Organometallic titanocene-gold compounds as potential chemotherapeutics in renal cancer. Study of their protein kinase inhibitory properties.

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Supporting Information

Contents:

1. Crystallographic Data for Compound 3

Table S1. Crystal Data and Structure Refinement for compound 3

formula	$C_{40}H_{36}Au_2Cl_8O_4P_2Ti$
fw	1368.06
T [K]	293 (2)
λ ($Mo_{K\alpha}$)[Å]	0.71073
crystal system	Triclinic
space group	P-1
a [Å]	10.298(2)
b [Å]	14.924(3)
c [Å]	15.579(3)
α [°]	84.15(3)
β [°]	80.48(3)
γ [°]	79.55(3)
V [Å]^3	2315.7(8)
Z	2
D_{calcd} (g cm^{-3})	1.962
μ (mm^{-1})	7.057
GOF	1.067
$R_1[I > 2\sigma]$	0.0599
wR_2(all data)	0.1593

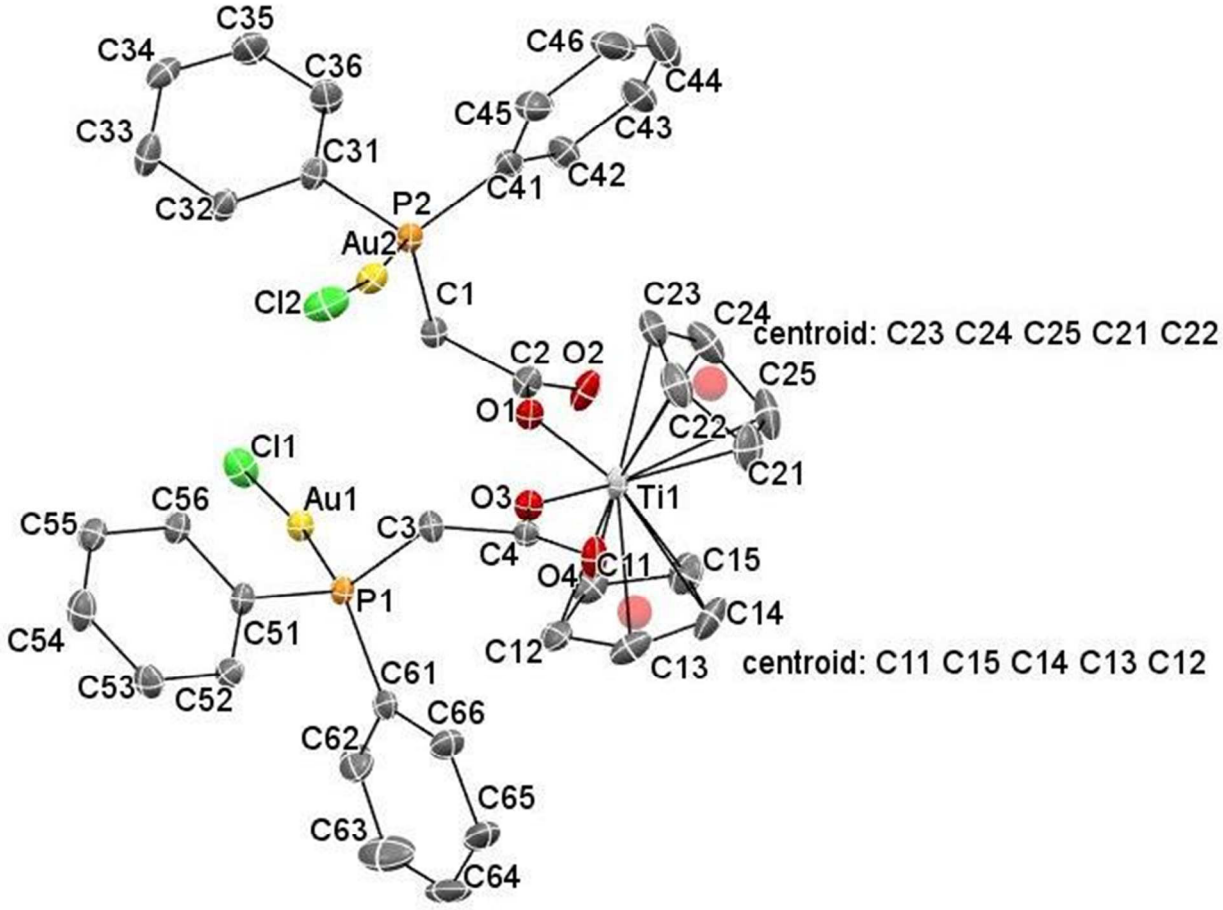

Figure S1. ORTEP views of the molecular structure of **3** showing the labelling scheme. Labelling for hydrogen atoms is omitted for clarity.

2. ^{1}H, $^{31}P\{^{1}H\}$ and $^{13}C\{^{1}H\}$ NMR spectra for compounds 4, 5, and 7 in $CDCl_3$

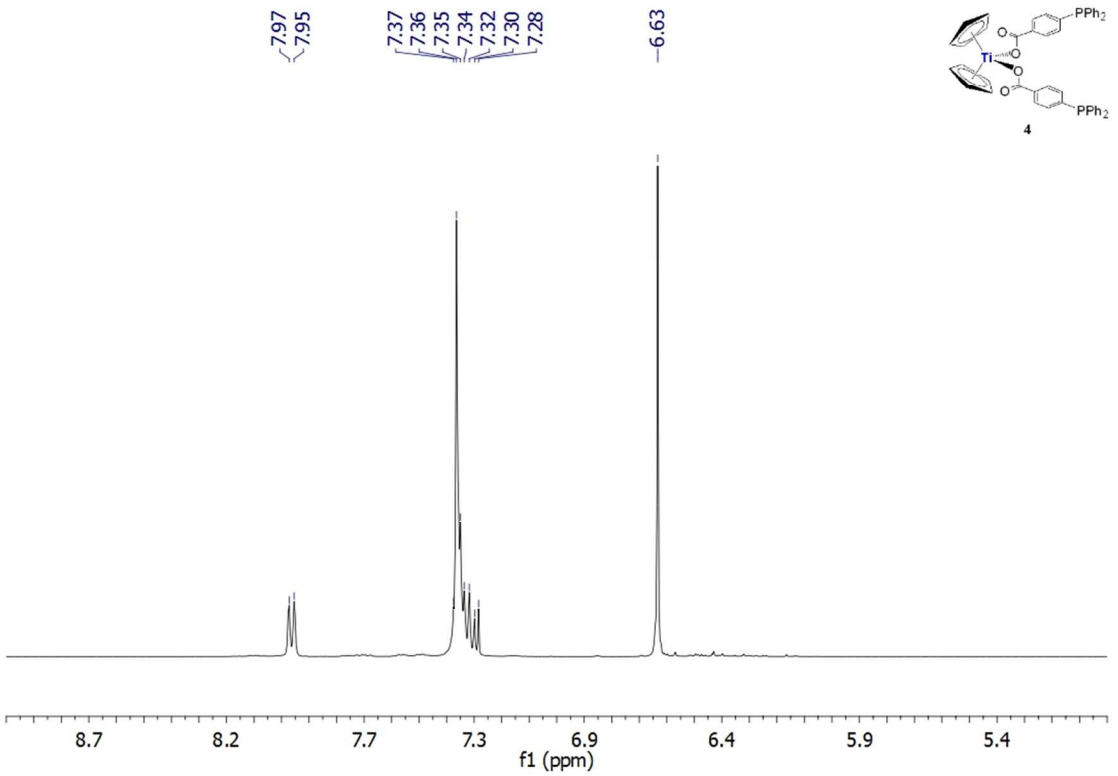

Figure S2. ^{1}H NMR spectrum of compound **4** in $CDCl_3$.

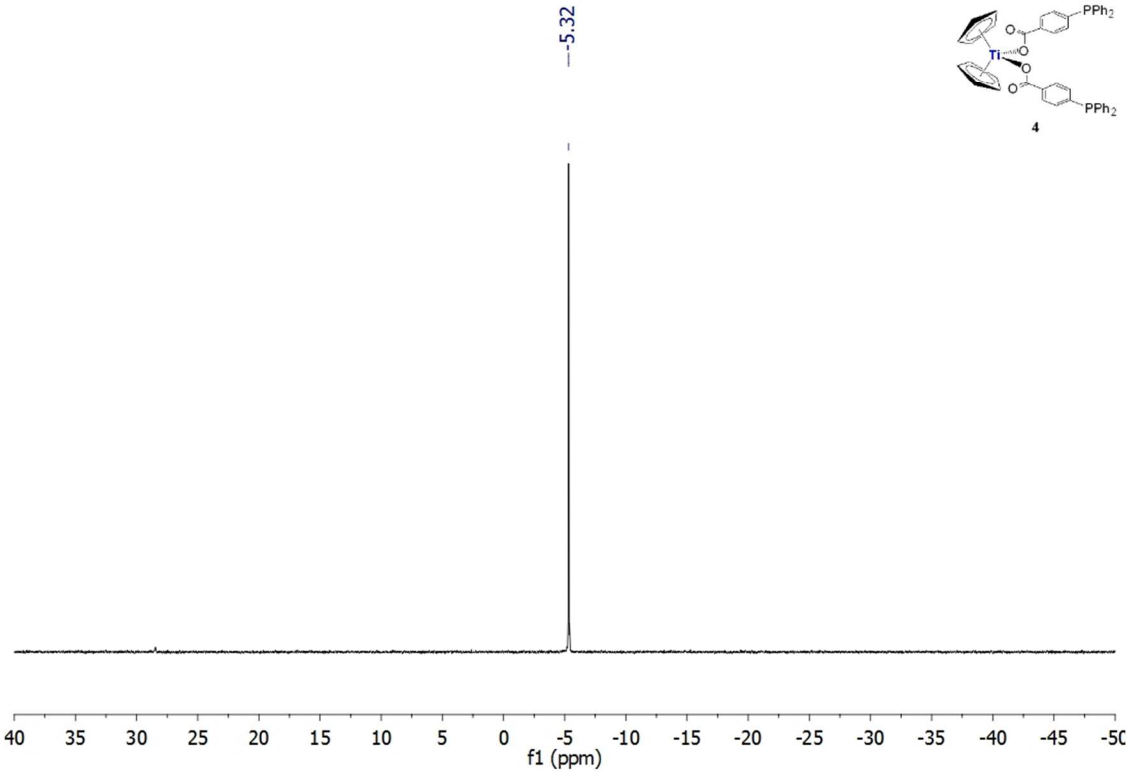

Figure S3. $^{31}P\{^{1}H\}$ NMR spectrum of compound **4** in $CDCl_3$.

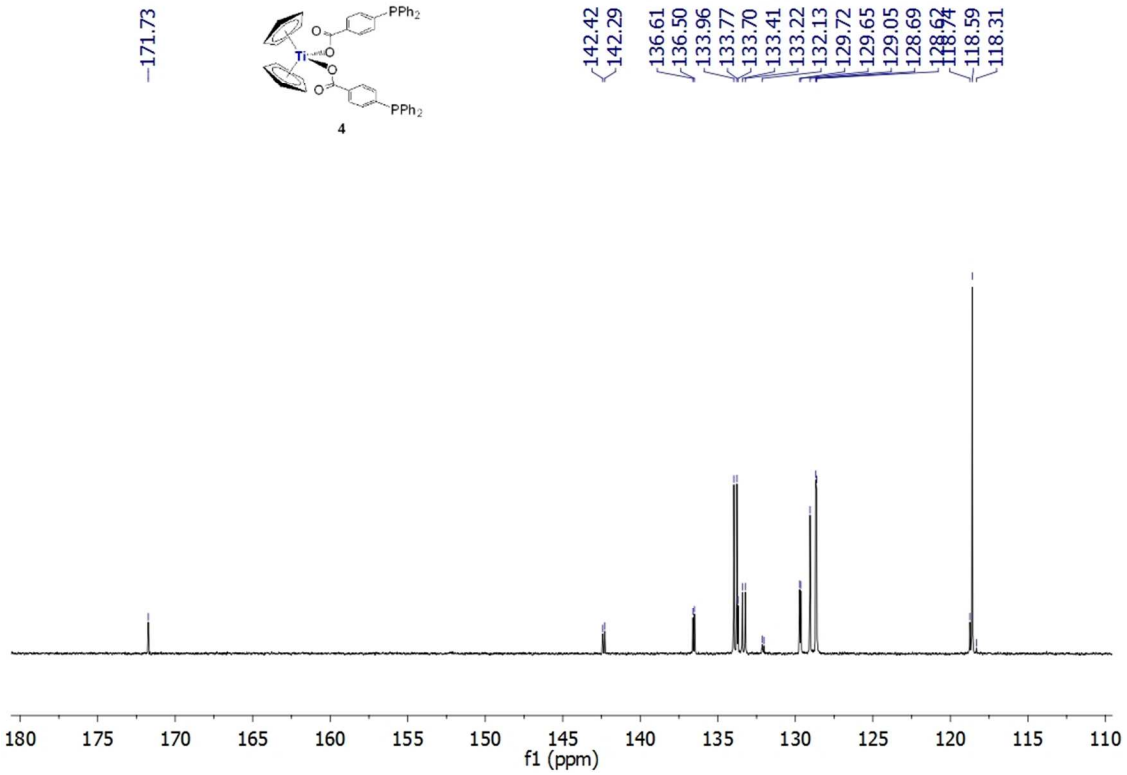

Figure S4. $^{13}C\{^{1}H\}$ NMR spectrum of compound **4** in $CDCl_3$.

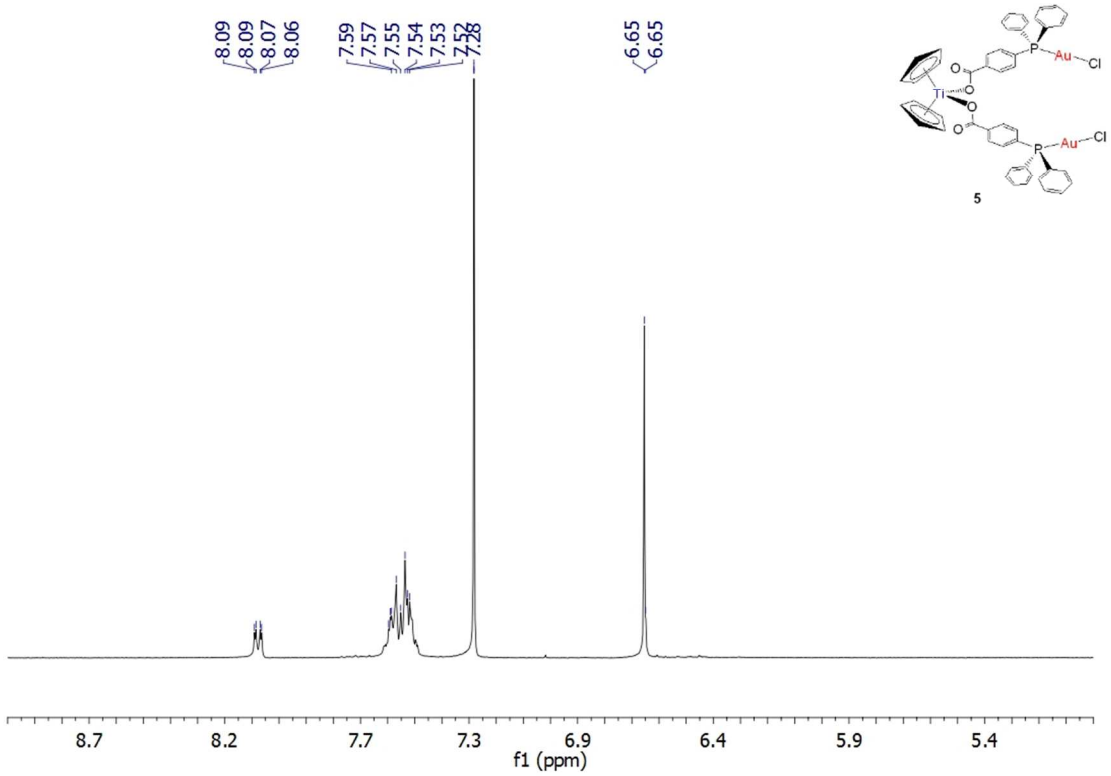

Figure S5. ^{1}H NMR spectrum of compound **5** in $CDCl_3$.

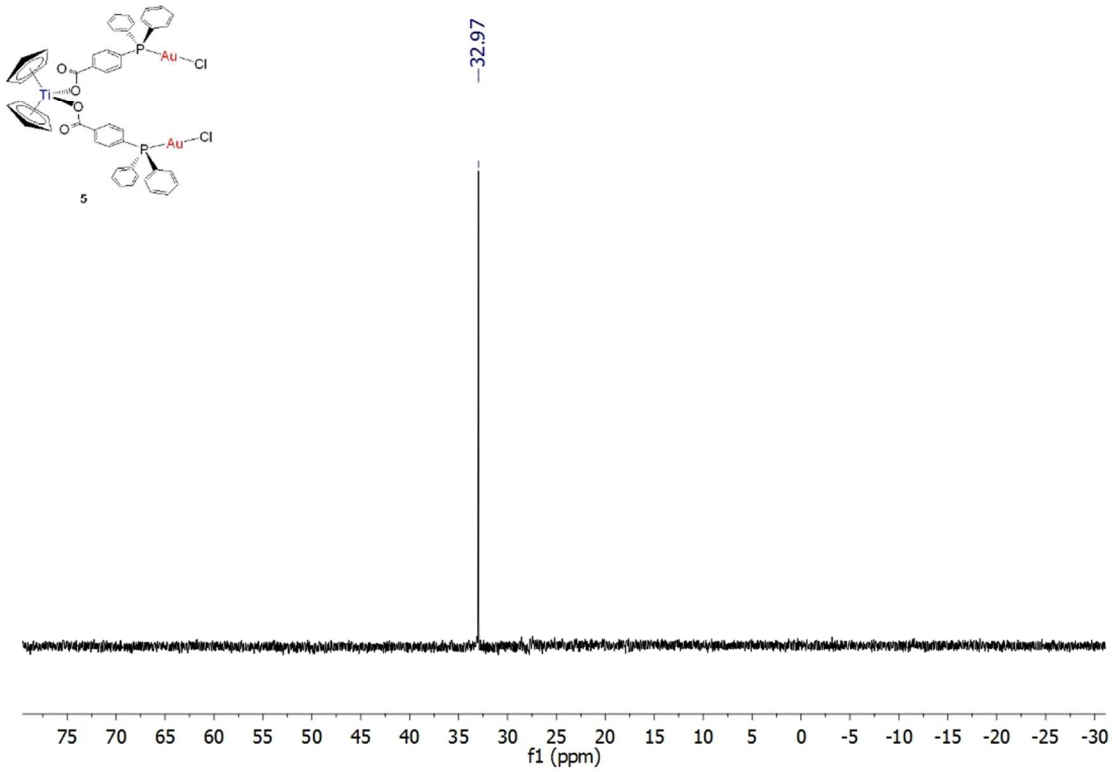

Figure S6. $^{31}P\{^{1}H\}$ NMR spectrum of compound **5** in $CDCl_3$.

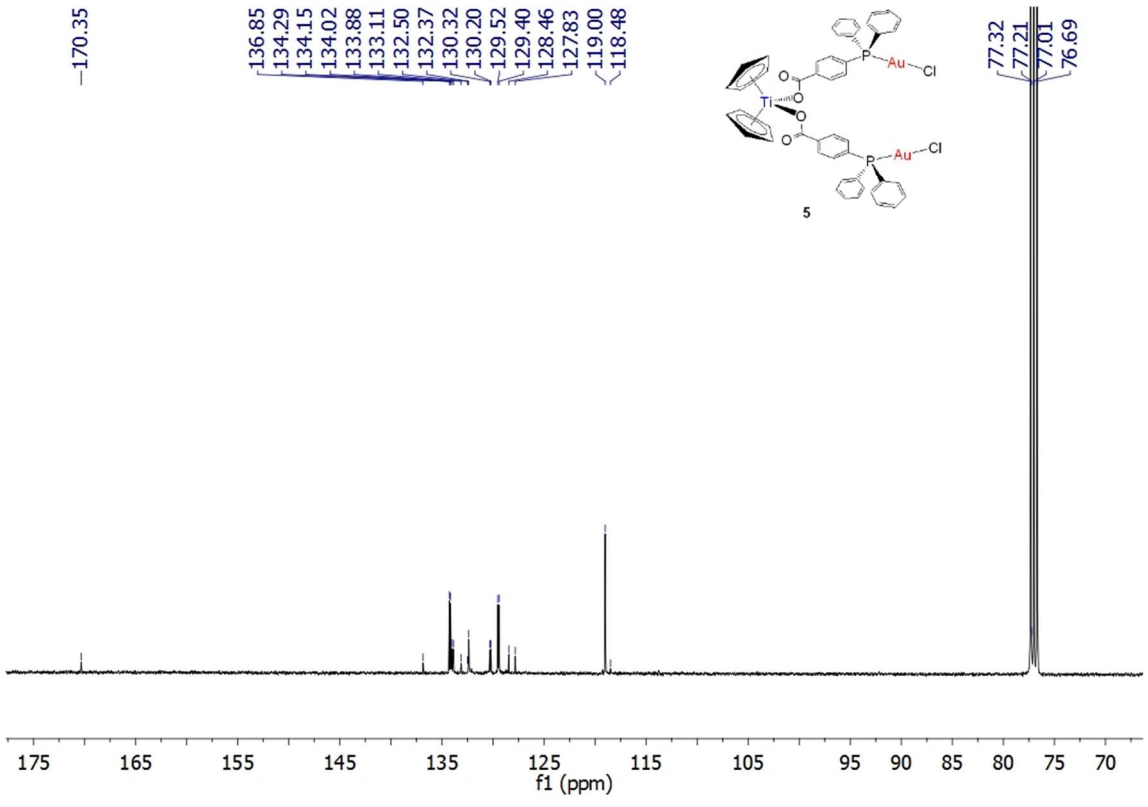

Figure S8. $^{13}C\{^{1}H\}$ NMR spectrum of compound **5** in $CDCl_3$.

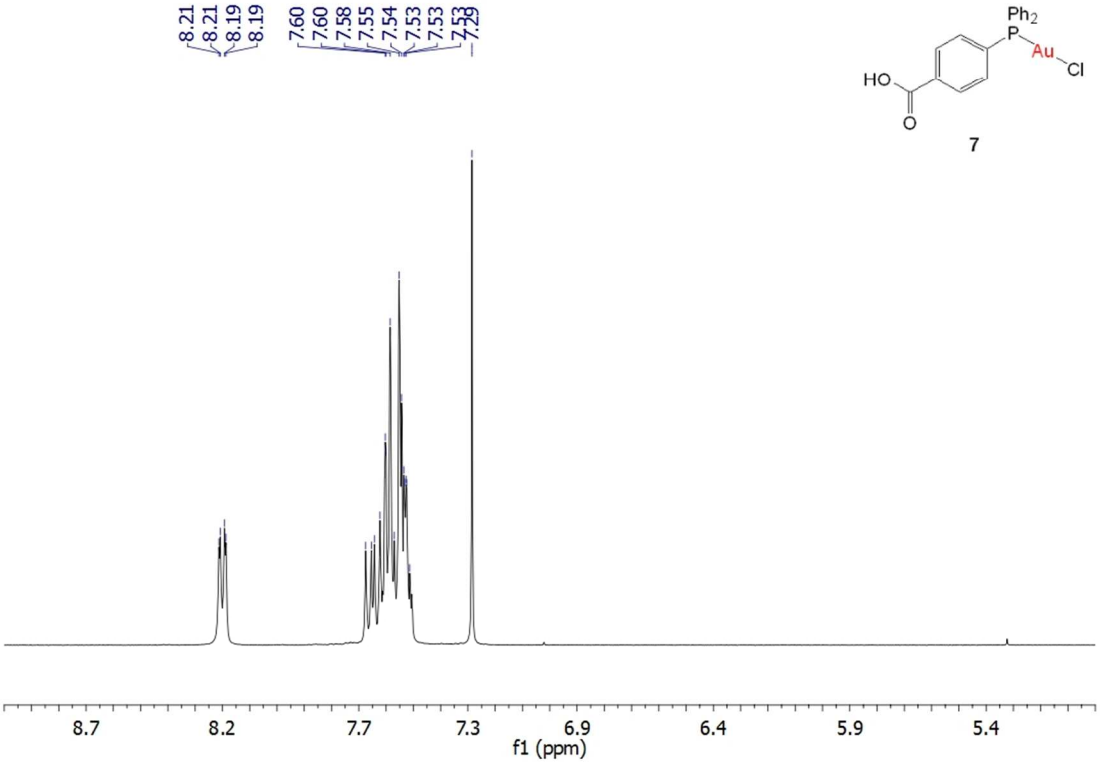

Figure S9. ^{1}H NMR spectrum of compound **7** in $CDCl_3$.

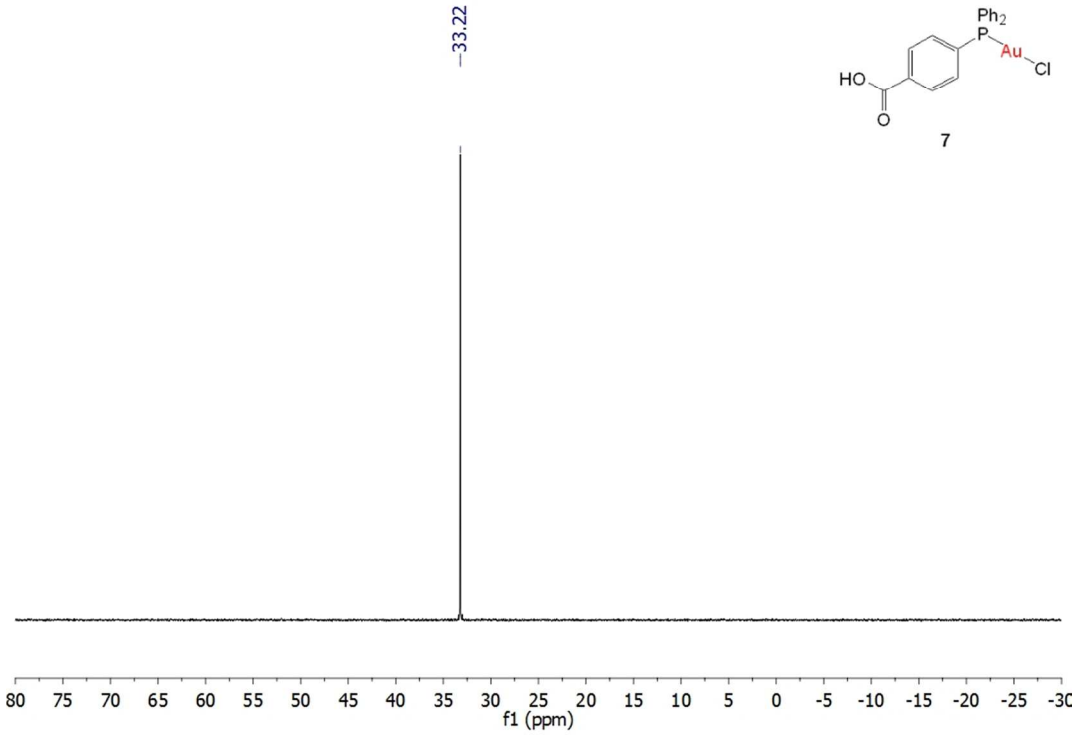

Figure S10. $^{31}P\{^{1}H\}$ NMR spectrum of compound **7** in $CDCl_3$.

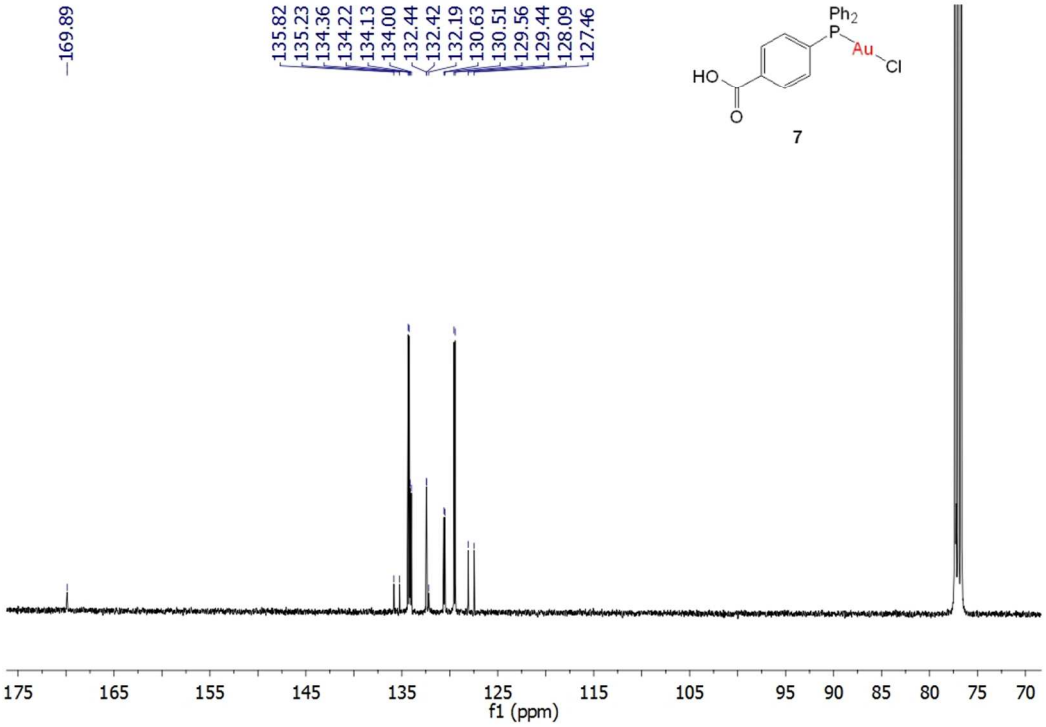

Figure S11. $^{13}C\{^{1}H\}$ NMR spectrum of compound **7** in $CDCl_3$.

3. Stability of compounds 3 and 5 in d^6-DMSO and in d^6-DMSO:D_2O (50:50) solution overtime assessed by $^{31}P\{^1H\}$and ^1H NMR spectroscopy.

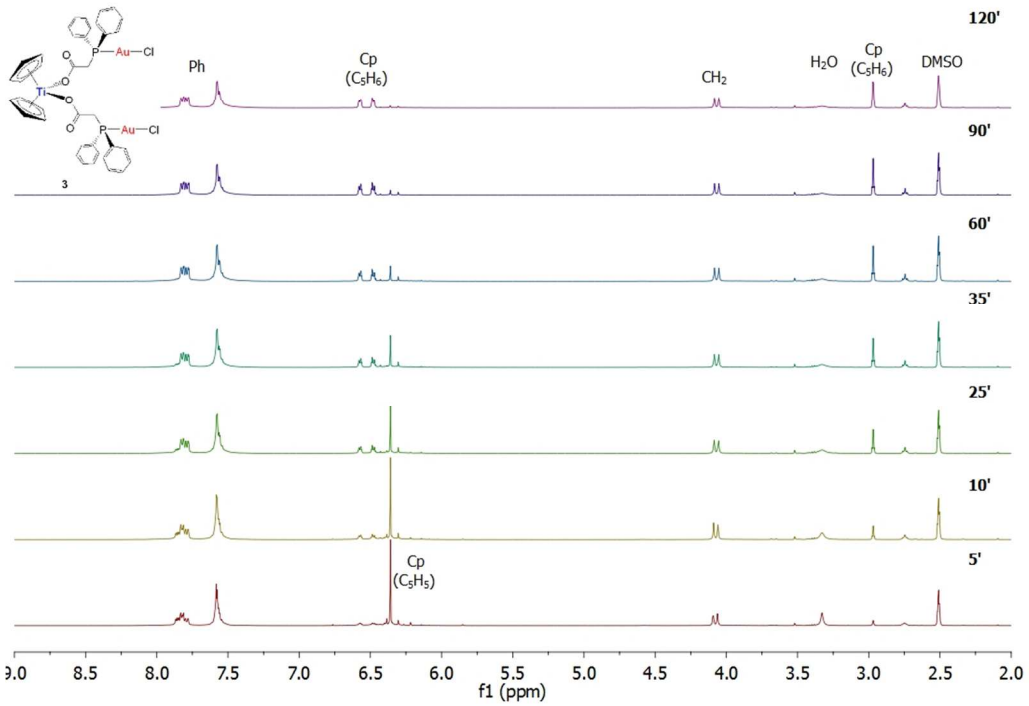

Figure S12. ^1H NMR spectrum in DMSO-d^6. Decomposition of compound **3** over time. $t_{½}$=10'.

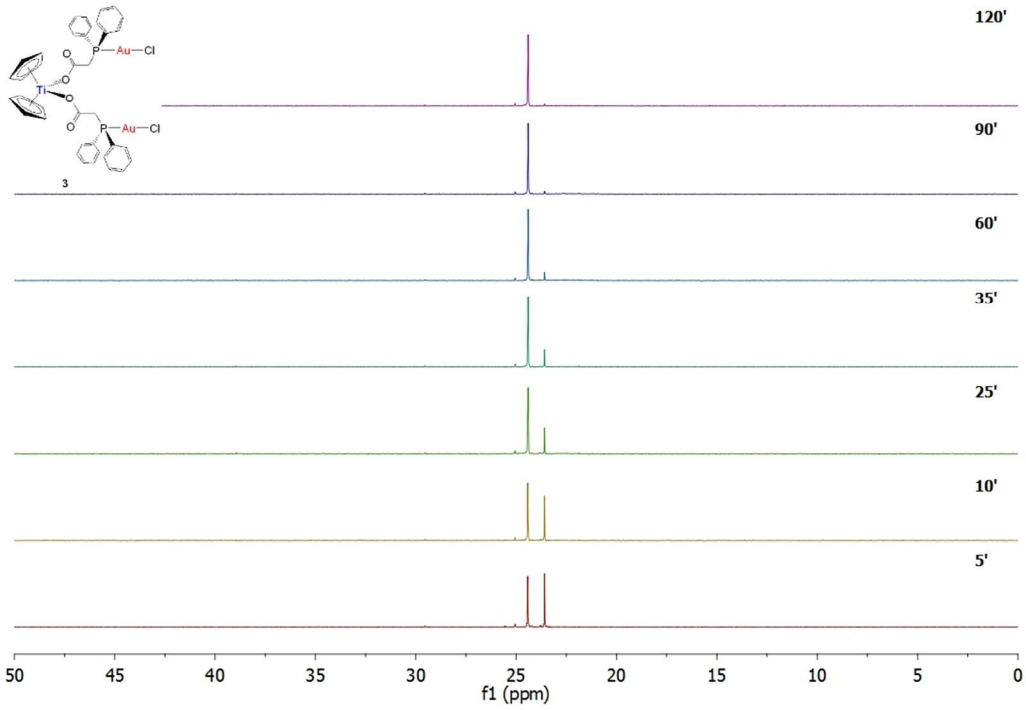

Figure S13. $^{31}P\{^1H\}$ NMR spectrum in DMSO-d^6. Decomposition of compound **3** over time. $t_{½}$=5'.

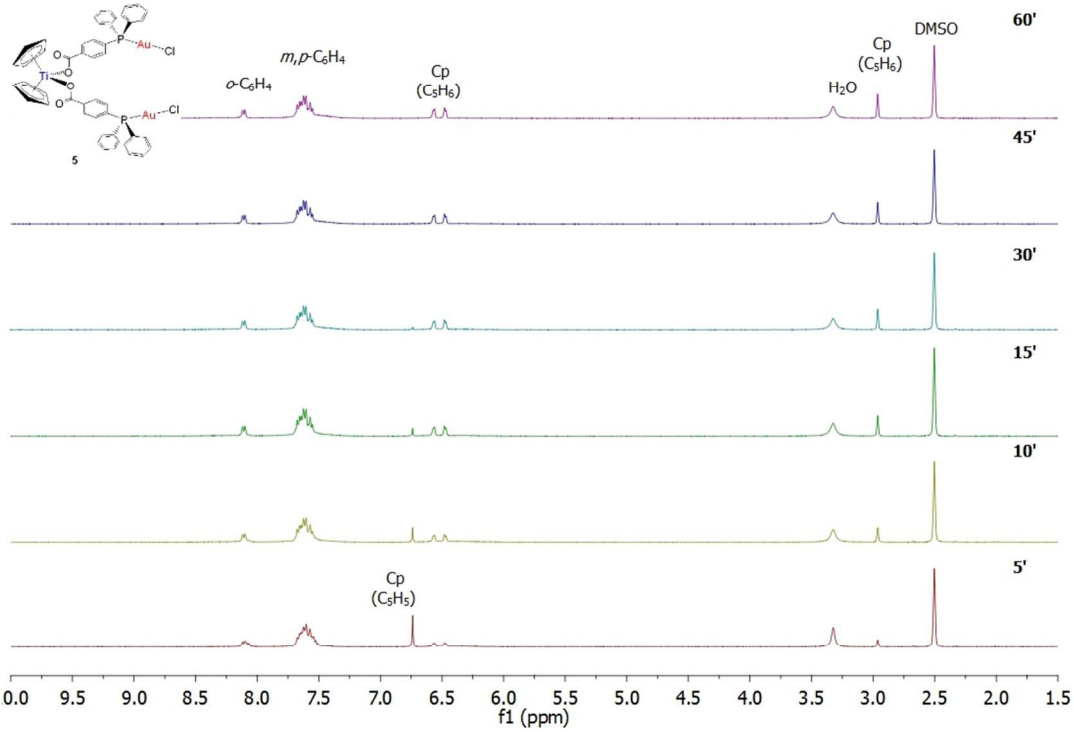

Figure S14. ^{1}H NMR spectrum in DMSO-d^{6}. Decomposition of compound **5** over time. $t_{½}$=5'.

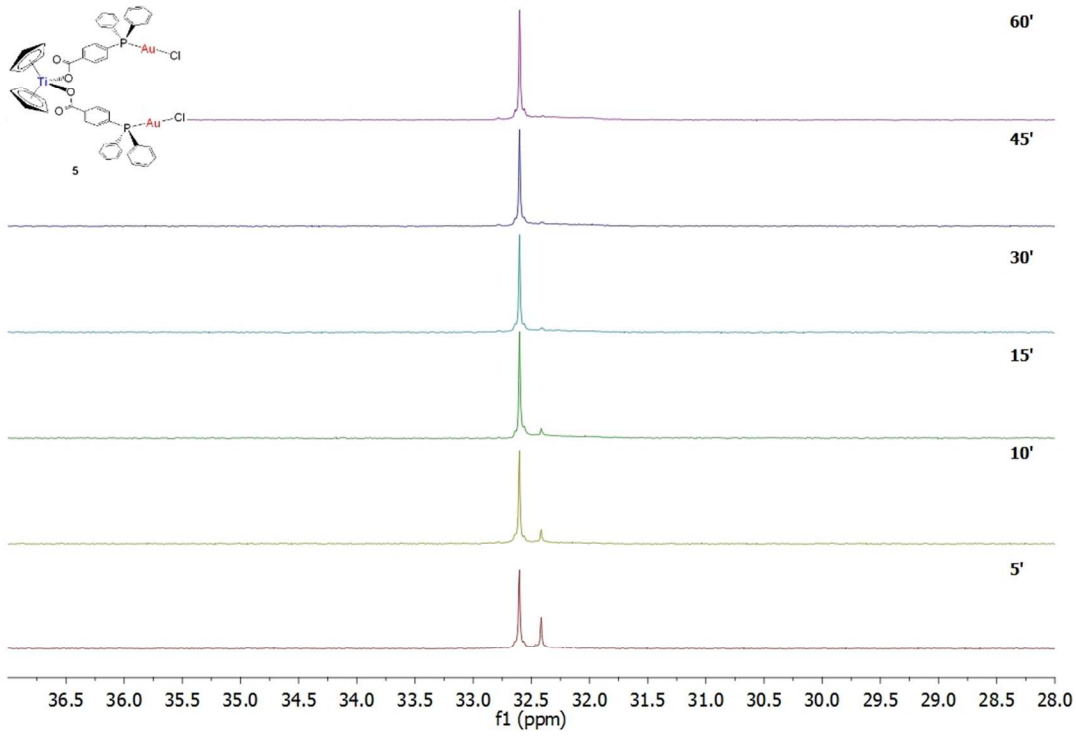

Figure S15. $^{31}P\{^{1}H\}$ NMR spectrum in DMSO-d^{6}. Decomposition of compound **5** over time. $t_{½}$<5'.

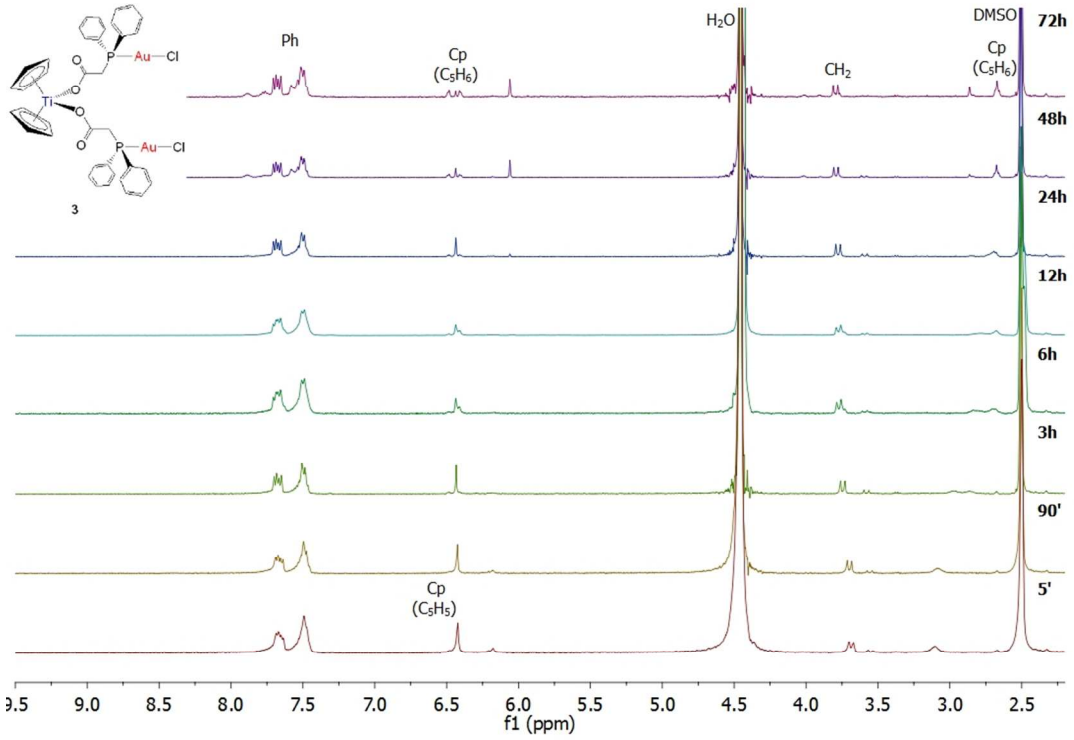

Figure S16. ^{1}H NMR spectrum in 50:50 DMSO-d^{6}/D_2O. Decomposition of compound **3** over time. $t_{½}$=24h.

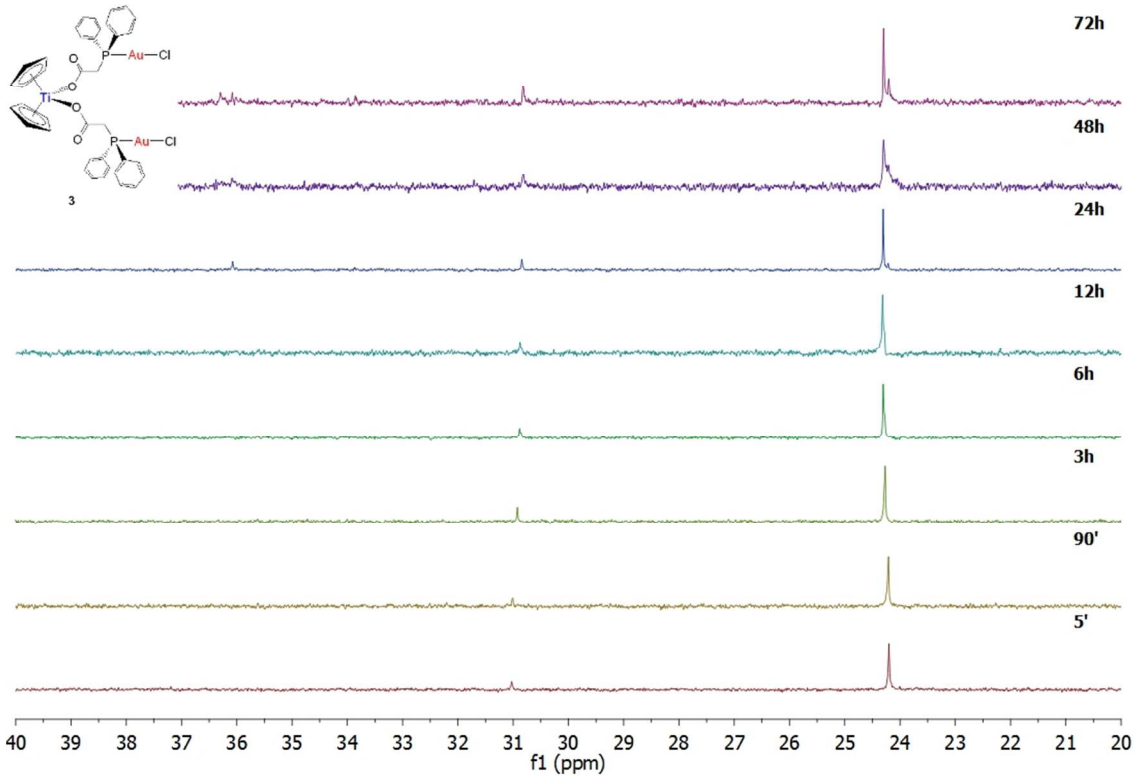

Figure S17. ^{31}P{^{1}H} NMR spectrum in 50:50 DMSO-d^{6}/D_2O. Decomposition of compound **3** over time. $t_{½}$=48h.

4. UV-Vis spectra of compounds 3 and 5 in CH_2Cl_2, in DMSO and in 1%DMSO-PBS solution overtime.

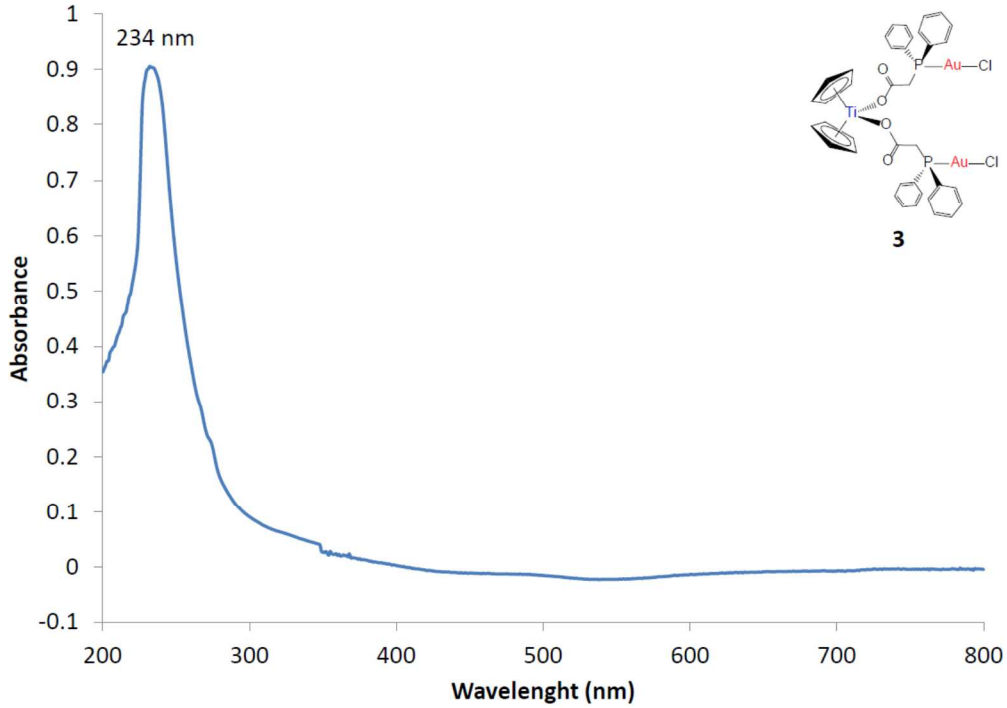

Figure S18. UV-visible spectrum of compound **3** ($3.2x10^{-5}$ M) in dichloromethane.

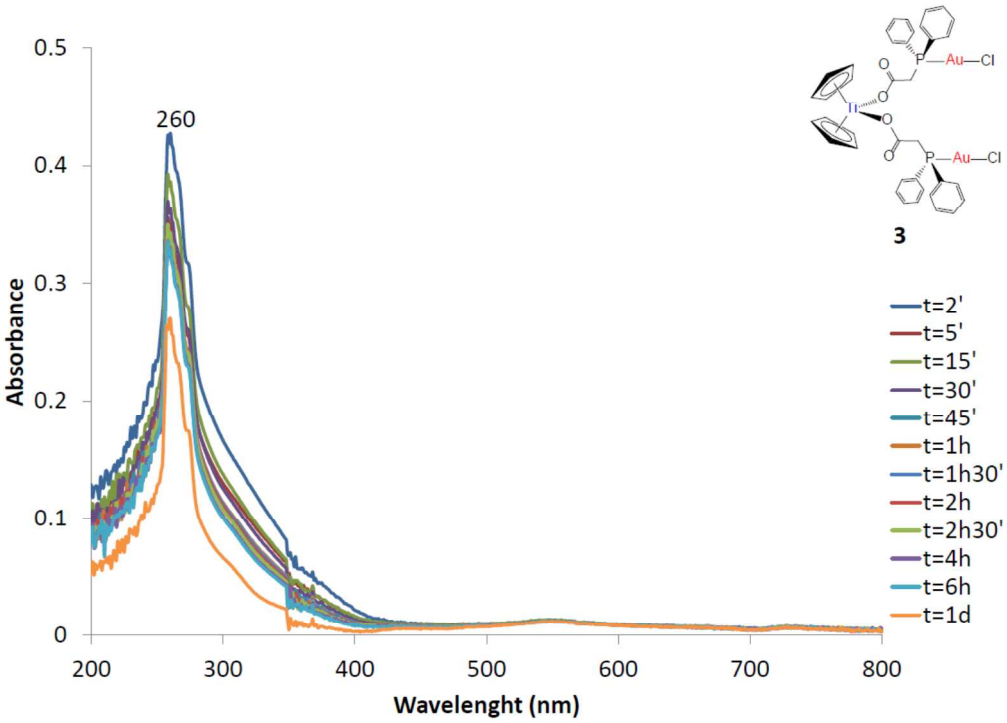

Figure S19. UV-visible spectrum of compound **3** ($3.2x10^{-5}$ M) in DMSO recorded over time.

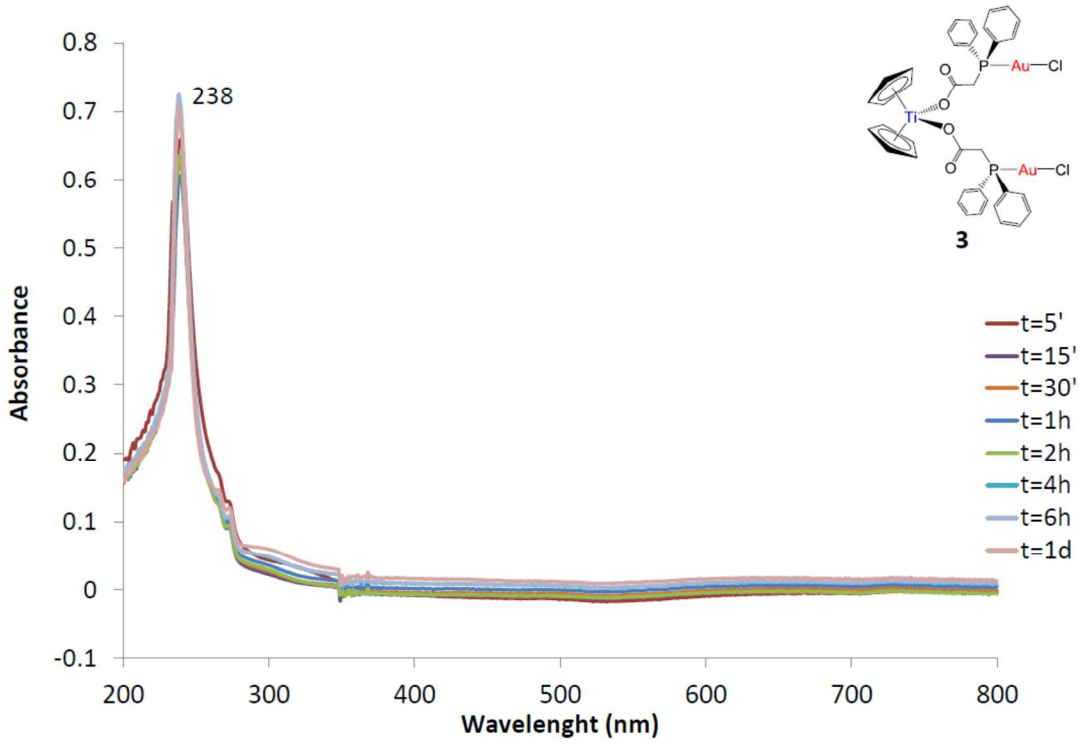

Figure S20. UV-visible spectrum of compound **3** ($3.2x10^{-5}$ M) in 1:99 DMSO/PBS-1X (pH 7.4) recorded over time, incubation at RT.

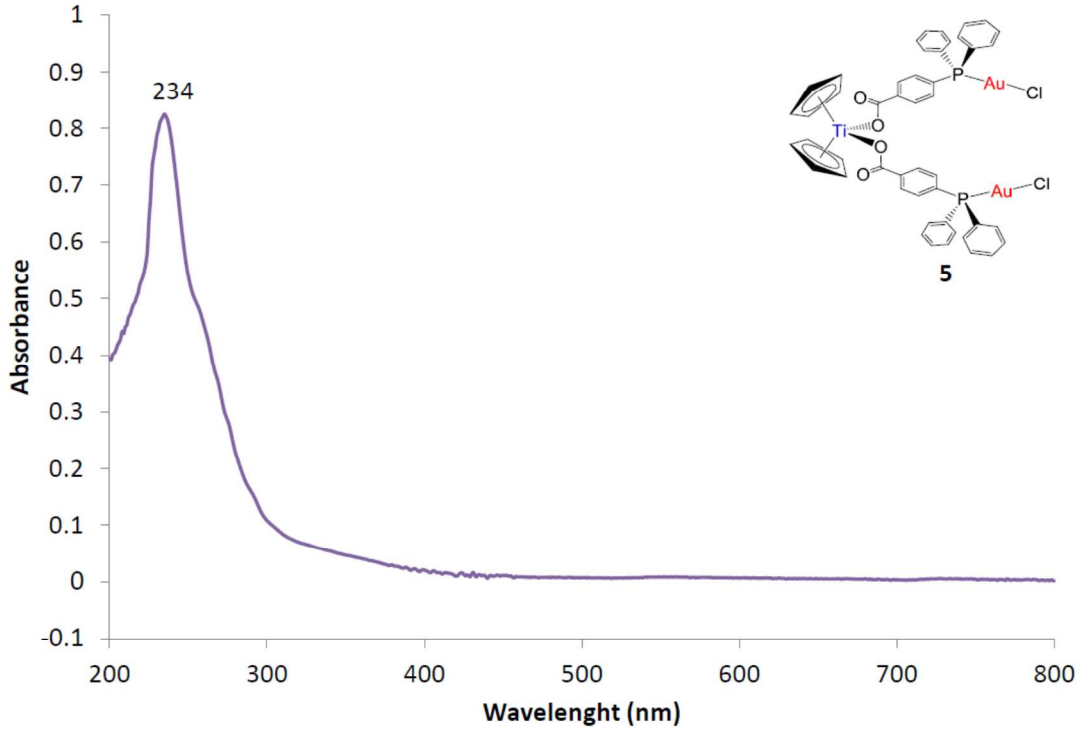

Figure S21. UV-visible spectrum of compound **5** ($5.6x10^{-5}$ M) in dichloromethane.

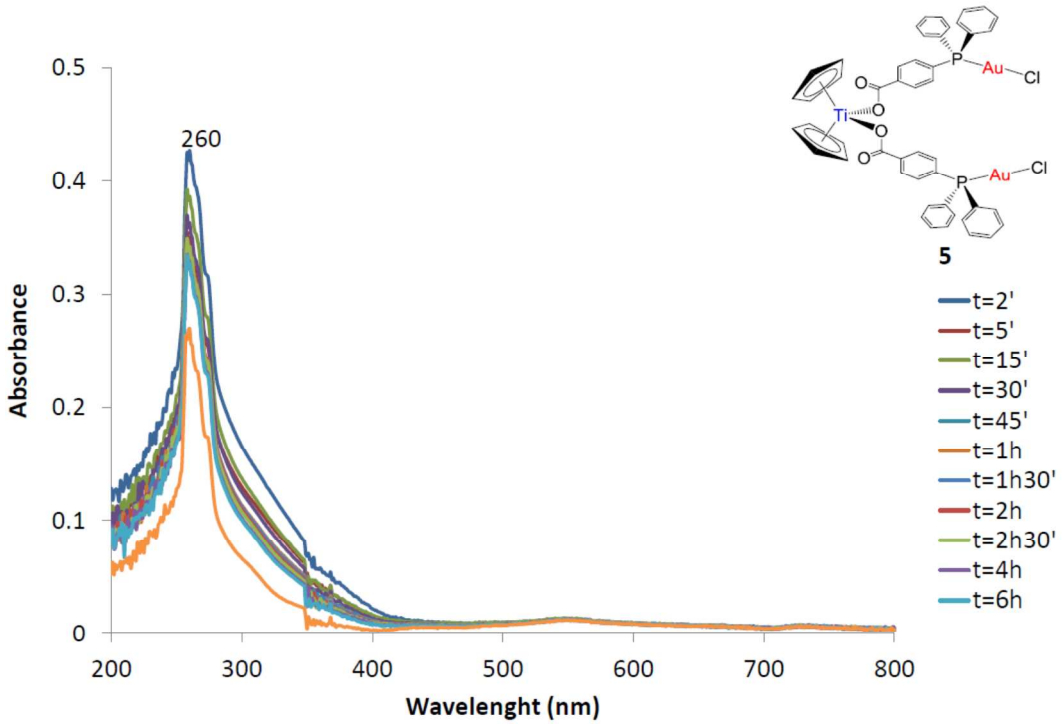

Figure S22. UV-visible spectrum of compound **5** ($5.6x10^{-5}$ M) in DMSO recorded over time.

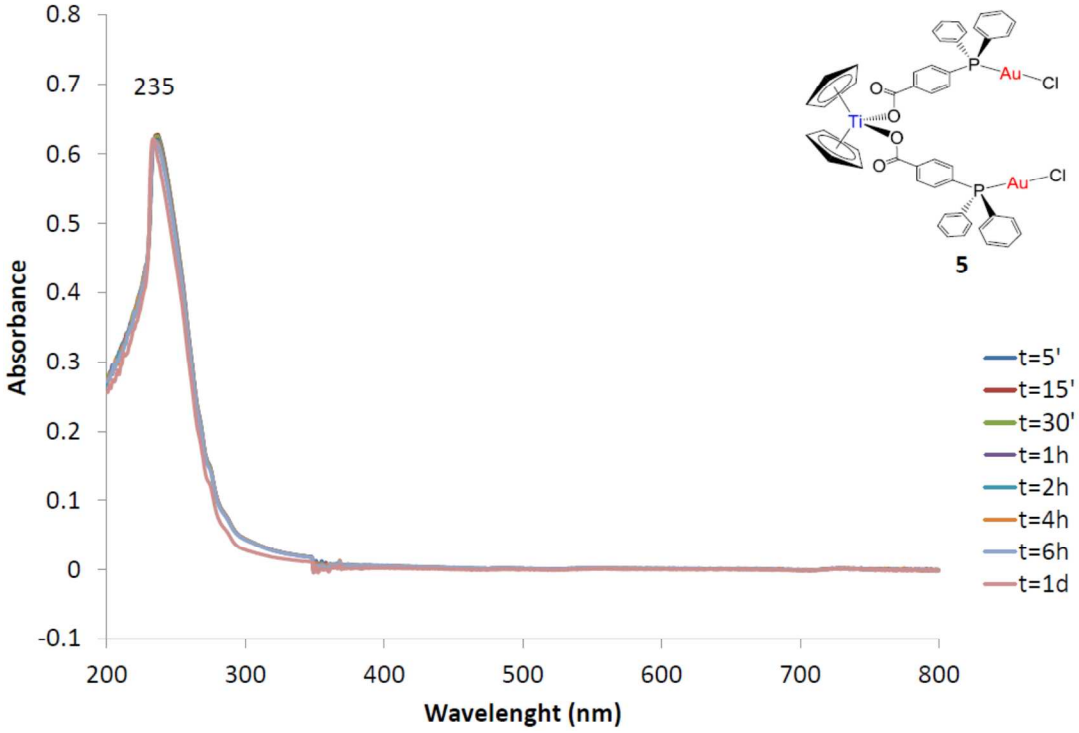

Figure S23. UV-visible spectrum of compound **5** ($5.6x10^{-5}$ M) in 1:99 DMSO/PBS-1X (pH 7.4) recorded over time, incubation at RT.

5. Mass spectra (ESI+) of compounds 3 and 5 in 1%DMSO-PBS solution overtime (24 h)

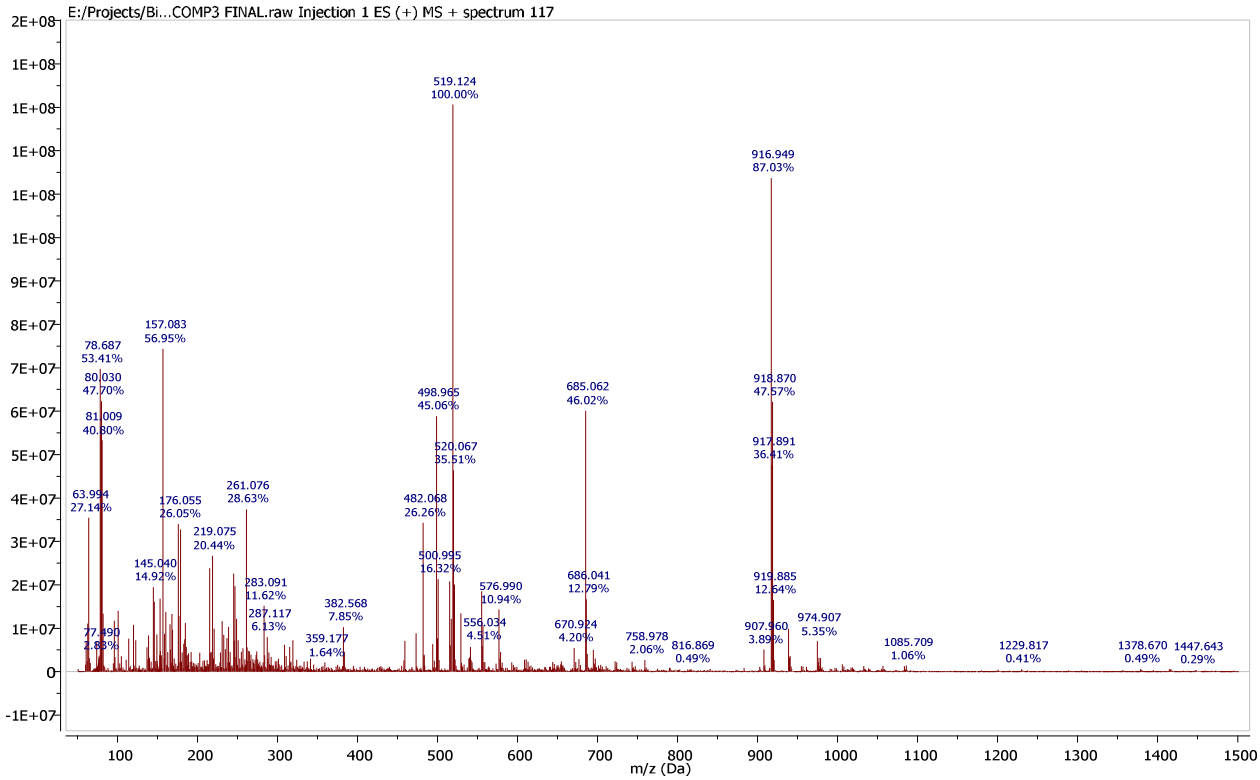

Figure S24. MS ESI+ of compound **3** in 1%DMSO-PBS solution at t=0.

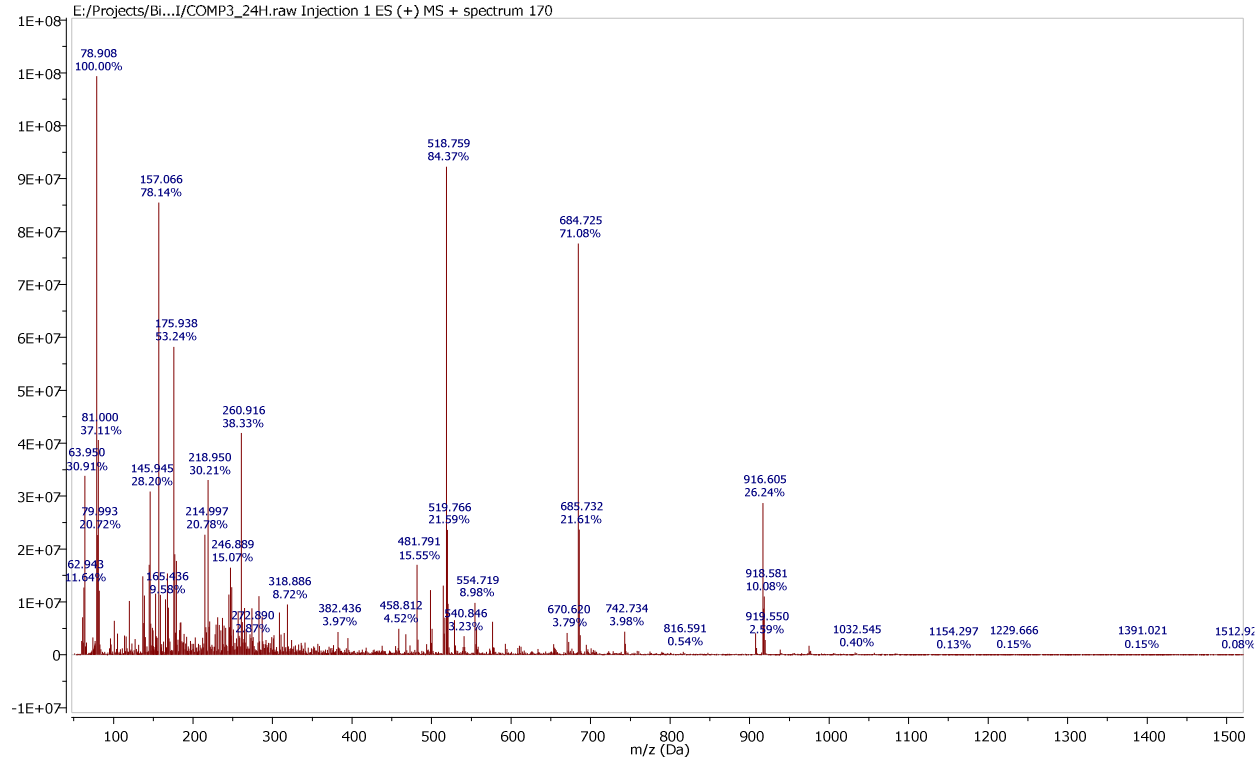

Figure S25. MS ESI+ of compound **3** in 1%DMSO-PBS solution at t=24h.

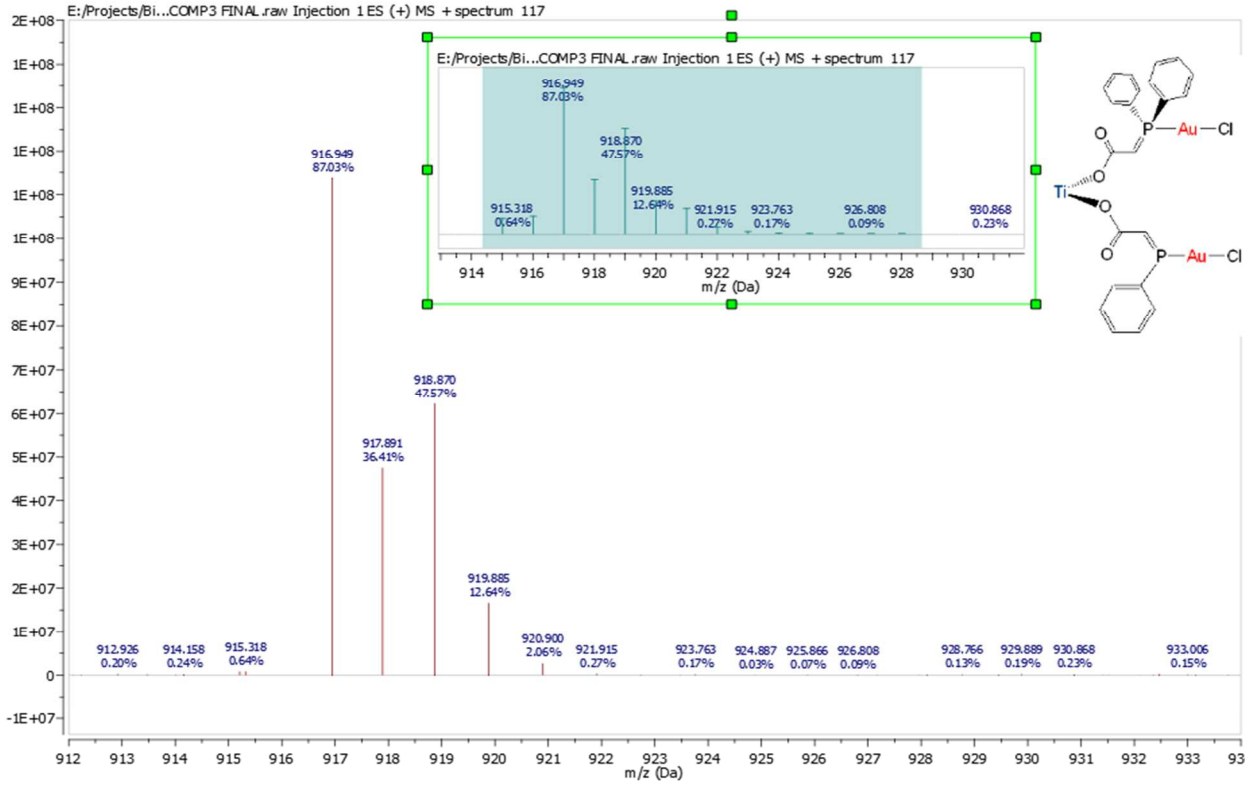

Figure S26. Magnification of peak at [m/z]: 916.0 $[Ti\{AuL\}\{AuL'\}Cl_2]^{2+}$ (L= PPh_2-CH-CO; L'= PPh-CH-CO_2) in MS ESI+ of compound **3** in 1%DMSO-PBS solution at t=0. Insert: theoretical isotopic distribution.

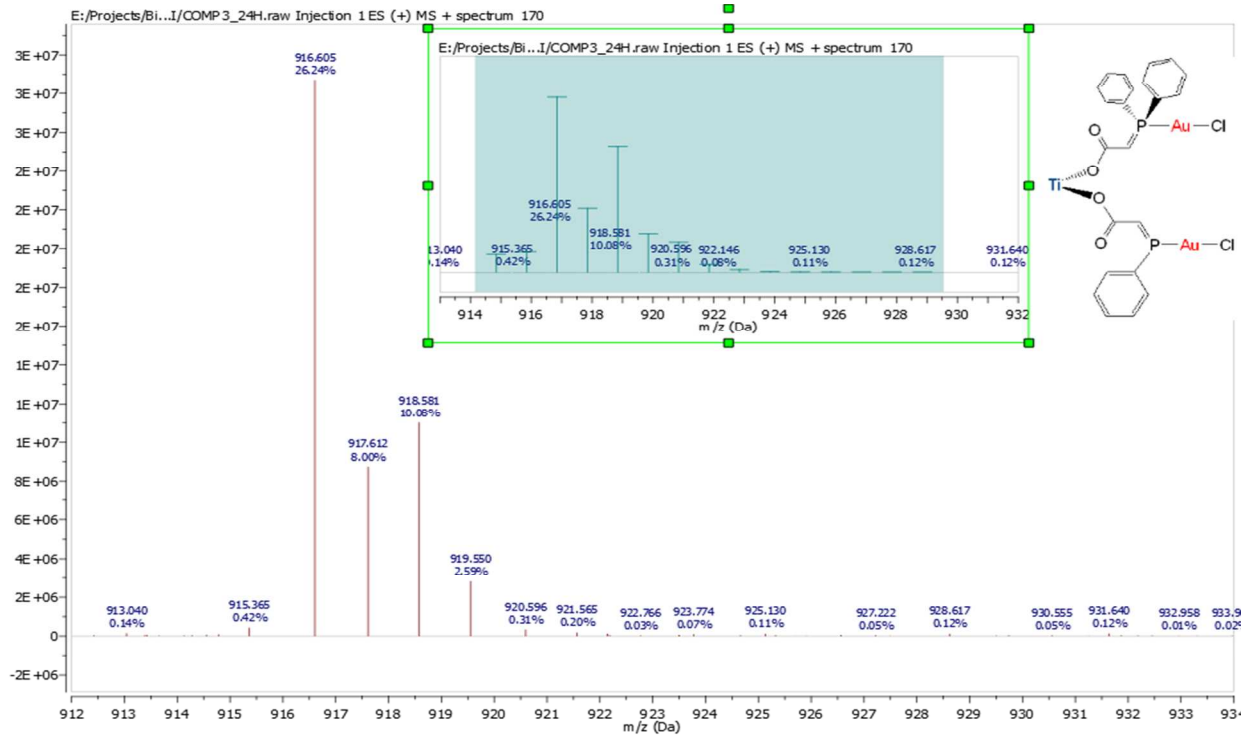

Figure S27. Magnification of peak at [m/z]: 916.0 $[Ti\{AuL\}\{AuL'\}Cl_2]^{2+}$ (L= PPh_2-CH-CO; L'= PPh-CH-CO_2) in MS ESI+ of compound **3 in** 1%DMSO-PBS solution at t=24h. Insert: theoretical isotopic distribution.

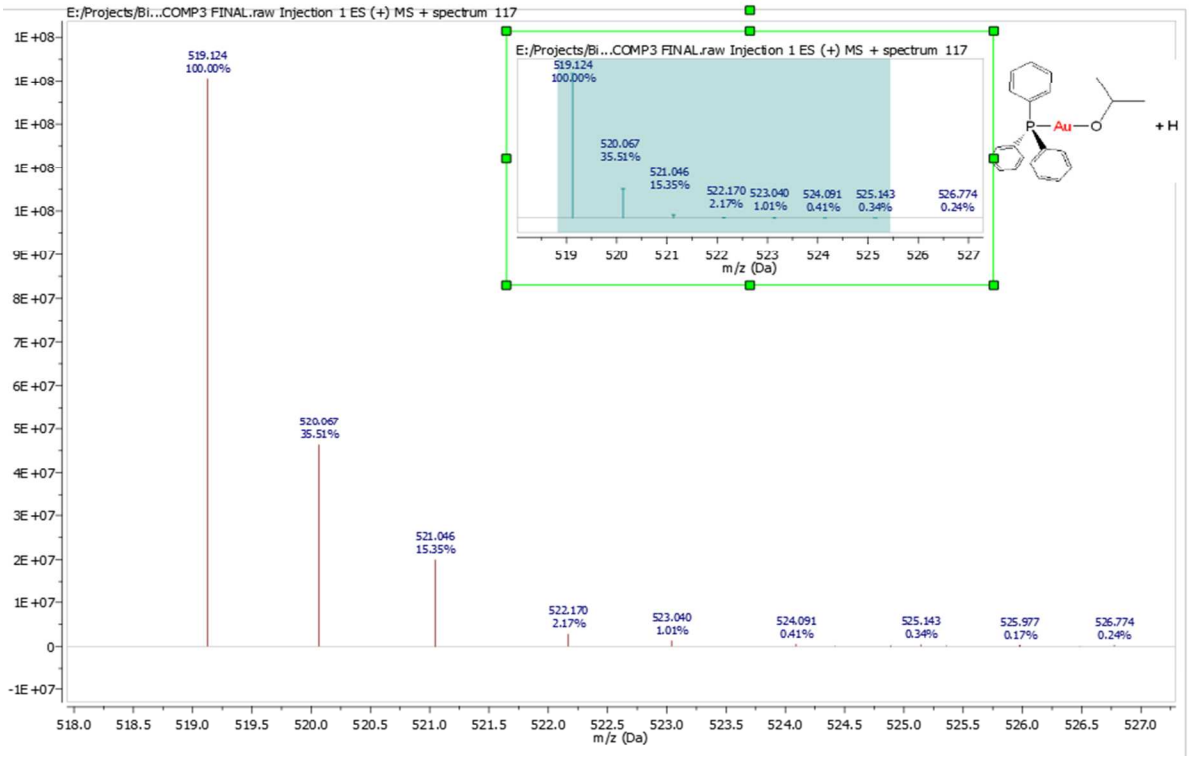

Figure S28. Magnification of peak at [m/z]: 519. 0 $[Au\{(CH_3)_2CHO\}\{PPh_3\} + H]^+$ in MS ESI+ of compound **3** in 1%DMSO-PBS solution at t=0. Insert: theoretical isotopic distribution.

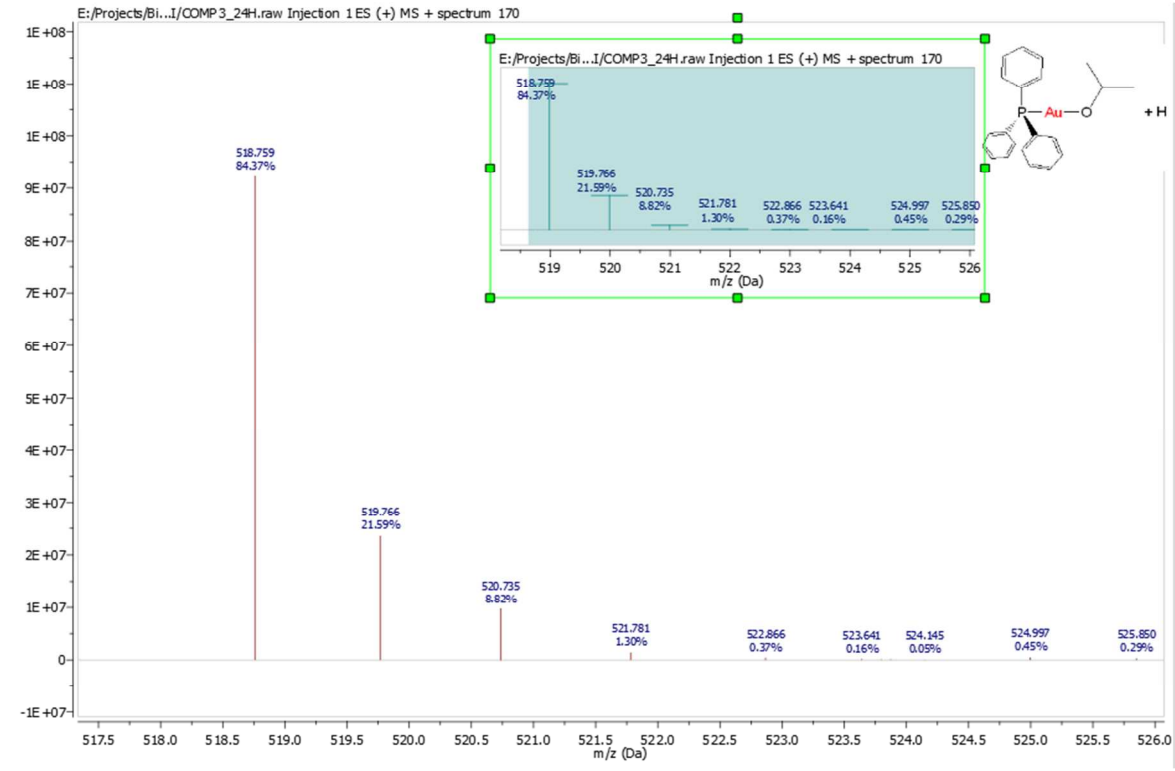

Figure S29. Magnification of peak at [m/z]: 519.0 $[Au\{(CH_3)_2CHO\}\{PPh_3\} + H]^+$ in MS ESI+ of compound **3** 1%DMSO-PBS solution at t=24h. Insert: theoretical isotopic distribution.

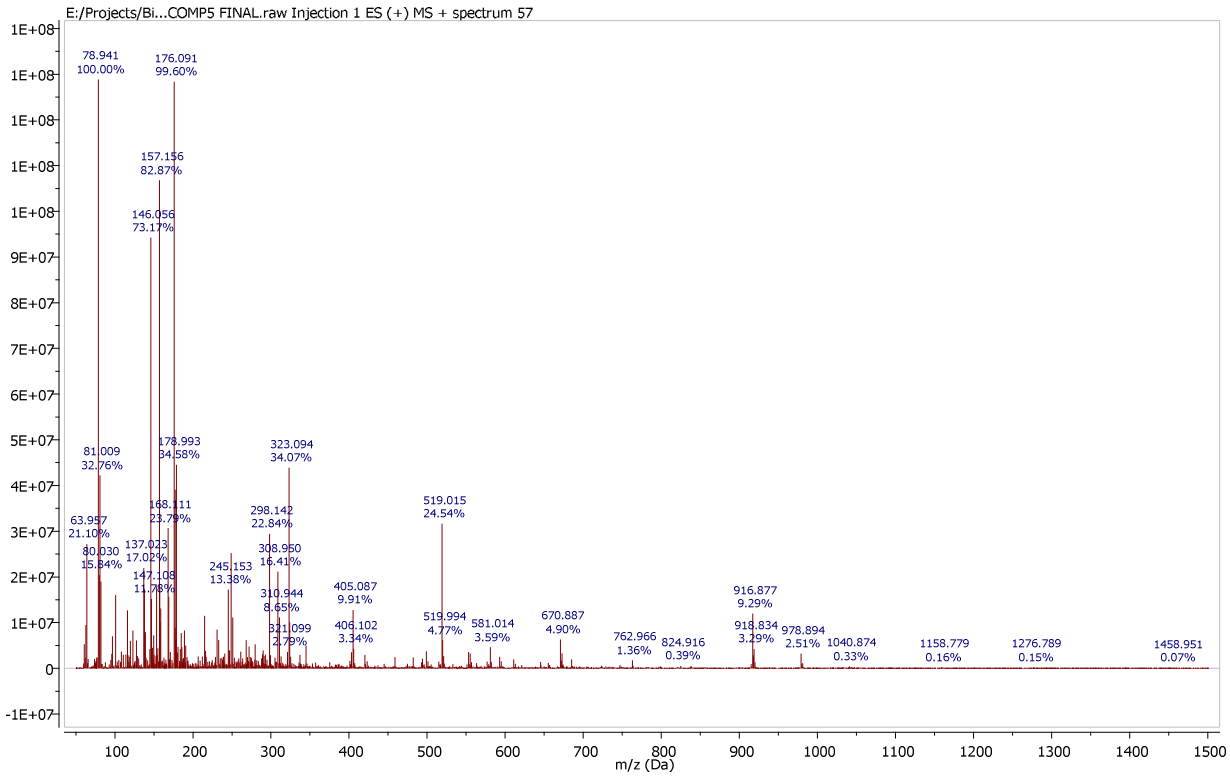

Figure S30. ESI+ mass spectra of compound **5** in 1%DMSO-PBS solution at t=0.

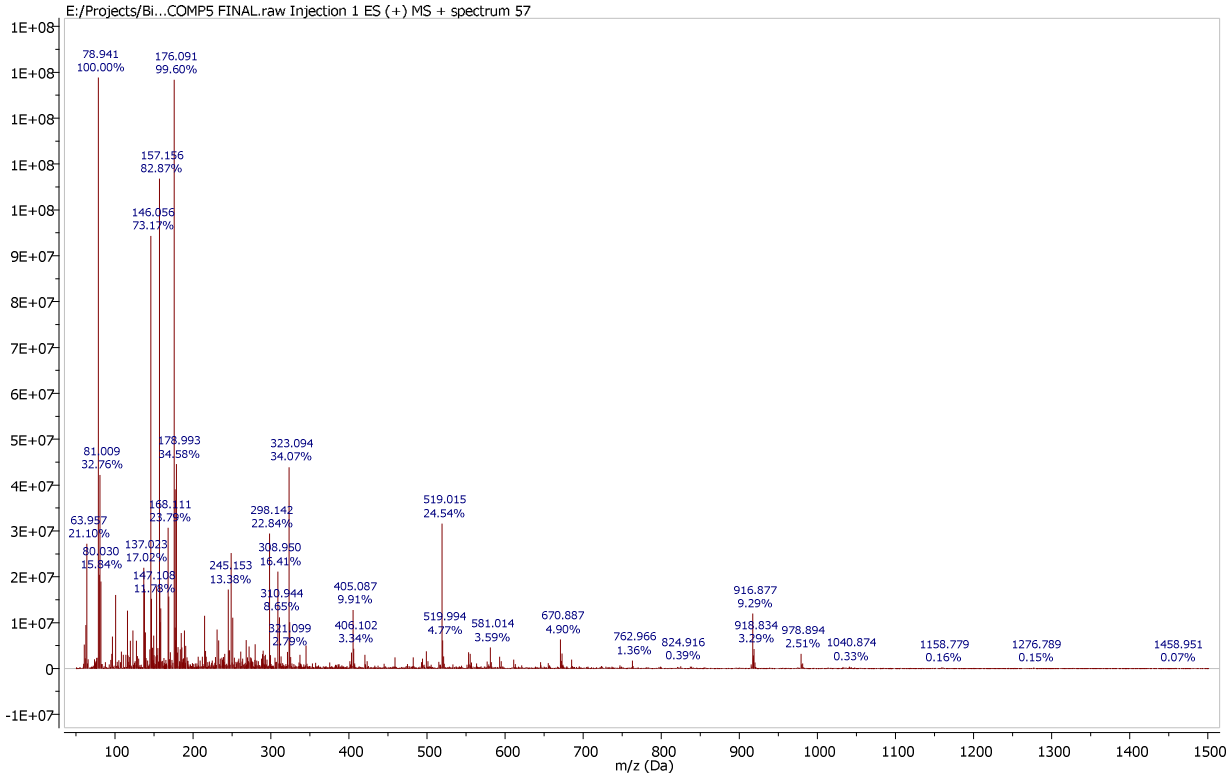

Figure S31. ESI+ mass spectra of compound **5** in 1%DMSO-PBS solution at t=24h.

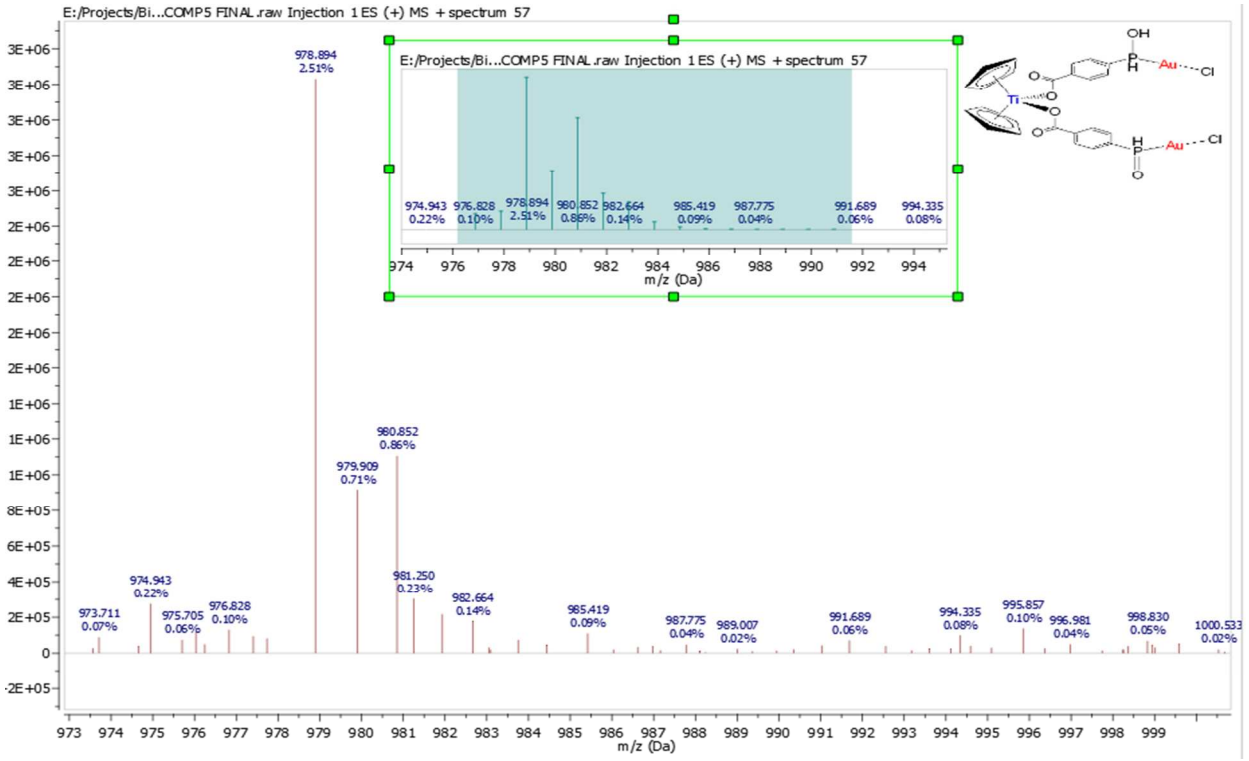

Figure S32. Magnification of peak at [m/z]: 916.0 $[Cp_2Ti\{AuClL\}\{AuClL'\}]^+$ (L= PHO-C_6H_4-CO_2; L'= PH(OH)-C_6H_4-CO_2) in MS ESI+ of compound **5** in 1%DMSO-PBS solution at t=0. Insert: theoretical isotopic distribution.

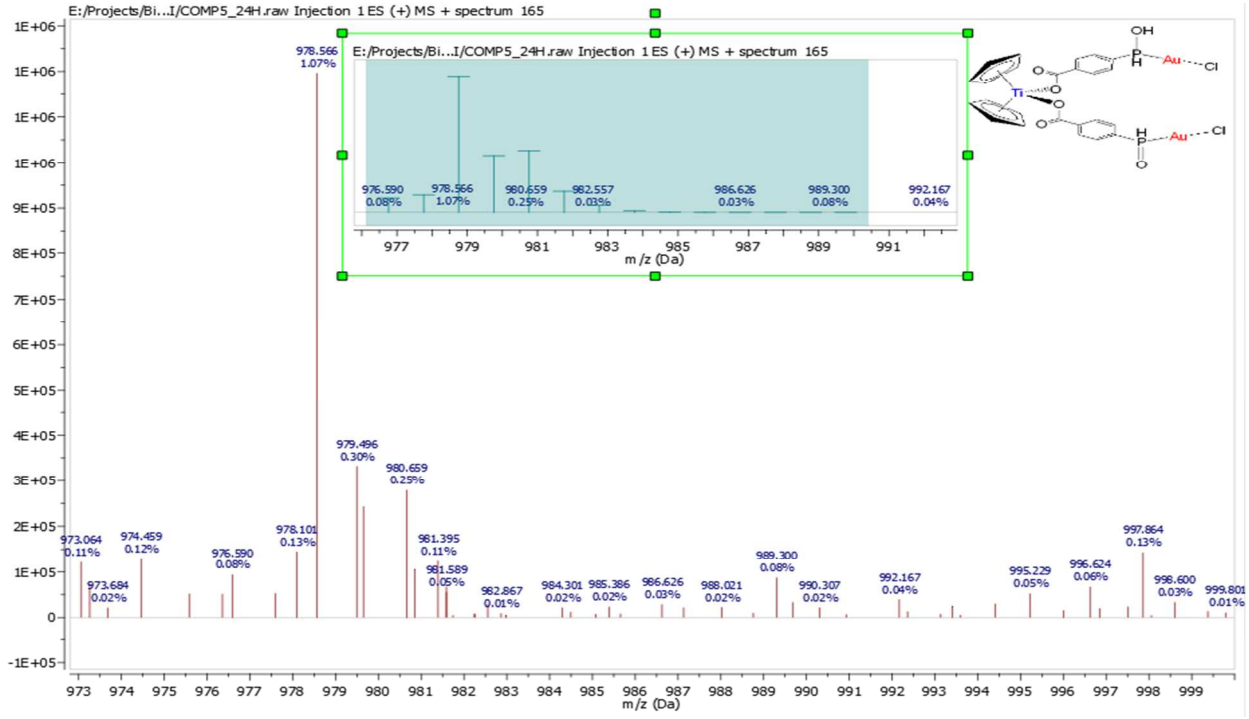

Figure S33. Magnification of peak at [m/z]: 916.0 $[Cp_2Ti\{AuClL\}\{AuClL'\}]^+$ (L= PHO-C_6H_4-CO_2; L'= PH(OH)-C_6H_4-CO_2) in MS ESI+ of compound **5** in 1%DMSO-PBS solution at t=24h. Insert: theoretical isotopic distribution.

6. Cell death experiments (Annexin V/PI assay) for compound 3 at 1 and 12 h and compound 5 at 12 and 24 h

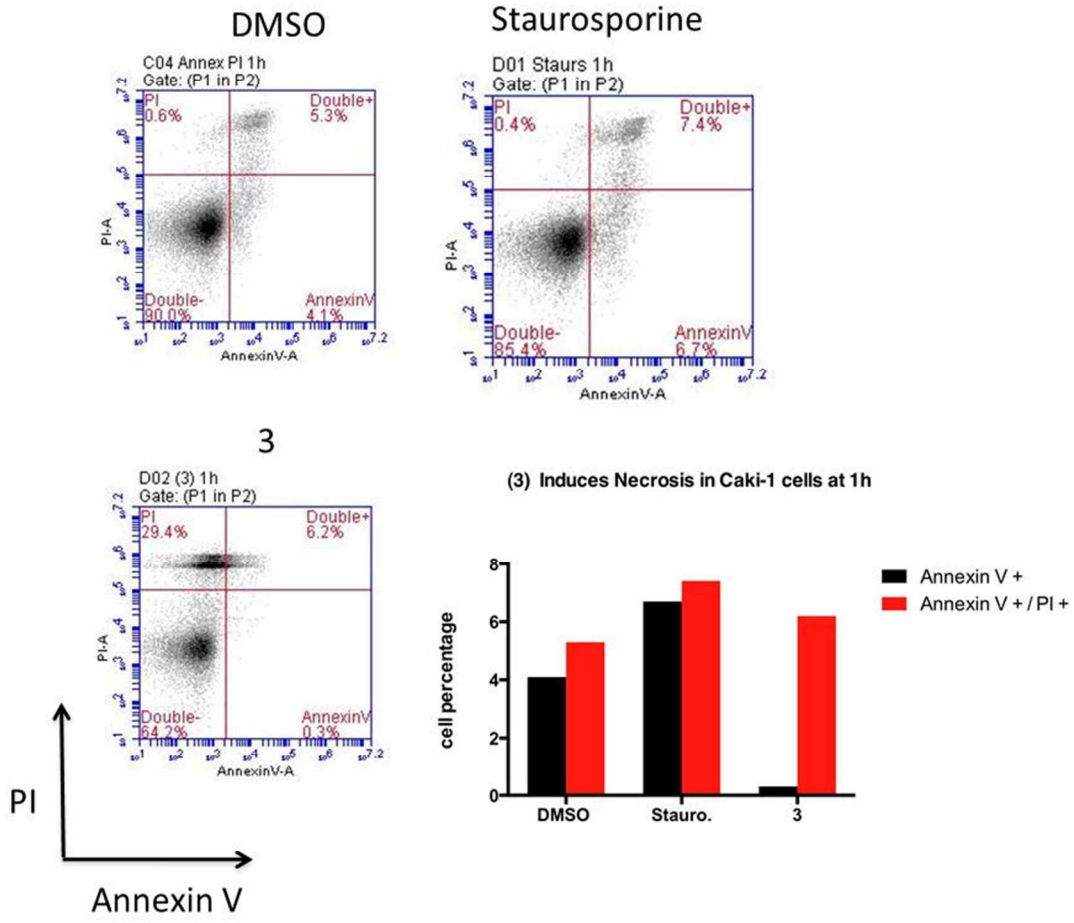

Figure S34. Cell death assays on Caki-1 cells induced by **3** (10 μM) measured by using two-colour flow cytometric analysis, after 1 h of incubation. 1%DMSO is vehicle alone control and Staurosporine is a known inducer of apoptosis as positive control.

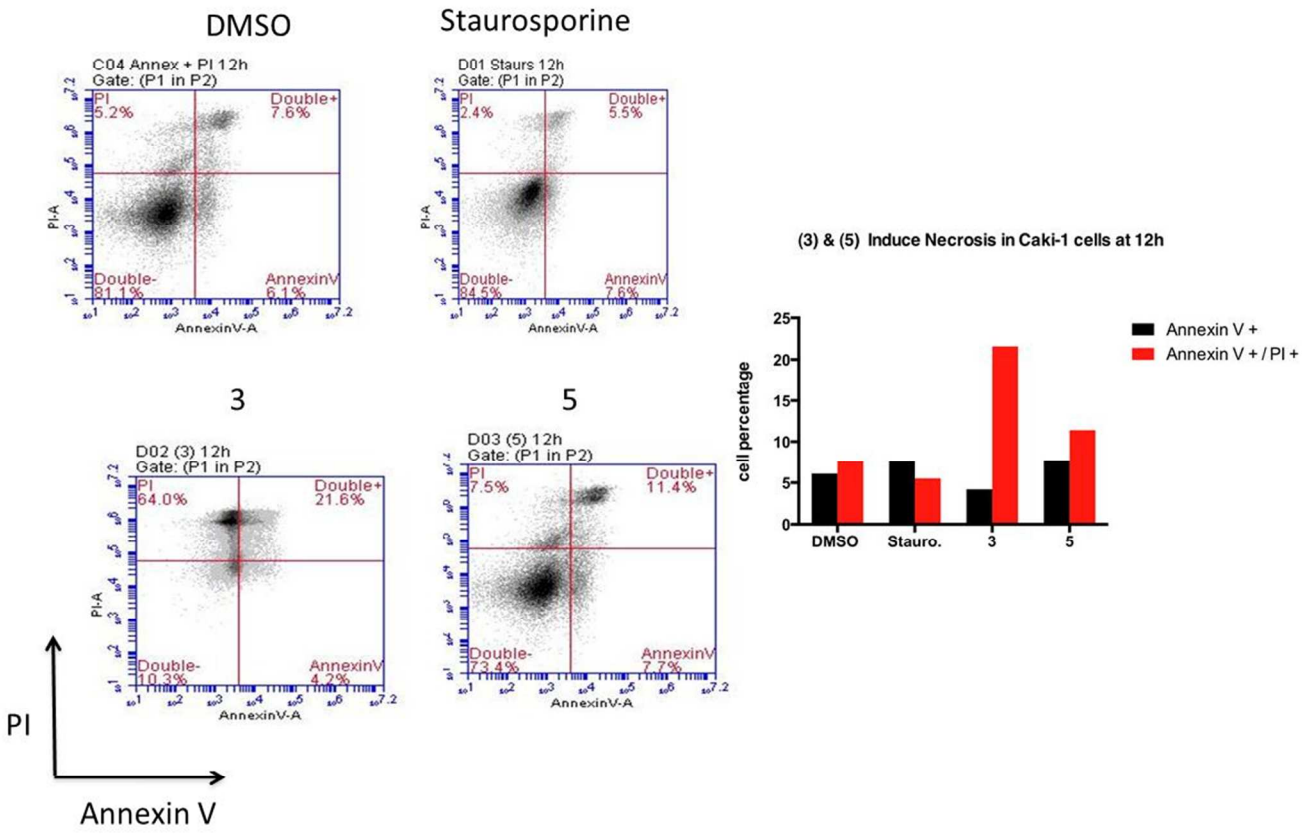

Figure S35. Cell death assays on Caki-1 cells induced by **3** and **5** (10 μM) measured by using two-colour flow cytometric analysis, after 12 h of incubation. 1%DMSO is vehicle alone control and Staurosporine is a known inducer of apoptosis as positive control.

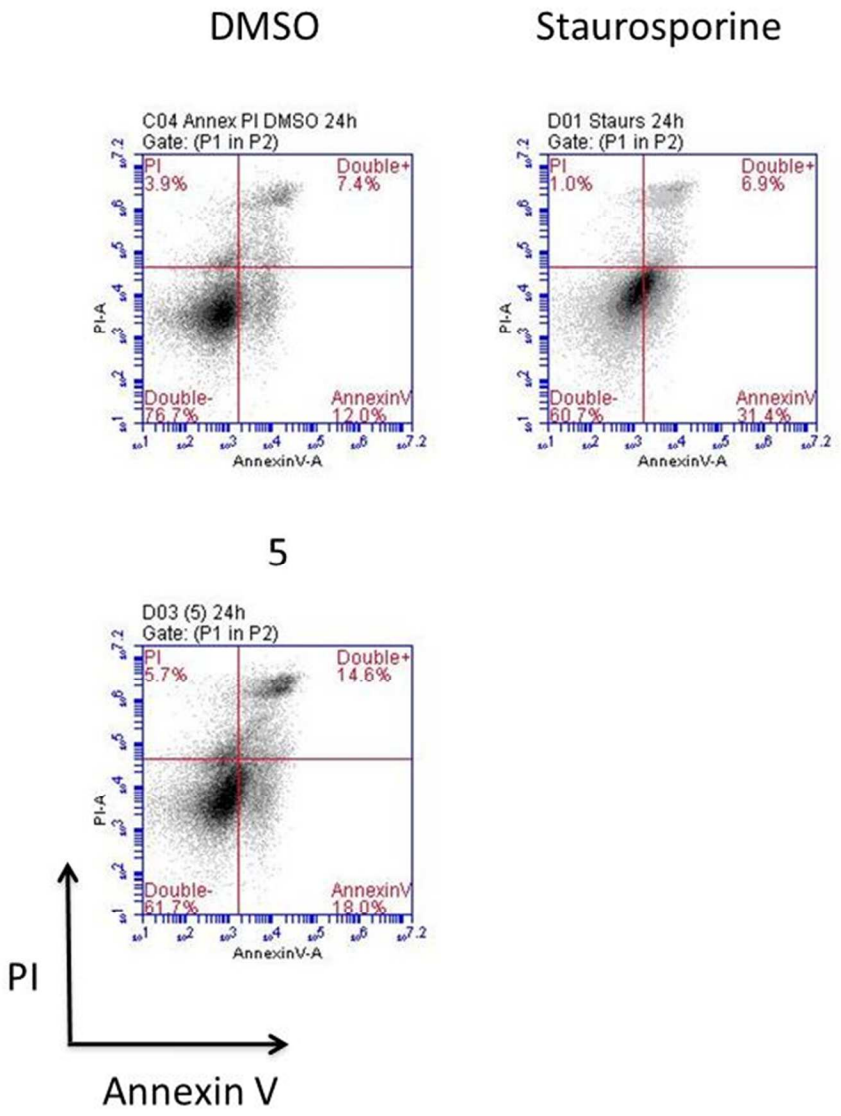

Figure S36. Cell death assays on Caki-1 cells induced by **5** (10 μM) measured by using two-colour flow cytometric analysis, after 24 h of incubation. 1%DMSO is vehicle alone control and Staurosporine is a known inducer of apoptosis as positive control.