Supporting Information

A Systematic Evaluation of the Metabolism and Toxicity of Thiazolidinone and Imidazolidinone Heterocycles

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1. Characterization of FMMH derivatives

The chemical shifts are given in ppm, using the proton solvent residue signal ($CDCl_3$: δ 7.26; DMSO-d_6: δ 2.50; CD_3OD: δ 3.34) as a reference in the ^1H NMR spectrum. The deuterium coupled signal of the solvent was used as reference in the ^{13}C NMR spectrum ($CDCl_3$: δ 77.23; DMSO-d_6: δ 39.52; CD_3OD: δ 49.20). The following abbreviations were used to describe the signals: s = singlets, d = doublet, t = triplet, q = quartet, quin = quintet, m = multiplet, br = broad signal.

2-(4-oxo-2-thioxothiazolidin-3-yl) acetic acid (1). Compound **1** was synthesized following a literature procedure. Mp 146.5-148.5 °C (lit.[28] 145-148 °C). ^1H NMR (400 MHz, DMSO-d_6) δ 4.41 (s, 2H), 4.56 (s, 2H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 36.0, 44.8, 167.3, 173.8, 202.8. MS (ESI) *m/z* 189.9 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 190.9711, found: 190.9715.

(*Z*)-5-benzylidene-2-thioxothiazolidin-4-one (2). Orange solid. Yield = 58 %. Mp 209.9-210.2 °C (lit.[29] 208-210°C). ^1H NMR (400 MHz, DMSO-d_6) δ 7.48 – 7.62 (m, 5H), 7.65 (s, 1H), 13.85 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 125.6, 129.5,130.5, 130.7, 131.6, 133.0, 169.5, 195.8. MS (ESI) *m/z* 220.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 220.9969, found: 220.9974.

(*Z*)-5-propylidene-2-thioxothiazolidin-4-one (3). Yellow solid. Yield = 98 %. Mp 92.0-93.0 °C. ^1H NMR (400 MHz, DMSO-d_6) δ 1.07 (t, J = 8.0 Hz, 3H), 2.19 (quin, J = 8.0 Hz, 2H), 6.90 (t, J = 8.0 Hz, 1H), 12.37 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 12.6, 25.2, 126.6, 139.0, 166.6, 168.1. MS (ESI) *m/z* 172.3.

(*Z*)-2-(5-benzylidene-4-oxo-2-thioxothiazolidin-3-yl) acetic acid (4). Yellow solid. Yield = 62 %. Mp 246.8-247.1 °C (lit.[28] 247-248 °C). ^1H NMR (400 MHz, DMSO-d_6) δ 4.74 (s, 2H),

7.54 – 7.61 (m, 3H), 7.69 (m, 2H), 7.91 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 45.1, 121.9, 129.6, 130.8, 131.2, 132.8, 133.9, 166.4, 167.2, 193.3. MS (ESI) *m/z* 278.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 279.0024, found: 279.0027.

(*Z*)-5-(2-hydroxybenzylidene)-2-thioxothiazolidin-4-one (22). Yellow solid. Yield = 70 %. Mp 218-219 °C (lit.[30] 224-225 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.94-6.98 (m, 2H), 7.30-7.36 (m, 2H), 7.85 (s, 1H), 10.64 (s, 1H), 13.70 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 119.9, 124.0, 127.1, 129.2, 132.73 157.5, 169.8, 196.1. MS (ESI) *m/z* 236.2 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 236.9918, found: 236.9924.

(*Z*)-5-(4-hydroxybenzylidene)-2-thioxothiazolidin-4-one (23). Yellow solid. Yield = 50 %. Mp 269-270 °C (lit.[30] 279-280 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.93 (d, J = 12.0 Hz, 2H), 7.47 (d, J = 12.0 Hz, 2H), 7.56 (s, 1H), 10.41 (s, 1H), 13.69 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 116.5, 121.0, 124.0, 132.3, 133.0, 160.3, 169.6 195.6. MS (ESI) *m/z* 236.3 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 235.9845, found: 235.9848.

(*Z*)-5-(3-hydroxybenzylidene)-2-thioxothiazolidin-4-one (24). Yellow solid. Yield = 50 %. Mp 236-237 °C (lit.[30] 237-239 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.87-6.90 (m, 1H), 6.97 (m, 1H), 7.02-7.04 (m, 1H), 7.33 (d, J = 8.0 Hz, 1H), 7.50 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 116.1, 117.9, 121.8, 126.2, 130.4, 131.1, 134.3, 157.9, 170.6, 196.4. MS (ESI) *m/z* 236.3 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 235.9845, found: 235.9853

2-(2,4-dioxothiazolidin-3-yl) acetic acid (5). Yellow solid. Yield = 17 %. Mp 151.8-152.7 °C. ^{1}H NMR (400 MHz, CD_3OD) δ 4.19 (s, 2H), 4.34 (s, 2H). ^{13}C NMR (100 MHz, CD_3OD) δ 34.7, 42.8, 169.9, 173.0, 173.5. MS (ESI) *m/z* 174.0 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 173.9867; found 173.9866.

(*Z*)-5-benzylidenethiazolidine-2,4-dione (6). Pale yellow solid. Yield = 33 %. Mp 252.8-254.2°C (lit.[31] 247-249 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.44 – 7.57 (m, 3H), 7.57 –

7.62 (m, 2H), 7.78 (s, 1H), 12.62 (br,1H).^{13}C NMR (100 MHz, DMSO-d_6) δ 124.1, 129.3, 129.9, 130.3, 131.4, 133.2, 168.0, 168.2. MS (ESI) *m/z* 203.9 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 204.0125; found 204.0128.

(*Z*)-5-propylidenethiazolidine-2,4-dione (7). Yellow solid. Yield = 54 %. Mp 69.0-70.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.07 (t, J = 8.0 Hz, 3H), 2.20 (quin, J = 8.0 Hz, 2H), 6.80 (t, J = 8.0 Hz, 1H), 13.58 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 12.5, 25.6, 129.0, 139.5, 168.2, 196.6. MS (ESI) *m/z* 156.4 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 157.0197; found 157.0199.

(*Z*)-2-(5-benzylidene-2,4-dioxothiazolidin-3-yl)acetic acid (8). White solid, Yield = 73 %. Mp 213.3-213.9 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.38 (s, 2H), 7.55 (m, 3H), 7.64 – 7.69 (m, 2H), 8.00 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 42.3, 120.7, 129.4, 130.2, 130.9, 132.8, 133.9, 165.0, 166.9, 167.9. MS (ESI) *m/z* 262.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 262.0180; found 262.0175.

Ethyl 2-(5-oxo-2-thioxoimidazolidin-1-yl) acetate (30). . Yellow solid. Yield = 48 %. ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.28 (t, J = 8.0 Hz, 3H), 3.86 (br, 2H), 4.03 (m, 2H), 4.19 (m, 2H), 5.79 (br, 1H), 7.34 (br, 1H).

2-(5-oxo-2-thioxoimidazolidin-1-yl) acetic acid (9). Yellow solid. Yield = 94 %. Decomposed at 300 °C (lit.[32] decomposed at 138 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.25 (s, 2H), 4.51 (s, 2H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 42.0, 49.1, 168.4, 172.4, 184.9. MS (ESI) *m/z* 261.2 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 174.0099; found 174.0101.

(*Z*)-5-benzylidene-2-thioxoimidazolidin-4-one (10). Brown solid. Yield = 69 %. Mp 267.0-267.3 °C (lit.[33] 268-270 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 6.49 (s, 1H), 7.34 – 7.46 (m, 3H), 7.71 – 7.79 (m, 2H), 12.16 (br, 1H), 12.37 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ

111.5, 127.7, 128.8, 129.2, 130.1, 132.3, 165.8, 179.3. MS (ESI) *m/z* 203.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 203.0285; found 203.0284.

(*Z*)-5-propylidene-2-thioxoimidazolidin-4-one (11). Yellow solid. Yield = 85 %. Mp 197.0-198.0 °C (lit.[34] 197-200 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 1.01 (t, J = 8.0 Hz, 3H), 2.27 (quin, J = 8.0 Hz, 2H), 5.65 (t, J = 8.0 Hz) 11.89-12.06 (br, 2H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.5, 20.4, 118.3, 130.9, 164.9, 178.5. MS (ESI) *m/z* 158.8 $[M-H]^+$. HRMS (ESI, $[M-H]^-$) calcd ($C_{10}H_8N_2OS$) 156.0357; found 156.0362.

(*Z*)-2-(4-benzylidene-5-oxo-2-thioxoimidazolidin-1-yl) acetic acid (12). Yellow solid. Yield = 37 %. Decomposed at 300 °C (lit.[35] 257 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.50 (s, 2H), 6.69 (s, 1H), 7.42-7.47 (m, 3H), 7.79-7.81 (m, 2H). ^{13}C NMR (100 MHz, DMSO-d_6) 41.8, 113.5, 126.1, 128.8, 129.6, 130.4, 132.1, 163.6, 168.1, 178.4. MS (ESI) *m/z* 262.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 262.0412, found: 262.0418.

Methyl 2-(2,5-dioxoimidazolidin-1-yl) acetate (31). White solid. Yield = 22 %. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.53 (br, 1H), 4.28 (s, 2H), 4.07 (d, J = 1.0 Hz, 2H), 3.78 (s, 3H).

2-(2,5-dioxoimidazolidin-1-yl) acetic acid (13). Yellow solid. Yield = 83 %. Mp 190.0-190.4 °C (lit.[35] 190 °C). ^{1}H NMR (400 MHz, D_2O) δ 4.16 (s, 2H), 4.31 (s, 2H). ^{13}C NMR (100 MHz, D_2O) δ 39.5, 46.7, 158.7, 171.3, 174.4. MS (ESI) *m/z* 157.0 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 157.0255; found 157.0260.

(*Z*)-5-benzylideneimidazolidine-2,4-dione (14). White solid. Yield = 37 %. Mp 227.0-227.5 °C (lit.[36] 220-224 °C). ^{1}H NMR (400 MHz, Acetone-d_6) δ 6.53 (s, 1H) 7.39 (m, 3H), 7.63 (m, 2H), 9.44 (br, 1H), 10.05 (br, 1H). ^{13}C NMR (100 MHz, Acetone-d_6) 109.5, 129.4, 129.8, 130.1, 134.5, 155.6, 165.7. MS (ESI) *m/z* 187.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 188.0586; found: 188.0588.

(*Z*)-5-propylideneimidazolidine-2,4-dione (15).White solid. Yield = 37 %. Mp 190.0-191.0 °C (lit.[37]191-197 °C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 0.99 (t, J = 8.0 Hz, 3H), 2.15 (quin, J = 8.0 Hz, 2H), 5.49 (t, J = 8.0 Hz, 1H), 10.12 (br, 1H), 10.92 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 13.2, 19.4, 113.2, 129.9, 154.8, 164.4. MS (ESI) *m/z* 138.9 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 140.0586; found: 140.0581.

(*Z*)-2-(4-benzylidene-2,5-dioxoimidazolidin-1-yl)acetic acid (16). White solid. Yield = 37 %. Mp 175.0-176.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 4.20 (s, 2H), 6.60 (s, 1H), 7.36-7.45 (m, 3H), 7.66 (d, J = 8.0 Hz, 2H), 10.92 (s, 1H) . ^{13}C NMR (100 MHz, DMSO-d_6) δ 39.5, 110.2, 126.4, 128.8, 128.9, 129.6, 132.6, 154.4, 163.6, 168.7.. MS (ESI) *m/z* 245.0 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 246.0641 found: 246.0644.

5-benzyl-2-thioxothiazolidin-4-one (17). Yellow solid. Yield = 54 %. Mp 169-170 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.17 (dd, J = 4.0, 12 Hz, 1H), 3.56 (dd, J = 4.0, 12 Hz, 1H), 4.62 (dd, J = 4.0, 8.0 Hz, 1H), 7.22-7.36 (m, 5H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 38.4, 56.7, 127.8, 129.0, 129.1, 135.6, 176.2, 199.8. MS (ESI) *m/z* 222.2 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 223.0126, found: 223.0126.

5-benzylthiazolidine-2,4-dione (18). White solid. Yield = 10 %. Mp 79-80 °C (lit.[25] 78.5-80.1°C). ^{1}H NMR (400 MHz, CD_3OD) δ 3.13, (dd, J = 4.0, 12 Hz, 1H), 3.47 (dd, J = 4.0, 12 Hz, 1H), 4.74 (dd, J = 4.0 Hz, 1H), 7.24-7.33 (m. 5H). ^{13}C NMR (100 MHz, CD_3OD) δ 30.7, 39.3, 54.5, 128.3, 129.6, 130.4, 139.0, 177.4. MS (ESI) *m/z* 206.2 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 207.0354, found: 207.0352.

5-benzyl-2-thioxoimidazolidin-4-one (19). Yellow solid. Yield = 65 %. Mp 180-181 °C (lit.[38]180-182°C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.97-2.99 (m, 2H), 4.55 (m, 1H), 7.16-7.29 (m, 5H), 10.05 (s, 1H), 11.43 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 35.6, 61.4,

126.8, 128.1, 129.6, 135.0, 175.7, 182.3. MS (ESI) *m/z* 205.1 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 206.0514, found: 206.0512.

5-(2-hydroxybenzyl)-2-thioxothiazolidin-4-one (25). Yellow solid. Yield = 35 %. Mp 186-187 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.94 (dd, J = 8.0, 12 Hz, 1H), 3.44 (dd, J = 4.0, 12 Hz, 1H), 4.98 (dd, J = 4.0, 12 Hz, 1H), 6.71-6.75 (m, 1H), 6.81 (d, J = 8.0 Hz, 1H), 7.06-7.10 (m, 2H), 9.66 (s, 1H), 13.17 (br, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 32.34, 54.44, 115.06, 118.94, 123.20, 128.38, 130.38, 155.36, 178.27, 203.67. MS (ESI) *m/z* 238.3 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 239.0075, found: 239.008.

5-(4-hydroxybenzyl)-2-thioxothiazolidin-4-one (26). Yellow solid. Yield = 30 %. Mp 209-210 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.01-3.22 (m, 2H), 4.90 (dd, J = 4.0, 8.0 Hz, 1H), 6.67 (d, J = 8.0 Hz, 2H), 7.00 (d, J = 8.0 Hz, 2H), 9.50 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 36.17, 56.69, 115.67, 126.93, 128.91, 130.77, 156.73, 178.70, 204.12. MS (ESI) *m/z* 238.2 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 238.0002, found: 238.0009.

5-(3-hydroxybenzyl)-2-thioxothiazolidin-4-one (27). Yellow solid. Yield = 30 %. Mp 190-191 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.97 (dd, J = 8.0, 12.0 Hz, 1H), 3.29 (dd, J = 8.0, 12.0 Hz, 1H), 4.68 (m, 1H), 6.58-6.62 (m, 3H), 7.00-7.04 (t, J = 8.0 Hz, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 37.46, 56.08, 113.92, 115.62, 119.88, 129.31, 137.91, 157.33, 177.97, 202.72. MS (ESI) *m/z* 238.3 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 238.0002, found: 238.0010.

(*R*)-5-benzylimidazolidine-2,4-dione (20). White solid. Yield = 61 %. Mp 181.0-182.0 °C (lit.[39]187-188°C). ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.88-2.97 (m, 2H), 4.31-4.34 (m, 1H), 7.18-7.30 (m, 5H), 7.90 (s, 1H), 10.41 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 36.4, 58.4, 126.6, 128.1, 129.7, 135.6, 157.1, 175.2. MS (ESI) *m/z* 189.4 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 190.0742, found: 190.0746.

(*S*)-5-benzylimidazolidine-2,4-dione (21). White solid. Yield = 53 %. Mp 181.0-182.0 °C (lit.[39] 187-188°C). ^{1}H NMR (400 MHz, DMSO-d_6) 2.88-2.97 (m, 2H), 4.31-4.34 (m, 1H), 7.18-7.30 (m, 5H), 7.90 (s, 1H), 10.42 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 36.4, 58.4, 126.6, 128.1, 129.7, 135.6, 157.1, 175.2. MS (ESI) *m/z* 189.4 $[M-H]^-$. HRMS (ESI-TOF) calcd for $[M-H]^-$: 190.0742, found: 190.0746.

Table S1. Precursor and product ions utilized for MRM analysis in determination of Phase I metabolic stability.

Compound	Precursor ion m/z (Q1)	Product ion m/z (Q3)	Retention time (min)
1	190.2	104.0 146.0	2.76
2	220.3	58.2	3.86
3	172.0	58.1	3.18
4	294.3	214.0 250.0	3.92
5	174.2	88.1 130.2	2.69
6	204.3	101.0 133.3	3.19
7	155.9	85.0 113.0	3.01
8	262.4	190.1 218.4	3.29
9	172.9	58.1 74.2	2.72

10	203.2	58.1 116.2	2.97
11	154.9	58.1 112.0	3.46
12	261.1	160.1 217.1	3.63
14	187.4	116.0	2.82
15	139.0	68.1 124.2	2.85
16	245.1	187.0 201.4	2.90
17	221.7	58.1 89.6	3.85
18	203.8	101.0 133.0	3.33
19	205.0	58.1 113.9	3.53
20 21	189.0	71.2 98.1	2.95
22	236.0	58.0 177.0	8.66
23	236.0	58.0 148.9	7.93
24	236.0	57.9 177.0	8.14
25	238.0	58	8.05
26	237.8	58.0 89.9	7.11
27	238.0	58	7.46

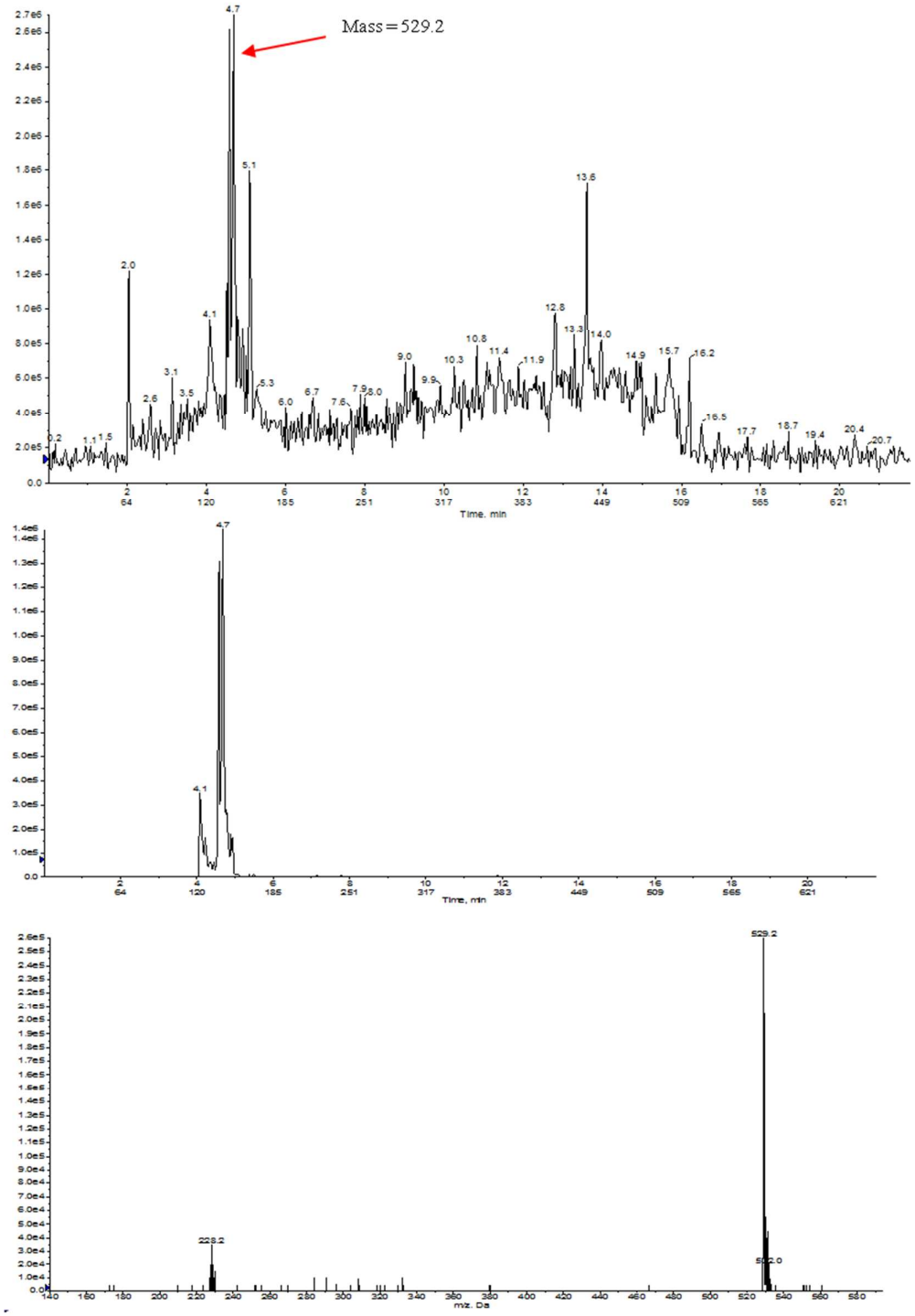

Figure S1. TIC, XIC trace, and neutral loss of 129 spectra of GSH trapped metabolites of compound **2**.

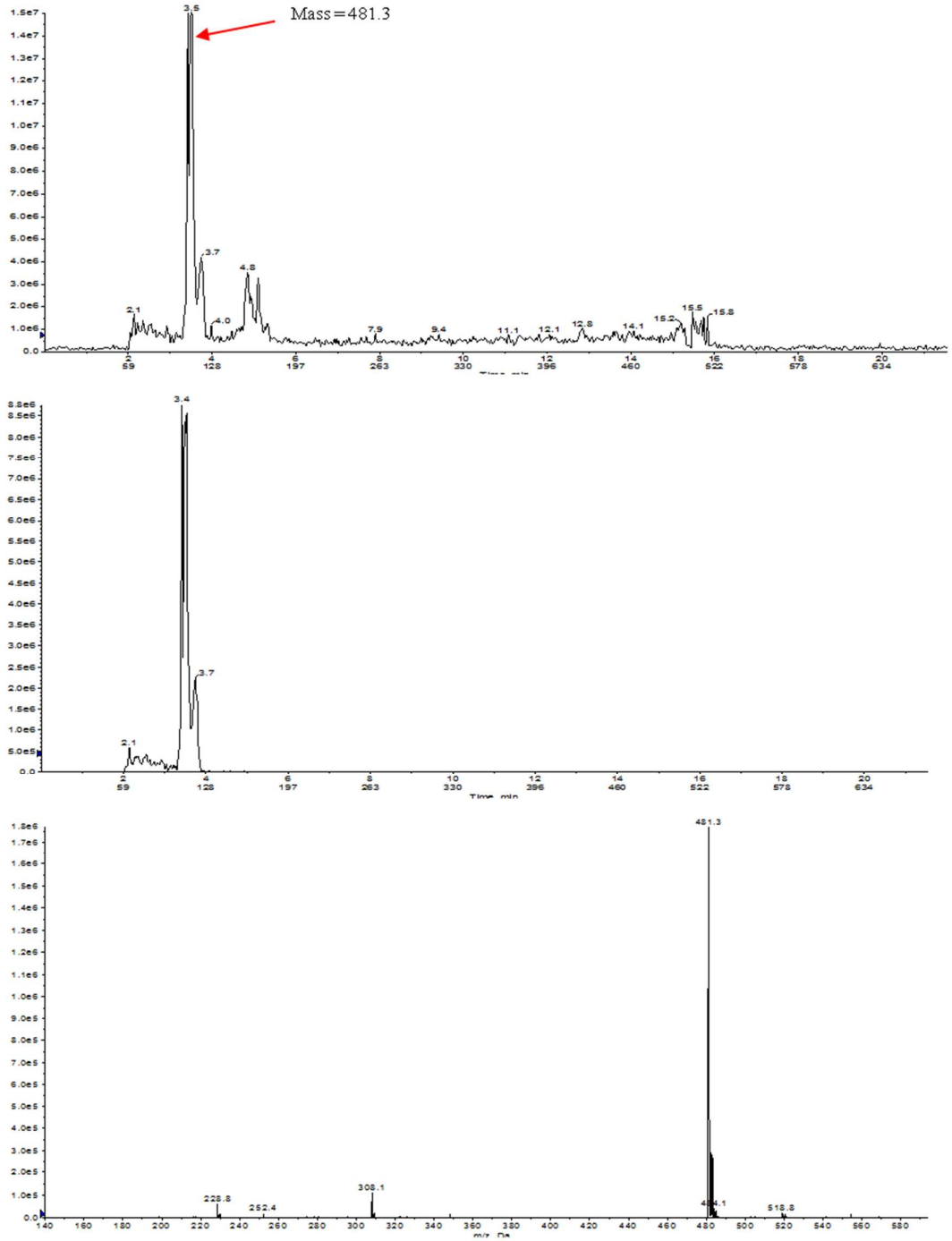

Figure S2. TIC, XIC trace, and neutral loss of 129 spectra of GSH trapped metabolites of compound **3**.

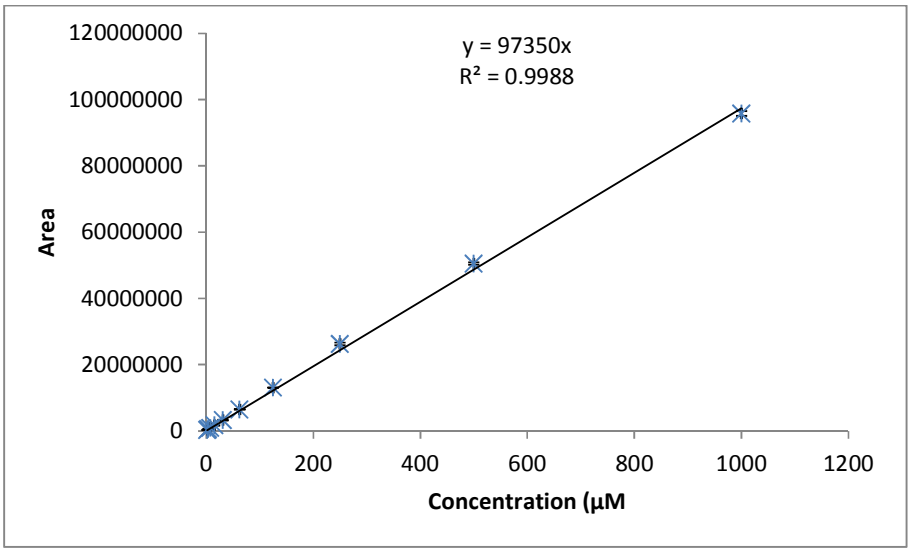

Figure S3. Standard calibration curve of dansyl-GSH (dGSH).