

Supplementary Materials

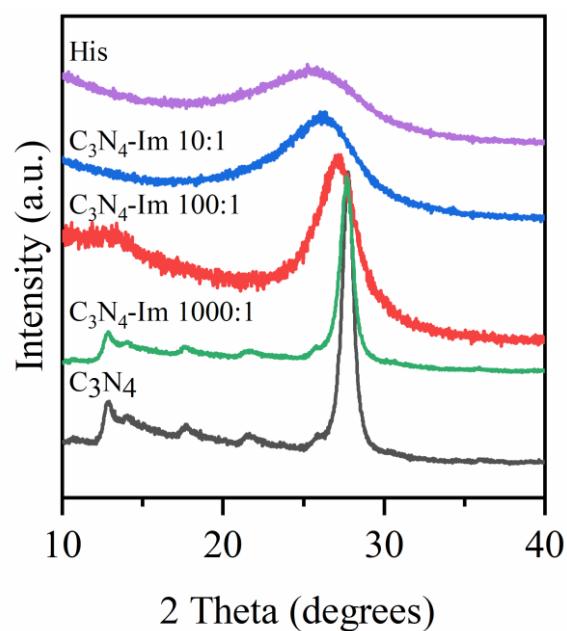


Figure S1. XRD pattern of histidine, g- C_3N_4 , and g- $\text{C}_3\text{N}_4\text{-Im}$ of different mass ratios between urea and histidine (10:1; 100:1; 1000:1) after calcination.

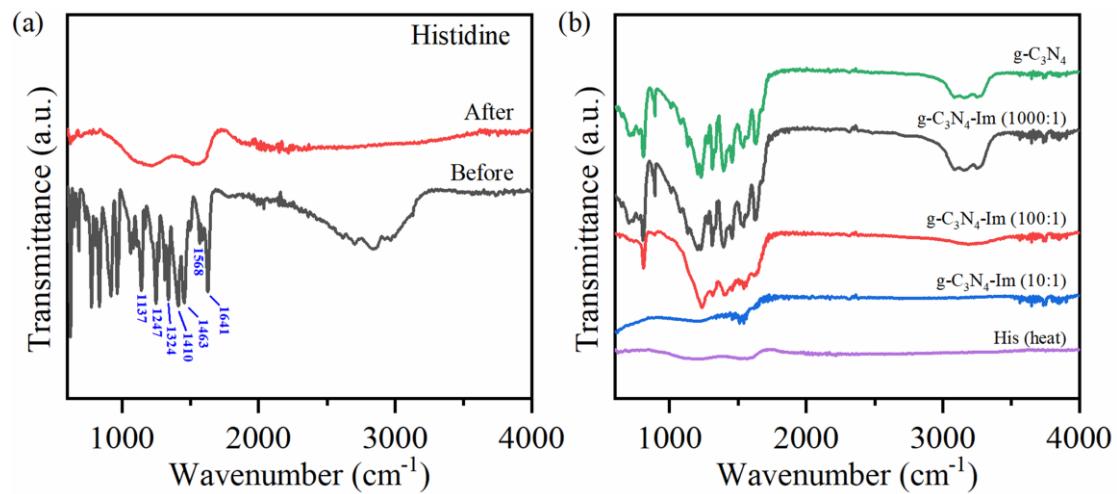


Figure S2. a) FTIR spectra of g-C₃N₄ before and after calcination; b) FTIR spectra of histidine and g-C₃N₄-Im of different mass ratios between urea and histidine (10:1; 100:1; 1000:1) after calcination.

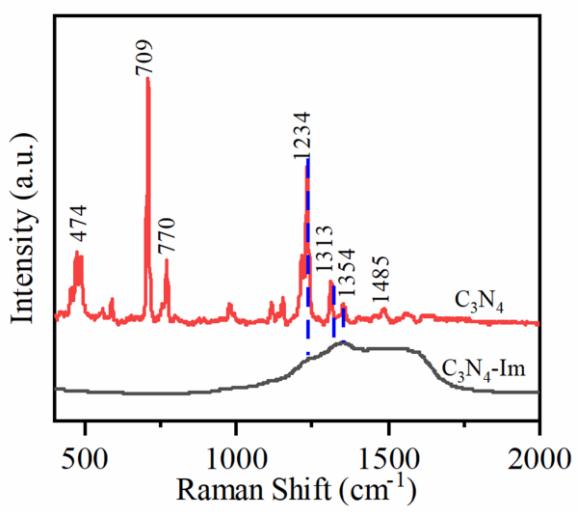


Figure S3. Raman spectra of heating the $\text{g-C}_3\text{N}_4$ and the $\text{g-C}_3\text{N}_4\text{-Im}$.

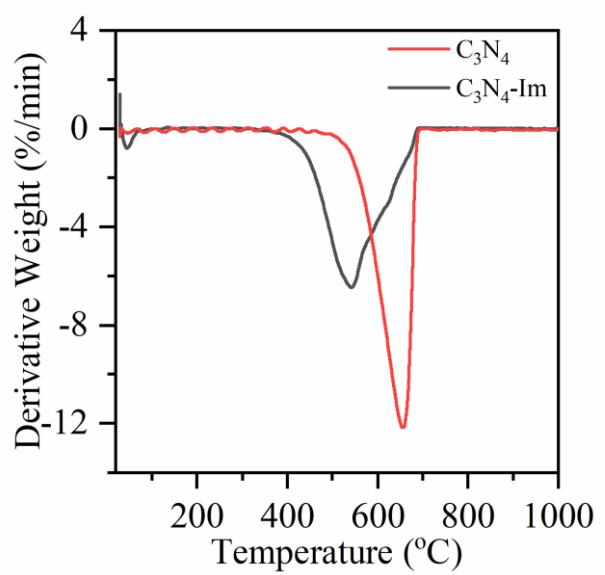


Figure S4. DTG curve of $\text{g-C}_3\text{N}_4$ and $\text{g-C}_3\text{N}_4\text{-Im}$.

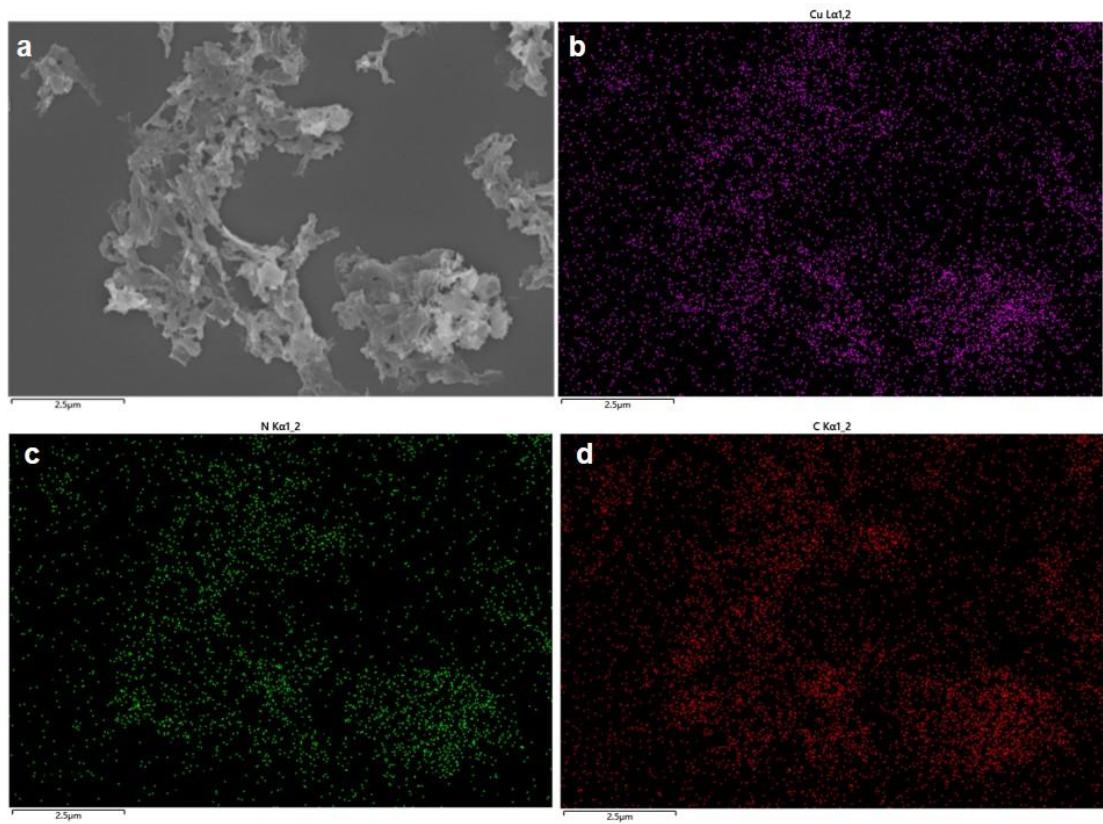


Figure S5. SEM image (a) and corresponding EDS mapping images (b-d) of g-C₃N₄-Im-Cu nanosheets.

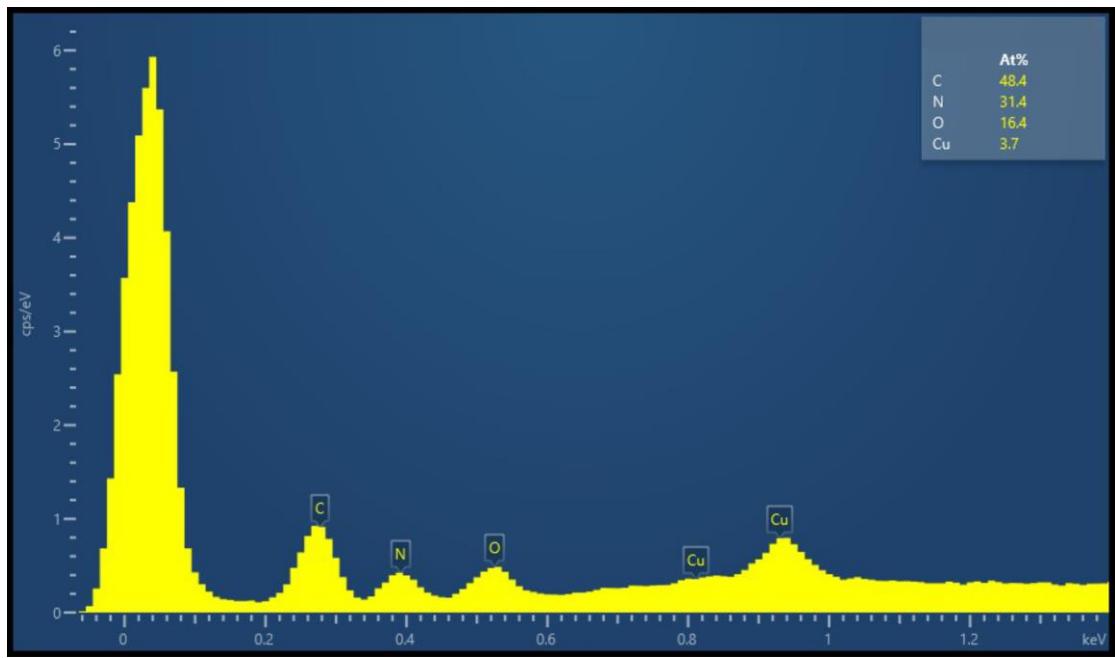


Figure S6. EDX spectrum of g-C₃N₄-Im-Cu nanosheets showing the content of C, N, Cu and O elements.

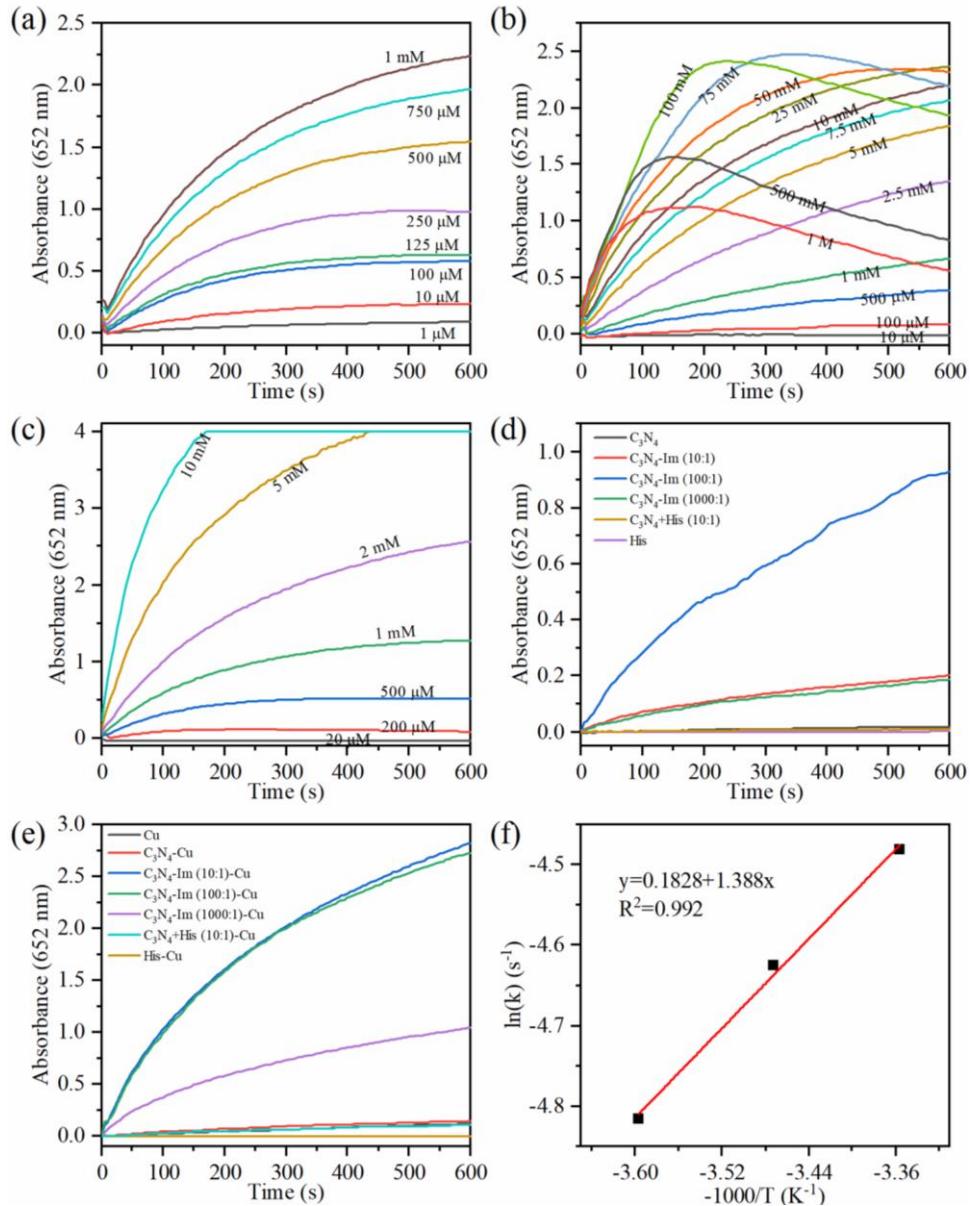


Figure S7. Optimal reaction conditions for peroxidase-like activity of g-C₃N₄-Im-Cu. The critical factors in the test of peroxidase-like activity of g-C₃N₄-Im-Cu were a) copper ion concentration, b) H₂O₂ concentration and c) TMB concentration. d) The time-dependent absorbance changes at 652 nm of TMB solutions in the presence of histidine, g-C₃N₄, and g-C₃N₄-Im of different mass ratios between urea and histidine (10:1; 100:1; 1000:1). e) Repeating the experiment in **Figure S5d** after copper ions coordination. In the d) and e), The mixed solution contains H₂O₂ 500 μl (100 mM), HAc/NaAc 1.5 ml (10 mM pH=7), catalyst 500 μL and TMB 500 μL (2 mM). f) The Arrhenius plot for H₂O₂ oxidation catalyzed by g-C₃N₄-Im-Cu, the test curves were calculated at the selected temperatures of 5 °C, 15 °C and 25 °C.

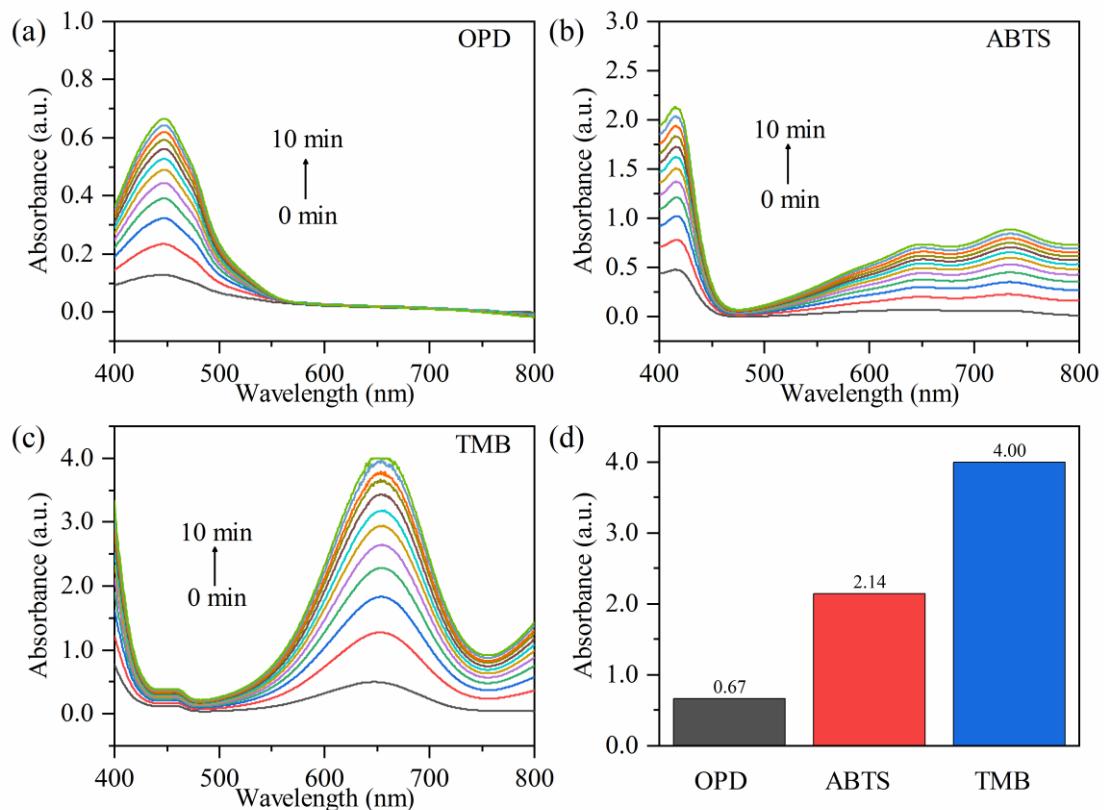


Figure S8. Peroxidase-like activity of $\text{g-C}_3\text{N}_4\text{-Im-Cu}$ with various substrates. UV-vis spectra of H_2O_2 and $\text{g-C}_3\text{N}_4\text{-Im-Cu}$ mixture in the presence of a) OPD, b) ABTS and c) TMB. d) Absorbance value of the absorption peaks of the three peroxidase substrates. The mixture contains H_2O_2 500 μl (100 mM), HAc/NaAc 1.5 ml (10mM pH=7), $\text{g-C}_3\text{N}_4\text{-Im-Cu}$ 500 μL (The concentrations of $\text{g-C}_3\text{N}_4\text{-Im}$ and Cu(II) ion are 0.25 mg/ml and 1 mM respectively) and peroxidase substrates 500 μL (2 mM).

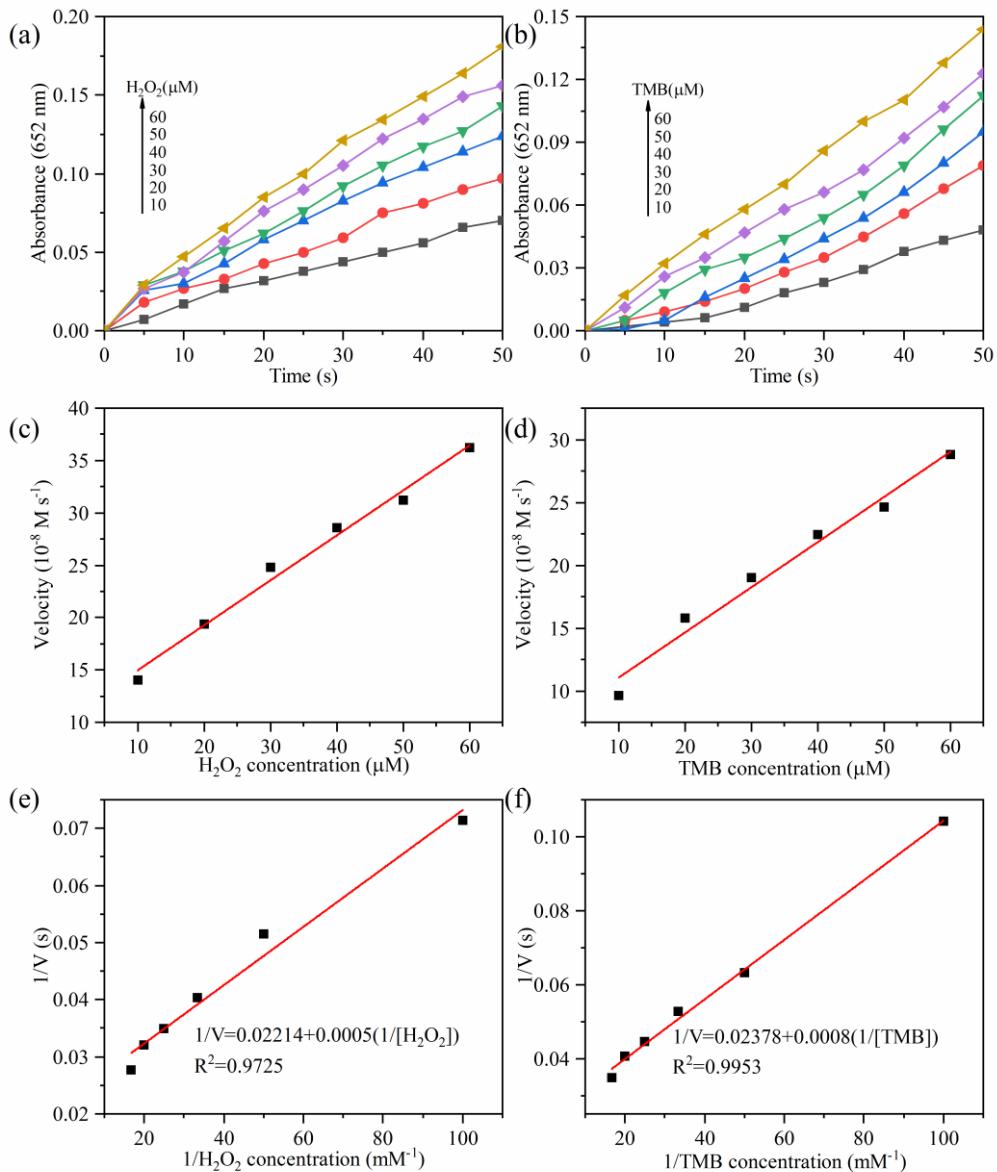


Figure S9. Steady-state kinetic assay of peroxidase-like activity of g-C₃N₄-Im-Cu. a-b) Time-dependent absorbance changes at 652 nm of TMB catalyzed by g-C₃N₄-Im-Cu in the presence of TMB or H₂O₂ with different concentrations. c-d) The velocity (V) of the reaction changes in the presence of TMB or H₂O₂ with different concentrations. e-f) Double reciprocal plots of activity of g-C₃N₄-Im-Cu in the presence of TMB or H₂O₂ with different concentrations. Experiments were carried out in 10 mM acetic acid-sodium acetate buffer (pH=7) using g-C₃N₄-Im-Cu as catalyst at 15 °C. a, c, e) H₂O₂ concentration was fixed at 100 mM and TMB concentration was varied. b, d, f) TMB concentration was fixed at 2 mM and H₂O₂ concentration was varied.

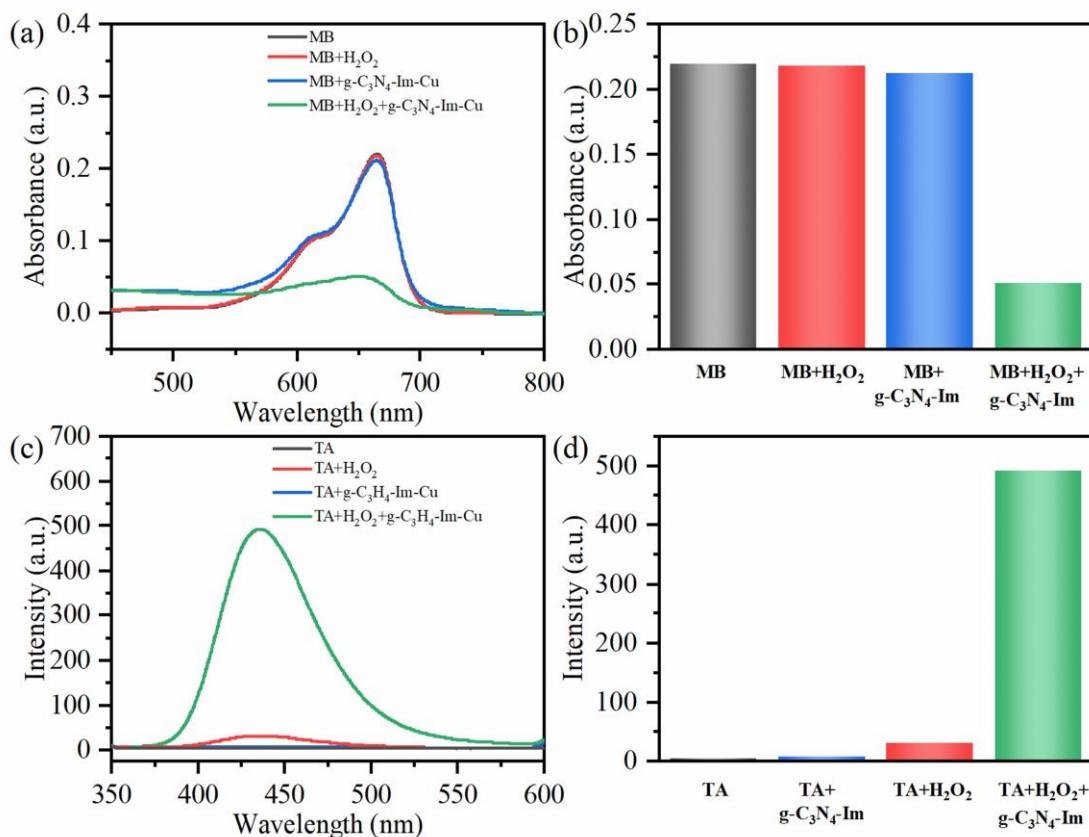


Figure S10. Detecting hydroxyl radical ($\bullet\text{OH}$) with UV-vis and fluorescence spectra. a-b) UV-vis spectra of MB, MB + H₂O₂, MB + g-C₃N₄-Im-Cu, MB + g-C₃N₄-Im-Cu + H₂O₂ after 0.5 h of reaction in acetic acid-sodium acetate buffer solution, respectively. The mixed solution contains H₂O₂ 500 μl (100 mM), HAc/NaAc 1.5 ml (10 mM, pH=7), g-C₃N₄-Im-Cu 500 μl and MB 500 μl (2 mM). c-d) Fluorescence spectra of acetic acid-sodium acetate buffer solution include TA, TA + H₂O₂, TA + g-C₃N₄-Im-Cu, TA + H₂O₂ + g-C₃N₄-Im-Cu after 12 h reaction. The mixed solution contains H₂O₂ 500 μl (100 mM), HAc/NaAc 1.5 ml (10 mM, pH=7), g-C₃N₄-Im-Cu 500 μl and TA 500 μl (2 mM).

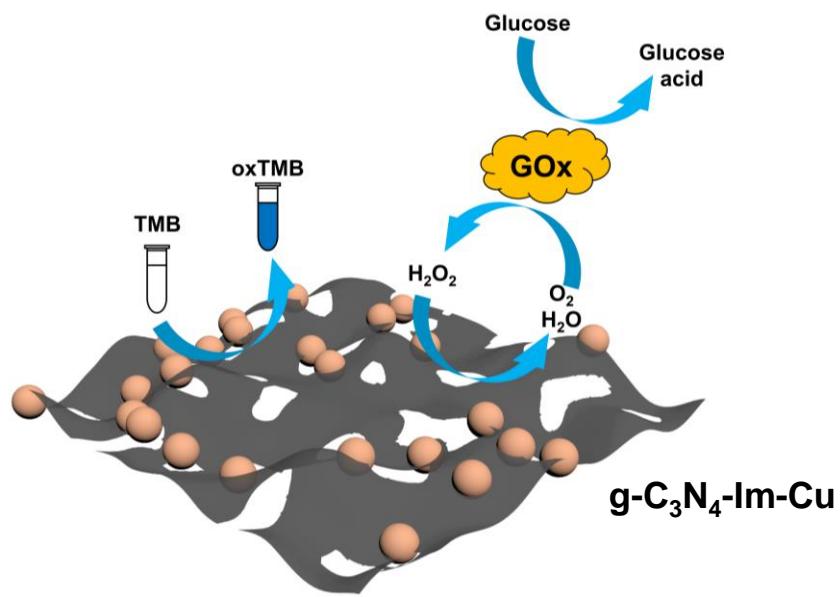


Figure S11. Schematic illustration of glucose detection with glucose oxidase (GOx) and $\text{g-C}_3\text{N}_4\text{-Im-Cu}$ catalyzed reactions.

Table S1. Activation energy calculation results.

T(K)	-1(K ⁻¹)	k(s ⁻¹)	Ea (kJ•mol ⁻¹)	A (s ⁻¹)
278	-0.0036	0.0081		
288	-0.00347	0.0098	11.54	1.2
298	-0.00336	0.01132		

Table S2. Activation energy for H₂O₂ oxidation catalyzed by different catalysts.

Number	Sample	Ea (kJ•mol ⁻¹)	Reference
1	ZrO ₂	33±1	[s1]
2	TiO ₂	34±1	[s2]
3	Y ₂ O ₃	44±5	[s2]
4	MoS ₂ /GO	24.64	[s3]
5	GO	28.8	[s4]
6	g-C ₃ N ₄ -Im-Cu	11.54	This work

Table.S3. Limit of detection (LOD) of various nanozymes for glucose.

No.	Catalysts	Limit of detection (μM)	Reference
1	$\text{Fe}_3\text{O}_4@\text{CeO}_2$	21	[s5]
2	Corrole- Fe_3O_4 nanocomposites	2.46	[s6]
3	Fe_3O_4 NPs	30	[s7]
4	Fe-porphyrin-based covalent organic framework (Fe-COF)	1	[s8]
5	Co_3O_4	0.32	[s9]
6	MoS_2/GO	0.086	[s3]
7	$\text{MoS}_2@\text{MgFe}_2\text{O}_4$	2	[s10]
8	MoO_3/C	10	[s11]
9	VS_2	1.5	[s12]
10	MnO_2 NFs	5	[s13]
11	GOD-GO/ MnO_2	17	[s14]
12	Carbon quantum dots (CQDs)	3	[s15]
13	$\text{GOx}@\text{ZIF-8}@\text{Fe-PDA}$	1.1	[s16]
14	MnO_2	10	[s17]
15	NiFe-LDHNS	50	[s18]
16	multielement-doped carbon dots (ME-CDs)	60	[s19]
17	CoO-OMC nanocomposite	68	[s20]
18	$\text{Fe}_3\text{O}_4@\text{SiO}_2-\text{NH}_2-\text{Au}@\text{Pd}_{0.30}\text{NPs}$	0.06	[s21]
19	rhodium nanoparticles (RhNPs)	0.75	[s22]
20	Ag@Au core/shell triangular nanoplates	800	[s23]

21	Au-Ni/g-C ₃ N ₄	1.7	[s24]
22	AgNPs/GQDs	0.03	[s25]
23	PtAg-multi-walled carbon nanotubes	600	[s26]
24	g-C ₃ N ₄	0.4	[s27]
25	Fe-g-C ₃ N ₄	0.5	[s28]
26	Cu NPs/g-C ₃ N ₄	0.37	[s29]
27	PdNPs/g-C ₃ N ₄	0.4	[s30]
28	g-C ₃ N ₄ -PdNPs	1	[s31]
29	Ru-C ₃ N ₄	0.1	[s32]
30	MnSe-g-C ₃ N ₄	8	[s33]
31	CuPd@H-C ₃ N ₄	0.1	[s34]
32	g-C ₃ N ₄ -Im-Cu	0.01	This work

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