Electronic Supplementary Information (ESI)

Nanoscale Covalent Organic Frameworks as Smart Carriers for Drug Delivery

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1. Materials and instruments

All reagents and chemicals were purchased commercially and used without further purifications unless otherwise stated. Specially, anhydrous 1,4-dioxane, anhydrous mesitylene, anhydrous THF and 4,4'-biphenyldiamine were purchased from Sigma-Aldrich. Reactions were monitored by TLC and visualized by a dual short wave/long wave UV lamp. Column flash chromatography was performed using 230-400 mesh silica gel.

1.1 NMR

NMR spectra were recorded by a Bruker BBFO-400 spectrometer with $CDCl_3$ as a solvent. Chemical shifts for ^1H NMR were reported as δ, parts per million, relative to the signal of $CDCl_3$ at 7.26 ppm. Chemical shifts for ^{13}C NMR were reported as δ, parts per million, relative to the centerline signal of the $CDCl_3$ triplet at 77.0 ppm. Proton and carbon assignments were established using spectral data of similar compounds. The abbreviations s, br. s, d, dd, br. d, ddd, t, q, br. q, m, and br. m stand for the resonance multiplicity singlet, broad singlet, doublet, doublet of doublets, broad doublet, doublet of doublet of doublets, triplet, quartet, broad quartet, multiplet and broad multiplet, respectively.

1.2 Thermogravimetric analysis (TGA) and Fourier transform infrared (FT-IR)

TGA was performed on a TGA 500 thermo gravimetric analyzer by heating the samples at 20 ºC min^{-1} to 1000 ºC in a nitrogen atmosphere (60 mL/min). FTIR spectra (KBr, Aldrich) were measured with a SHIMADZU IR Prestige-21 spectrometer. Samples were packed firmly to obtain transparent films.

1.3 PXRD measurement

PXRD studies were performed on a SHIMADZU XRD-6000 Labx diffractometer using Cu-Ka radiation at 40 kV and 300 mA with a scanning rate of $0.02^o\ s^{-1}$ (2θ) at room temperature.

1.4 UV absorption and fluorescence spectra

Absorption spectra were recorded on UV-3600 UV−vis−NIR spectrophotometer (Shimadzu), while emission spectra were recorded on RF-5301 PC spectrofluorophotometer (Shimadzu) with 1.0 cm path length cell.

1.5 TEM measurements

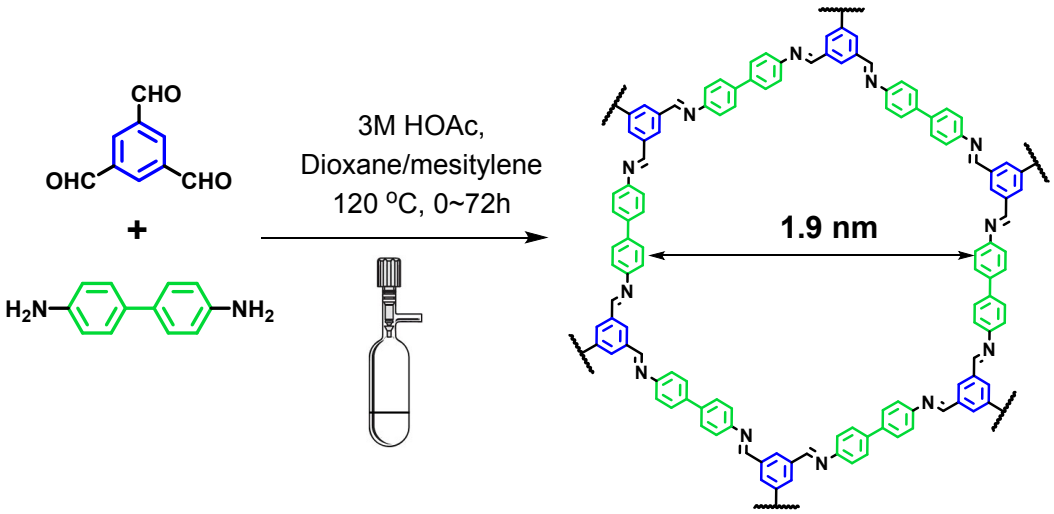
Transmission electron microscopy (TEM) images were measured on a JEM-1400 (JEOL) operated at 100-120 kV.

2. Experimental details

2.1 Synthesis of PI-2-COF [S1,S2]

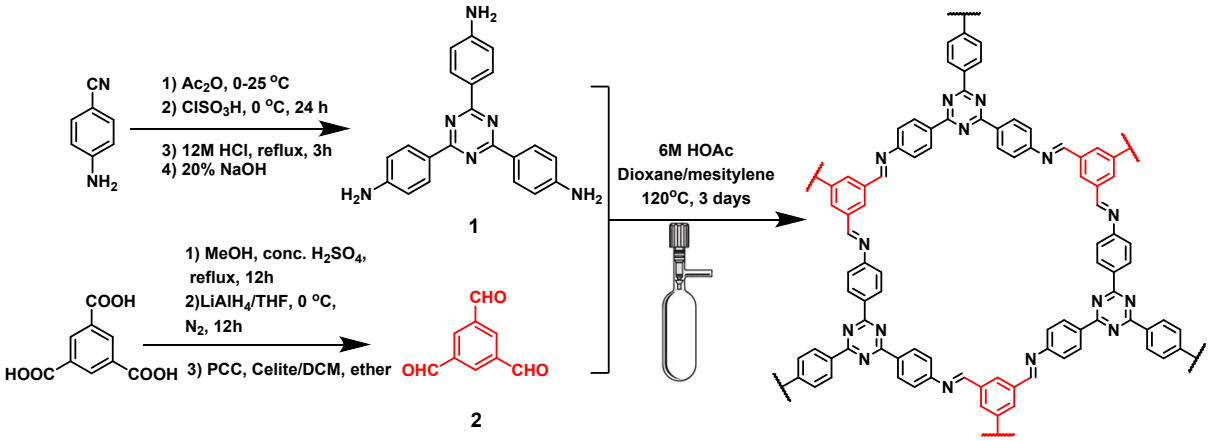
4,4'-Biphenyldiamine (83 mg, 0.3 mmol) and 1,3,5-triformylbenzene (48 mg, 0.2 mmol) were weighed into a 25 mL Schlenk storage tube (SynthwareTM, OD28 x L120mm, high vaccum valve size 0-8mm, with PTFE o-ring & wiper), dissolved in mixed solvent (3 mL, 1,4-dioxane:mesitylene = 1:1) and sonicated for 10 min. After that, aqueous HOAc (0.3 mL, 6 mol/L) was added to the mixture. The tube was degassed by three freeze-pump-thaw cycles. Finally, the tube was sealed off, heated at 120 ºC in an oven at the same time, and left undisturbed for 72 h. The precipitates

were isolated by centrifugation, washed with anhydrous THF for 3 times, and dried at 120 ºC under vacuum for 24 h, to give a yellow powder with up to 90 % isolation yields. The synthetic procedure is shown below.



2.2 Synthesis of PI-3-COF [S1,S2]

2,4,6-Tris(4-aminophenyl)-1,3,5-triazine (**1**) and 1,3,5-triformylbenzene (**2**) were synthesized according to the reported procedures.[S3] The ^{1}H NMR and ^{13}C NMR spectra matched well with those reported ones. Then, compounds **1** (70 mg, 0.2 mmol) and **2** (48 mg, 0.2 mmol) were placed in a 25 mL Schlenk storage tube, which were dissolved in a mixture solvent (10 mL, 1,4-dioxane : mesitylene = 10:1, v:v), and sonicated for 10 min. After that, aqueous HOAc (0.75 mL, 6M) was added, and the mixture was degassed by three freeze-pump-thaw cycles. Finally, the tube was sealed, heated at 120 ºC in an oven, and left undisturbed for 72 h. The precipitate was collected through centrifugation, which was washed with anhydrous THF for 3 times, and then dried at 150 °C under vacuum for 24 h. A yellow powder was obtained in 90% isolation yield. The synthetic procedure is shown below.



2.3 Theoretical calculations

Density functional theory calculations were performed with the Gaussian 09 program[S4] using the B3LYP functional. All-electron double-ξ valence basis sets with polarization functions 6-31G* were used for all atoms. Geometry optimizations were performed with full relaxation of all atoms in gas phase without solvent effects.

3. Structural modeling[S4]

3.1 **Table S1.** Simulated data of **PI-2-COF**

Space group	P6/M (No.175)		
Calculated unit cell	a=b=29.6551 Å, c=3.4469 Å; α=β=90°, γ=120°		
atom	x	y	z
N1	0.39261	0.58453	-0.00000
C2	0.31638	0.61442	-0.00000
C3	0.40696	0.63276	-0.00000
C4	0.42476	0.56216	-0.00000
C5	0.47820	0.59097	-0.00000
C6	0.50731	0.56690	-0.00000
C7	0.48422	0.51309	-0.00000
C8	0.43027	0.48490	-0.00000
C9	0.40138	0.50916	-0.00000
C10	0.36893	0.64967	-0.00000
H11	0.63982	0.51368	-0.00000
H12	0.59104	0.55607	-0.00000
H13	0.54795	0.59176	-0.00000
H14	0.49825	0.63209	0.00000
H15	0.44681	0.66156	-0.00000
H16	0.42609	0.72915	-0.00000

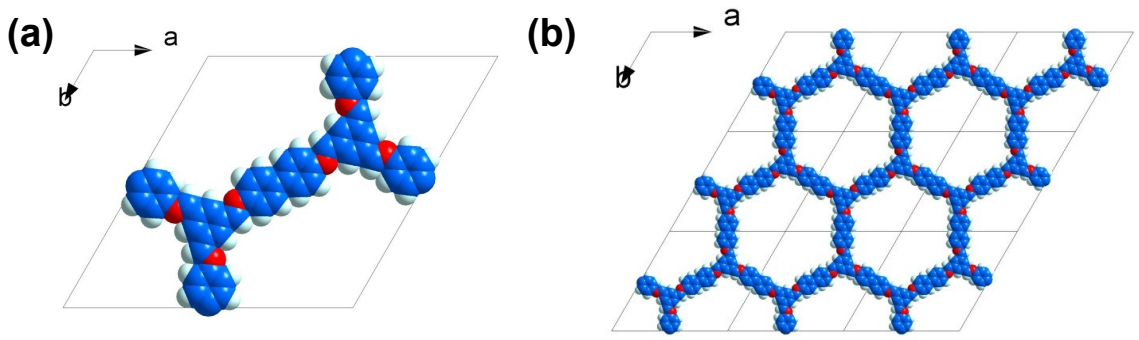


Figure S1. (a) Crystal lattice of the unit cell simulated in an eclipsed arrangement (top view in AB plane) for PI-2-COF; (b) Packing structure of 9 unit cells simulated in an eclipsed arrangement (top view in AB plane).

3.2 Table S2. Simulated data of **PI-3-COF**

Space group	P-6 (No.174)		
Calculated unit cell	a=b=18.1590 Å, c=3.4970 Å; α=β=90°, γ=120°		
atom	x	y	z
C1	0.15464	0.56364	3.50000
N2	0.10738	0.59749	3.50000
C3	-0.15691	0.43862	3.50000
C4	-0.10235	0.40528	3.50000
H5	-0.12561	0.34534	3.50000
C6	-0.01667	0.45691	3.50000
H7	0.01947	0.43286	3.50000
C8	0.01824	0.54298	3.50000
C9	-0.03574	0.57862	3.50000
H10	-0.01218	0.63874	3.50000
C11	-0.12258	0.52527	3.50000
H12	-0.15920	0.54868	3.50000
C13	0.36741	0.75401	0.50000
C14	0.28036	0.70135	0.50000
H15	0.23669	0.72988	0.50000
C16	0.61655	0.36657	0.50000
N17	0.70015	0.41959	0.50000

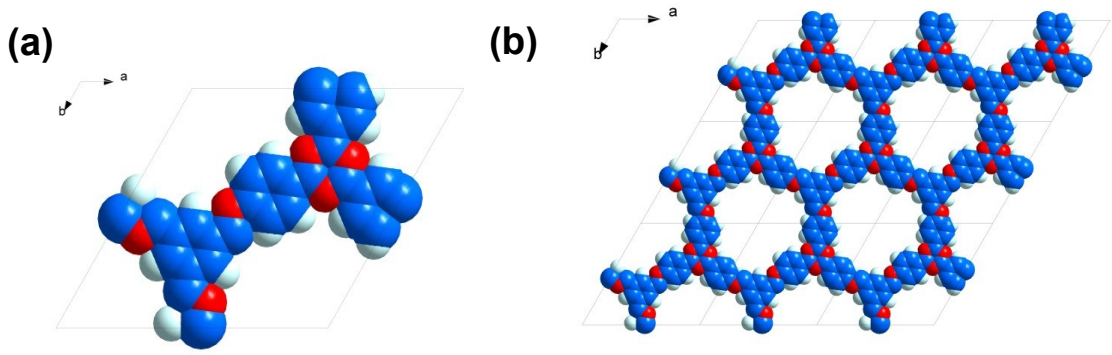


Figure S2. (a) Crystal lattice of the unit cell simulated in an eclipsed arrangement (top view in AB plane) for PI-2-COF; (b) Packing structure of 9 unit cells simulated in an eclipsed arrangement (top view in AB plane).

4. Physical properties

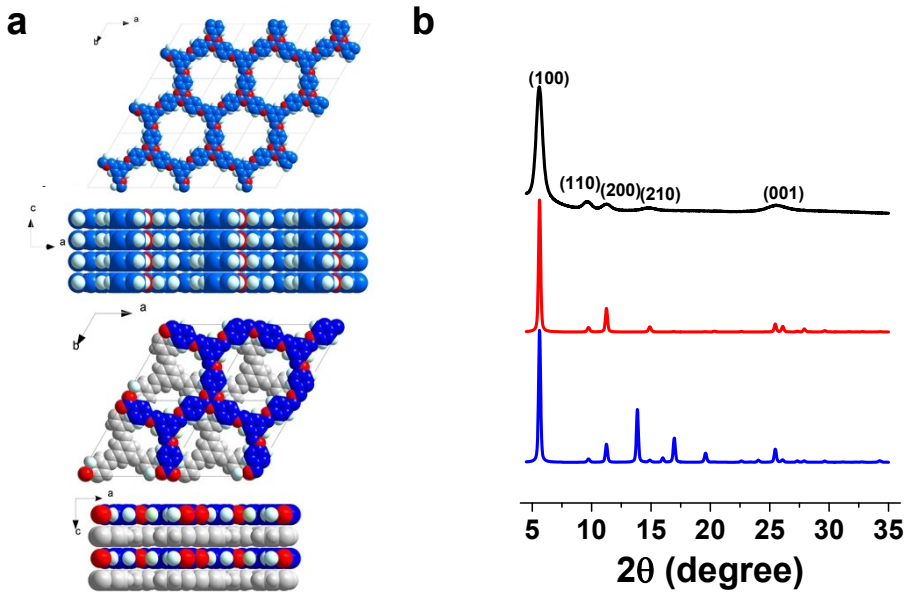


Figure S3. Theoretical calculation and practical PXRD of PI-3-COF: (a) packing structure simulated in an eclipsed arrangement (top view in AB plane); (b) experimental PXRD pattern of PI-3-COF (black curve), simulated PXRD pattern for eclipsed structure (red curve), and simulated PXRD pattern for staggered structure (blue curve).

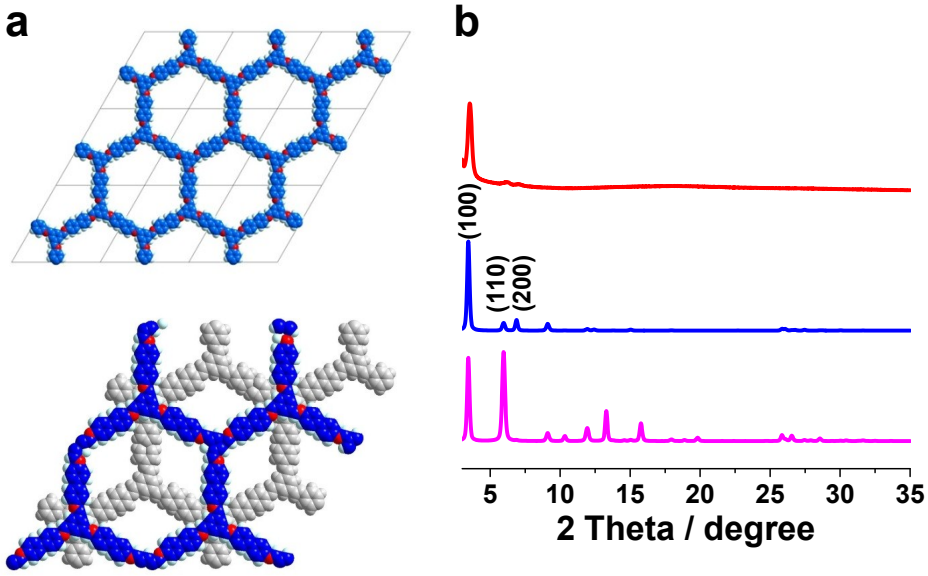


Figure S4. Theoretical calculation and practical PXRD of PI-2-COF: (a) packing structure simulated in an eclipsed (upper) and staggered structure (down); (b) experimental PXRD pattern of PI-2-COF (red curve), and simulated patterns of eclipsed (blue curve) and staggered (pink curve) structures.

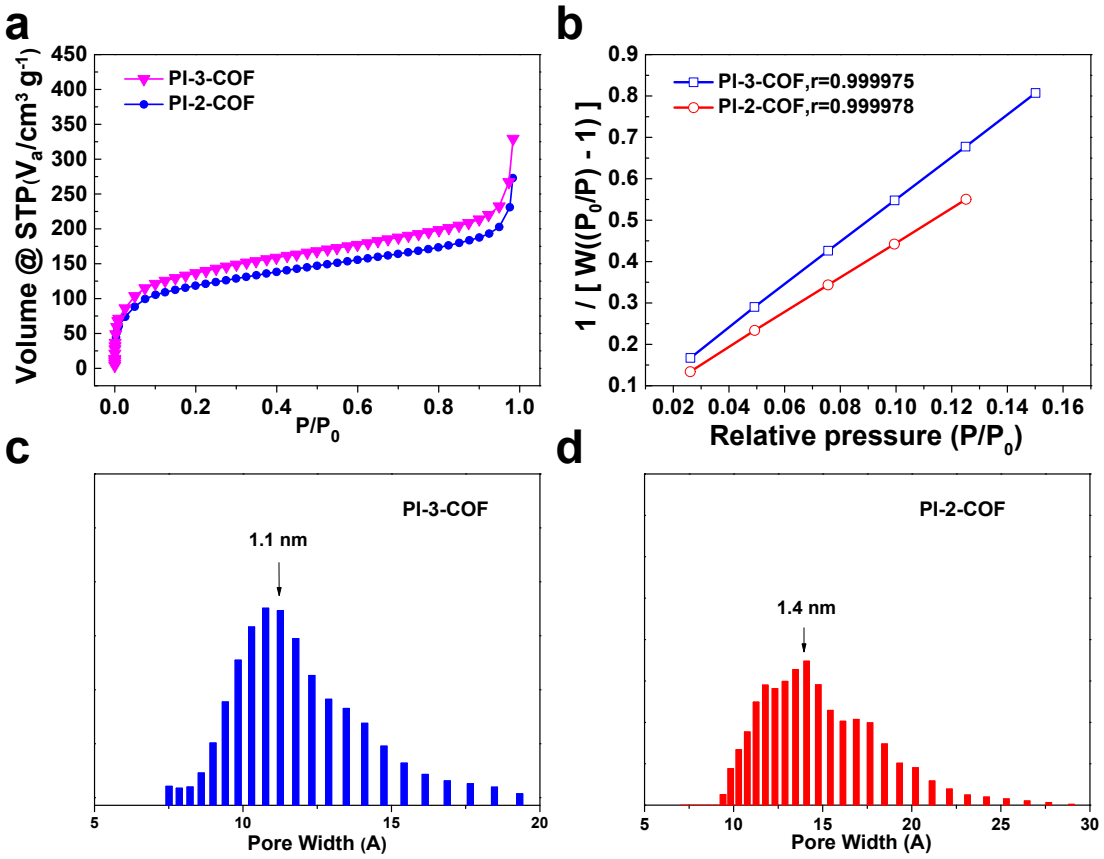


Figure S5. (a) N_2 adsorption isotherms and (b) BET results of the COFs; (c,d) Pore-size distribution plots of two COFs calculated from experimental N_2 adsorption isotherms using the nonlocal density functional theory (NLDFT).

5. *T*-plot method

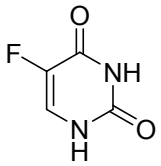
Low-pressure gas sorption measurements were performed by using Quantachrome Instruments Autosorb-iQ (Boynton Beach, Florida USA) with the extra-high pure gases. The as-synthesized COFs were immersed in CH_2Cl_2 (30 mL) for 3 days, during which CH_2Cl_2 was replaced for three times. Then, the samples were moved into a sample cell and dried under vacuum at 60 °C by using the "out gasser" function for 8 h before the measurements.

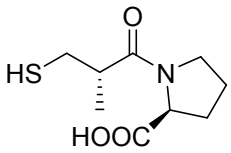
Table S3. T-plot method for microporous and mesoporous volumes.

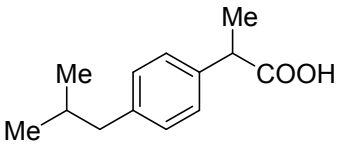
	PI-3-COF	PI-2-COF
[a]Total pore volume (cc/g)	0.662	1.138
[b]Micropore volume (cc/g)	0.498	0.853
[c]Mesopore volume (cc/g)	0.164	0.285
[b]Micropore area (m^2/g)	929.275	1286.788
[b]External surface area (m^2/g)	59.557	339.219

The data was determined by N_2 adsorption/desorption isotherms at 77K: [a] calculated by the point of the highest adsorption; [b] determined by the t-plot method, where the contribution points are from 0.35-0.5 in P/P_0; [c] obtained by the difference between the total pore volume and micropore volume.

6. Drug loading and release







5-fluorouracil (5-FU) **Captopril** **Ibuprofen (IBU)**

Three drugs, *i.e.*, 5-fluorouracil (5-FU), captopril and ibuprofen (IBU) were used for drug loading and release studies (their chemical structures are shown above). The drug was loaded by immersing solvent-free COF samples in hexane solution of drug with a certain concentration. A typical procedure for loading drug in COFs was as follows: COF (50 mg) was suspended in hexane solution of drug (5 mL, 0.1 M) under stirring for 6h, while preventing evaporation of hexane by covering with a cap. The drug-loaded sample was separated from solution by vacuum filtration, washed with hexane, and dried at room temperature.

A typical procedure for releasing drug from drug-loaded COF was as follows: the drug-loaded COF (10 mg) was placed in a vial and dipped in 2 mL of phosphate buffered saline (PBS, pH = 7.4, standard buffer solution from Sigma) at 37 °C. At predetermined time intervals, the dissolution medium was replaced with 2 mL of fresh PBS, and the withdrawn medium was used to determine the drug concentration. The drug concentration was analyzed by UV-Vis spectrophotometry with the help of a calibration curve. The release study was continued until no drug was detectable in the withdrawn PBS.

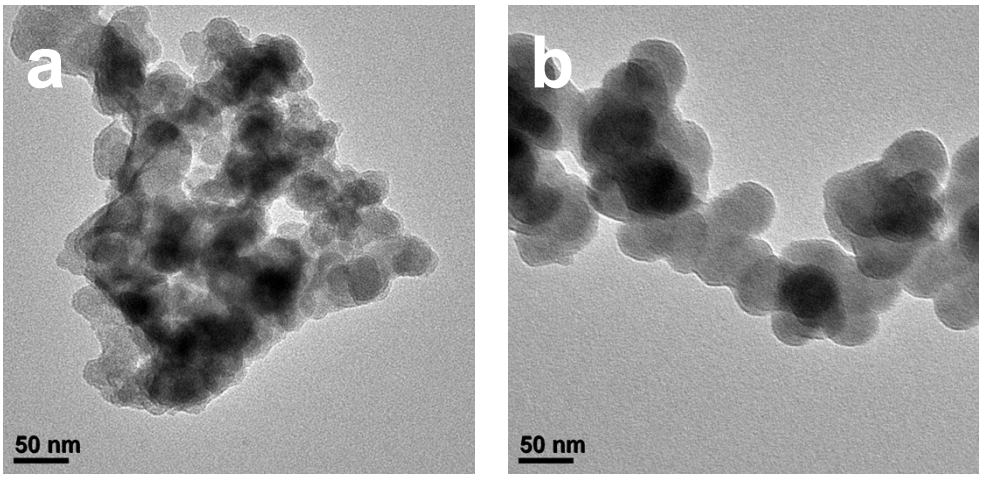


Figure S6. TEM images of the two COFs in PBS solution with a bit DMSO as an additive: (a) PI-3-COF and (b) PI-2-COF.

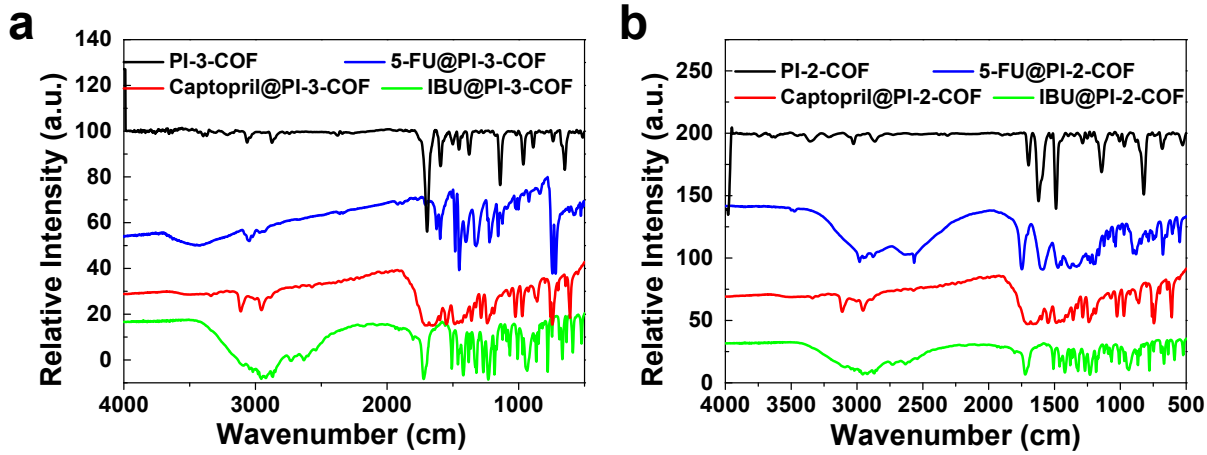


Figure S7. (a,b) FTIR spectra of two COFs (black curve) and drug loaded COFs: 5-FU (blue curve), captopril (red curve), and IBU (green curve).

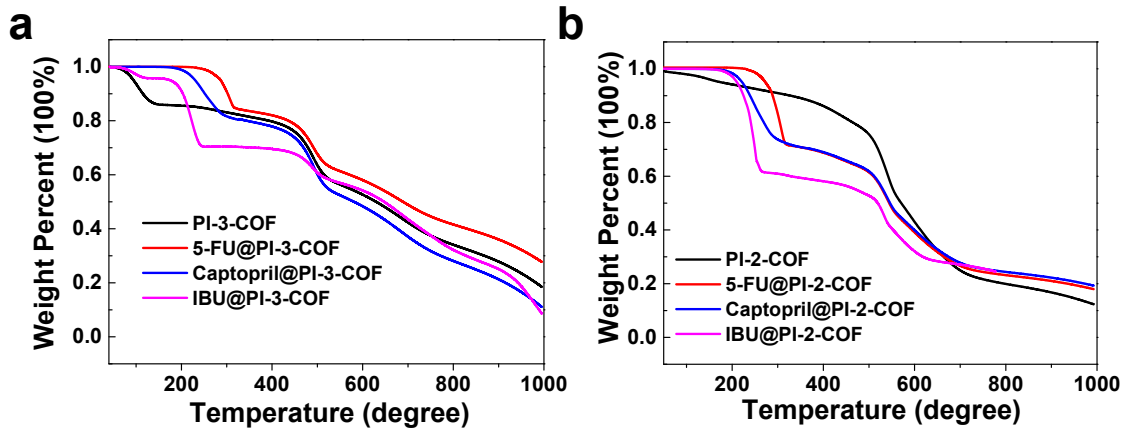


Figure S8. (a,b) TGA curves of two COFs (black curve) and drug loaded COFs: 5-FU (red curve), captopril (blue curve), and IBU (pink curve).

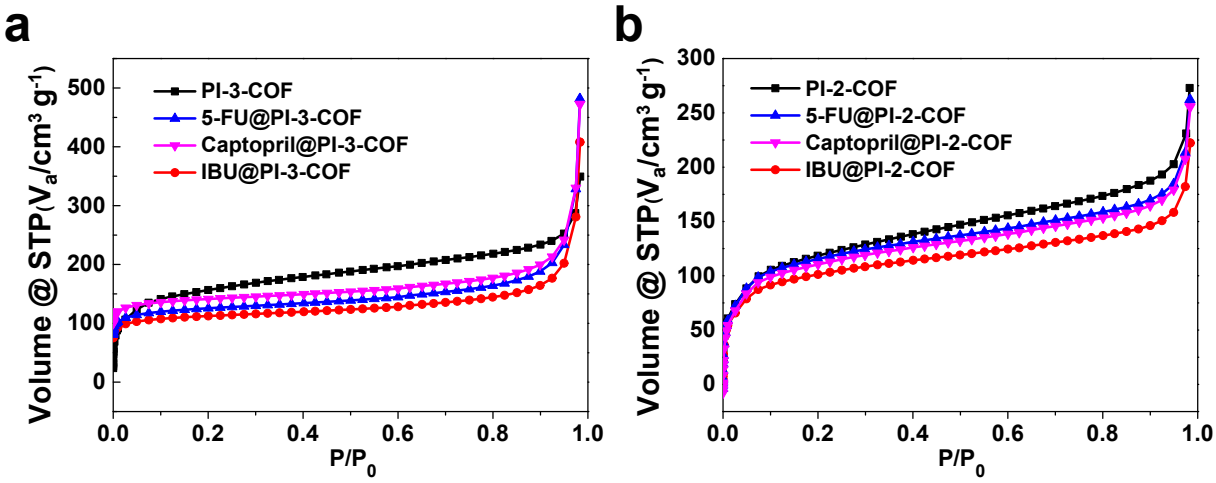


Figure S9. (a,b) N_2 adsorption isotherms of two COFs (black curve) and drug loaded COFs: 5-FU (blue curve), captopril (pink curve), and IBU (red curve) at 77 K.

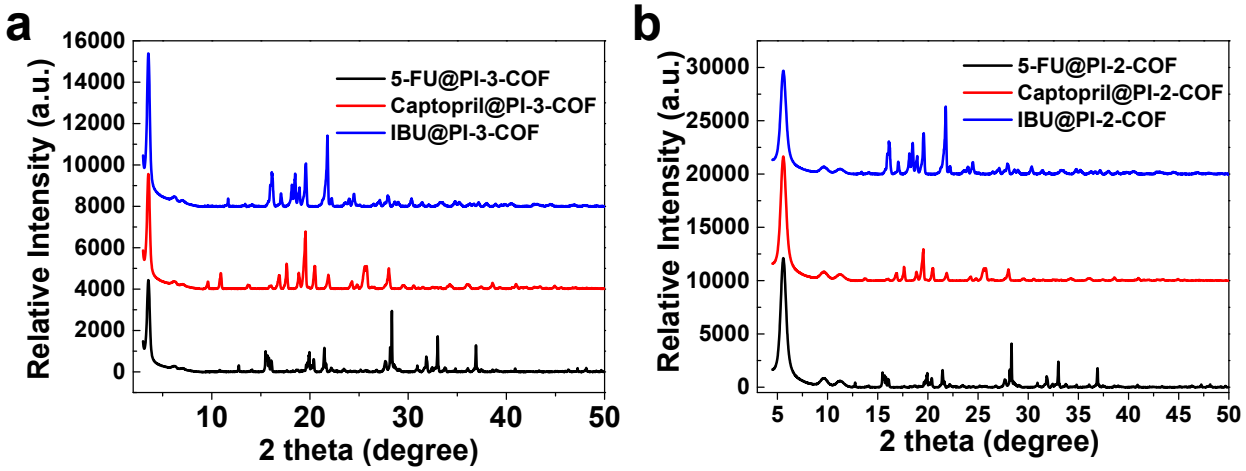


Figure S10. (a,b) PXRD patterns of drug loaded COFs: 5-FU (black curve), captopril (red curve), and IBU (blue curve).

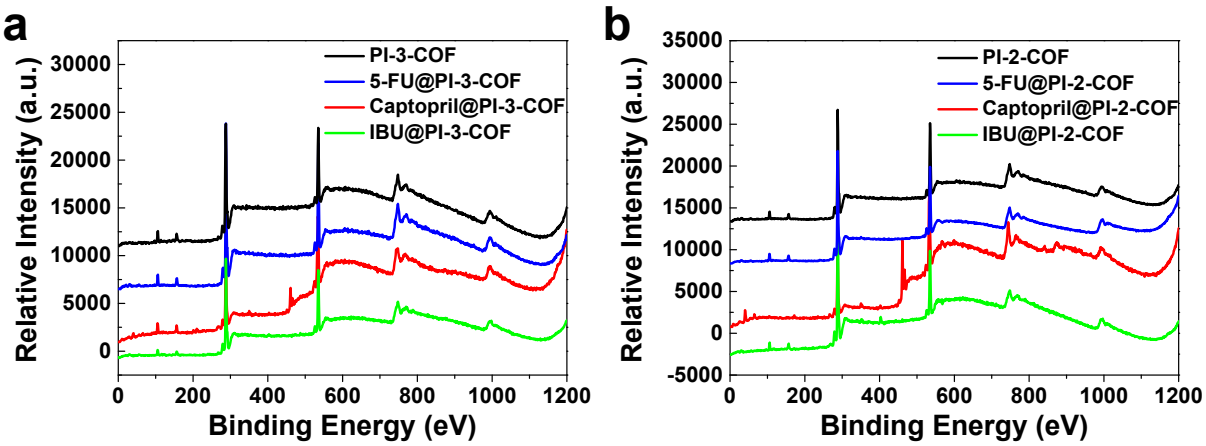


Figure S11. (a,b) XPS spectra of two COFs (black curve) and drug loaded COFs: 5-FU (blue curve), captopril (red curve), and IBU (green curve).

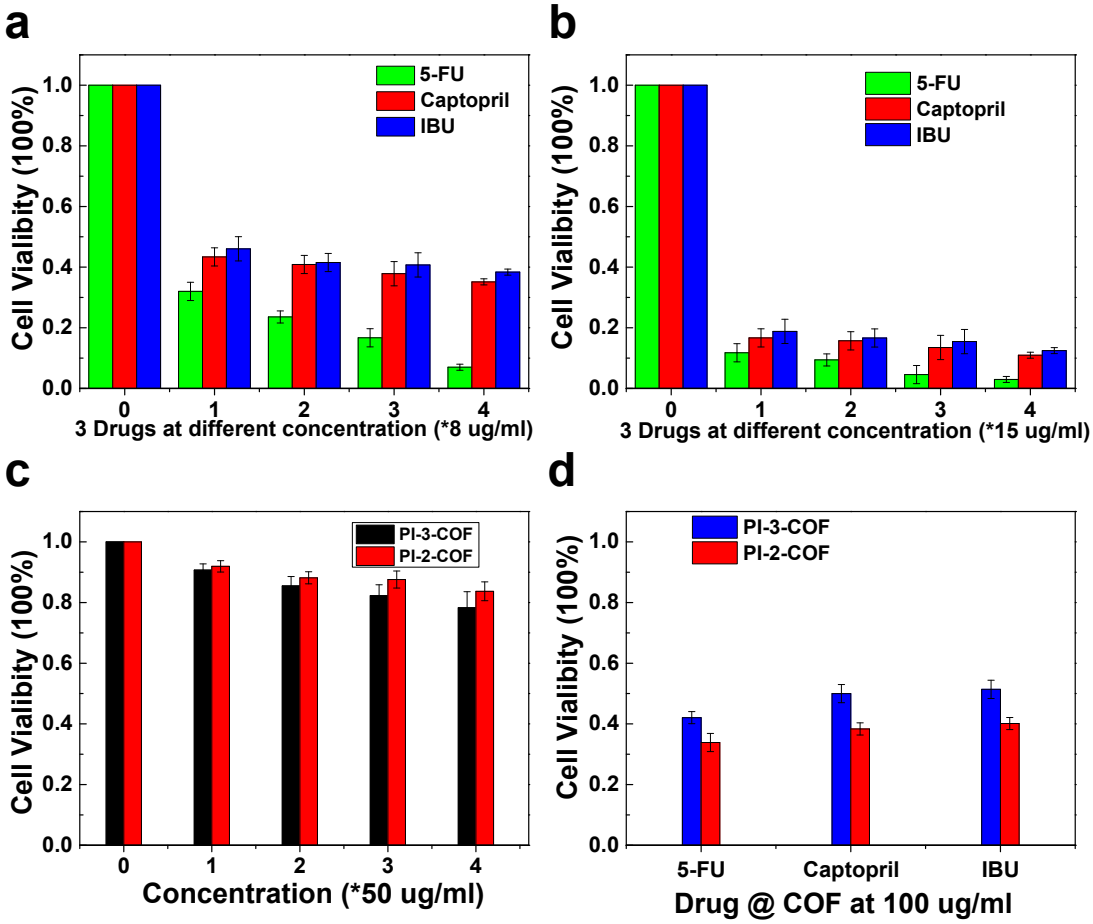


Figure S12. (a,b) *In vitro* cell cytotoxicity of three drugs incubated for 24 h, respectively. x axis represents the concentrations of drugs. The corresponding concentrations are 0, 8 μg,16 μg, 24 μg and 32 μg for (a), and 0, 15 μg, 30 μg, 45 μg and 60 μg for (b); (c) *In vitro* cell cytotoxicity of two COFs and their concentration range was 50~200 mg/mL; (d) *In vitro* cell cytotoxicity of drug loaded COFs against MCF-7 cells at 100 μg/mL for 24 h. The concentration of loaded drugs depends on the COF loading capacity, i.e., 16% and 30% for PI-3-COF and PI-2-COF, respectively.

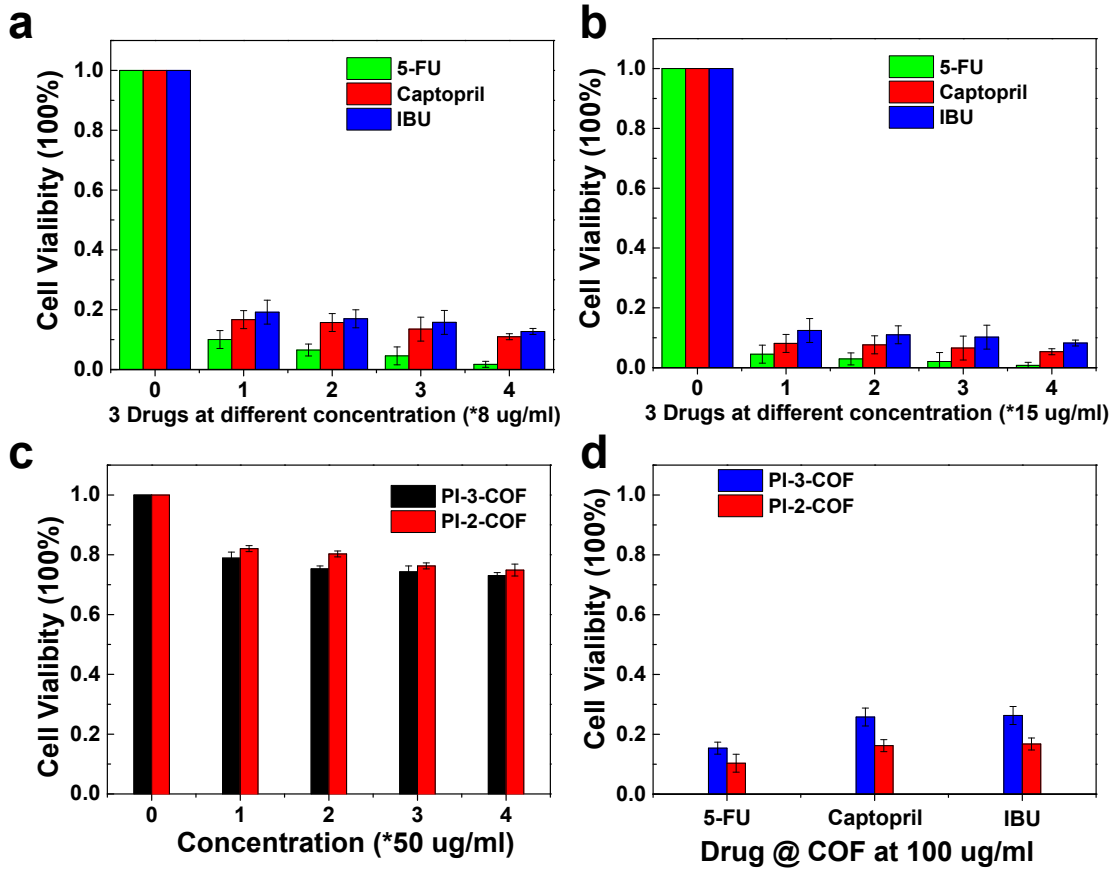


Figure S13. (a,b) *In vitro* cell cytotoxicity of three drugs incubated for 48 h, respectively. x axis represents the concentrations of drugs. The corresponding concentrations are 0, 8 μg,16 μg, 24 μg and 32 μg for (a), and 0, 15 μg, 30 μg, 45 μg and 60 μg for (b); (c) *In vitro* cell cytotoxicity of two COFs and their concentration range was 50~200 mg/mL; (d) *In vitro* cell cytotoxicity of drug loaded COFs against MCF-7 cells at 100 μg/mL for 48 h. The concentration of loaded drugs depends on the COF loading capacity, i.e., 16% and 30% for PI-3-COF and PI-2-COF, respectively.

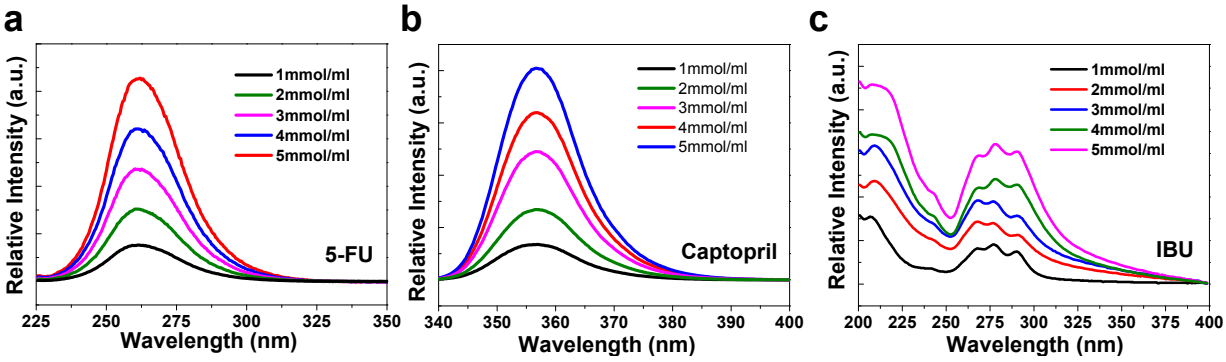


Figure S14. UV−vis spectra of three drugs in PBS: (a) 5-FU, (b) captopril and (c) IBU (green curve), where the tests were performed upon the concentration change from 1 mM/mL to 5 mM/mL.

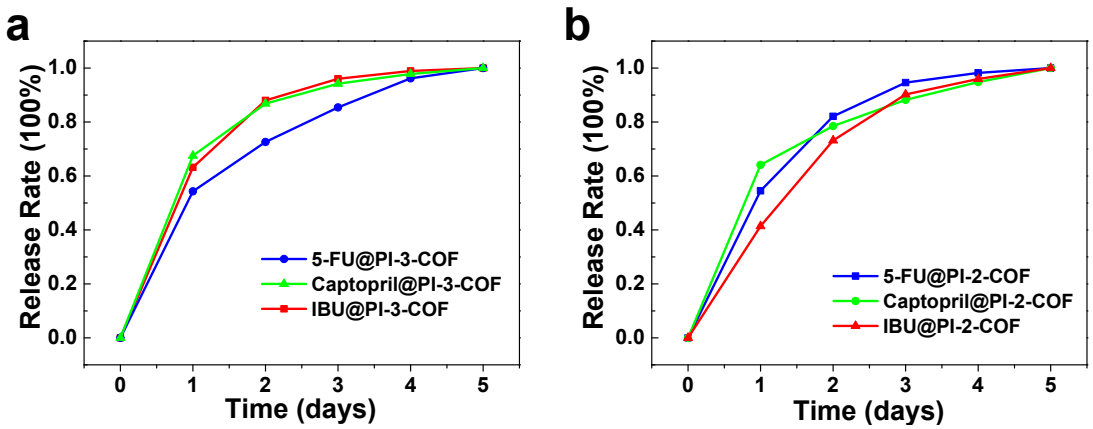


Figure S15. Release profiles of drug loaded (a) PI-3-COF and (b) PI-2-COF: 5-FU (blue curve), captopril (green curve) and IBU (red curve).

7. References

(S1) Q. Gao, L. Bai, Y. Zeng, P. Wang, X. Zhang, R. Zou and Y. Zhao, *Chem. Eur. J.*, 2015, **21**, 16818–16822.

(S2) Q. Gao, L. Bai, X. Zhang, P. Wang, P. Li, Y. Zeng, R. Zou and Y. Zhao, *Chin. J. Chem.*, 2015, **33**, 90-94.

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(S4) M. J. Frisch, G. W. Trucks, H. B. Schlegel, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, G. Scalmani, V. Barone, B. Mennucci, G. A. Petersson, H. Nakatsuji, M. Caricato, X. Li, H. P. Hratchian, A. F. Izmaylov, J. Bloino, G. Zheng, J. L. Sonnenberg, M. Hada, M. Ehara, K. Toyota, R. Fukuda, J. Hasegawa, M. Ishida, T. Nakajima, Y. Honda, O. Kitao, H. Nakai, T. Vreven, J. A., Jr. Montgomery, J. E. Peralta, F. Ogliaro, M. Bearpark, J. J. Heyd, E. Brothers, K. N. Kudin, V. N. Staroverov, R. Kobayashi, J. Normand, K. Raghavachari, A. Rendell, J. C. Burant, S. S. Iyengar, J. Tomasi, M. Cossi, N. Rega, J. M. Millam, M. Klene, J. E. Knox, J. B. Cross, V. Bakken, C. Adamo, J. Jaramillo, R. Gomperts, R. E. Stratmann, O. Yazyev, A. J. Austin, R. Cammi, C. Pomelli, J. W. Ochterski, R. L. Martin, K. Morokuma, V. G. Zakrzewski, G. A. Voth, P. Salvador, J. J. Dannenberg, S. Dapprich, A. D. Daniels, Ö. Farkas, J. B. Foresman, J. V. Ortiz, J. Cioslowski and D. J. Fox, Gaussian 09, Revision A.02, Gaussian, Inc., Wallingford CT, 2009.