

**Electronic Supporting Information for:**

**Novel enzyme-functionalized covalent organic frameworks for  
colorimetric sensing of glucose in body fluids and drinks**

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**1. Supplementary Materials**

2-hydroxybenzene-1, 3, 5-tricarbaldehyde (HTA) was purchased from TCI. 4,4'-  
diamino-3,3'-biphenyldicarboxylic acid (DBA) was bought from Ariel chemical  
technology Co. Ltd. (China). Glucose oxidase (GOX) and horseradish peroxidase  
(HRP) were purchased from Zengbo Biological Technology Co. Ltd.. 1-ethyl-3-(3-  
dimethylaminopropyl)carbodiimide (EDC), N-hydroxysuccinimide (NHS), 3,3',5,5'-  
tetramethylbenzidine (TMB), fructose, mannose, sucrose, galactose and glucose were

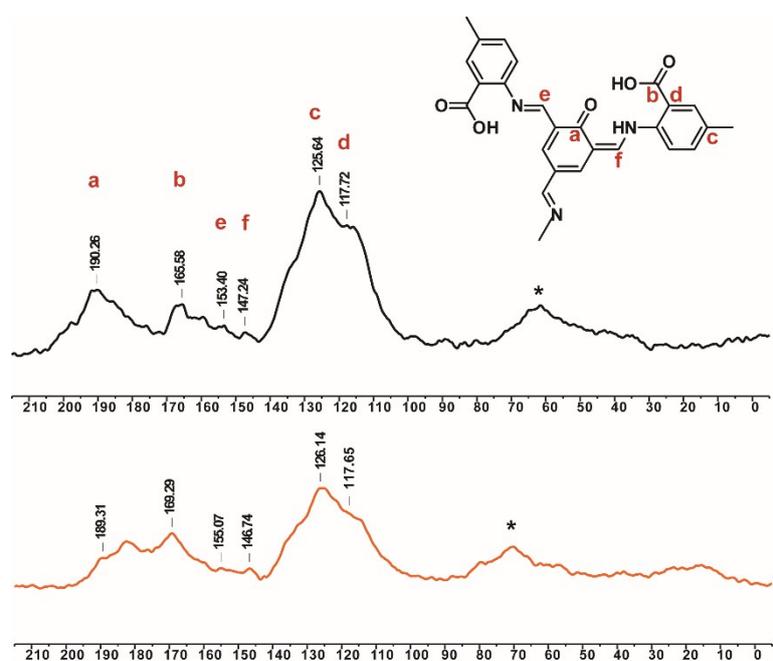
bought from Energy Chemical. All the chemicals mentioned in this work were utilized without further purification. The BCA protein assay kit (P0010) was purchased from Beyotime Biotechnology Company. O-toluidine (O-T) method kit (TC0716) was purchased from Leagene Biotechnology Company. Serum and diabetic urine were supported by volunteers from the Affiliated Hospital of Shandong University of Traditional Chinese Medicine. The green scream drink was bought from Nongfu Spring.

## **2. Instruments**

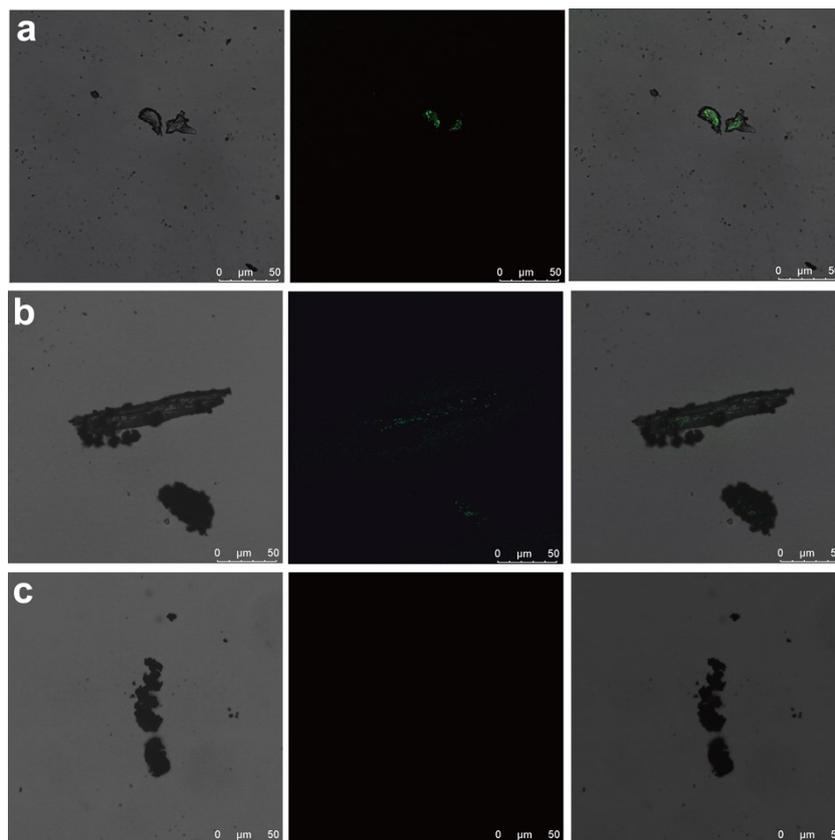
The powder X-ray diffraction (PXRD) pattern was obtained on D8 ADVANCE with Cu K $\alpha$  radiation ( $\lambda = 1.5405 \text{ \AA}$ ) with a  $2\theta$  range from  $2^\circ$  to  $40^\circ$  at room temperature. Bruker ALPHA FT-IR Spectrometer was exploited to record the Fourier transform infrared (FT-IR) spectroscopy at  $500\text{-}4000 \text{ cm}^{-1}$  range.  $^{13}\text{C}$  Solid state NMR spectra were collected on a Bruker Avance III 400 MHz machine. Thermogravimetric analysis (TGA) was performed on TGA/SDTA851e in  $\text{N}_2$  atmosphere at  $10 \text{ }^\circ\text{C min}^{-1}$  from  $25 \text{ }^\circ\text{C}$  to  $800 \text{ }^\circ\text{C}$ . Scanning electron microscopy (SEM) images were conducted on a SUB010 instrument. Transmission electron microscopy (TEM) analysis was recorded on Hitachi HT7700 electron microscope. The Brunauer-Emmett-Teller (BET) surface areas were tested on an ASAP 2020/TriStar 3000 (Micromeritics) at  $77 \text{ K}$ . Ultraviolet-visible (UV-Vis) spectra was performed on a Hitachi U-4100 spectrophotometer. Confocal fluorescence images were taken by CS SP8 confocal laser scanning microscopy. The Circular dichroism (CD) spectra were recorded on a

J-815 spectropolarimeter (Jasco, Japan).

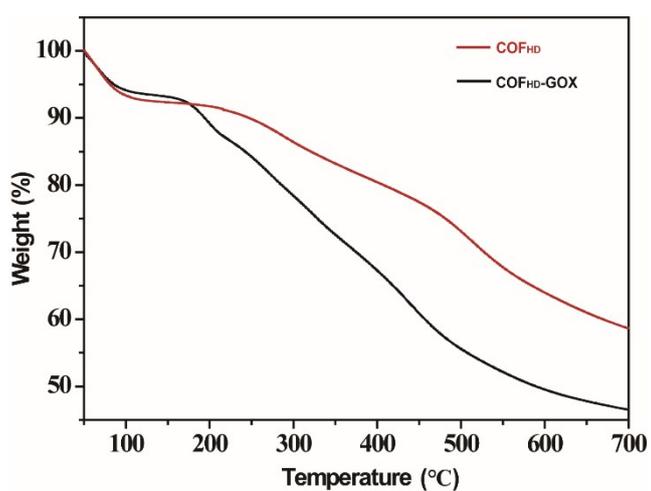
### 3. Supplementary Figures and Tables



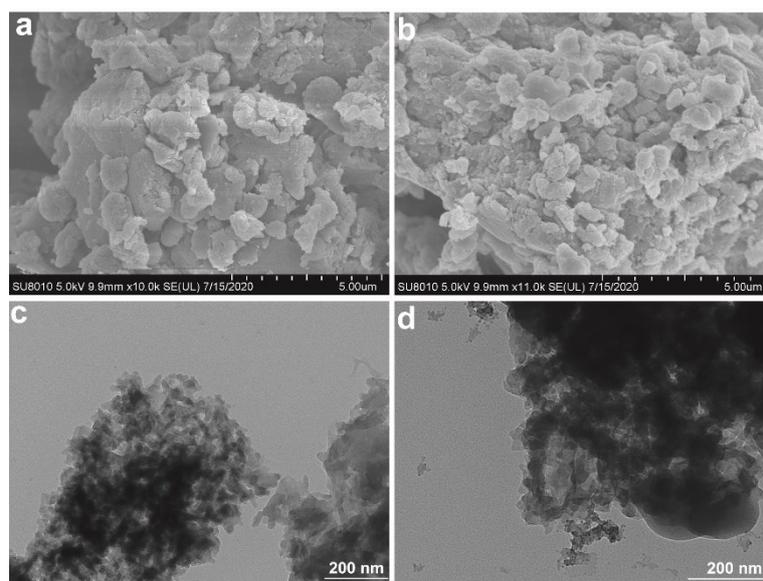
**Figure S1.** The solid-state  $^{13}\text{C}$  NMR spectra of  $\text{COF}_{\text{HD}}$  (black curve) and  $\text{COF}_{\text{HD}}\text{-GOX}$  (orange curve).



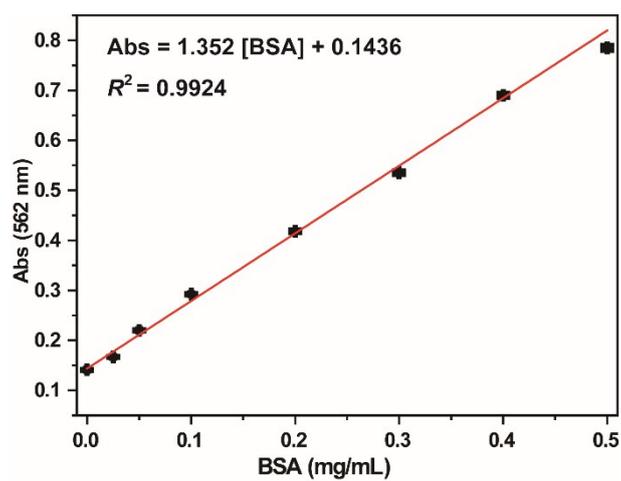
**Figure S2** Confocal microscopy images of (a) FITC labeled GOX; (b) COF<sub>HD</sub>-GOX (GOX was labeled with FITC in advance and then fluorescent GOX was post-modified on COF<sub>HD</sub>); (c) COF<sub>HD</sub>.



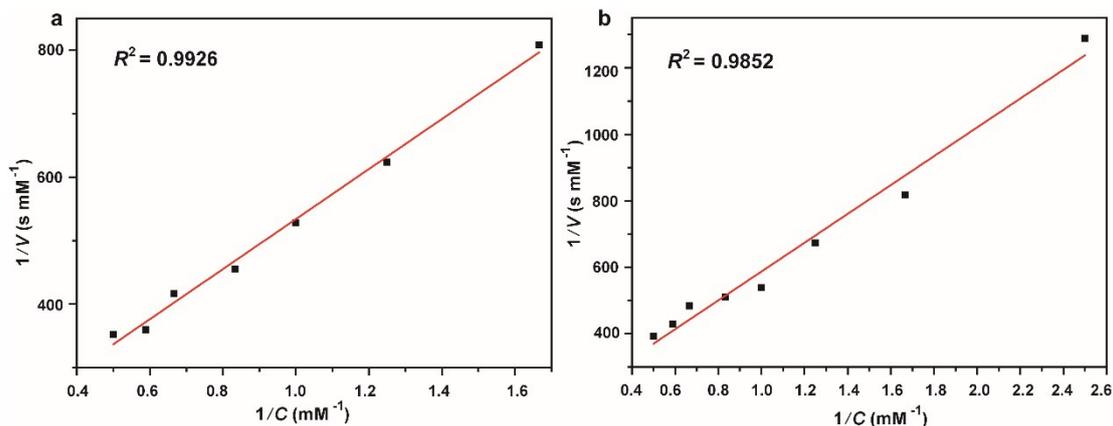
**Figure S3.** The TGA curves of COF<sub>HD</sub> (red curve) and COF<sub>HD</sub>-GOX (black curve).



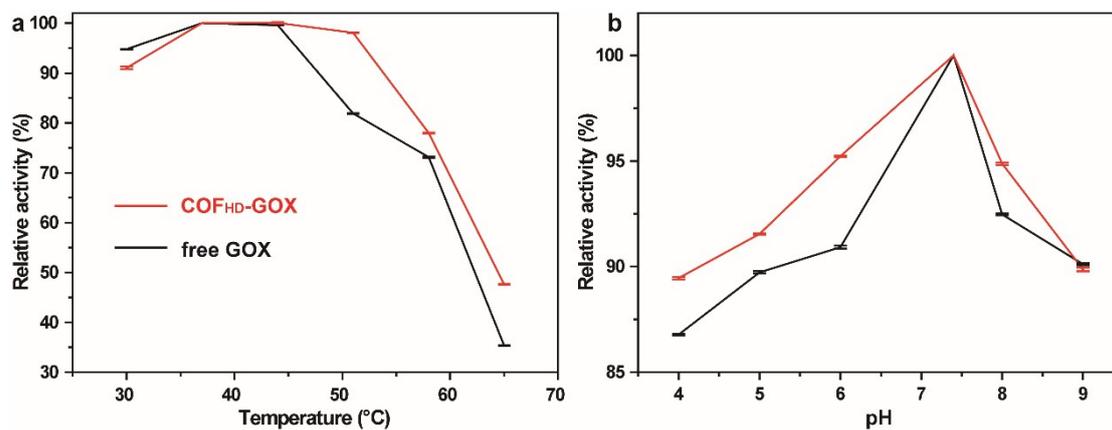
**Figure S4.** SEM image of (a) COF<sub>HD</sub>; (b) COF<sub>HD</sub>-GOX. The TEM image of (c) COF<sub>HD</sub>; (d) COF<sub>HD</sub>-GOX.



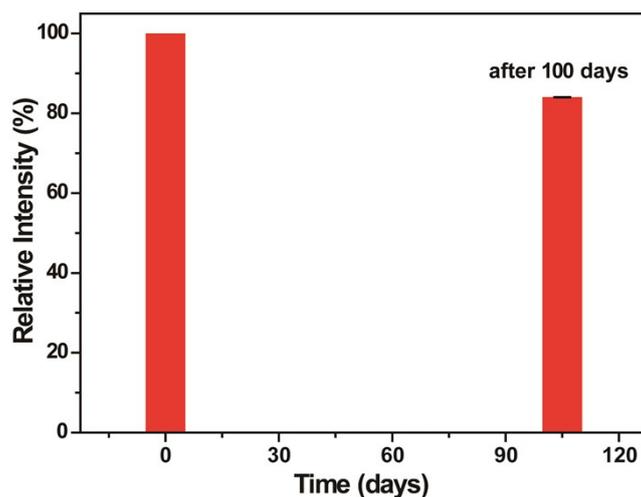
**Figure S5.** The standard curve by BCA Protein Assay Kit. The error bars represent the standard deviation of three measurements.



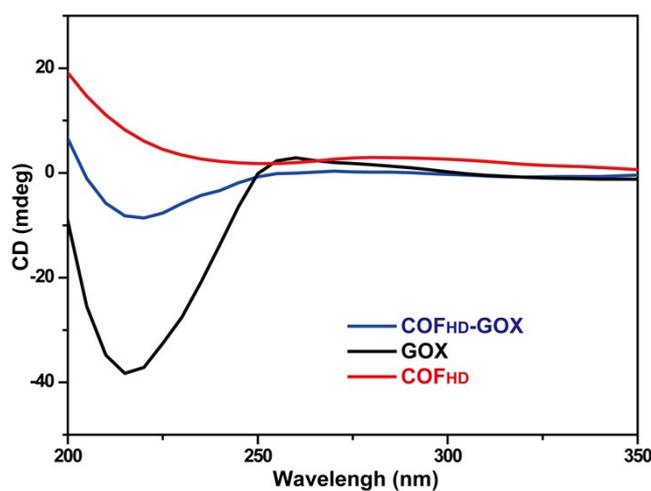
**Figure S6.** Double reciprocal plots of activity of (a) free GOX and (b)  $\text{COF}_{\text{HD}}\text{-GOX}$  with the concentration of glucose.



**Figure S7.** Effect of (a) temperature and (b) pH on the activity of  $\text{COF}_{\text{HD}}\text{-GOX}$  (red line) and free GOX (black line). The error bars represent the standard deviation of three measurements.



**Figure S8.** The storage stability of COF<sub>HD</sub>-GOX. After kept in PBS for 100 days, the relative activity is still more than 85%.



**Figure S9.** The CD spectra of GOX, COF<sub>HD</sub> and COF<sub>HD</sub>-GOX (0.5 mg/mL in PBS, pH 7.4).

P2/m			
a = b = 29.8715 Å, c = 3.388Å			
$\alpha = \beta = 90.^\circ$ and $\gamma = 60.63^\circ$			
Atom	u	v	w
H1	0.4693	0.71337	0.5
H2	0.2011	0.82243	0.5
H3	0.3205	0.96564	0.5
C4	0.063	0.00015	0.5

C5	0.028	0.98173	0.5
C6	0.0468	0.92813	0.5
C7	0.0997	0.89284	0.5
C8	0.1347	0.9121	0.5
C9	0.1156	0.96581	0.5
C10	0.5706	0.97929	0.5
C11	0.6041	0.99962	0.5
C12	0.5833	0.05399	0.5
C13	0.5298	0.08611	0.5
C14	0.4972	0.06502	0.5
C15	0.5172	0.01133	0.5
C16	0.1165	0.83683	0.5
O17	0.1638	0.80304	0.5
O18	0.0832	0.8219	0.5
C19	0.6601	0.96264	0.5
O20	0.6761	0.91426	0.5
O21	0.6928	0.97827	0.5
N22	0.8109	0.12231	0.5
C23	0.7773	0.10421	0.5
C24	0.628	0.27461	0.5
C25	0.6002	0.12744	0.5
C26	0.6347	0.14623	0.5
C27	0.6155	0.19961	0.5
C28	0.648	0.22134	0.5
C29	0.7015	0.18986	0.5
C30	0.7244	0.13598	0.5
C31	0.6913	0.11205	0.5
N32	0.5768	0.30758	0.5
C33	0.4798	0.44883	0.5
C34	0.5003	0.39496	0.5
C35	0.5543	0.36281	0.5
C36	0.5859	0.38556	0.5
C37	0.5647	0.43918	0.5
C38	0.5114	0.47148	0.5
N39	0.3835	0.92387	0.5
O40	0.7104	0.0651	0.5
C41	0.536	0.6257	0.5

O42	0.5841	0.59482	0.5
O43	0.5199	0.67405	0.5
H44	0.0482	0.0436	0.5
H45	0.019	0.91226	0.5
H46	0.1439	0.98112	0.5
H47	0.5877	0.93548	0.5
H48	0.5127	0.12991	0.5
H49	0.4538	0.09152	0.5
H50	0.792	0.06077	0.5
H51	0.6549	0.29162	0.5
H52	0.5571	0.15553	0.5
H53	0.5722	0.22693	0.5
H54	0.7274	0.20809	0.5
H55	0.4362	0.47429	0.5
H56	0.6295	0.36013	0.5
H57	0.5909	0.45707	0.5
H58	0.3412	0.94935	0.5

**Table S1.** Atomistic coordinates for COF<sub>HD</sub> optimized by Forcite method.