ELECTRONIC SUPPLEMENTARY INFORMATION

Investigations on Reactivity, Stability and Biological Activity of Halido (NHC)gold(I) Complexes

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1. Characterization of **2**, **3**, **4** and **5**

1.1 ^{1}H NMR spectra

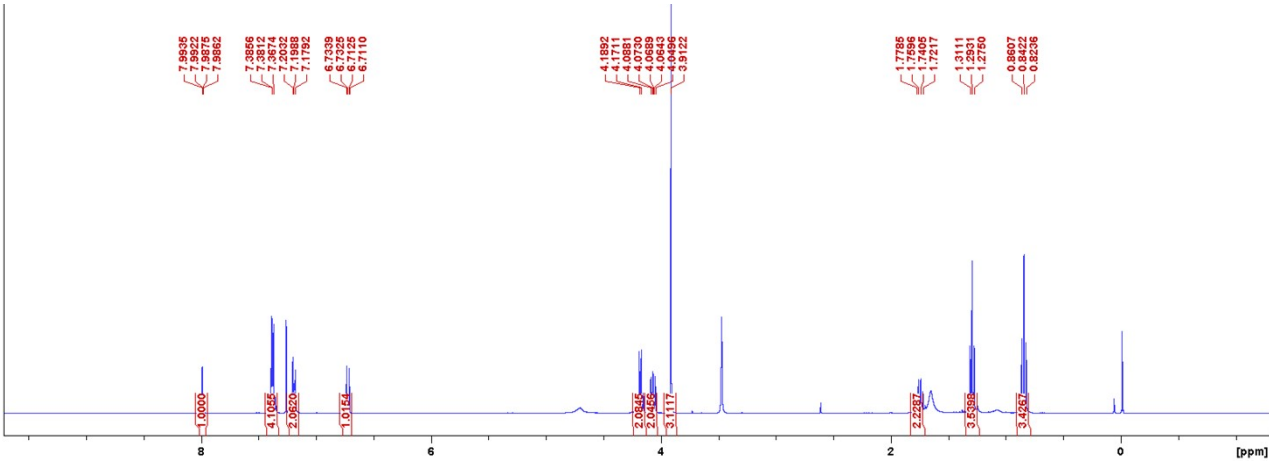

Figure S1. ^{1}H NMR spectrum (400 MHz) of **2** recorded in $CDCl_3$.

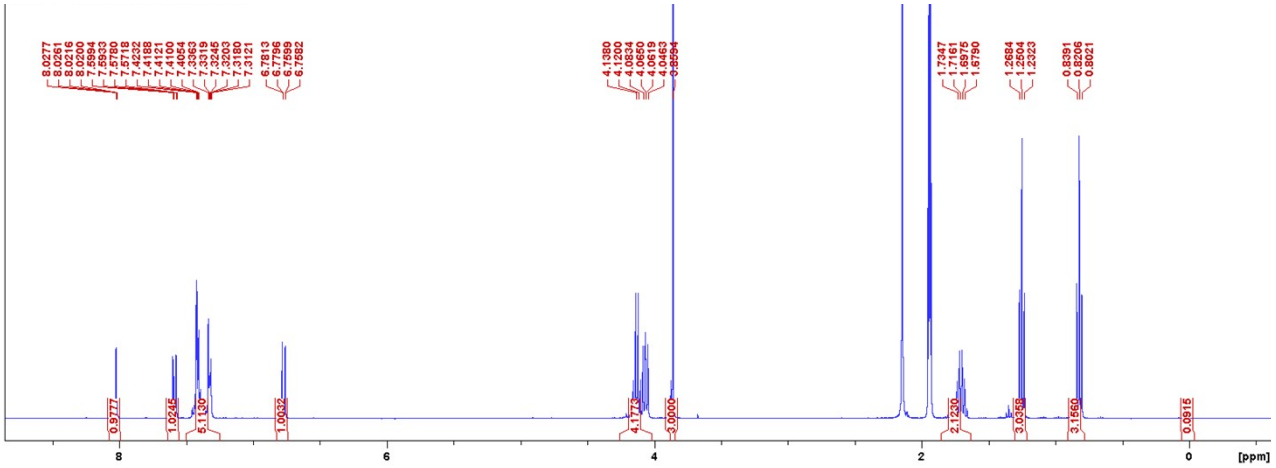

Figure S2. ^{1}H NMR spectrum (400 MHz) of **3** recorded in CD_3CN.

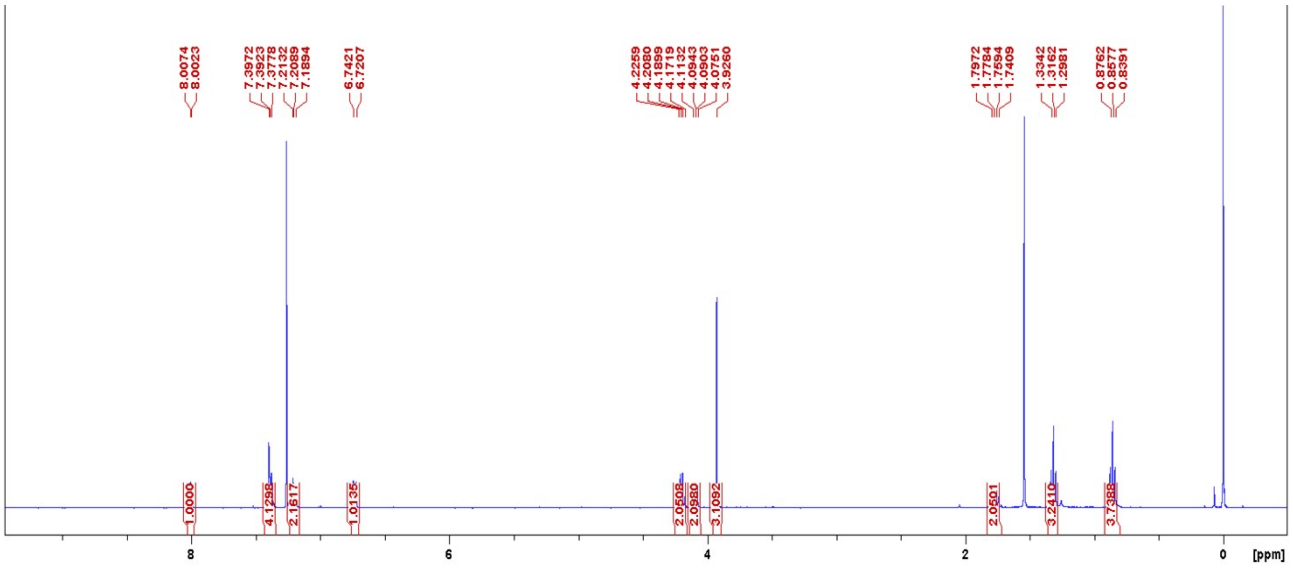

Figure S3. ^{1}H NMR spectrum (400 MHz) of **4** recorded in $CDCl_3$.

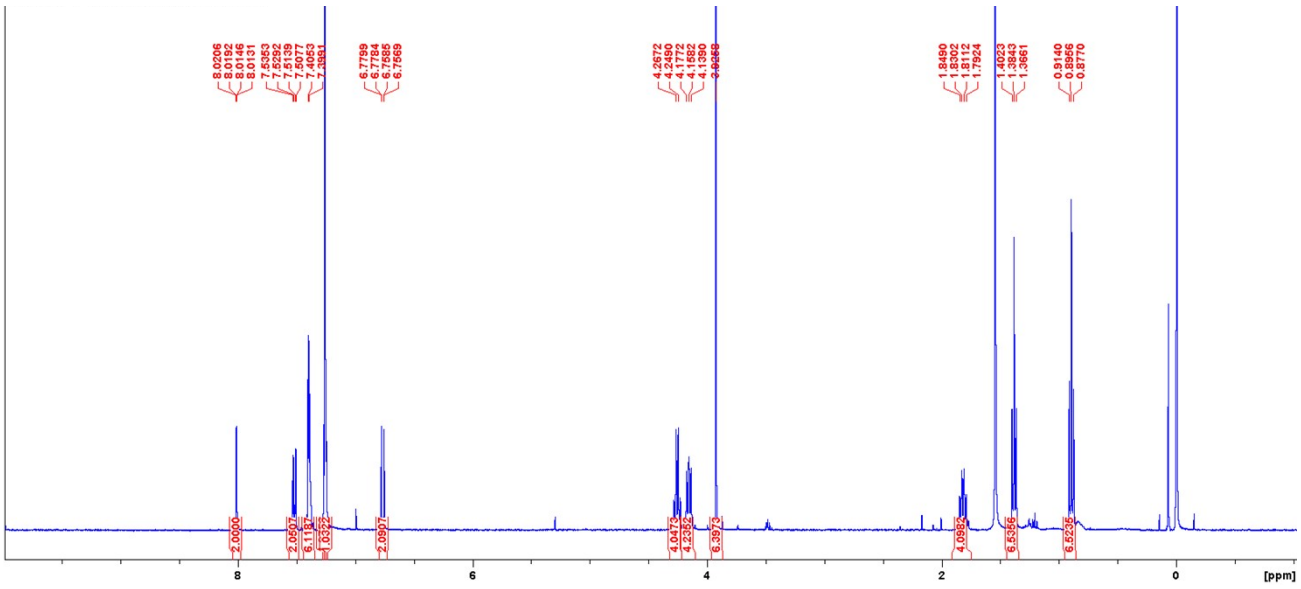

Figure S4. ^{1}H NMR spectrum (400 MHz) of **5** recorded in $CDCl_3$.

1.2 ^{13}C NMR spectra

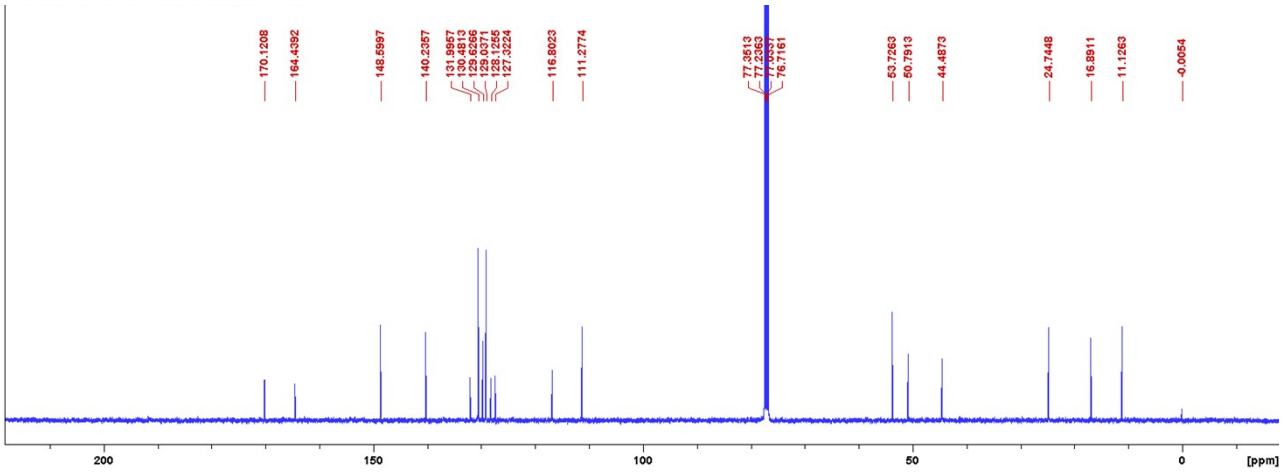

Figure S5. ^{13}C NMR spectrum (100 MHz) of **2** recorded in $CDCl_3$.

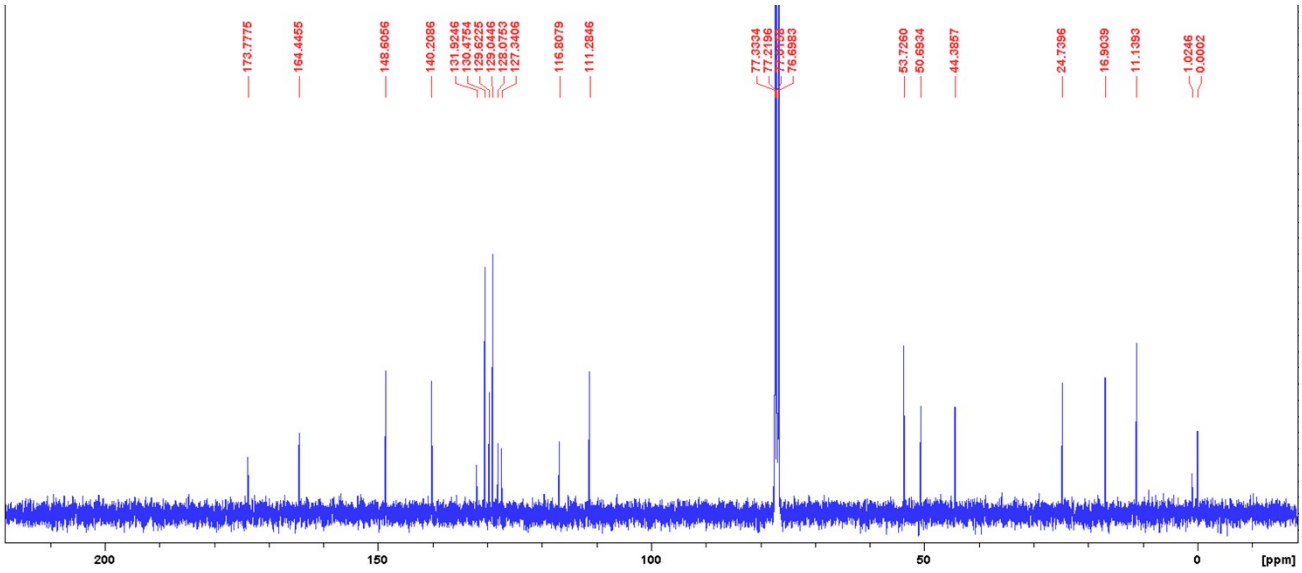

Figure S6. ^{13}C NMR spectrum (100 MHz) of **3** recorded in $CDCl_3$.

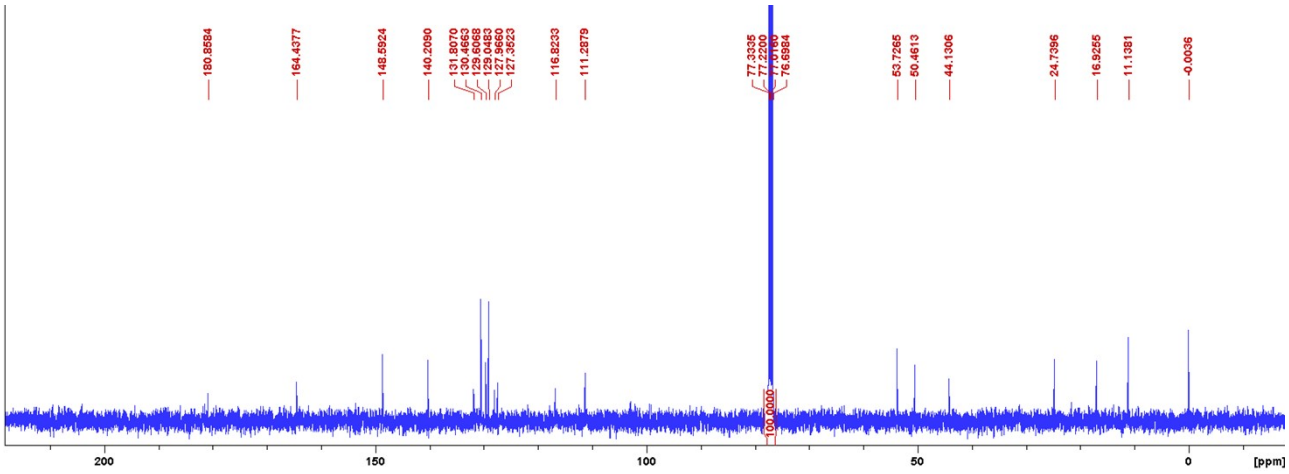

Figure S7. ^{13}C NMR spectrum (100 MHz) of **4** recorded in $CDCl_3$.

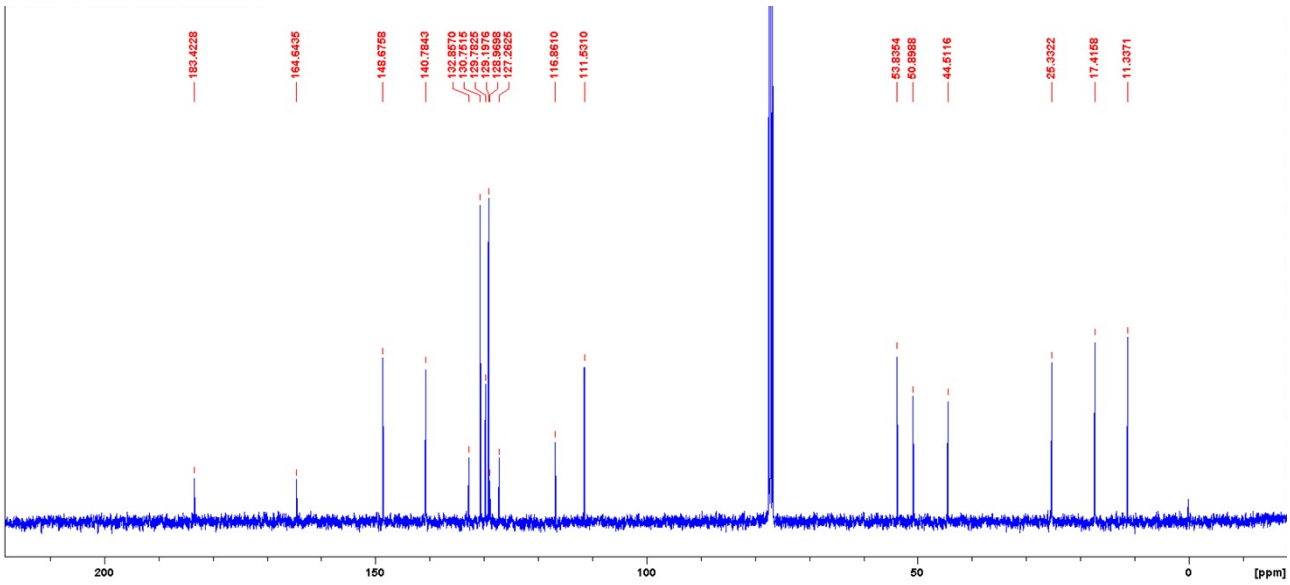

Figure S8. ^{13}C NMR spectrum (100 MHz) of **5** recorded in $CDCl_3$.

1.3 UV-Vis spectra

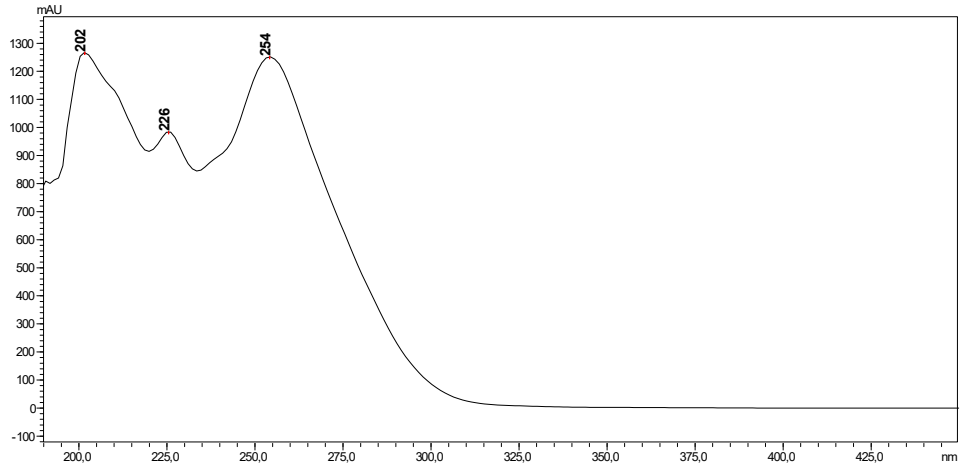

Figure S9. UV-Vis spectrum of **2**.

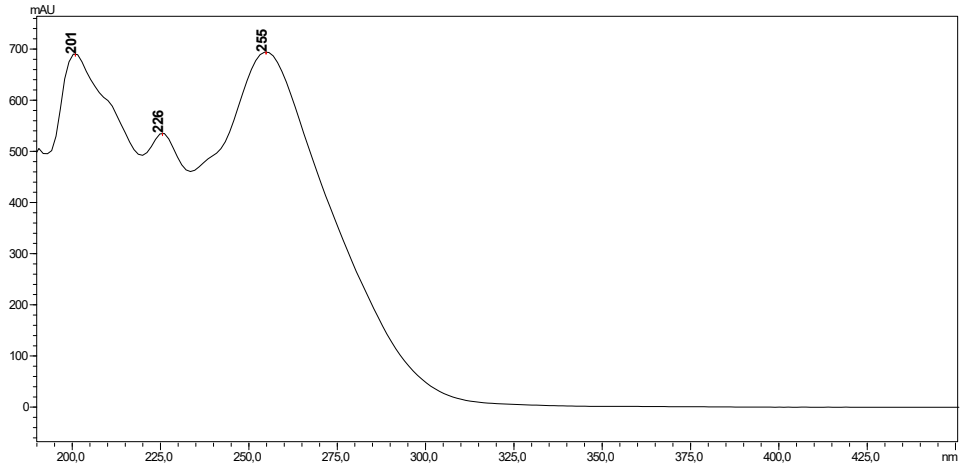

Figure S10. UV-Vis spectrum of **3**.

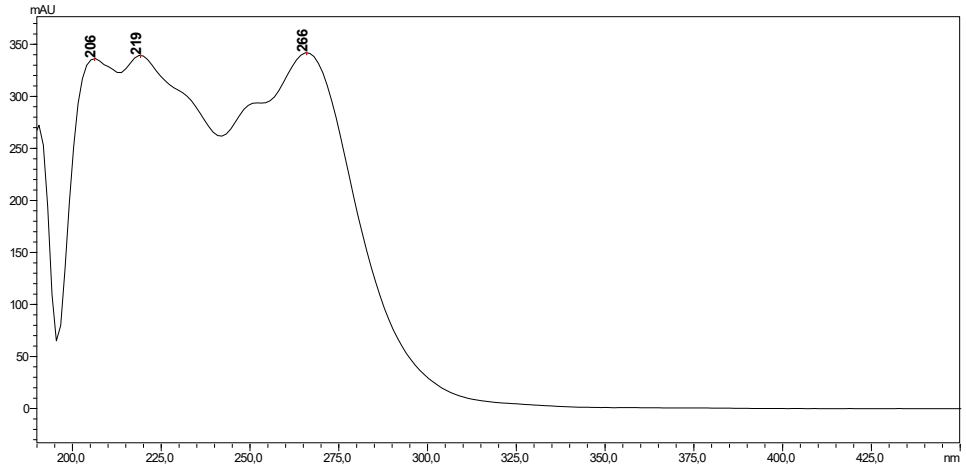

Figure S11. UV-Vis spectrum of **4**.

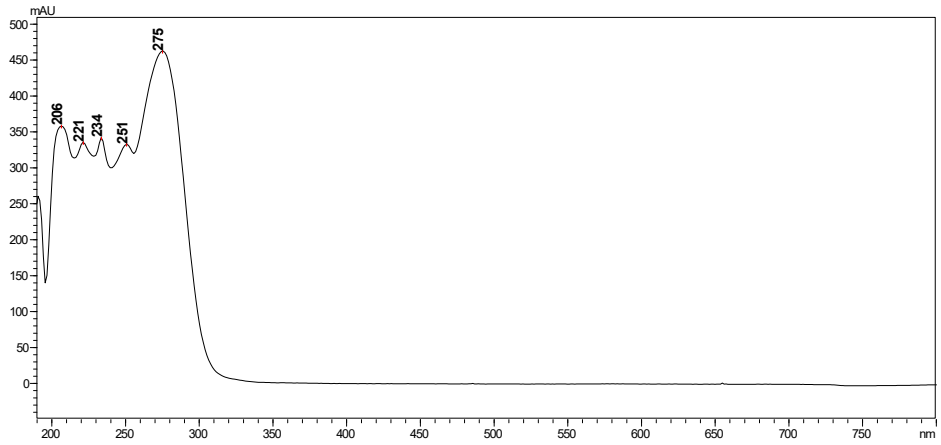

Figure S12. UV-Vis spectrum of **5**.

1.4 Electrospray ionization mass spectra

10 µL of the HPLC samples, taken at various time points, were diluted with a 0.1% formic acid containing ACN/water = 50/50 (v/v) mixture. Subsequently, the samples were analyzed on an Orbitrap Elite mass spectrometer (Thermo Fisher Scientific, Waltham, MA, USA) using direct infusion and the HESI source in positive mode.

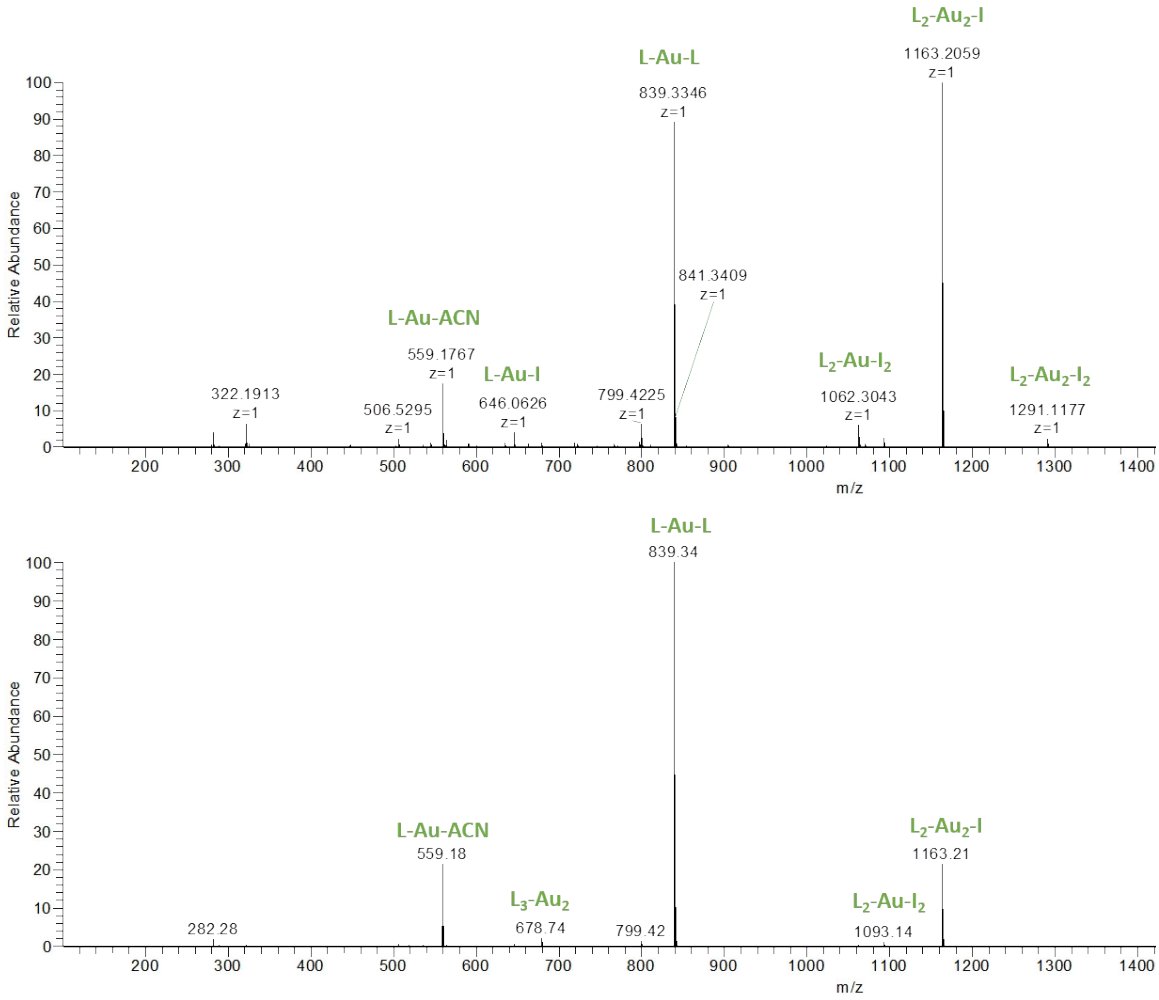

Figure S13. ESI-MS spectrum of **4** at t = 0 h in pure ACN (top) and ACN/water mixture = 50/50 (v/v) (bottom).

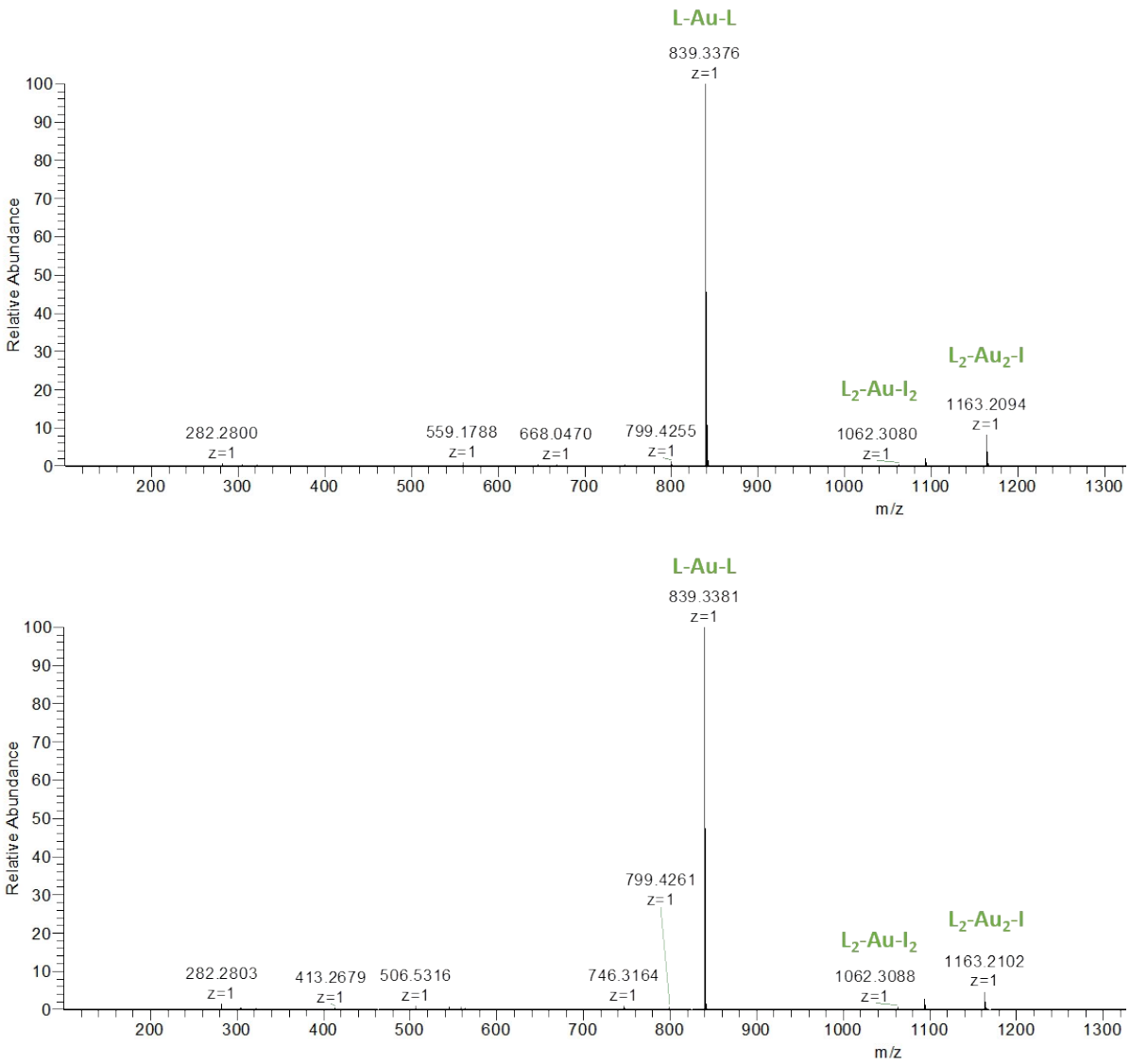

Figure S14. ESI-MS spectra of **4** at t = 24 h in pure ACN (top) and ACN/water mixture = 50/50 (v/v) (bottom).

2. Data of reactivity studies

Table S1. Relative peak areas [%] of **2**, **3**, **4** and **5** in ACN, ACN/water, ACN/water + 0.9%NaCl, ACN/water + KI, determined by HPLC-UV-Vis.

	t = 0h				t = 0.5 h					t = 8h					t = 72h					
ACN	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**			**2**	**3**	**4**	**5**		
2 (0.25mM)	96.95			3.05	96.85			3.15	96.09			3.91			96.23			3.77		
3 (0.25mM)		99.65		0.35		99.50		0.50		99.14		0.86				98.69		1.31		
4 (0.25mM)			98.74	1.26			98.09	1.91			91.45	8.55					83.57	16.43		
ACN/water	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**		**(7)**	**2**	**3**	**4**	**5**	**(6)**	**(7)**
2 (0.25mM)	95.23			4.77	94.89			5.11	94.54			5.46			90.13			9.87		
3 (0.25mM)		98.92		1.08		95.27		4.73		83.14		13.01		3.85		64.11		25.12		10.77
4 (0.25mM)			93.96	6.04			76.01	23.99			74.40	25.60					47.50	44.41	8.09	
ACN/water + 0.9% NaCl	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**			**2**	**3**	**4**	**5**		
2 (0.25mM)	97.68			2.32	97.80			2.20	97.89			2.11			97.96			2.04		
3 (0.25mM)	99.74	0.00		0.26	99.77	0.00		0.23	99.50	0.00		0.50			99.07	-		0.93		
4 (0.25mM)	55.36		42.88	1.76	52.83		38.41	8.77	51.32		35.24	13.44			51.35		34.79	14.07		
4 (1.0 mM)	30.29		64.92	4.79	25.50		55.91	17.47	25.66		56.35	17.36			23.11		58.87	17.72		
ACN/water + KI	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**	**2**	**3**	**4**	**5**	**6**		**2**	**3**	**4**	**5**	**6**	
2 (0.25 mM) + 1 eq. KI	0.00		95.81	4.19	0.00		79.02	20.98	0.00		75.80	24.20			0.00		75.31	24.69		
3 (1.0 mM) + 20 eq. KI		0.00	89.38	10.62		0.00	68.84	31.16		0.00	71.69	28.31				0.00	72.21	27.79		
3 (0.25mM) + 1 eq. KI		15.80	78.78	5.42			80.20	19.80			40.61	39.44	19.95				30.96	47.36	21.68	
4 (0.25 mM) + 1 eq. KI			98.09	1.91			84.24	15.76			76.12	23.88					75.24	24.76		

3. HPLC chromatograms obtained during the reactivity studies

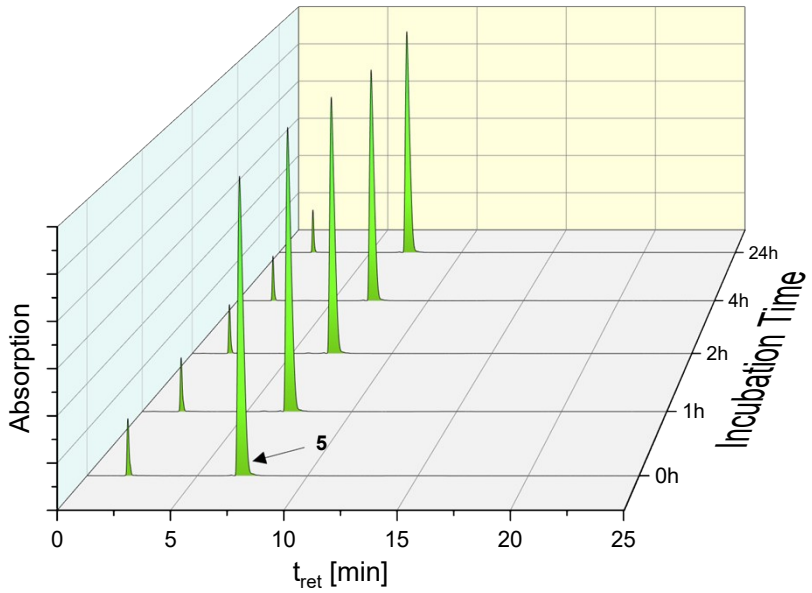

Figure S15. HPLC chromatogram of **5** in ACN/water mixture = 50/50 (v/v) with 1 eq. of KI.

4. Biological activity

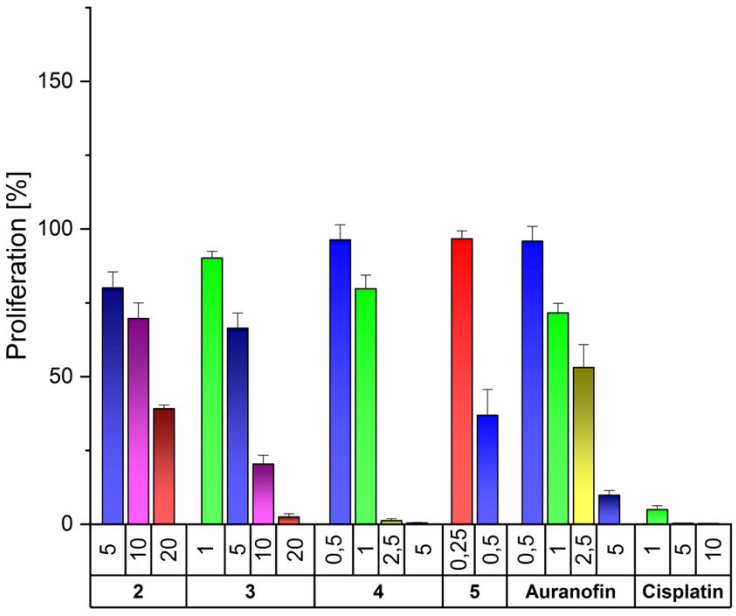

Figure S16. Anti-proliferative activity of complexes **2**, **3**, **4** and **5** in HL-60 cancer cells at different concentrations after 72 h incubation. Auranofin and Cisplatin served as reference. Values are given in µM. Proliferation in the absence of the compounds was set to 100%. The mean proliferation ± standard deviation was calculated from three independent experiments.

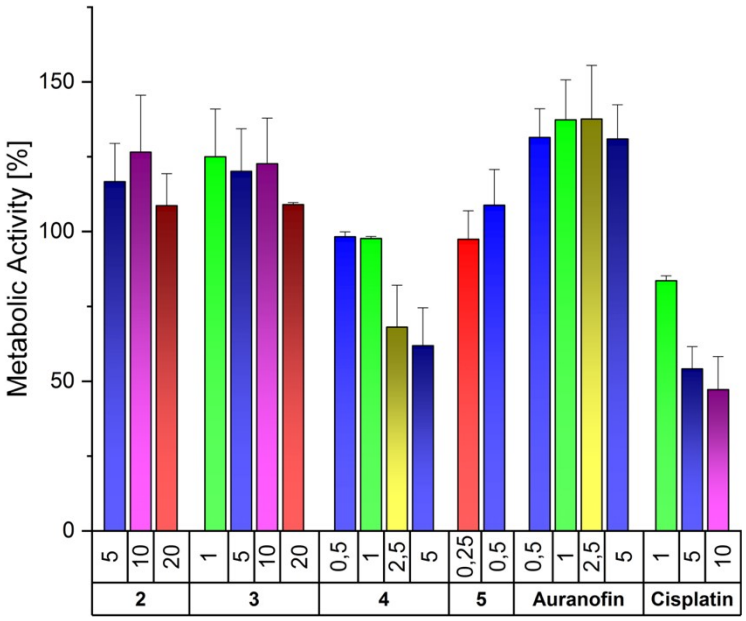

Figure S17. Metabolic activity of complexes **2**, **3**, **4** and **5** after 72 h incubation in HL-60 cells at different concentrations. Auranofin and Cisplatin served as reference. The metabolic activity in the absence of the compounds was set to 100%. The mean ± standard deviation was calculated from three independent experiments.

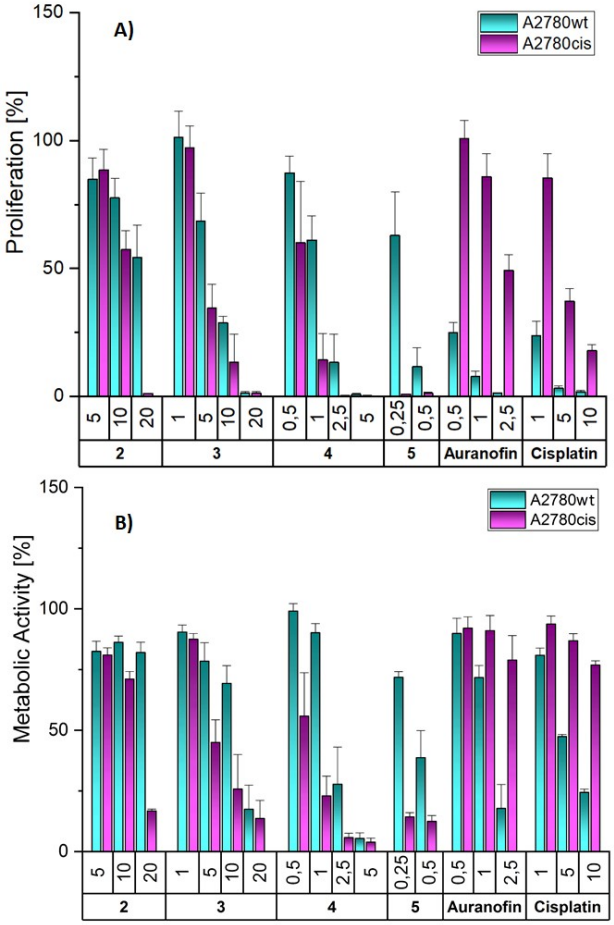

Figure S18. Antiproliferative (A) and antimetabolic activity (B) of **2**, **3**, **4**, and **5** at different concentrations in A2780wt (blue) and A2780cis (purple) cells after 72 h of incubation. Auranofin and Cisplatin served as reference. Compound concentrations are given in µM. Proliferation in the absence of the compounds was set at 100%. The mean value + standard error was calculated from three independent experiments.

5. Mass spectra, HPLC chromatograms and UV-Vis spectra of experiments with Sec, NAC, and Cys

5.1 HPLC chromatogram and UV-Vis spectra of **3** with Sec

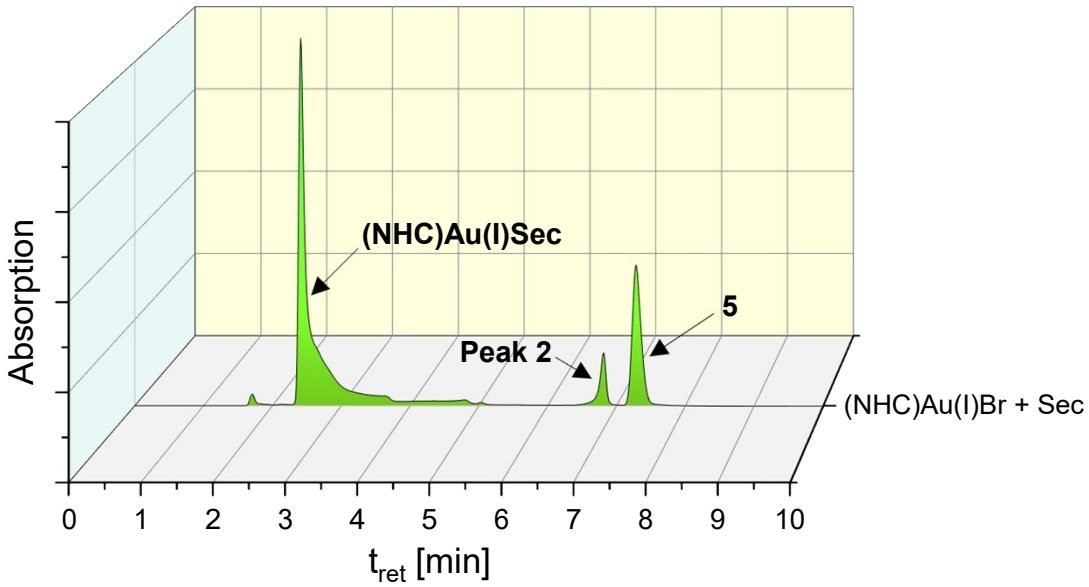

Figure S19. HPLC chromatogram of **3** in ACN/water mixture = 50/50 (v/v) with 1 eq. Sec taken after 5 min.

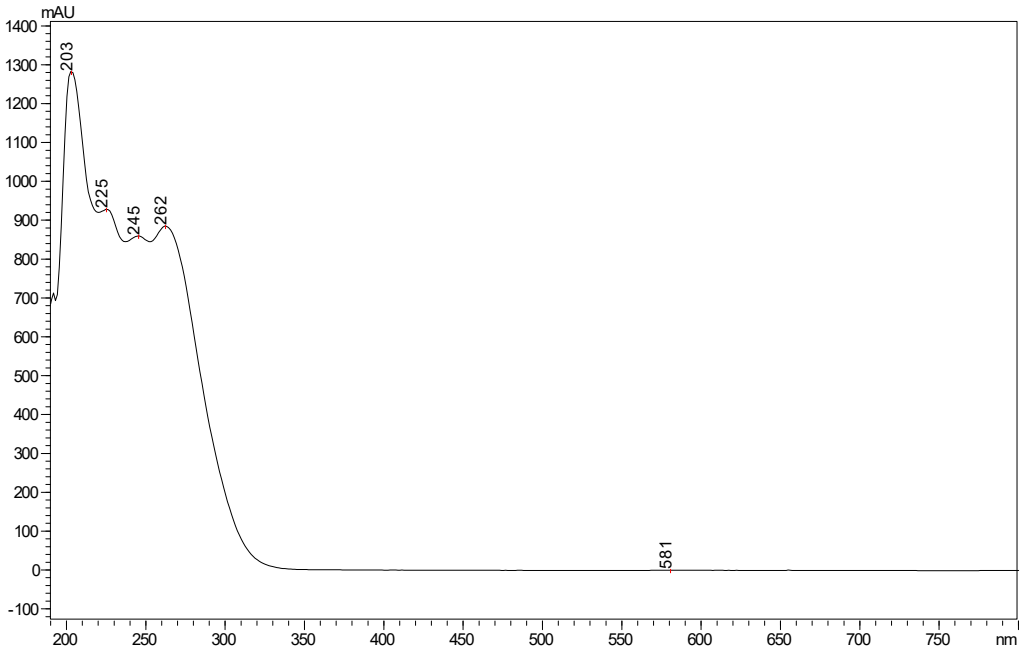

Figure S20. On-line UV-Vis spectrum of (NHC)Au(I)Sec (taken from the HPLC chromatogram **3** + Sec (Figure S19)).

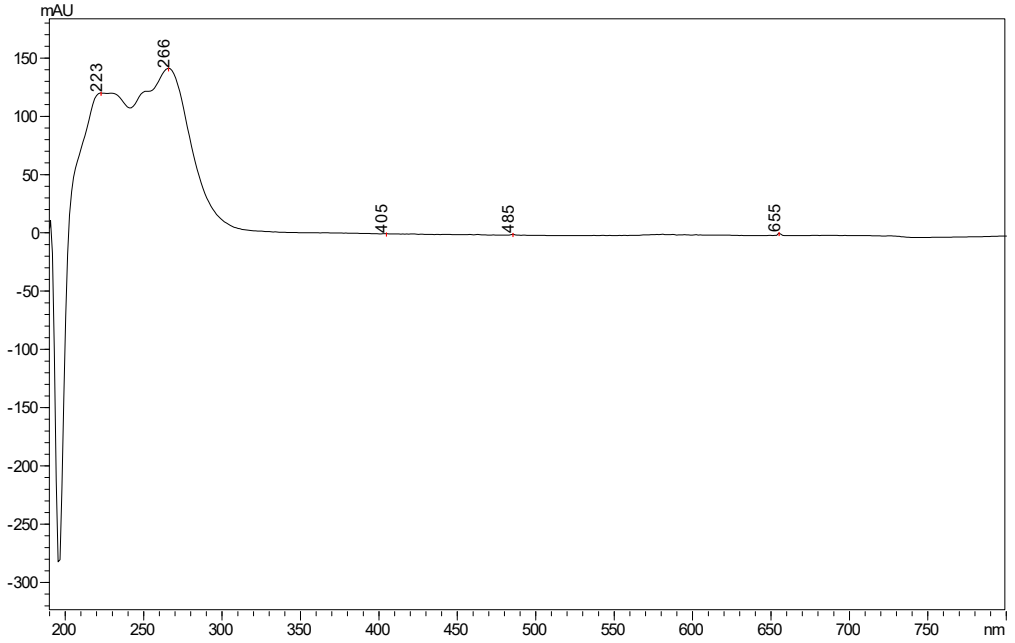

Figure S21. UV-Vis spectrum of peak 2 at t_{ret} = 6.81 min (taken from the HPLC chromatogram **3** + Sec (Figure S19)).

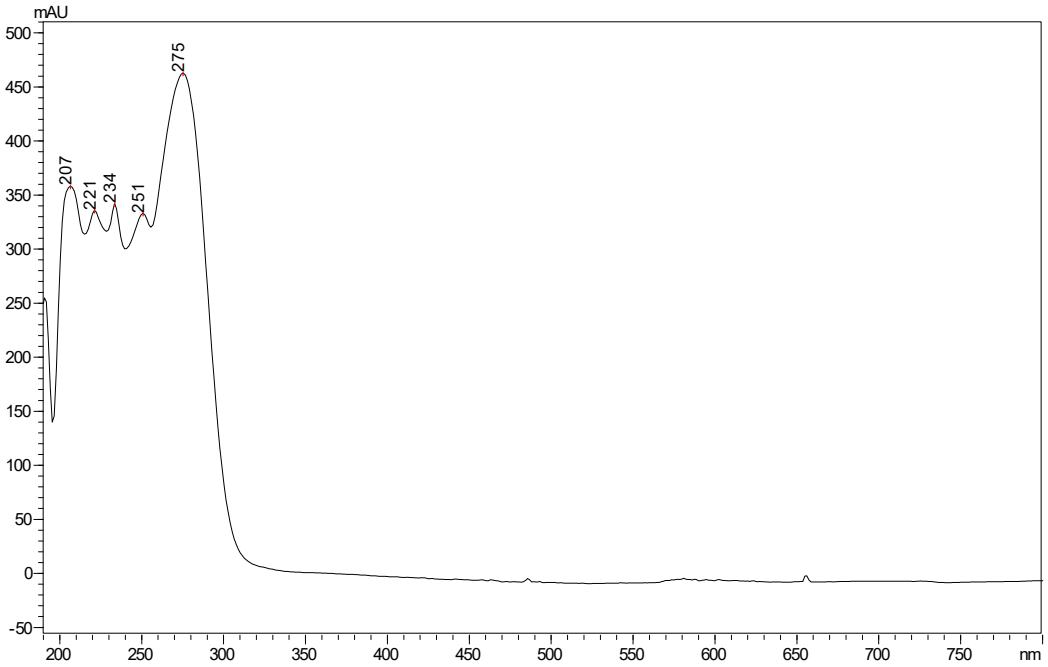

Figure S22. UV-Vis spectrum of **5** (taken from the HPLC chromatogram **3** + Sec (Figure S19)).

5.2 Mass spectra of experiments with **3** and Sec

10 µL of the HPLC samples, incubated for different lengths of time, were diluted with a 0.1% formic acid containing ACN/water = 50/50 (v/v) mixture. Subsequently, the samples were analysed on an Orbitrap Elite mass spectrometer (Thermo Fisher Scientific, Waltham, MA, USA) using direct infusion and the HESI source in positive mode.

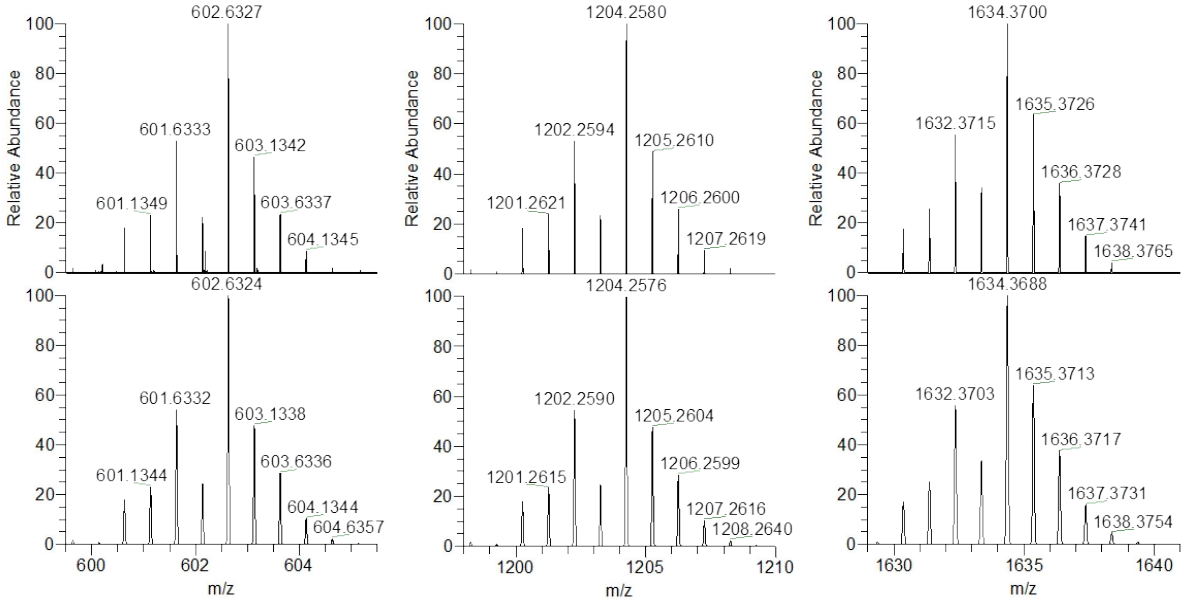

Figure S23. Experimental (top) and simulated (bottom) isotopic distributions of Se-containing species $(NHC)_2Au_2Sec$ (m/z 602 and 1204) and $(NHC)_3Au_3Se$ (m/z 1634).

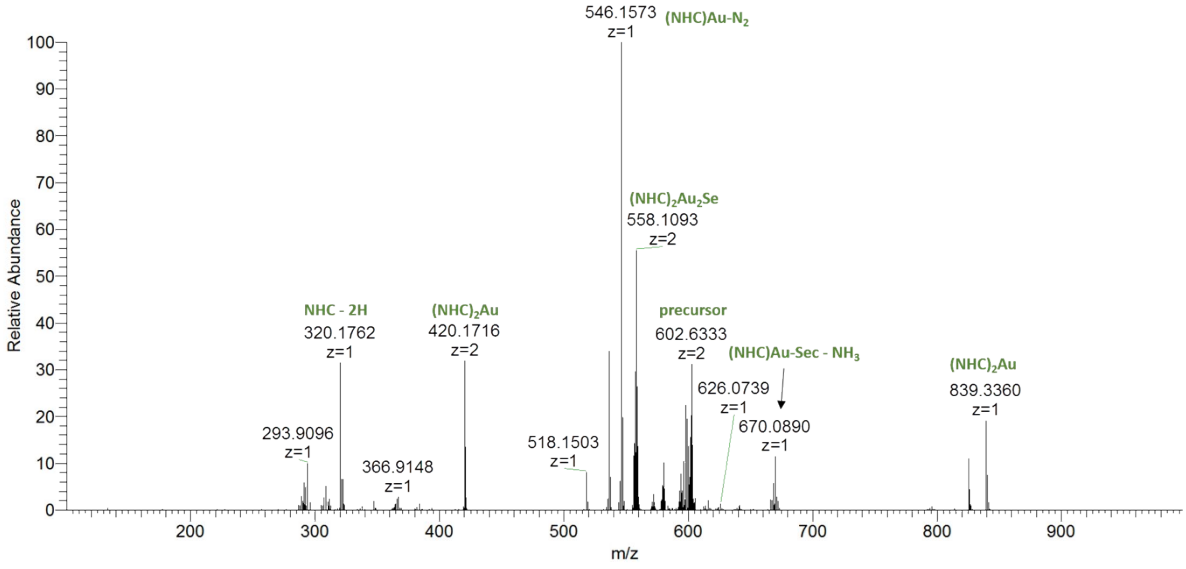

Figure S24. HCD fragmentation spectrum of m/z 602.

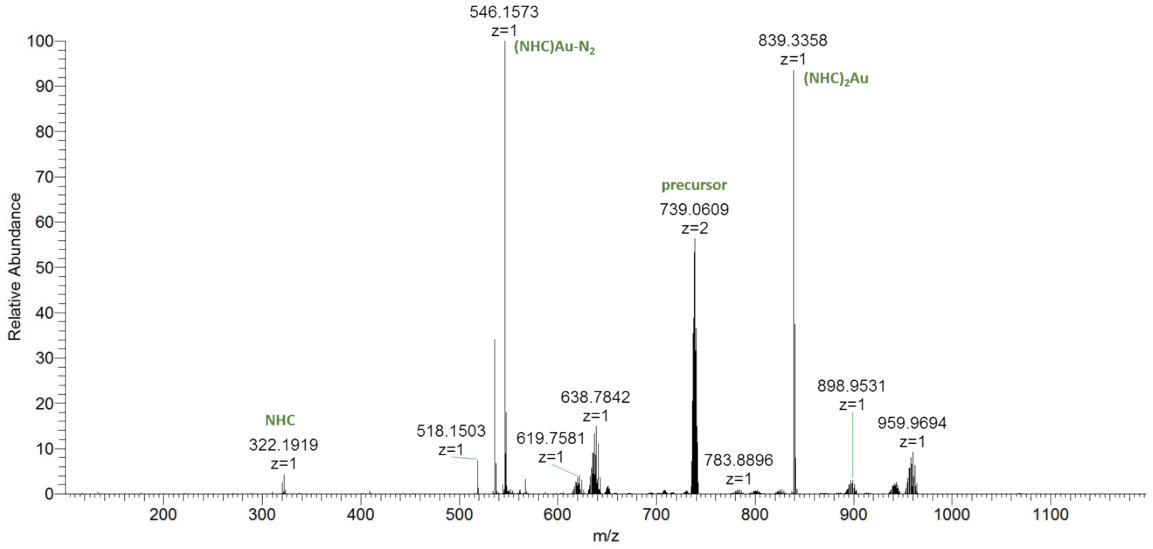

Figure S25. HCD fragmentation spectrum of m/z 739, which could not be identified. According to the isotopic distribution the species contains more than one Se. It may be a side product of an incomplete Sec reduction.

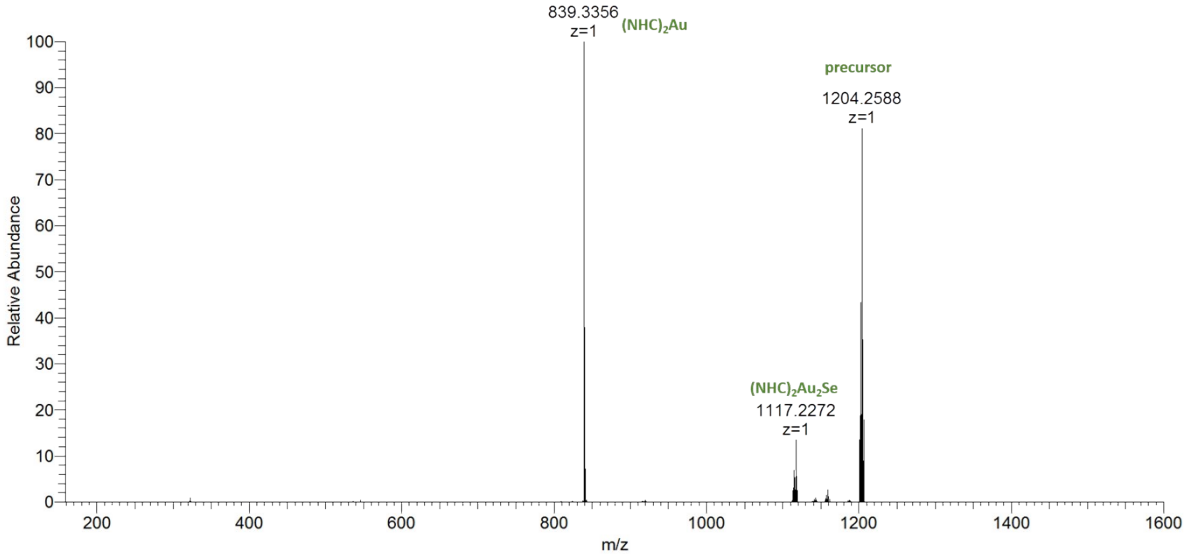

Figure S26. HCD fragmentation spectrum of m/z 1204.

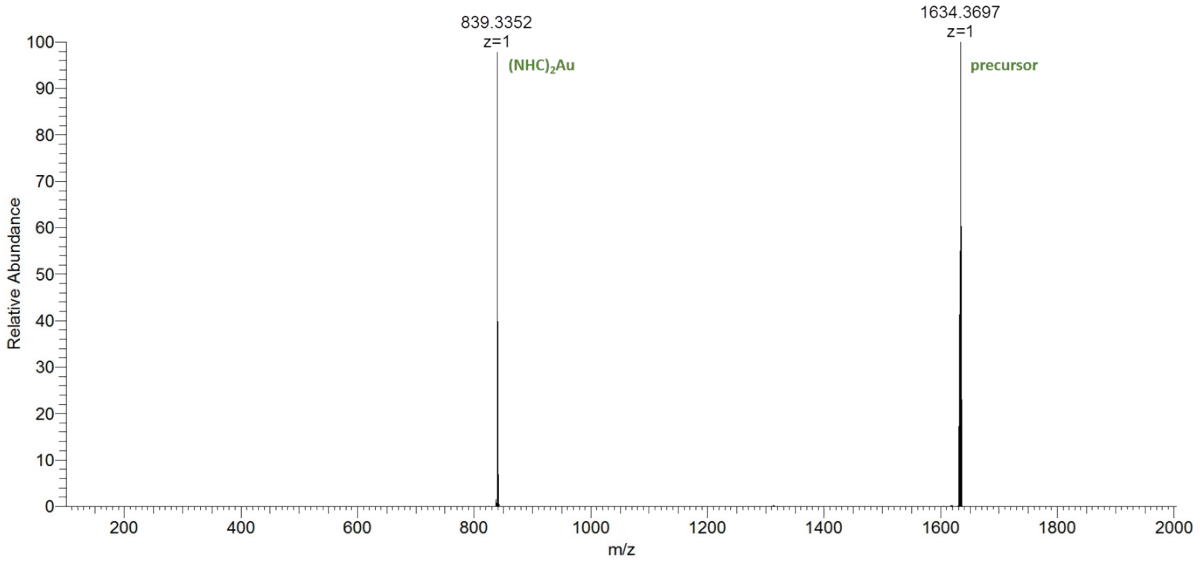

Figure S27. HCD fragmentation spectrum of m/z 1634.

5.3 HPLC chromatograms of **5** with Sec

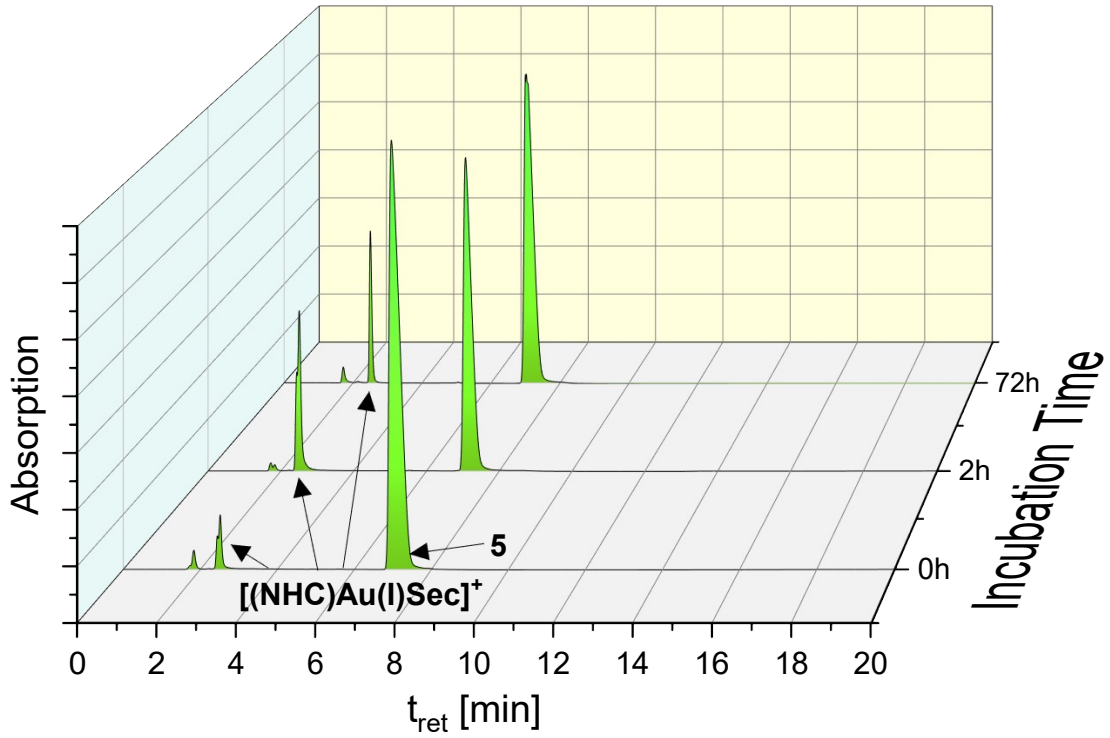

Figure S28. HPLC chromatograms of **5** in ACN/water mixture = 50/50 (v/v) with 1 eq. Sec.

5.4 HPLC chromatograms of **3** with NAC or Cys

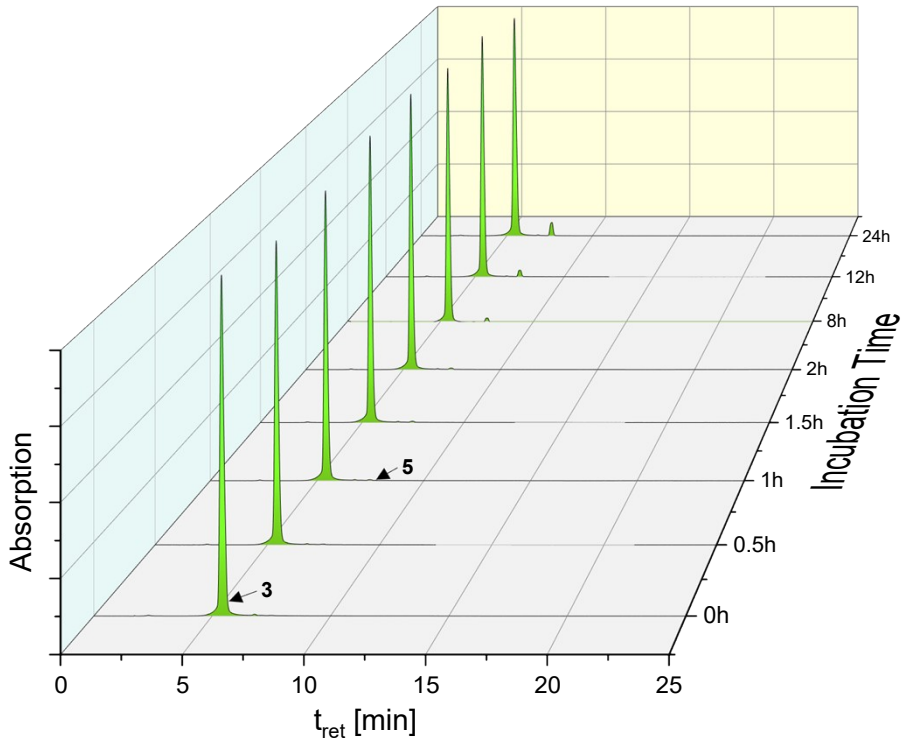

Figure S29. HPLC chromatograms of **3** in ACN/water mixture = 50/50 (v/v) with 1 eq. NAC.

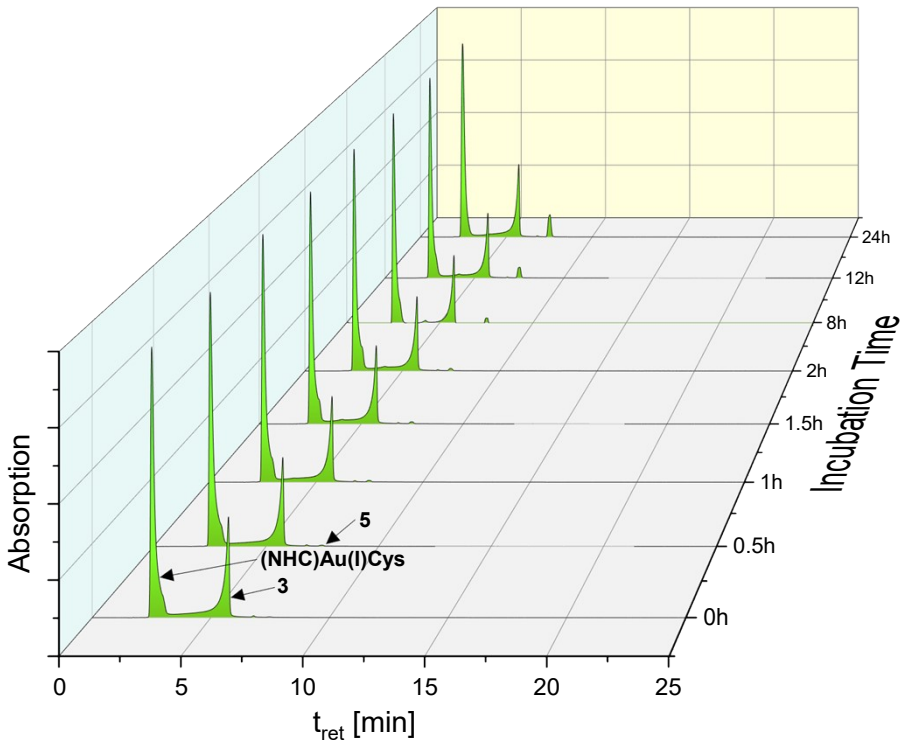

Figure S30. HPLC chromatograms of **3** in ACN/water mixture = 50/50 (v/v) with 1 eq. Cys.