

**Platinum Glutamate Acid Complex as a Peroxidase Mimic: High Activity, Controllable Chemical Modification, and Application in Biosensor**

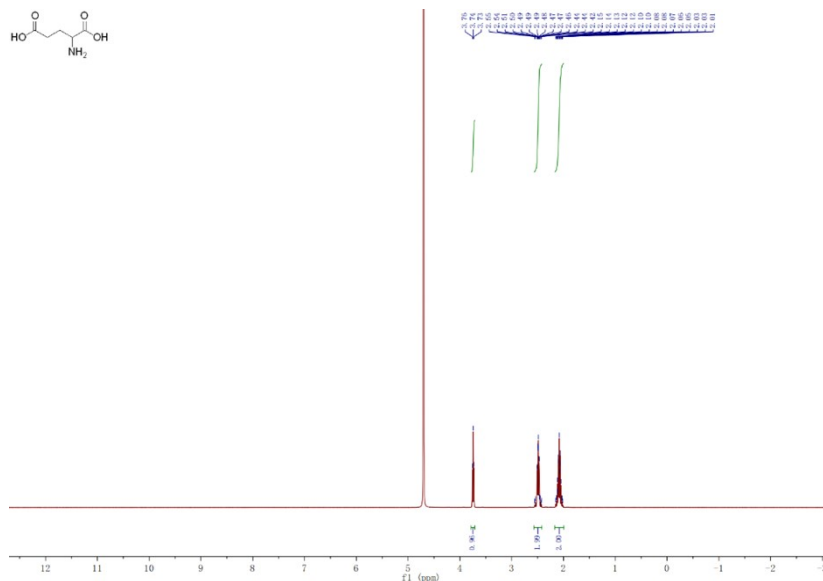
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### **Determination of the Michaelis-Menten constant**

Ethanol was employed as a solvent to prepare a 5mM solution of TMB, while water served as the solvent for the precise formulation of a 1mM solution of Glu-Pt. A comprehensive range of H<sub>2</sub>O<sub>2</sub> concentrations (5, 4.5, 4, 3.5, 3, 2.5, 2, 1.5, 1, 0.5, 0.2mM) was prepared. Each well of a microplate received 80μL of Glu-Pt and 80μL of TMB, separately. Subsequently, 40μL of H<sub>2</sub>O<sub>2</sub> at varying concentrations was meticulously added to each well. The final concentrations in the microplate were 2mM for TMB, 0.4mM for Glu-Pt, and diverse concentrations of H<sub>2</sub>O<sub>2</sub> (1, 0.9, 0.8, 0.7, 0.6, 0.5, 0.4, 0.3, 0.2, 0.1, 0.04mM). Each concentration set underwent a meticulous triplicate parallel experimentation. The microplate underwent uniform shaking, initiating an 8-minute reaction period. Following this, the absorbance at 652 nm was precisely measured using a microplate reader. C/V curves and double reciprocal curves were adeptly generated to gain comprehensive insights into the reaction dynamics. The determination of the Michaelis-Menten constant ( $K_m$ ) was carried out to provide a quantitative measure of the enzymatic activity.

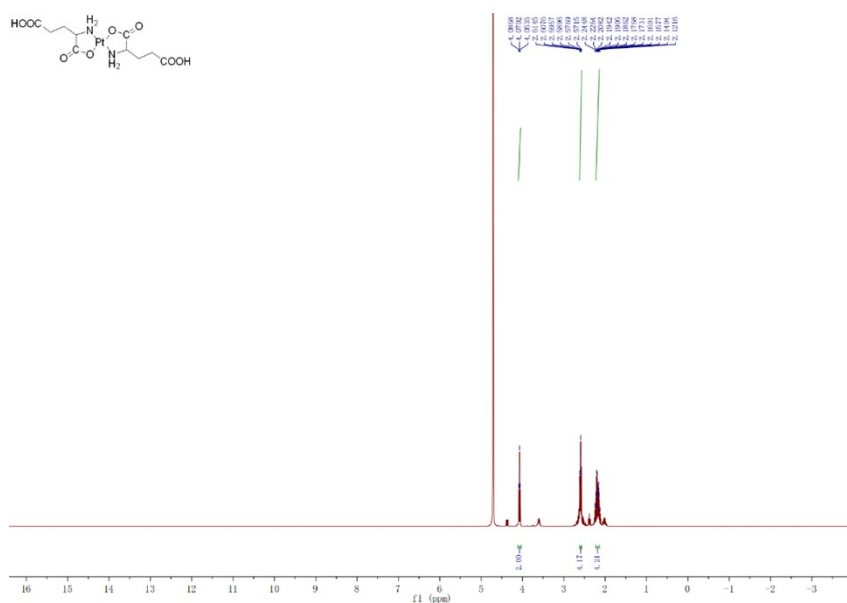
### Glutamic acid

$^1\text{H}$  NMR (400 MHz  $\text{D}_2\text{O}$ ):  $\delta$  3.74 (t,  $J = 6.4$  Hz, 1H), 2.55–2.42 (m, 2H), 2.15–2.01 (m, 2H).

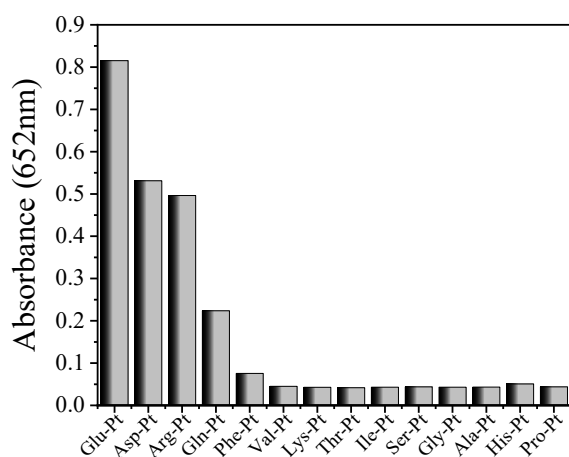


### Platinum glutamic acid complex:

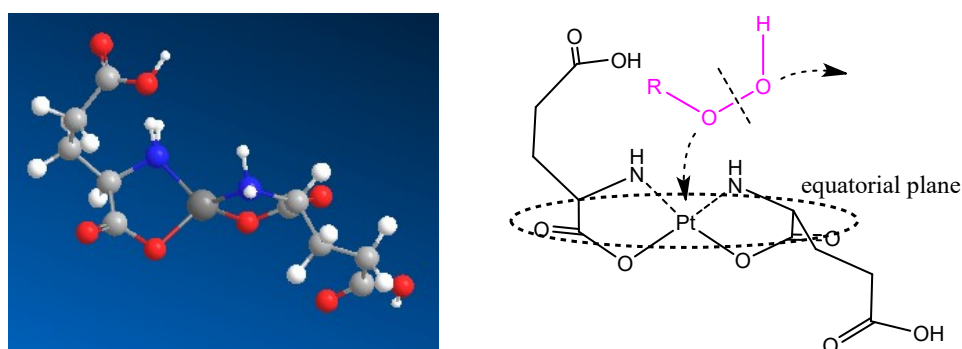
$^1\text{H}$  NMR (400 MHz  $\text{D}_2\text{O}$ ):  $\delta$  4.07 (t,  $J = 6.7$  Hz, 2H), 2.61–2.57 (m, 4H), 2.24–2.12 (m, 4H).



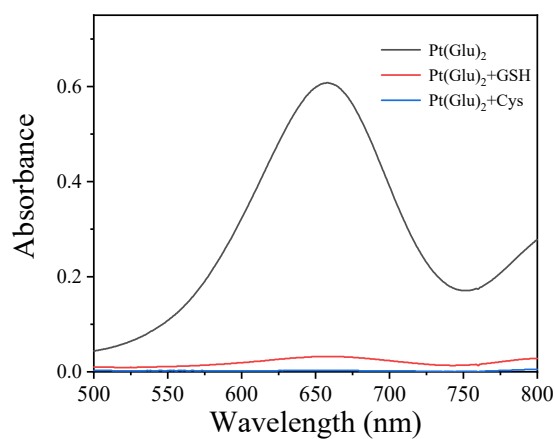
**Fig. S1.**  $^1\text{H}$  400 MHz NMR spectra of glutamic acid (top) and platinum glutamic acid complex (bottom).



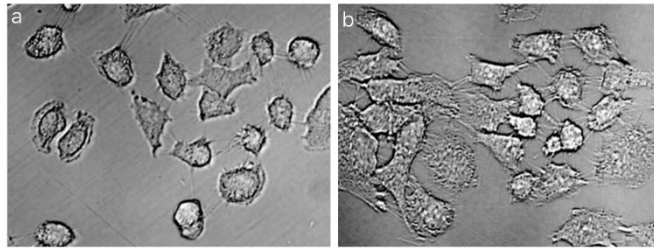
**Fig. S2.** The comparison of peroxidase-like activity of platinum complexes formed with amino acids.



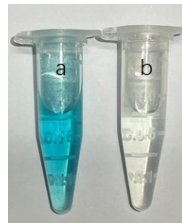
**Fig. S3.** The structure of  $\text{Pt}(\text{Glu})_2$  (right) and a schematic representation of its coordination with  $\text{H}_2\text{O}_2$  (left).



**Fig. S4.** The absorption spectra obtained from the  $\text{H}_2\text{O}_2$  and TMB in the presence of  $\text{Pt}(\text{Glu})_2$ ,  $\text{Pt}(\text{Glu})_2+\text{GSH}$ , and  $\text{Pt}(\text{Glu})_2+\text{cysteine}$ .



**Fig. S5.** Bioimaging HCCLM3 incubated with the mixture of GOx and Pt(Glu)<sub>2</sub> (a) and GOx chemically modified with Pt(Glu)<sub>2</sub>(b).



**Fig. S6.** Color change upon the addition of cholesterol in the ChOx-Pt(Glu)<sub>2</sub>/TMB (a) and ChOx/TMB (b).

**Table S1.** Comparison of nanozyme-based detection methods for cholesterol detection.

Sensing system	Analytical method	Linear range ( $\mu\text{M}$ )	LOD ( $\mu\text{M}$ )	Reference
Nanoporous Gold	Electrochemistry	50-6000	8.4	1
ChOx/CS-GR/GCE	Differential Pulse Voltammetry	5-1000	0.715	2
carbon dot/hemoglobin	Fluorometry	0-800	56	3
N-GQDs/Chromium Picolinate	Fluorometry	0-520	0.4	4
Boron nitride nanosheet@CuS	Colorimetry	10-100	2.9	5
Fe <sub>3</sub> O <sub>4</sub> magnetic nanoparticles	Colorimetry	2-50	0.8	6
Polypyrrole NPs	Colorimetry	10-100	3.5	7
Pt(Glu) <sub>2</sub>	Colorimetry	0.1-80	0.56	This work

**Table S2.** Comparison of other detection methods for ALP detection

Sensing system	Analytical method	Linear range(U/L)	LOD (U/L)	Reference
CdSe nanoparticles	Electrochemistry	2-25	2	8
Lumin-SiNPs	Electrochemistry	5-50	0.8	9
Dopamine-resorcinol	Fluorometry	0.1-6.0	0.07	10
O-phenylenediamine - Ascorbic acid 2- phosphate	Fluorometry	0.1-30	0.06	11
N-CDs	Fluorometry	2.5-45	0.4	12
Cu Alkyne-azide cycloaddition and DNA- templated CuNPs	Fluorometry	100-1600	50	13
Ce <sup>4+</sup> -TMB	Colorimetry	50-250	2.3	14
Fe <sub>55</sub> -N-C	Colorimetry	0.05-20	0.03	15
Fe(II)-phenanthroline	Colorimetry	0-220	0.94	16
AuNPs-cystine	Colorimetry	0.2-20	0.2	17

Cu- MOF/Pyrophosphate	Colorimetry	1-34	0.19	18
<b>Polydiacetylenes</b> liposomes	Colorimetry	10-200	2.8	19
Cu-C <sub>3</sub> N <sub>4</sub> -550	Colorimetry	0.4-20	0.32	20
FeCo NPs	Colorimetry	0.6-10	0.49	21
<b>Fe-N-C single-atom</b> <b>nanozymes</b>	Colorimetry	0.1-1.5	0.05	22
Pt(Glu) <sub>2</sub>	Colorimetry	0.5-12	0.09	This work

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