

## SUPPLEMENTAL INFORMATION

**Table S1:** MS characterizations, yields and HPLC purification conditions of LW01158, LW01160, LW01186, and LW02002.

Compound name	HPLC conditions	Retention time (min)	Yield (%)	Calculated mass (m/z)	Found (m/z)
LW01158	26% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	14.1	32	[M+2H] <sup>2+</sup> 799.4	[M+2H] <sup>2+</sup> 799.6
LW01160	23% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	10.6	24	[M+2H] <sup>2+</sup> 799.4	[M+2H] <sup>2+</sup> 1599.2
LW01186	29% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	12.1	8	[M+2H] <sup>2+</sup> 806.4	[M+2H] <sup>2+</sup> 806.8
LW02002	27% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	14.5	17	[M+2H] <sup>2+</sup> 806.4	[M+2H] <sup>2+</sup> 806.7

**Table S2:** MS characterizations, yields and HPLC purification conditions of Ga-LW01158, Ga-LW01160, Ga-LW01186, and Ga-LW02002.

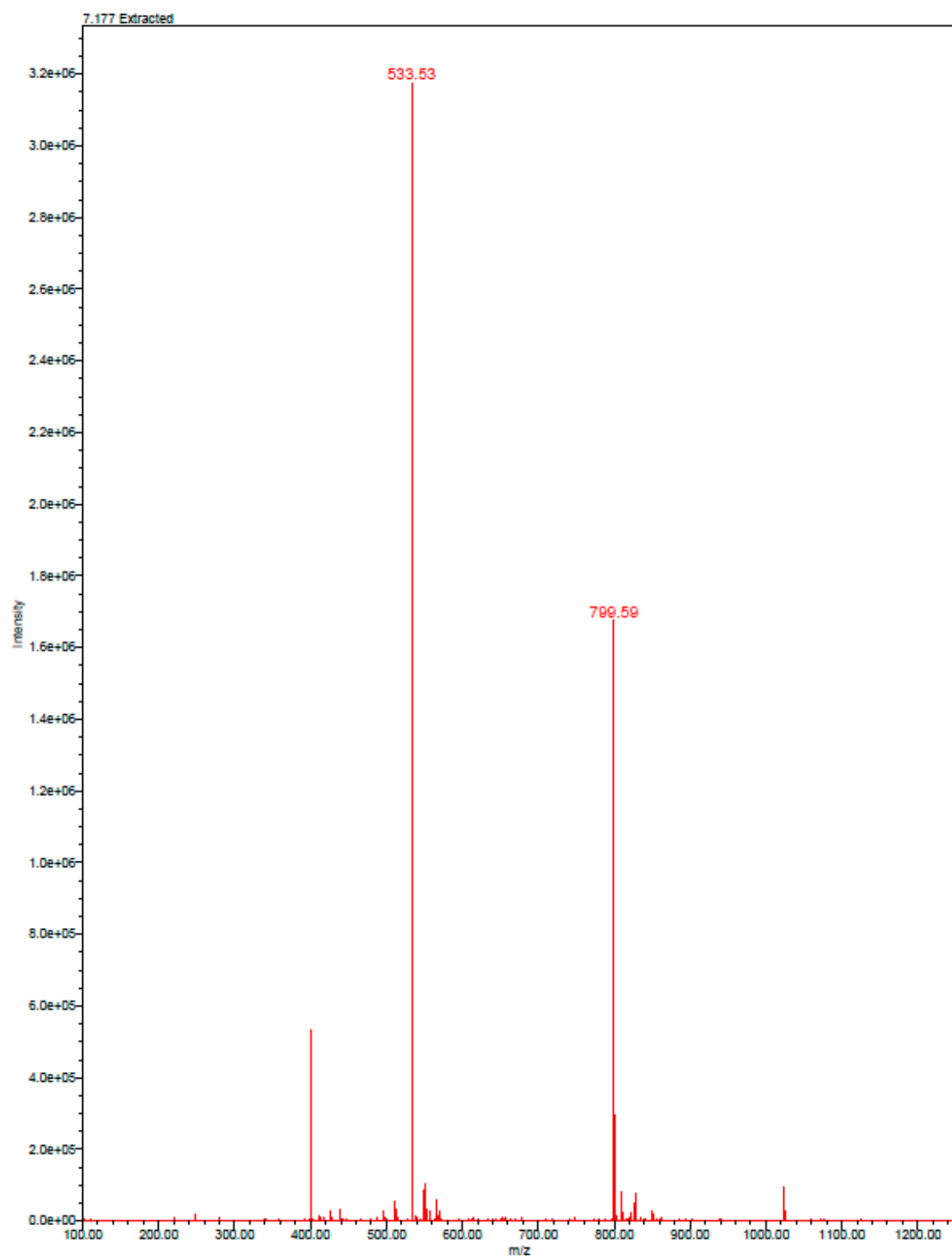
Compound name	HPLC conditions	Retention time (min)	Yield (%)	Calculated mass (m/z)	Found (m/z)
Ga-LW01158	26% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	13.2	76	[M+2H] <sup>2+</sup> 832.9	[M+2H] <sup>2+</sup> 833.4
Ga-LW01160	25% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	9.3	81	[M+2H] <sup>2+</sup> 832.9	[M+2H] <sup>2+</sup> 833.6
Ga-LW01186	29% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	13.5	81	[M+2H] <sup>2+</sup> 839.9	[M+2H] <sup>2+</sup> 839.5
Ga-LW02002	27% CH <sub>3</sub> CN and 0.1% TFA in H <sub>2</sub> O; flow rate 4.5 mL/min	16.3	77	[M+2H] <sup>2+</sup> 839.9	[M+2H] <sup>2+</sup> 839.4

**Table S3:** HPLC conditions for the purification and quality control of <sup>68</sup>Ga-labeled LW01158, LW01186, and LW02002. FA: formic acid.

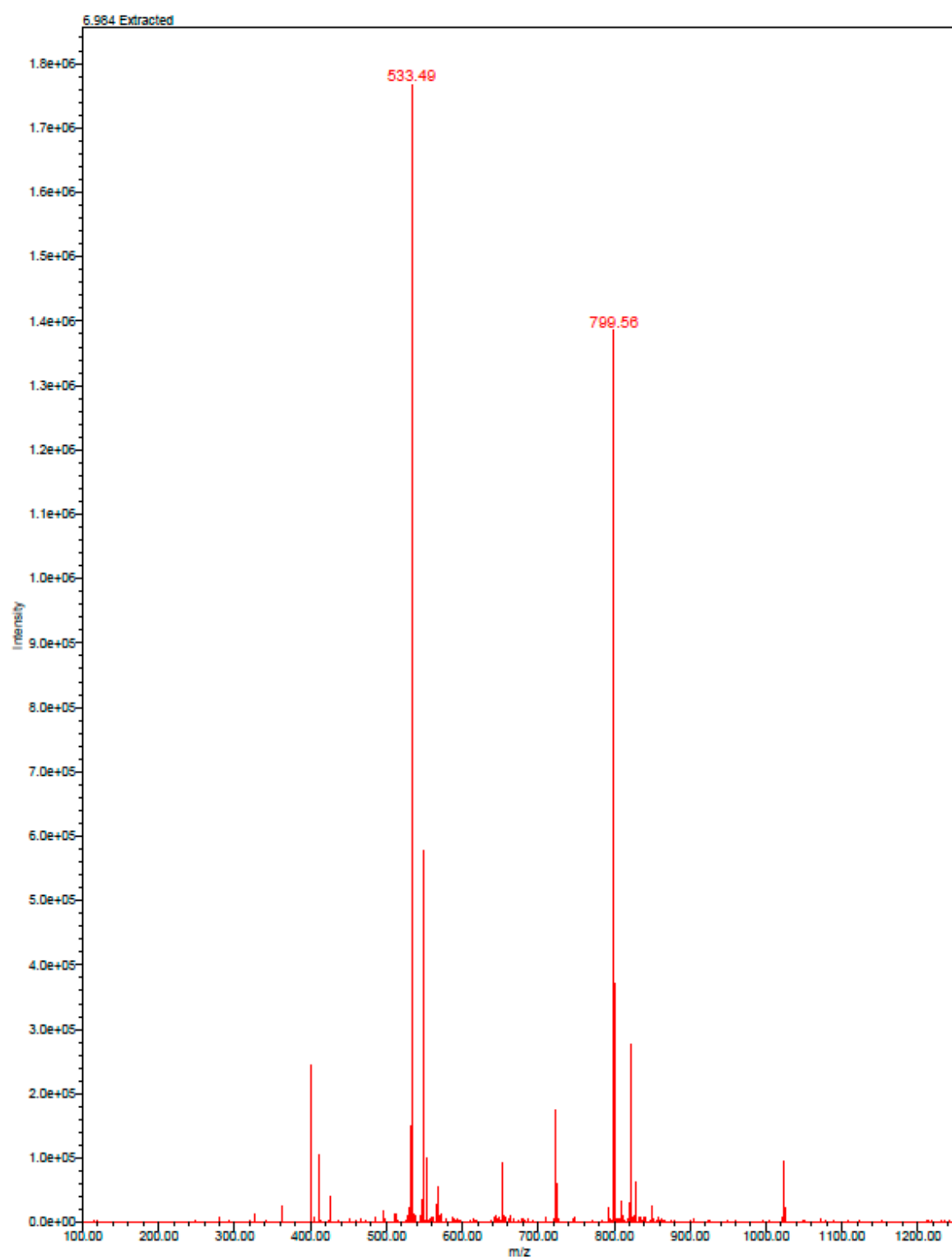
Compound name	HPLC conditions		Retention time (min)
[ <sup>68</sup> Ga]Ga-LW01158	Semi-Prep	20% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 4.5 mL/min	12.5
	QC	21.5% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 2.0 mL/min	6.1
[ <sup>68</sup> Ga]Ga-LW01186	Semi-Prep	22% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 4.5 mL/min	11.6
	QC	24% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 2 mL/min	5.5
[ <sup>68</sup> Ga]Ga-LW02002	Semi-Prep	20% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 4.5 mL/min	14.3
	QC	21.5% CH <sub>3</sub> CN and 0.1% FA in H <sub>2</sub> O; flow rate 2.0 mL/min	6.7

**Table S4:** Biodistribution (mean  $\pm$  SD, n = 4) and uptake ratios of  $^{68}\text{Ga}$ -labeled GRPR-targeted tracers in PC-3 tumor-bearing mice. The biodistribution data of [ $^{68}\text{Ga}$ ]Ga-TacsBOMB2 and [ $^{68}\text{Ga}$ ]Ga-RM2 have been reported previously [1], and are included here for comparison. The mice in the blocked group were co-injected with 100  $\mu\text{g}$  of Ga-LW01158. \*, \*\*, and \*\*\* indicate  $p < 0.05$ ,  $< 0.01$  and  $< 0.001$ , respectively, when comparing the 1 h and 1 h blocked data of [ $^{68}\text{Ga}$ ]Ga-LW01158.

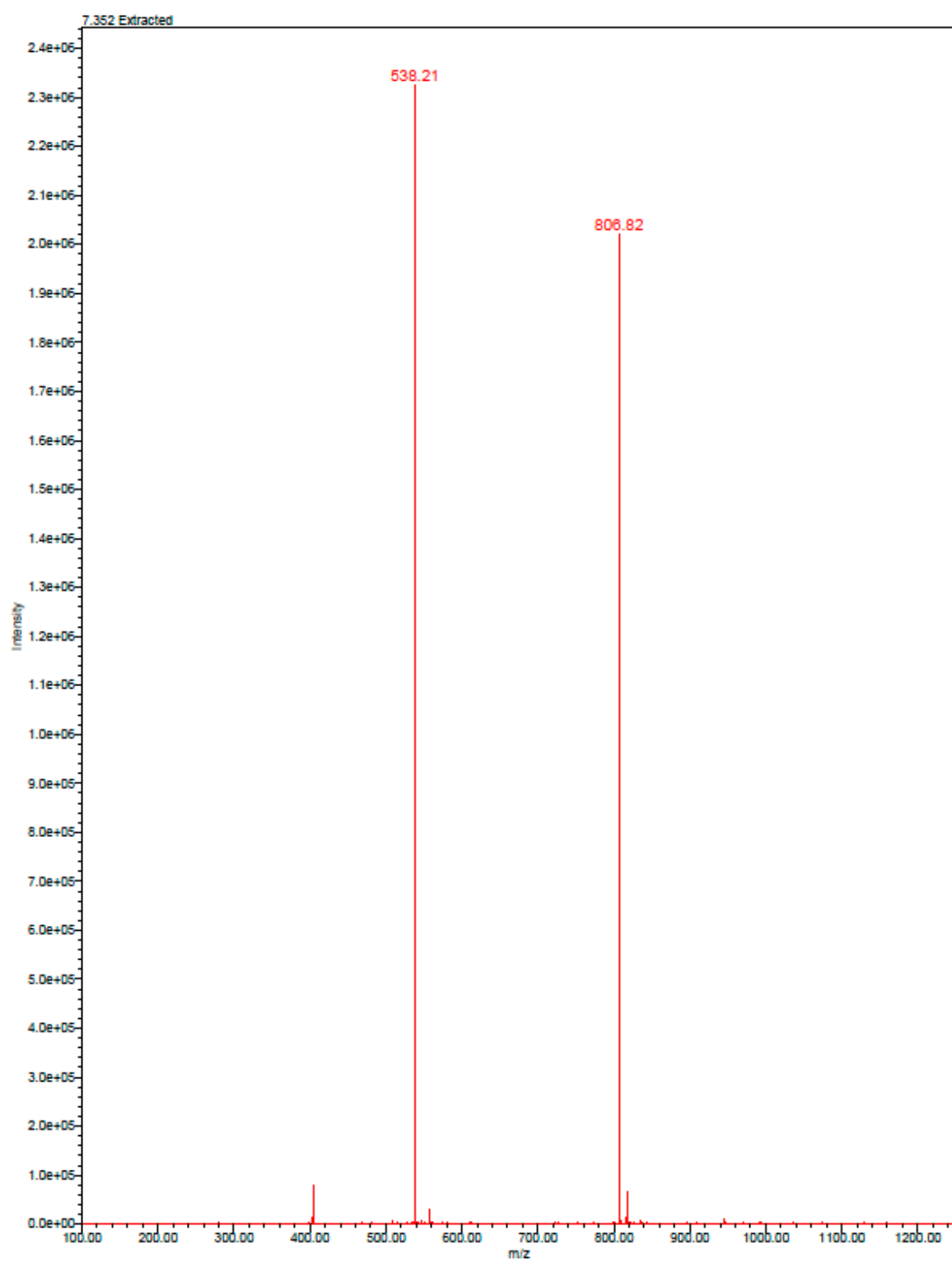
Tissue (%ID/g)	[ $^{68}\text{Ga}$ ]Ga- TacsBOMB2 1 h	[ $^{68}\text{Ga}$ ]Ga- LW01158 1 h	[ $^{68}\text{Ga}$ ]Ga- LW01158 1 h blocked	[ $^{68}\text{Ga}$ ]Ga- LW01186 1 h	[ $^{68}\text{Ga}$ ]Ga- LW02002 1 h	[ $^{68}\text{Ga}$ ]Ga- RM2 1 h
Blood	0.76 $\pm$ 0.21	1.14 $\pm$ 0.15	2.82 $\pm$ 0.90*	1.83 $\pm$ 0.31	0.94 $\pm$ 0.08	0.64 $\pm$ 0.10
Fat	0.09 $\pm$ 0.03	0.07 $\pm$ 0.01	0.27 $\pm$ 0.10**	0.16 $\pm$ 0.03	0.07 $\pm$ 0.01	0.05 $\pm$ 0.03
Testes	0.19 $\pm$ 0.05	0.24 $\pm$ 0.01	0.66 $\pm$ 0.24*	0.31 $\pm$ 0.05	0.20 $\pm$ 0.10	0.18 $\pm$ 0.03
Small intestine	1.04 $\pm$ 0.30	2.46 $\pm$ 0.30	1.17 $\pm$ 0.39**	2.54 $\pm$ 0.44	0.66 $\pm$ 0.06	5.08 $\pm$ 1.05
Large intestine	0.37 $\pm$ 0.16	1.41 $\pm$ 0.38	0.90 $\pm$ 0.57	1.27 $\pm$ 0.21	0.54 $\pm$ 0.29	2.19 $\pm$ 0.67
Spleen	0.47 $\pm$ 0.17	0.60 $\pm$ 0.42	0.67 $\pm$ 0.21	1.09 $\pm$ 0.33	0.46 $\pm$ 0.13	0.44 $\pm$ 0.26
<b>Pancreas</b>	<b>2.81 <math>\pm</math> 0.78</b>	<b>12.0 <math>\pm</math> 1.41</b>	<b>0.66 <math>\pm</math> 0.28***</b>	<b>14.1 <math>\pm</math> 1.90</b>	<b>2.36 <math>\pm</math> 0.36</b>	<b>41.9 <math>\pm</math> 10.1</b>
Stomach	0.32 $\pm$ 0.08	1.30 $\pm$ 0.41	0.42 $\pm$ 0.15**	0.83 $\pm$ 0.13	0.32 $\pm$ 0.11	3.87 $\pm$ 2.80
Liver	2.61 $\pm$ 0.70	4.33 $\pm$ 0.22	4.25 $\pm$ 0.64	22.1 $\pm$ 3.19	1.06 $\pm$ 0.24	0.84 $\pm$ 0.55
Adrenal glands	0.57 $\pm$ 0.40	1.37 $\pm$ 0.35	1.06 $\pm$ 1.06	2.75 $\pm$ 0.74	0.58 $\pm$ 0.07	3.01 $\pm$ 0.91
Kidneys	2.51 $\pm$ 0.59	2.98 $\pm$ 0.34	21.1 $\pm$ 11.0*	4.58 $\pm$ 0.66	3.19 $\pm$ 0.36	2.57 $\pm$ 0.48
Heart	0.27 $\pm$ 0.08	0.34 $\pm$ 0.05	1.13 $\pm$ 0.62*	0.56 $\pm$ 0.09	0.33 $\pm$ 0.06	0.19 $\pm$ 0.03
Lungs	0.75 $\pm$ 0.52	1.19 $\pm$ 0.32	3.74 $\pm$ 0.94**	3.82 $\pm$ 1.00	5.42 $\pm$ 3.13	0.62 $\pm$ 0.26
<b>PC-3 tumor</b>	<b>10.2 <math>\pm</math> 2.27</b>	<b>11.2 <math>\pm</math> 0.65</b>	<b>2.18 <math>\pm</math> 0.56***</b>	<b>5.87 <math>\pm</math> 0.64</b>	<b>8.32 <math>\pm</math> 1.20</b>	<b>10.5 <math>\pm</math> 2.03</b>
Bone	0.19 $\pm$ 0.06	0.13 $\pm$ 0.01	0.39 $\pm$ 0.19*	0.23 $\pm$ 0.02	0.15 $\pm$ 0.03	0.11 $\pm$ 0.03
Muscle	0.15 $\pm$ 0.05	0.20 $\pm$ 0.05	0.41 $\pm$ 0.15*	0.24 $\pm$ 0.04	0.15 $\pm$ 0.02	0.14 $\pm$ 0.06
Brain	0.05 $\pm$ 0.03	0.03 $\pm$ 0.00	0.08 $\pm$ 0.02**	0.06 $\pm$ 0.01	0.06 $\pm$ 0.03	0.03 $\pm$ 0.01
Tumor/bone	61.3 $\pm$ 25.0	86.6 $\pm$ 12.0	6.18 $\pm$ 1.67***	25.6 $\pm$ 4.94	58.8 $\pm$ 15.2	96.5 $\pm$ 27.1
Tumor/muscle	70.1 $\pm$ 14.2	58.0 $\pm$ 12.5	5.53 $\pm$ 0.84***	24.9 $\pm$ 4.63	56.4 $\pm$ 9.92	80.8 $\pm$ 27.5
Tumor/blood	14.0 $\pm$ 3.48	9.91 $\pm$ 1.38	0.79 $\pm$ 0.11***	3.26 $\pm$ 0.59	8.93 $\pm$ 1.49	16.5 $\pm$ 3.06
Tumor/kidney	4.10 $\pm$ 0.46	3.76 $\pm$ 0.36	0.11 $\pm$ 0.03***	1.26 $\pm$ 0.10	2.62 $\pm$ 0.33	4.13 $\pm$ 0.73
Tumor/pancreas	3.70 $\pm$ 0.55	0.94 $\pm$ 0.15	3.43 $\pm$ 0.58***	0.42 $\pm$ 0.04	3.60 $\pm$ 0.86	0.25 $\pm$ 0.04



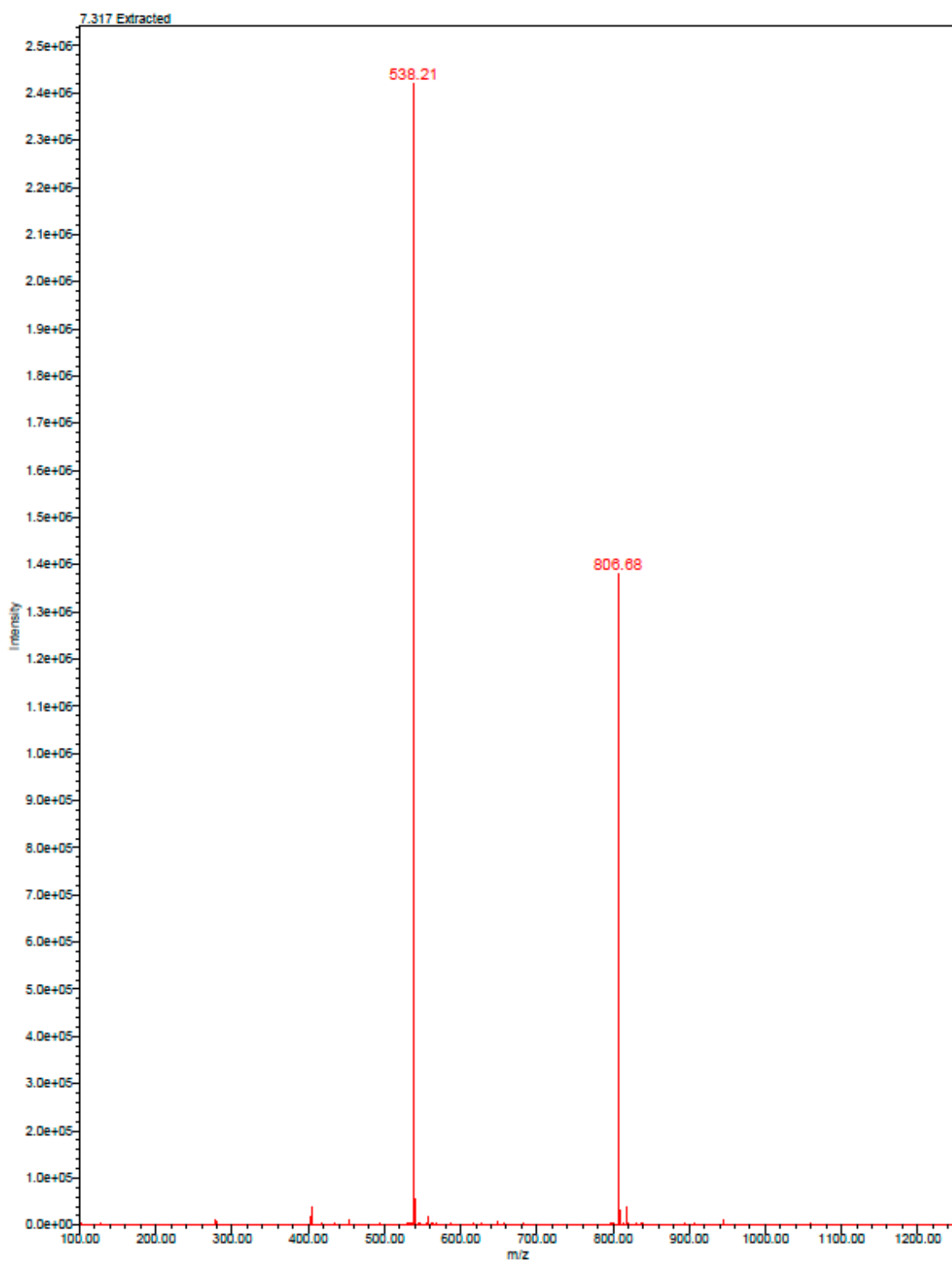
**Figure S1:** The MS spectrum of LW01158: calculated  $[M+2H]^{2+}$  (m/z) 799.4; found 799.6.



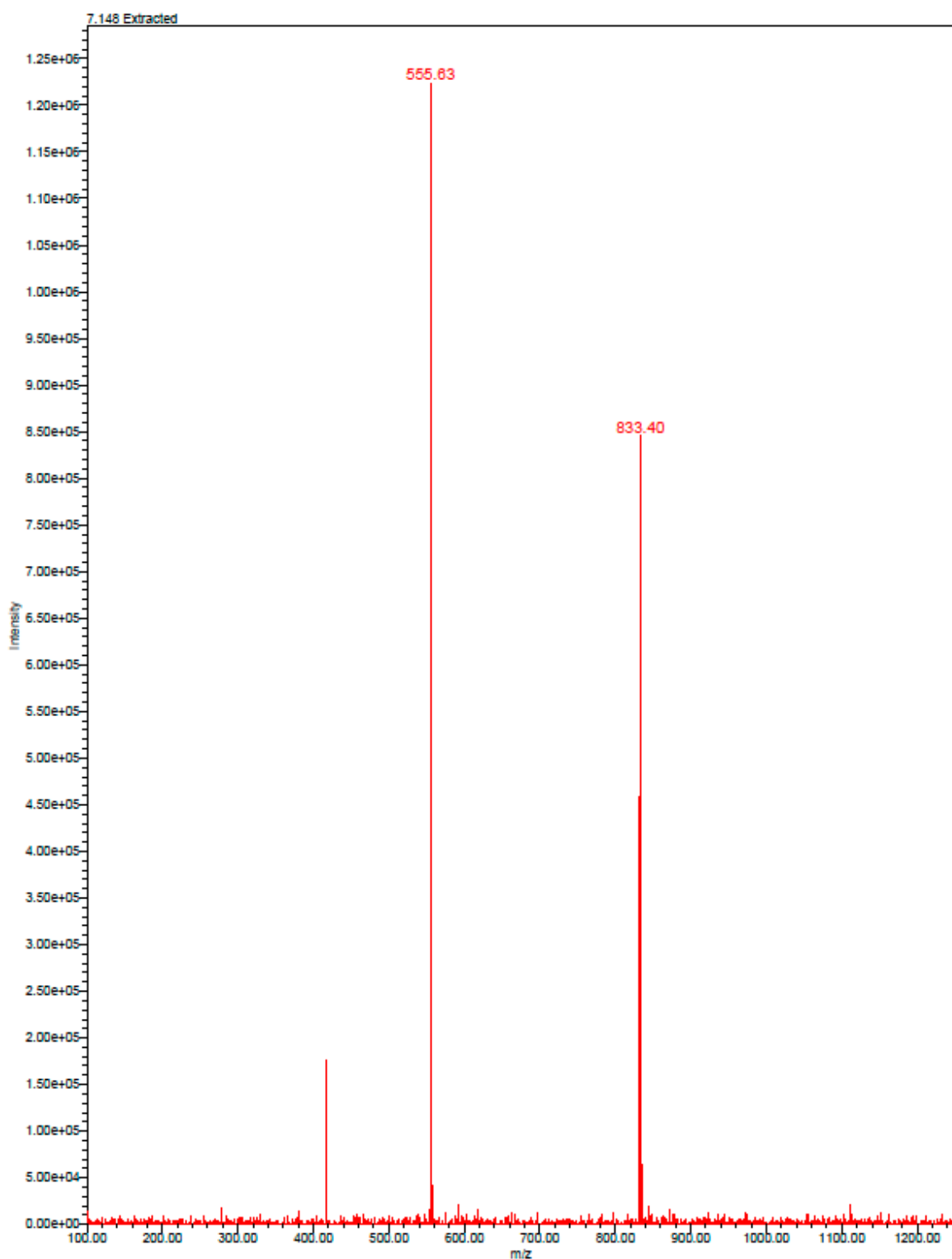
**Figure S2:** The MS spectrum of LW01160: calculated  $[M+2H]^{2+}$  (m/z) 799.4; found 799.6.



**Figure S3:** The MS spectrum of LW01186: calculated  $[M+2H]^{2+}$  (m/z) 806.4; found 806.8.

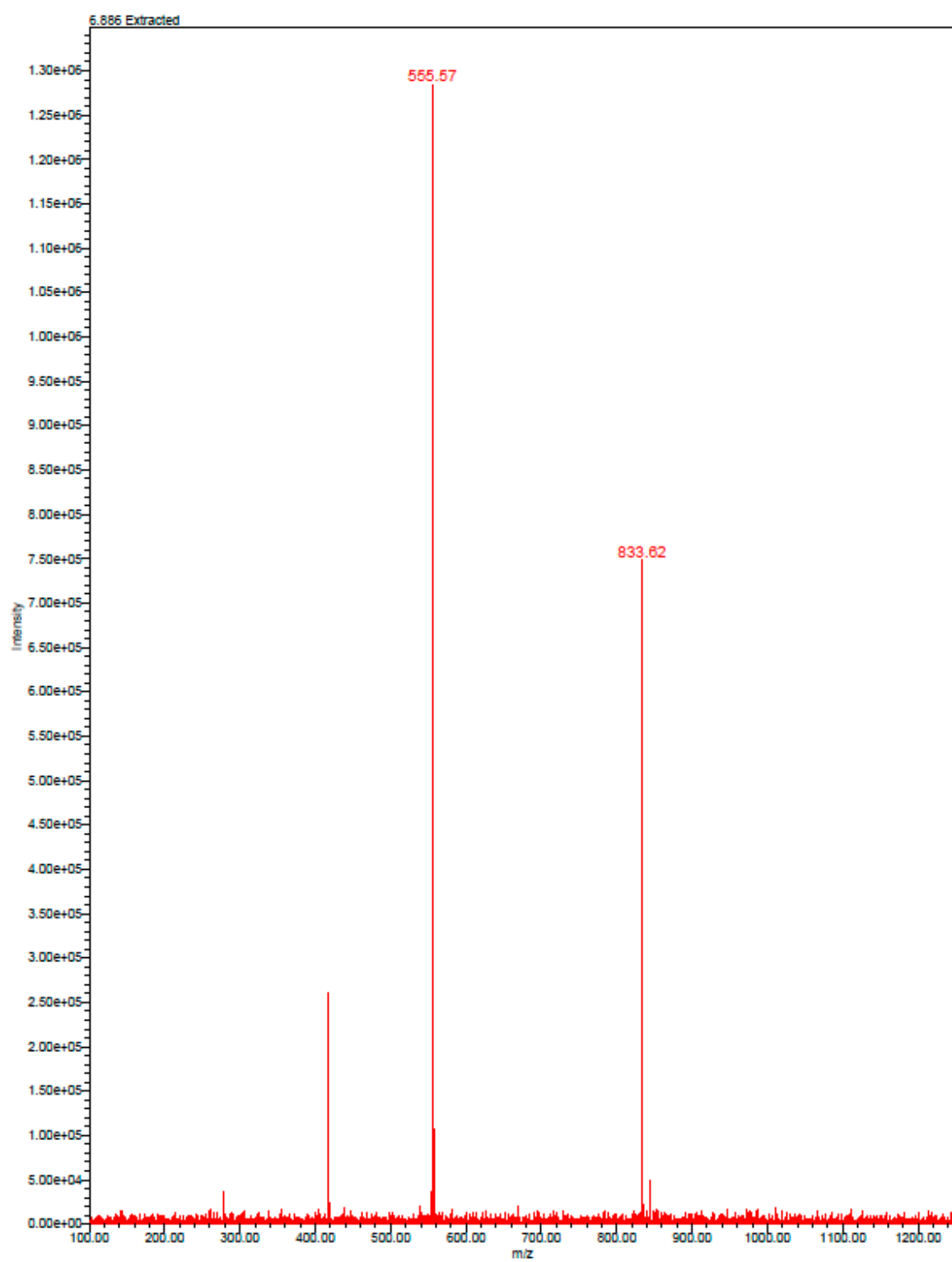


**Figure S4:** The MS spectrum of LW02002: calculated  $[M+2H]^{2+}$  (m/z) 806.4; found 806.7.

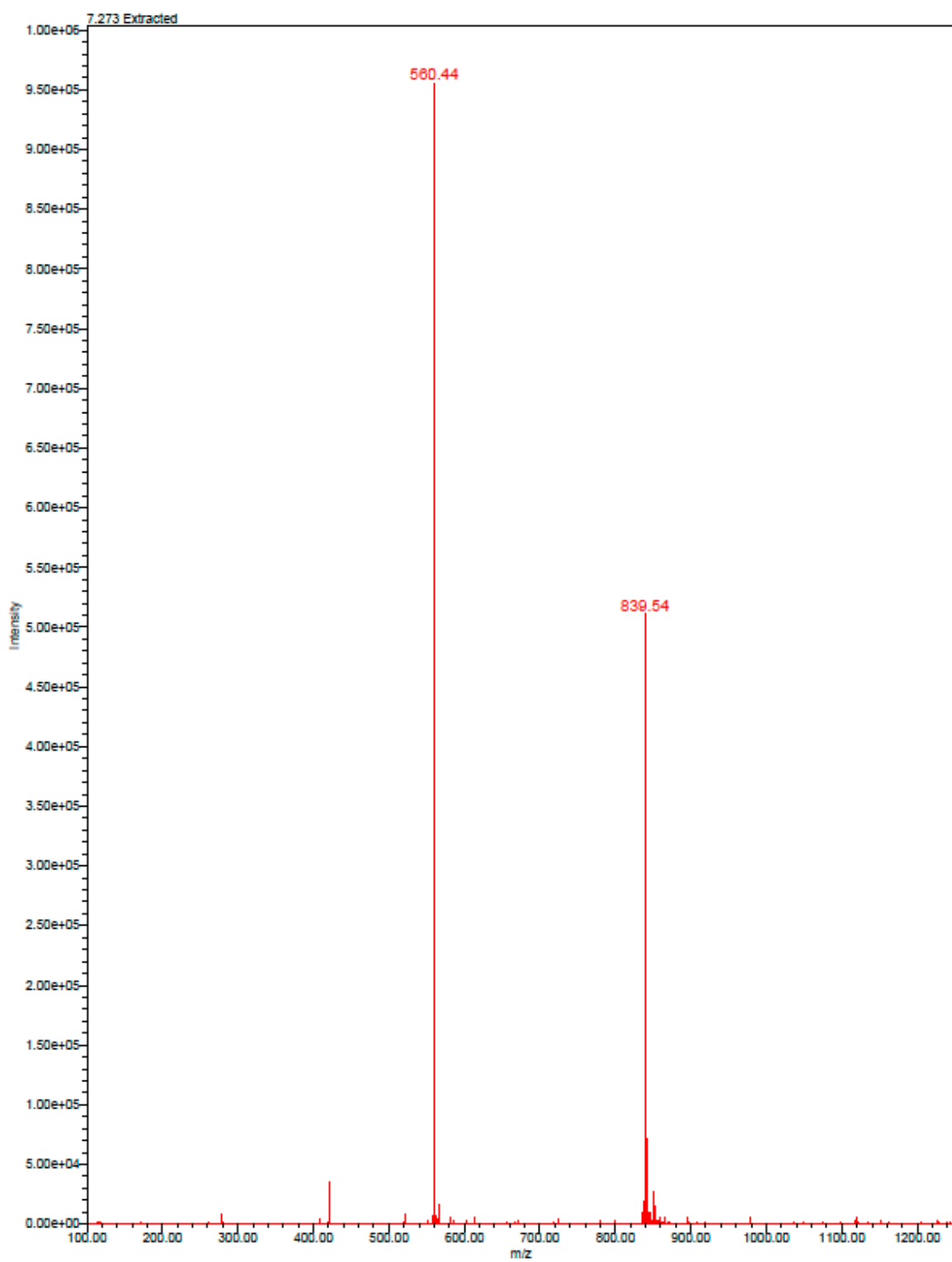


**Figure S5:** The MS spectrum of Ga-LW01158: calculated  $[M+2H]^{2+}$  (m/z) 832.9; found 833.4.

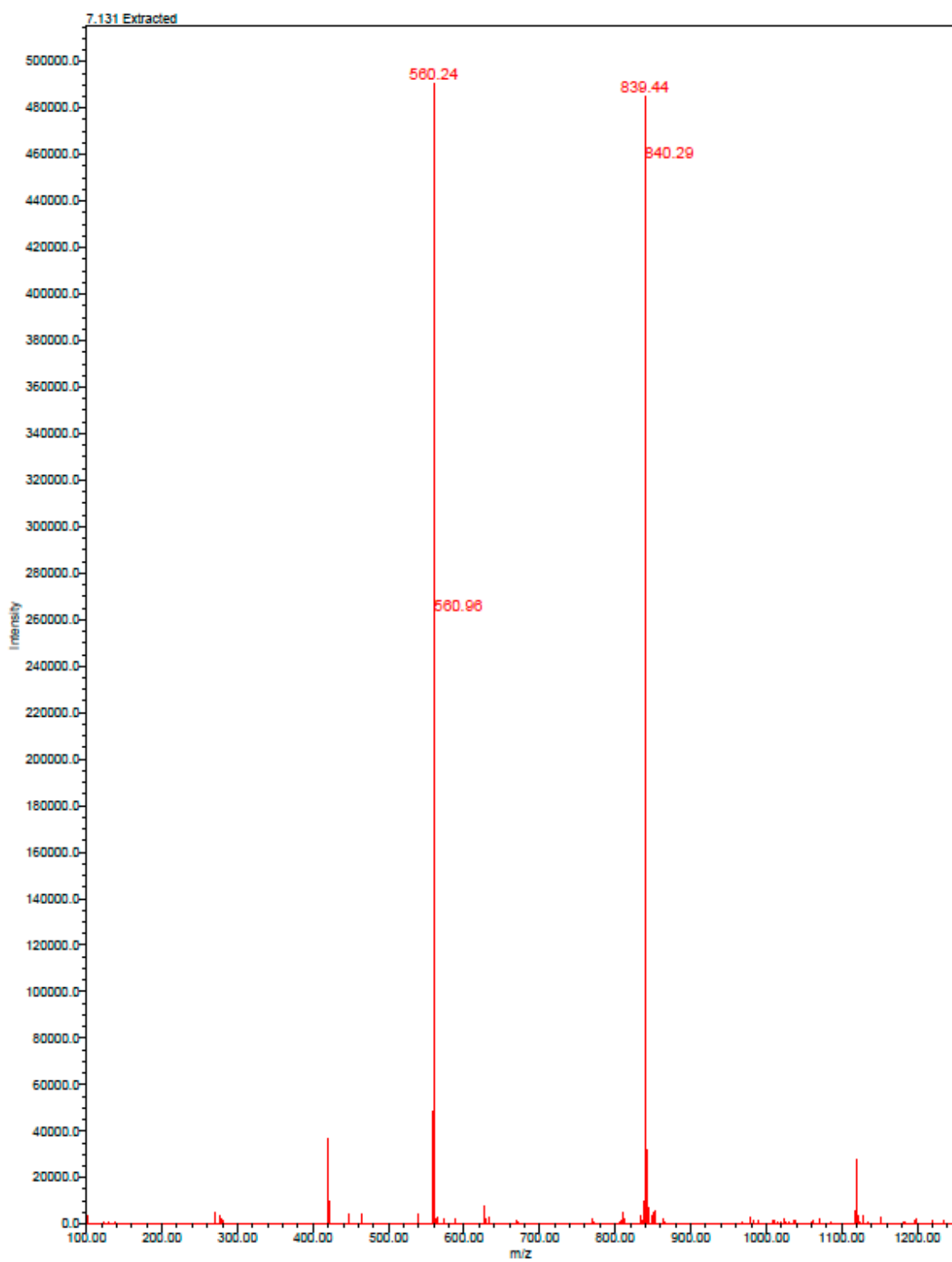




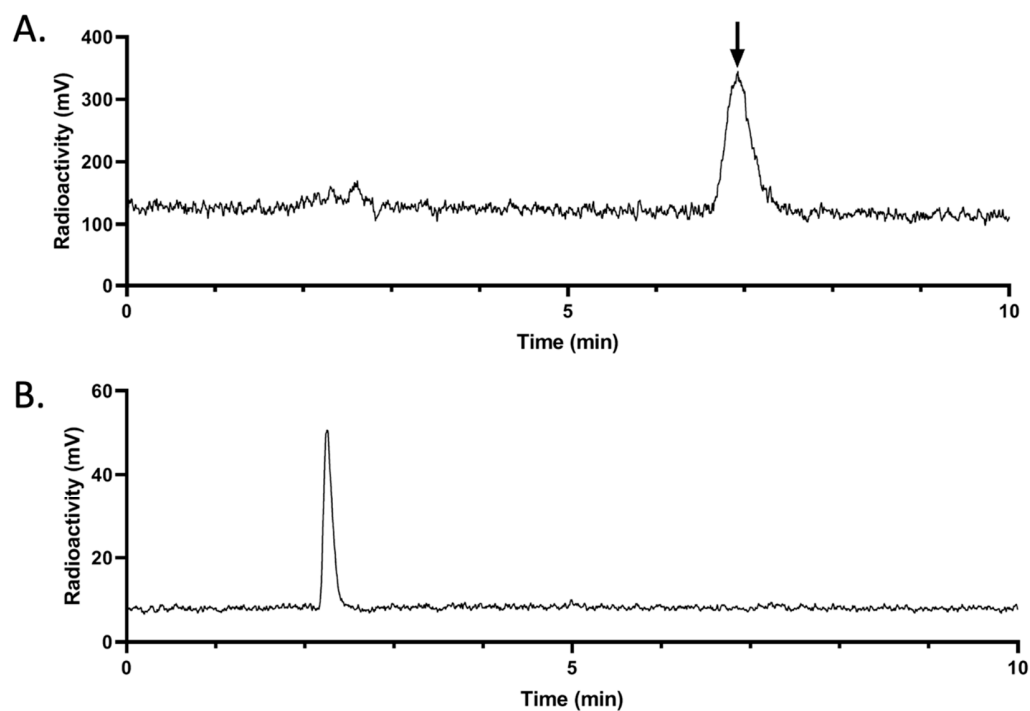
**Figure S6:** The MS spectrum of Ga-LW01160: calculated  $[M+2H]^{2+}$  (m/z) 832.9; found 833.6.



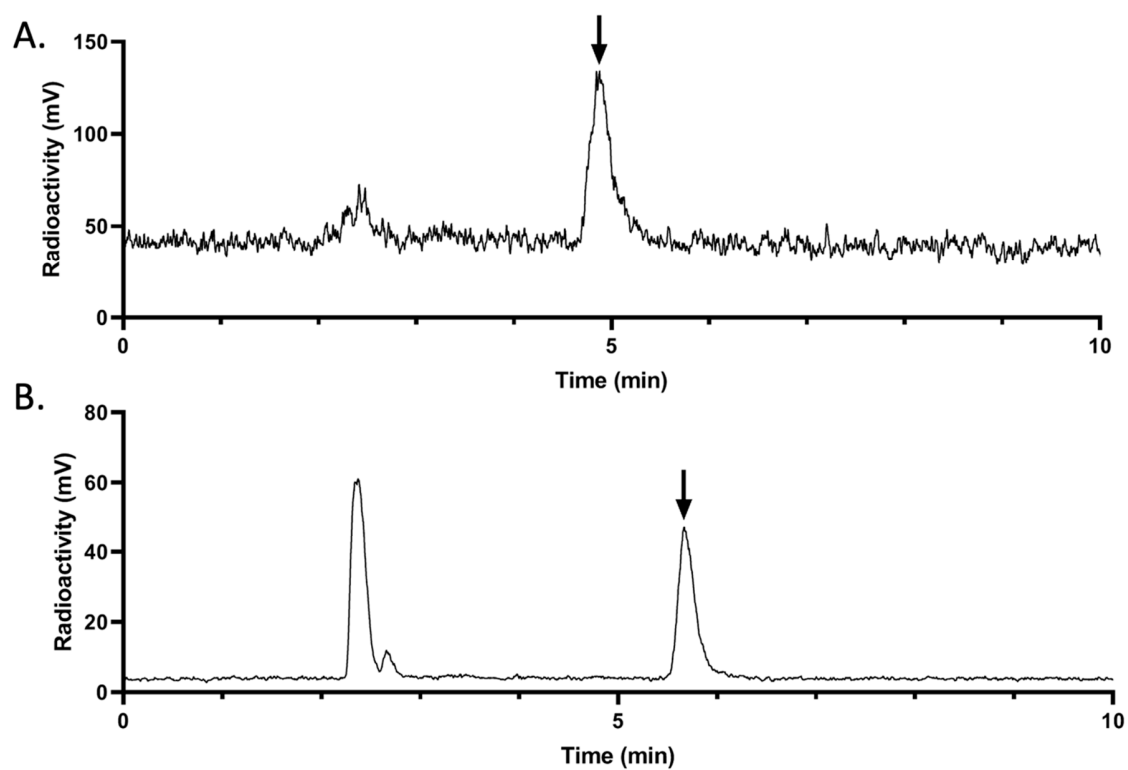
**Figure S7:** The MS spectrum of Ga-LW01186: calculated  $[M+2H]^{2+}$  (m/z) 839.9; found 839.5.



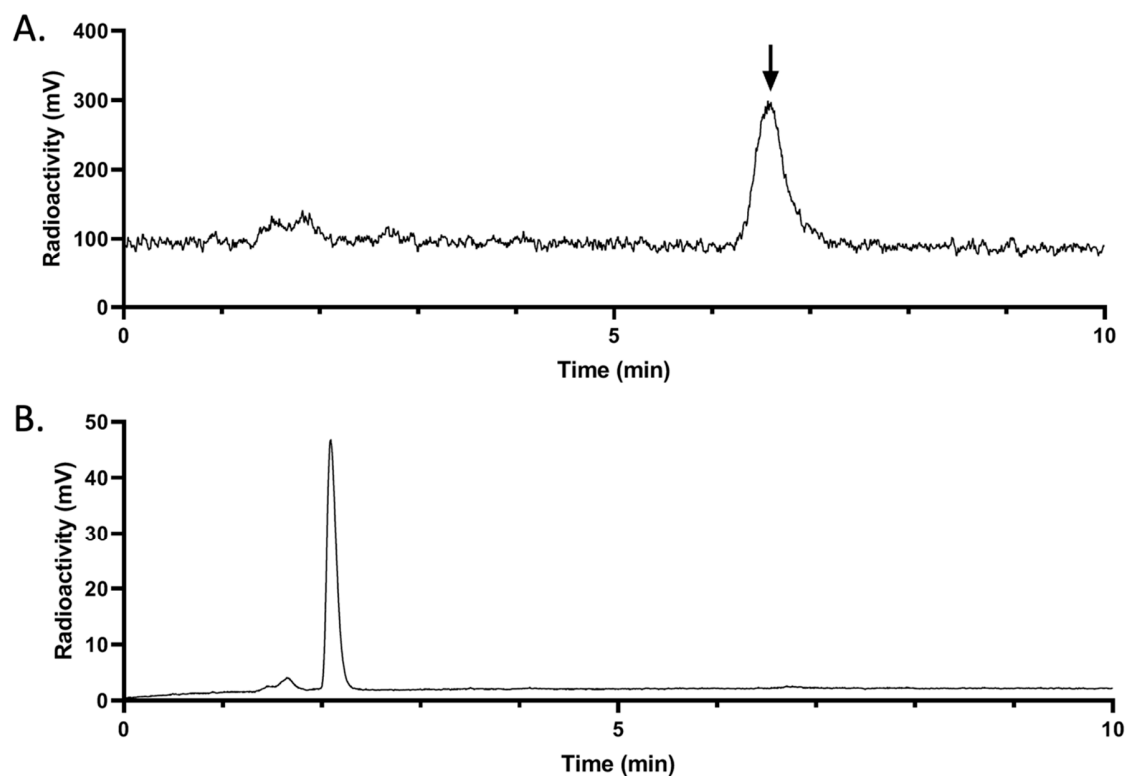
**Figure S8:** The MS spectrum of Ga-LW02002: calculated  $[M+2H]^{2+}$  (m/z) 839.9; found 839.4.



**Figure S9:** Representative radio-HPLC chromatograms from analysis of intact fraction of [ $^{68}\text{Ga}$ ]Ga-LW01158 in (A) mouse plasma and (B) urine samples collected at 15 min post-injection. The peak pointed by an arrow is the intact tracer.



**Figure S10:** Representative radio-HPLC chromatograms from analysis of intact fraction of [ $^{68}\text{Ga}$ ]Ga-LW01186 in (A) mouse plasma and (B) urine samples collected at 15 min post-injection. The peak pointed by an arrow is the intact tracer.



**Figure S11:** Representative radio-HPLC chromatograms from analysis of intact fraction of [<sup>68</sup>Ga]Ga-LW02002 in (A) mouse plasma and (B) urine samples collected at 15 min post-injection. The peak pointed by an arrow is the intact tracer.

Reference:

1. Wang, L.; Zhang, Z.; Merkens, H.; Zeisler, J.; Zhang, C.; Roxin, A.; Tan, R.; Bénard, F.; Lin, K.-S., <sup>68</sup>Ga-Labeled [Leu<sup>13</sup>ψThz<sup>14</sup>]Bombesin(7–14) derivatives: Promising GRPR-targeting PET tracers with low pancreas uptake. *Molecules*. **2022**, *27*, 3777.