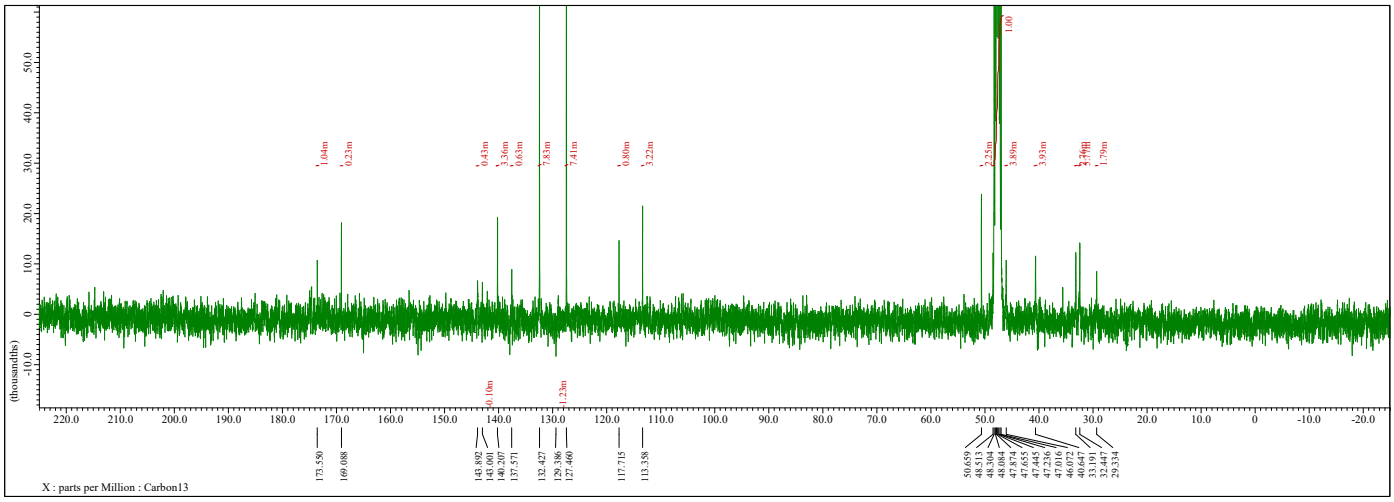


Figure S43. 1D 1H and 13C NMR spectrum of compound 14j

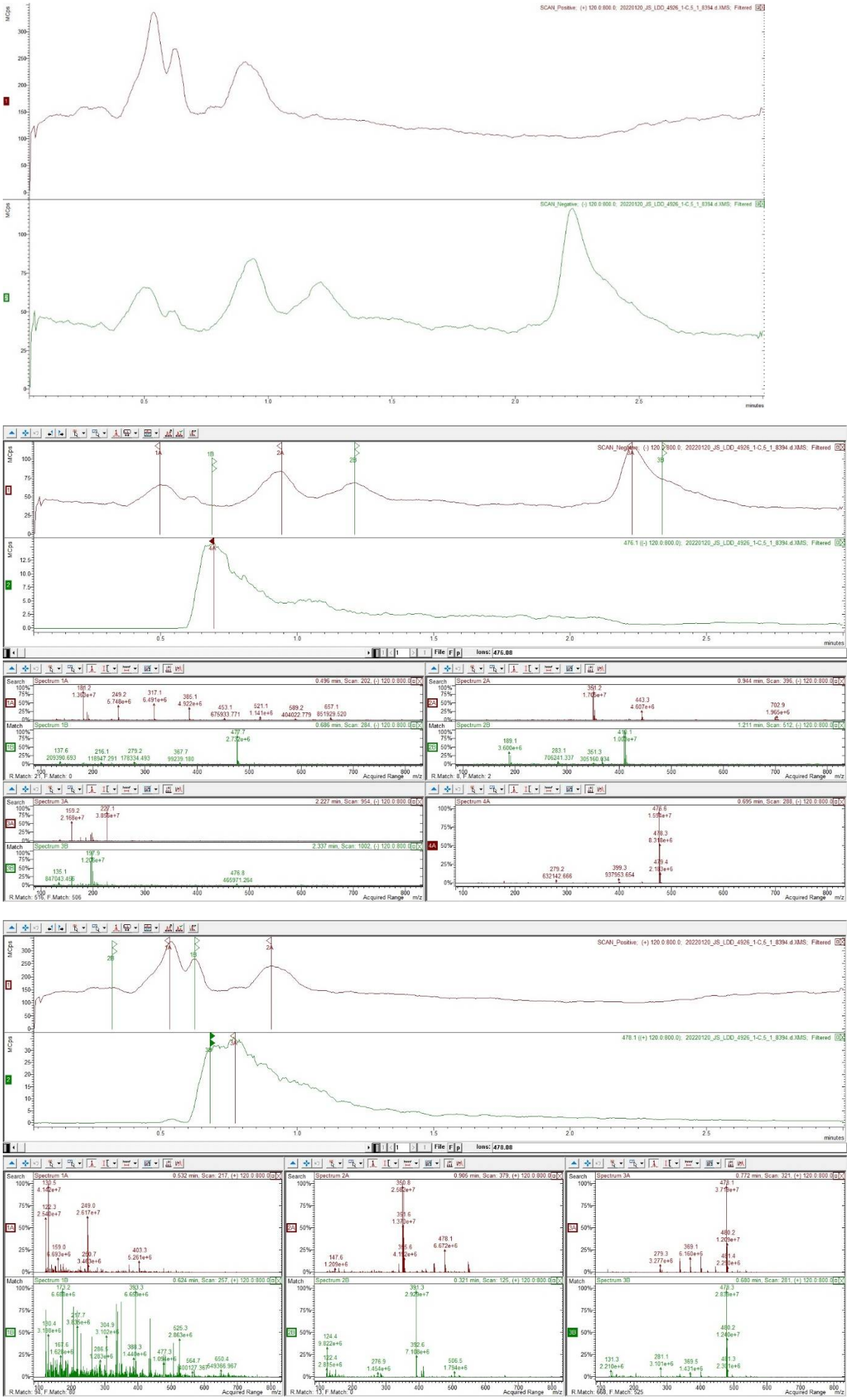


Figure S44. LC/MS spectrum of compound 14j

6-chloro-2-(trifluoromethyl)-5-((1-(4-(trifluoromethyl)benzoyl)piperidin-4-yl)amino)-1H-benzo[d]imidazole-4,7-dione (14k) Following the general procedure for the synthesis of **14a-k**, the substitution reaction of **9b** with **13k** afforded **14k**. purple powder. **14k** fully dissolved in 600 μL of the methanol-d4 and a drop of acetone. Yield 72%; ^{1}H NMR (400 MHz, METHANOL-d4) δ ppm 1.64 (d, J=6.87 Hz, 1 H) 1.72 (d, J=11.45 Hz, 1 H) 2.04 (br. s., 2 H) 2.21 (d, J=6.87 Hz, 1 H) 2.99 - 3.15 (m, 1 H) 3.15 - 3.29 (m, 1 H) 3.67 (d, J=12.37 Hz, 1 H) 4.65 (d, J=14.20 Hz, 1 H) 4.76 (s, 1 H) 7.64 (m, J=7.79 Hz, 2 H) 7.79 (m, J=8.24 Hz, 2 H); ^{13}C NMR (100 MHz, METHANOL-d4) δ 173.51, 169.56, 143.96, 139.61, 136.98, 131.58, 131.25, 127.26, 125.48, 125.44, 50.81, 46.14, 40.66, 2 carbon peaks in 173.51 ppm overlapped and 2 carbon peaks low signal in 140~145 ppm; LC/MS (ESI, m/z) 519.3 $[M - H]^-$ 521.4 $[M + H]^+$.

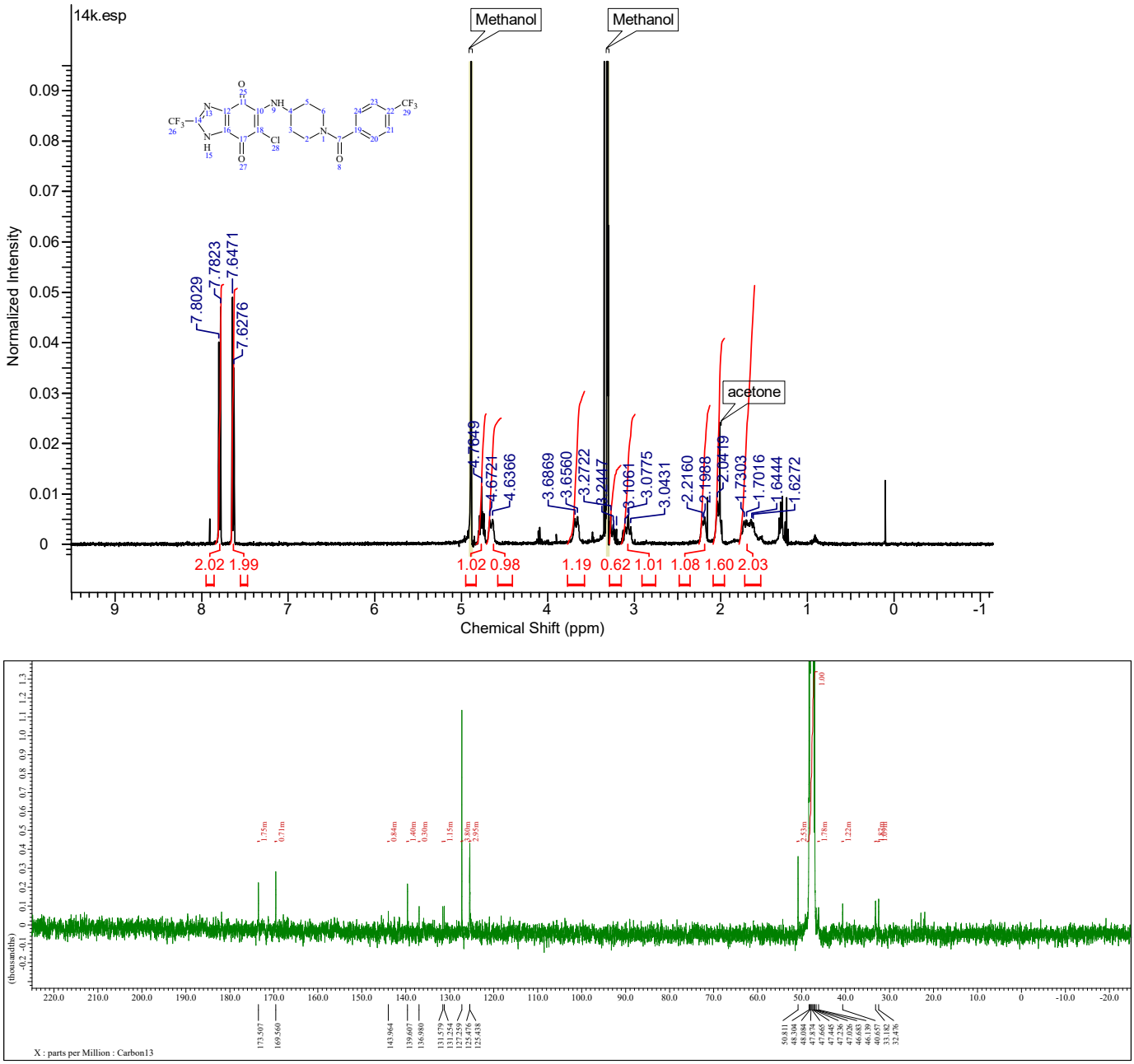


Figure S45. 1D 1H and 13C NMR spectrum of compound 14k

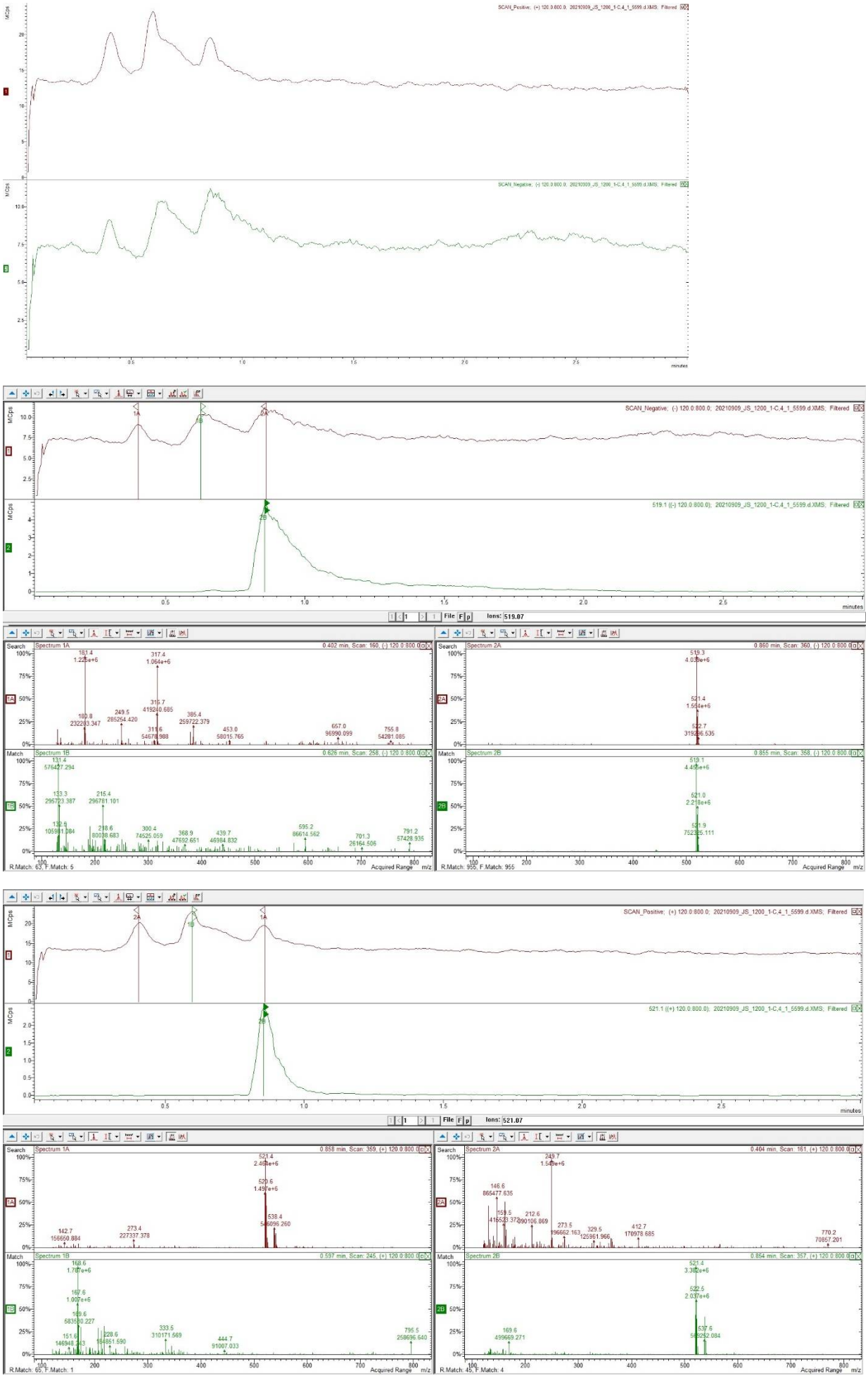


Figure S46. LC/MS spectrum of compound 14k

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