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Electronic Supporting Information

Liquid-phase Growth of Platinum Nanoparticles on Molybdenum Trioxide Nanosheets: An Enhanced Catalyst with Intrinsic Peroxidase-like Catalytic Activity

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Experimental Section

Chemicals. Bulk α-MoO_3 powder was purchased from Stem Chemicals Inc. (MA). Potassium tetrachloroplatinate (II) (99.99% trace metals basis), glucose, fructose, lactose, maltose, 3,3',5,5'-tetramethylbenzidine (TMB), glucose oxidase (GOx, from Aspergillus Niger) and human serum samples were obtained from Sigma-Aldrich. Hydrogen peroxide (H_2O_2) (50%) and ethanol (99%) were obtained from Aik Moh Paints & Chemicals Pte Ltd (Singapore). All chemicals were used as received without further purification. The deionised water was purified using Milli-Q System (Millipore, Billerica, MA).

Preparation of 2D MoO_3 nanosheets. Liquid exfoliation method was used to prepare the 2D MoO_3 nanosheets.[1] Briefly, 0.2 g of bulk α-MoO_3 powder was grounded with 0.2 mL of acetonitrile for 15 min. The powder was then dispersed in a mixture of ethanol and water (25 mL, v/v = 1/1). After that, the mixed solution was probe-sonicated (Ultrasonic Processor GEX500) for 120 min at the power of 125 W, and then centrifuged at 3,000 r.p.m. for 20 min at room temperature. The blue supernatant containing a high concentration of 2D MoO_3 nanosheets was collected.

Synthesis of Pt-MoO_3 hybrid nanomaterials. After 2 mL of 0.1 mg mL^{-1} 2D MoO_3 nanosheets solution was centrifuged at 8,000 r.p.m. for 20 min, the supernatant was removed and the precipitate was re-dispersed in the mixture of 7.5 mL of ethanol and 0.5 mL of 1.0 mM K_2PtCl_4 in deionised water. This mixed solution in a 10 mL glass vial was then irradiated with a 150 W halogen lamp at 80% of its full intensity for 2 h, during which the glass vial was cooled by ice water to prevent the light-induced overheating. After the reaction, the resulting solution was centrifuged at 7,500 r.p.m. for 15 min, and the precipitated Pt-MoO_3 hybrid nanomaterials was re-dispersed in water solution before further characterization.

Characterization. A drop of a solution containing the 2D MoO_3 nanosheets or Pt-MoO_3 hybrid nanomaterials was dropped on a substrate and then naturally dried in air. Then the samples were characterized by TEM (JEM 2100F, JEOL) coupled with EDX elemental mapping (holey carbon-coated copper grid as substrate), AFM (Dimension 3100, Veeco) (Si as substrate), Raman spectroscopy (InVia Reflex Raman Microscope, Renishaw) (glass as substrate), and XRD (XRD-600, Shimadzu) (glass as substrate). The metal components were measured by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES, Optima 5300 DV, PerkinElmer).

Assay. Kinetic measurements were carried out in time course mode by monitoring the absorbance change at 652 nm using an automatic microplate reader (Infinite M200, Tecan). For the experiment of 2D MoO_3 nanosheets, 16 μg of MoO_3 nanosheets reacted with 300 μL solution containing 25 mM Na_2HPO_4 (pH 4.0, 40 ºC), 800 μM TMB (used as substrate), and H_2O_2 (concentration of 50 mM, unless otherwise stated). For the experiment of Pt-MoO_3 hybrid nanomaterials, 30 μL of Pt-MoO_3 (0.414 μg MoO_3 and 0.396 μg Pt) reacted with 300 μL solution containing 25 mM Na_2HPO_4 (pH 4.0, 40 ºC) and 800 μM TMB (used as substrate), and H_2O_2 (concentration of 5 mM, unless otherwise stated). The apparent kinetic parameters were calculated based on the function $v = V_{\max} \times [S]/(K_m + [S])$, where v is the initial velocity, $V_{\max}$ is the maximal reaction velocity, $[S]$ is the concentration of substrate, and K_m is the Michaelis constant.[2]

Glucose detection. This detection procedure was shown as follows: (1) 5 μL of 40 mg mL^{-1} GOx and 100 μL of glucose with different concentration in 0.5 mM Na_2HPO_4 buffer solution (pH 7.0) were incubated at 37 ºC for 1 h; (2) 15 μL of the above glucose reaction solutions were added into 100 μL of 2.4 mM TMB, 30 μL of the Pt-MoO_3 solution, and 155 μL of 25 mM Na_2HPO_4 buffer solution (pH 4.0). The mixed solution was used to perform the measurement. In control experiments, 5 mM maltose, 5 mM lactose, and 5 mM fructose were used for the test.

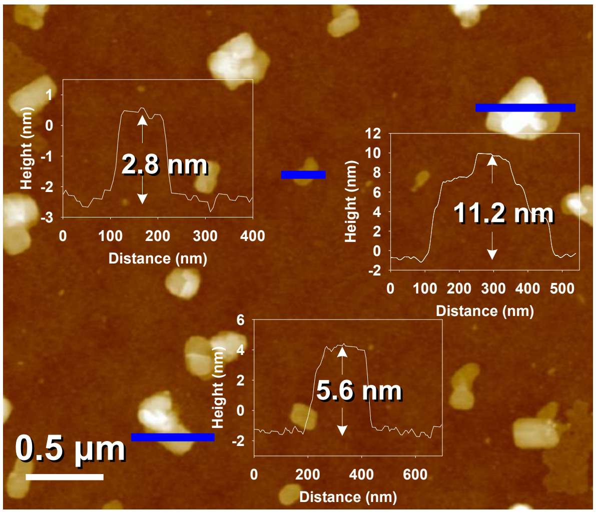

Figure S1. AFM image of the prepared 2D MoO_3 nanosheets.

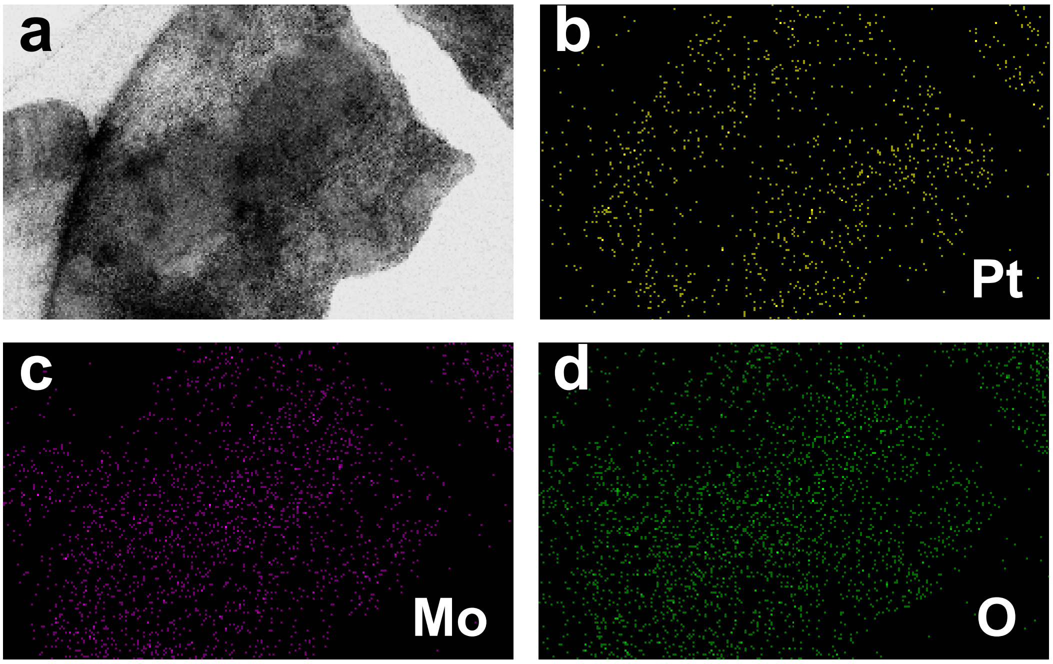

Figure S2. (a) A dark-field TEM image of a Pt-MoO_3 hybrid nanostructure, and its elemental mapping of (b) Pt, (c) Mo and (d) O.

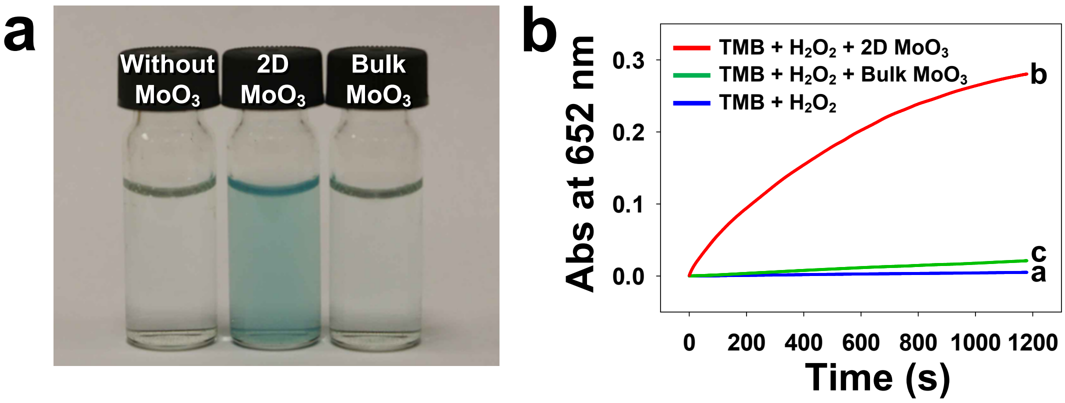

Figure S3. (a) Typical photograph of 800 μM TMB solution oxidized by 2D MoO_3 nanosheets in the presence of 50 mM H_2O_2 when incubated in 25 mM Na_2HPO_4 solution (pH 4.0) at 40 °C. From left to right: without MoO_3; 16 μg 2D MoO_3 nanosheets; and 16 μg bulk MoO_3 powder. (b) The corresponding time-dependent absorbance changes at 652 nm.

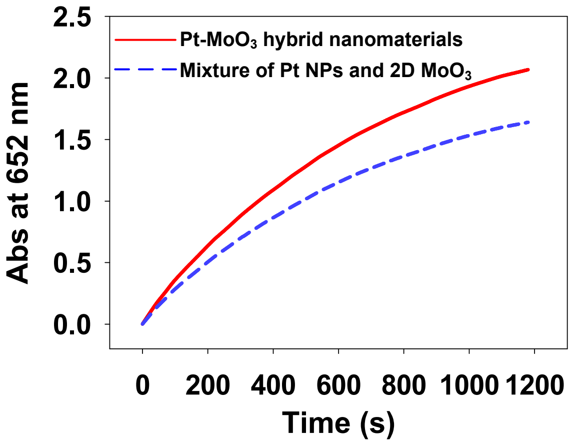

Figure S4. Time-dependent absorbance changes at 652 nm of 800 μM TMB solutions (5 mM H_2O_2) with two different catalysts: physical mixture of 0.414 μg MoO_3 nanosheets and 0.396 μg Pt NPs, and 30 μL of Pt-MoO_3 solution (0.414 μg MoO_3 and 0.396 μg Pt).

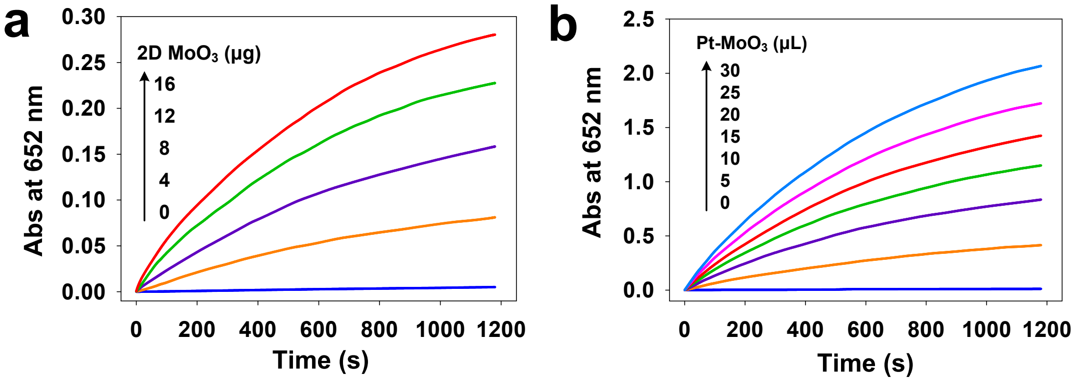

Figure S5. Time-dependent absorbance changes at 652 nm of 800 μM TMB solutions in the presence of (a) 2D MoO_3 nanosheets (50 mM H_2O_2) with different amount (from 0 to 16 μg) and (b) Pt-MoO_3 hybrid nanomaterials (5 mM H_2O_2) with different amount (from 0 to 30 μL). 30 μL of Pt-MoO_3 hybrid nanostructure is composed of 0.414 μg MoO_3 and 0.396 μg Pt.

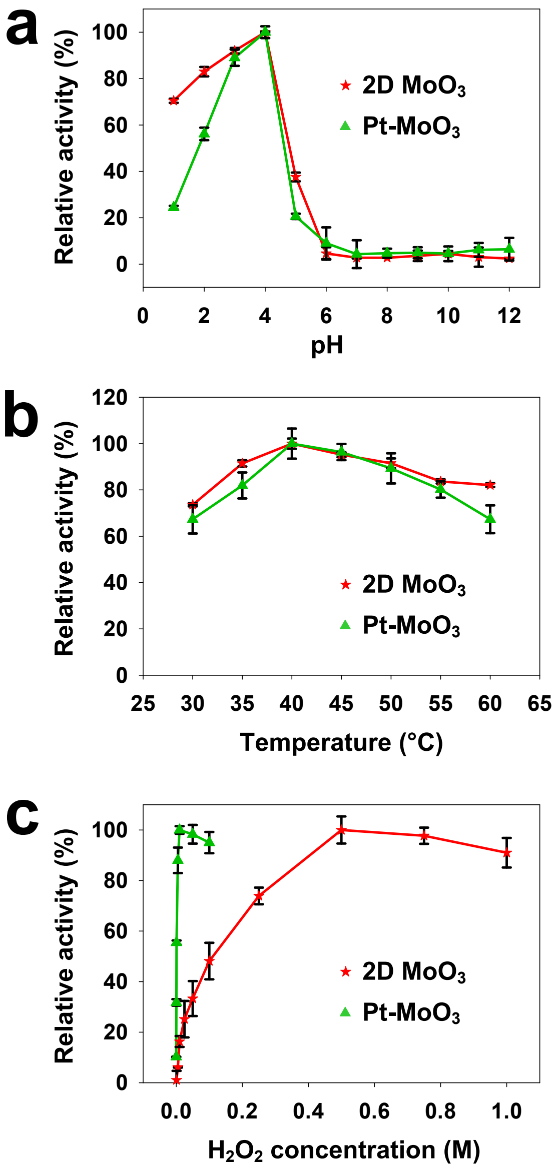

Figure S6. The peroxidase-like activity of 2D MoO_3 nanosheets and Pt-MoO_3 hybrid nanomaterials depends on pH (a), temperature (b), and H_2O_2 concentration (c). Experiments were carried out with MoO_3 nanosheets (16 µg) or Pt-MoO_3 hybrid nanomaterials (0.414 µg MoO_3 and 0.396 µg Pt) in 0.3 mL reaction solution using 25 mM Na_2HPO_4 solution with 800 µM TMB as substrate. The concentration of H_2O_2 was 50 mM for 2D MoO_3 nanosheets and 5 mM for Pt-MoO_3 at pH 4.0 and 40 °C, unless otherwise stated. The maximum point in each curve was set as 100%. The optimal catalytic environment for 2D MoO_3 nanosheets towards the TMB-H_2O_2 system is 25 mM Na_2HPO_4 solution (pH 4.0) with 500 mM H_2O_2 at 40 °C. The optimal catalytic environment for Pt-MoO_3 hybrid nanomaterials towards the TMB-H_2O_2 system is 25 mM Na_2HPO_4 solution (pH 4.0) with 5 mM H_2O_2 at 40 °C.

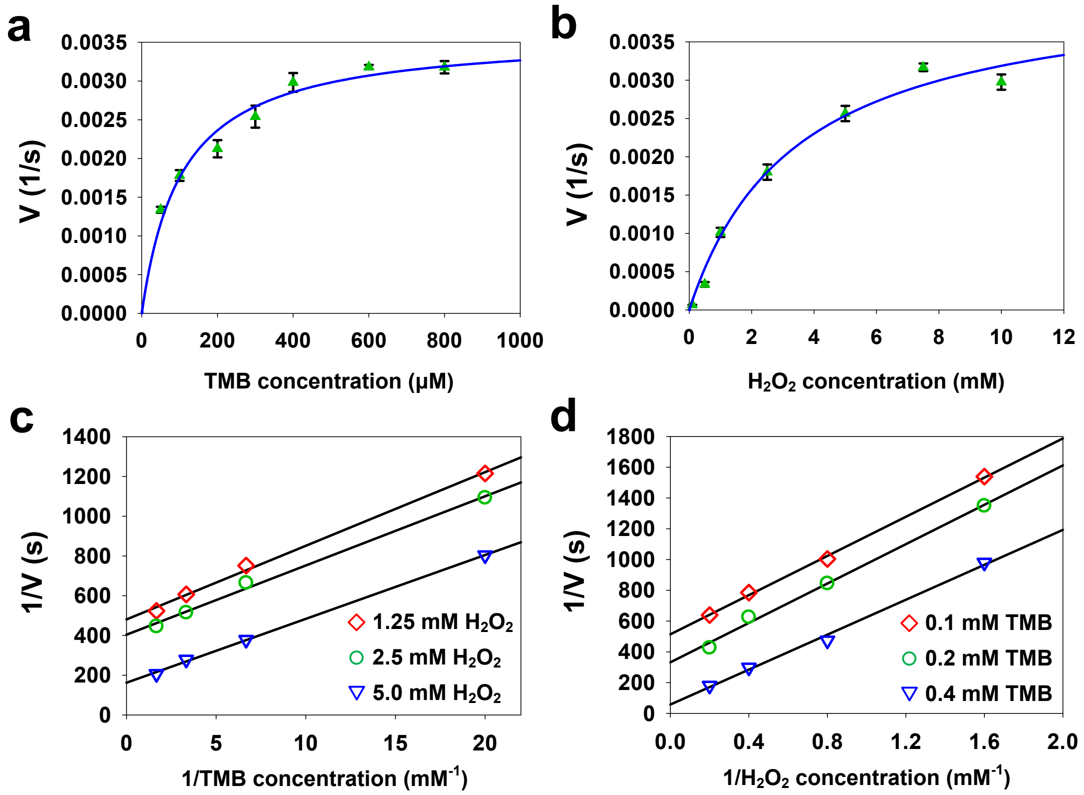

Figure S7. Steady state kinetic assay and catalytic mechanism of Pt-MoO_3 hybrid nanomaterials. The velocity (*v*) of reaction was measured with 30 μL of Pt-MoO_3 hybrid nanomaterials (0.414 μg MoO_3 and 0.396 μg Pt) in 0.3 mL of 25 mM Na_2HPO_4 solution (pH 4.0) at 40 °C. (a) The concentration of TMB was 800 μM and the H_2O_2 concentration was varied. (b) The concentration of H_2O_2 was 5 mM and the TMB concentration was varied. (c, d) Double-reciprocal plots of activity of Pt-MoO_3 hybrid nanomaterials at a fixed concentration of one substrate versus varying concentration of the second substrate for H_2O_2 and TMB.

Table S1. Comparison of the kinetic parameters of different catalysts and HRP.

Catalyst	Substrate	K_m [mM]	V_{max} [10^{-8} M s^{-1}]
2D MoO_3 nanosheets	TMB	0.093	3.041
	H_2O_2	109.5	5.284
Pt NPs	TMB	0.301	4.252
	H_2O_2	6.1	3.611
Mixture of 2D MoO_3 nanosheets and Pt NPs	TMB	0.296	4.263
	H_2O_2	5.9	3.625
Pt-MoO_3 hybrid nanomaterials	TMB	0.106	4.286
	H_2O_2	3.2	3.794
HRP[2]	TMB	0.434	10
	H_2O_2	3.7	8.7

K_m: Michaelis constant. V_{max}: maximal reaction velocity.

Figure S8. Steady state kinetic assay and catalytic mechanism of 2D MoO_3 nanosheets. The velocity (*v*) of reaction was measured with 16 μg MoO_3 nanosheets in 0.3 mL of 25 mM Na_2HPO_4 solution (pH 4.0) at 40 °C. (a) The concentration of TMB was 800 μM and the concentration of H_2O_2 was varied. (b) The concentration of H_2O_2 was 50 mM and the concentration of TMB was varied. (c, d) Double-reciprocal plots of activity of 2D MoO_3 nanosheets at a fixed concentration of one substrate versus varying concentration of the second substrate for H_2O_2 and TMB.

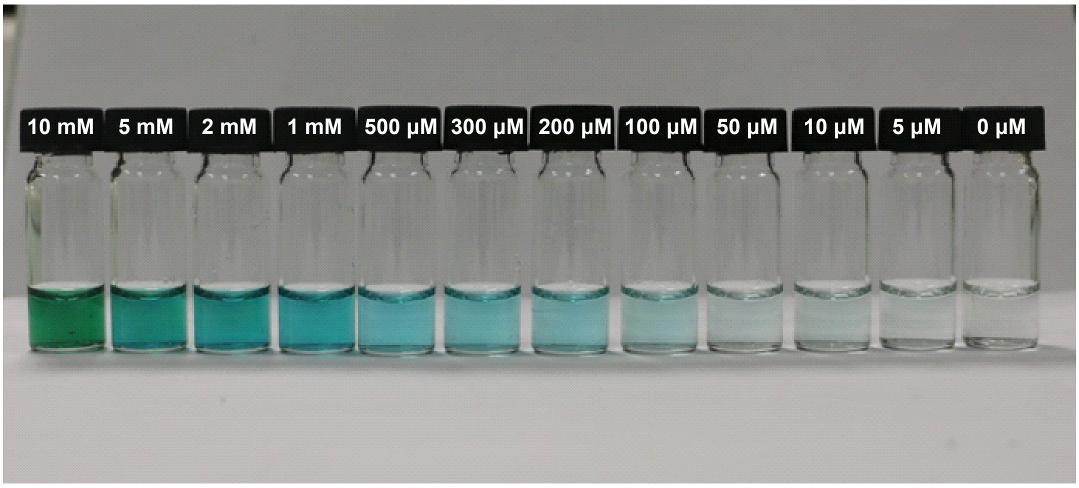

Figure S9. Photograph of the phosphate buffer solutions containing 120 μL of Pt-MoO_3 hybrid nanomaterials and 800 μM TMB with the addition of 60 μL of glucose reaction solutions (catalyzed by GOx). The glucose concentration decreases from left to right.

Table S2. Analysis of serum samples.

Serum sample[a]	Our colorimetric method (mM)[b]	Recovery test[d]		
		Added (mM)	Founded (mM)	Recovery (%)
No.1	3.61 ± 0.02[c]	1.0	0.93 ± 0.04	93.0
No.2	3.89 ± 0.09	1.0	0.95 ± 0.06	95.0
No.3	3.54 ± 0.07	1.0	0.91 ± 0.05	91.0

[a] All the samples were diluted 10-fold using 0.5 mM Na_2HPO_4 solution (pH 7.0). [b] All samples were analyzed using standard addition method (n = 3). [c] Mean value ± standard deviation (n = 3). [d] Recovery test was performed by addition of glucose standard solution into the serum samples with the same test procedure in the sample analysis.

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