Supplementary Information for

Antifouling binary liquid-infused membranes for biological sample pretreatment

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Experimental section

Materials

Phenobarbital was provided by Shanghai Chemical Reagent Co., Ltd (China). Methcathinone stock solution (1 mg/mL in methanol) was purchased from Cerilliant Co., Ltd (Round Rock, TX, USA). Trimethoprim stock solution (1 mg/mL in methanol), formic acid, 1-undecanol, 1-ethyl-2-nitrobenzene (ENB), tributyl phosphate (TBP), and bovine serum albumin were purchased from Aladdin Chemical Reagent Co., Ltd (Shanghai, China). Ammonium acetate, hydrochloric acid (HCl), and sodium hydroxide (NaOH) were supplied by Sinopharm Chemical Reagent Co., Ltd (China). Methanol with chromatographic purity grade was purchased from Tedia (Fairfield, OH, USA). A Milli-Q water purification system (Mollsheim, France) produced the ultrapure water that was used throughout this work. Luminol (2,3-aminophthalhydrazide)-H_2O_2 kit was provided by Phygene Biotechnology Co. Ltd (Fuzhou, China). Polypropylene (PP) membrane was supplied by Membrana (Wuppertal, Germany). Perfluoropolyether (PFPE) lubricant (Krytox GPL 100, DuPont) was purchased from the Chemours Company.

Preparation of the samples

Trimethoprim and methcathinone sample solutions (5 μg/mL) were prepared by diluting their stock solutions with 10 mM HCl aqueous solutions. Phenobarbital sample solution (5 μg/mL) was prepared by diluting the stock solution with 10 mM NaOH aqueous solution. Blood samples were prepared by mixing whole blood with 20 mM HCl or NaOH aqueous solutions containing 10 μg/mL trimethoprim, methcathinone, or phenobarbital, in which the volume ratios were 1:1.

Ethical Statement

Whole blood samples were obtained from the authors who were healthy and none of us had taken any medication around the time of sample collection. Venous blood (ca. 10 mL) was collected from the brachial vein of each person at each blood sampling time. The collected blood was stored in a -20 °C freezer and thawed for use before experiments.

This study was approved by the Ethics Committee of Tongji Medical College of Huazhong University of Science and Technology (Approval no. S064).

EME set-up and procedure

Self-made electromembrane extraction (EME) set-up was employed for the separation processes. The sample solution (0.8 mL) was loaded into a 2.0 mL PP centrifuge tube, which acted as the donor phase compartment. Meanwhile, organic solvent (10 μL) including undecanol (C11-OH), 1-ethyl-2-nitrobenzene (ENB), tributyl phosphate (TBP), and their mixture was used to adequately wet the PP membrane. Unless otherwise stated, the volume ratio of C11-OH and ENB (or TBP) was 1:1. The liquid-infused membrane was fixed at the bottom of a disposable 1-mL polyethylene Pasteur pipette while the acceptor solution was poured into the acceptor tube, which acted as the acceptor phase compartment. Afterward, two platinum wires (0.3 mm diameter, Xudong Instrument Co., Wuxi, China) were inserted into the sample and acceptor solutions, respectively, and were connected to an electrophoresis supply power (DYY-6C, Beijing LiuYi Instrument Factory, China). Finally, the sample and acceptor compartments were assembled together and the EME procedure was conducted under a voltage drive and 1000 rpm agitation. At the given extraction time, the sample and acceptor solutions were collected and quantitatively analyzed by high-performance liquid chromatography with an ultraviolet detector (HPLC-UV). Unless otherwise stated, the respective extraction time for water and blood samples is 30 and 20 min. To separate trimethoprim and methcathinone, positive and negative electrodes were connected to the sample and acceptor solutions, respectively. In contrast, a reversed voltage was appled to separate phenobarbital.

HPLC-UV analysis

An Agilent 1260 Infinity II system with a VWD detector G7114A was used for HPLC analysis. A Poroshell 120 EC-C18 column (4.6 mm × 150 mm, 4 μm packing) maintained at 40 °C was used for separation. Mobile phase A was water containing 5% methanol and 0.1% formic acid, and mobile phase B was methanolic solution containing 5% water and

0.1% formic acid. Chromatographic separation was performed with an isocratic elution (A:B = 60:40). The injection volume was 10 μL and the flow of mobile phase was 0.6 mL min^{-1}. The detection of phenobarbital and trimethoprim was conducted at 214 nm and the UV detector was operated at 240 nm to determine methcathinone.

Characterization

Porous structures of the PP membrane were observed by a field emission scanning electron microscope (TESCAN MIRA LMS). The samples were pretreated by Au-sputtered specimens to increase surface conductivity. Surface chemical compositions of the PP membrane were analyzed by X-ray photoelectron spectroscopy (PHI-Vesoprobe 5000 III). Contact angles, dynamic adhesive behaviors, and slippage of water and blood droplets were measured a JC2000D contact angle system (Shanghai Zhongchen, China). Infrared spectra were obtained using a Fourier transform micro-infrared spectrometer (ThermoFisher Nicolet IS50).

Luminol luminescent assay

Whole blood was mixed with ultrapure water at the same volume. Then, the PP membranes (5 mm × 5 mm) that were infused with C11-OH, ENB or TBP, and their mixture (v/v 1:1) were immersed in the blood sample for 20 min followed by washing with water. Next, the blood-fouling membranes were placed on a white ceramic plate, and 10 μL luminol solution diluted 1000 times with water was dropped on the membrane surfaces. Immediately, the surface fluorescence was observed using a smartphone (Huawei Mate 30 Pro) in dark.

Calculation of recovery

The recovery (R, %) of each analyte was calculated by the following equation:

$$R\ (\%) = \frac{n_A}{n_S} \times 100\% = \frac{C_A}{C_S} \times \frac{V_A}{V_S} \times 100\%$$

where n_S is the initial amount of analyte in the sample solution, n_A is the final amount of

analyte in the acceptor solution after the EME, C_S represents the initial concentration of analyte in the sample solution, and C_A indicates the final concentration of analyte in the acceptor solution after the EME. As the volumes of the sample (V_S) and acceptor (V_A) solutions were 0.8 and 0.2 mL, respectively, the equation can be expressed as follows:

$$R\ (\%) = \frac{C_A}{4C_S} \times 100\%$$

Theoretical calculation

Gauss16 was used for the optimization of the molecular structures and the calculation of the interaction energy. The structural optimization of all molecules was performed using the following methods: opt freq b3lyp/6-31+g(d,p) em=gd3bj. The calculation of interaction energy between molecules was conducted using the method b3lyp/6-31+g(d,p) counterpoise=2 em=gd3bj.

Supplementary Figures

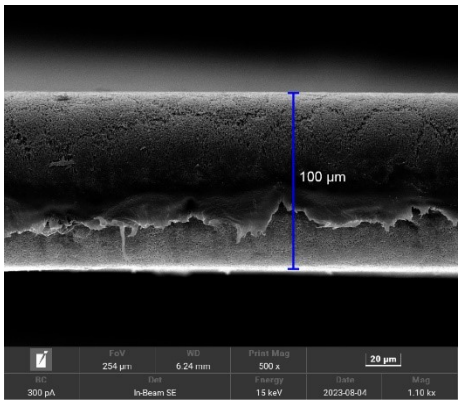

Fig. S1 Cross-section SEM image of the PP membrane. The thickness of the membrane is about 100 µm.

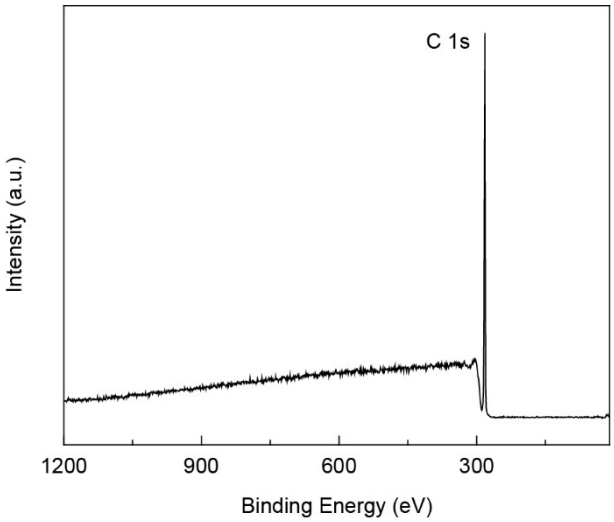

Fig. S2 C 1s XPS spectrum of the PP membrane.

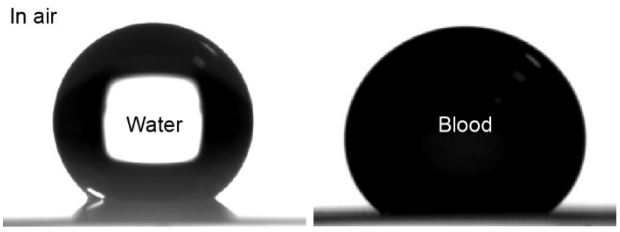

Fig. S3 Contact angles of water and blood droplets on the PP membrane in air.

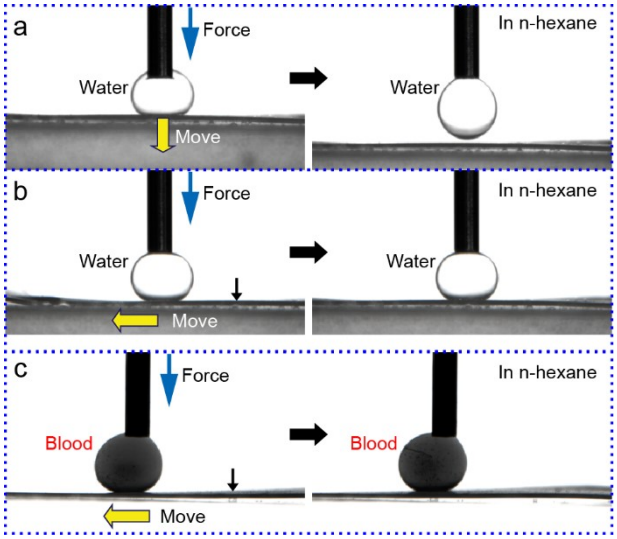

Fig. S4 Dynamic adhesive behaviors in n-hexane of water (a, b) and blood (c) droplets on the PP membrane at the vertical (a) and horizontal (b, c) directions.

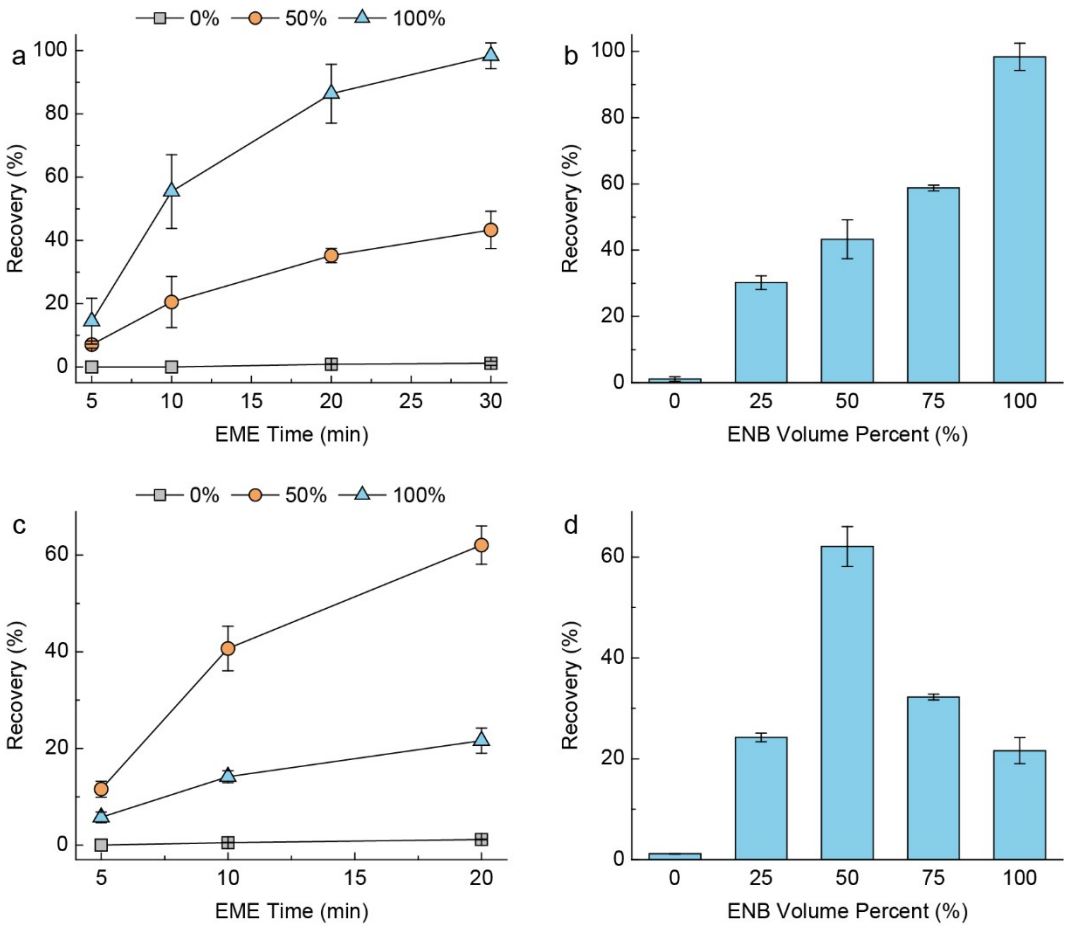

Fig. S5 Recoveries of trimethoprim separated from water (a, b) and blood (c, d) using the liquid-infused membranes. The infused liquid includes low polar solvent (C11-OH), specific solvent (ENB), and their mixture, in which the ENB volume percents are adjusted from 0 to 100%.

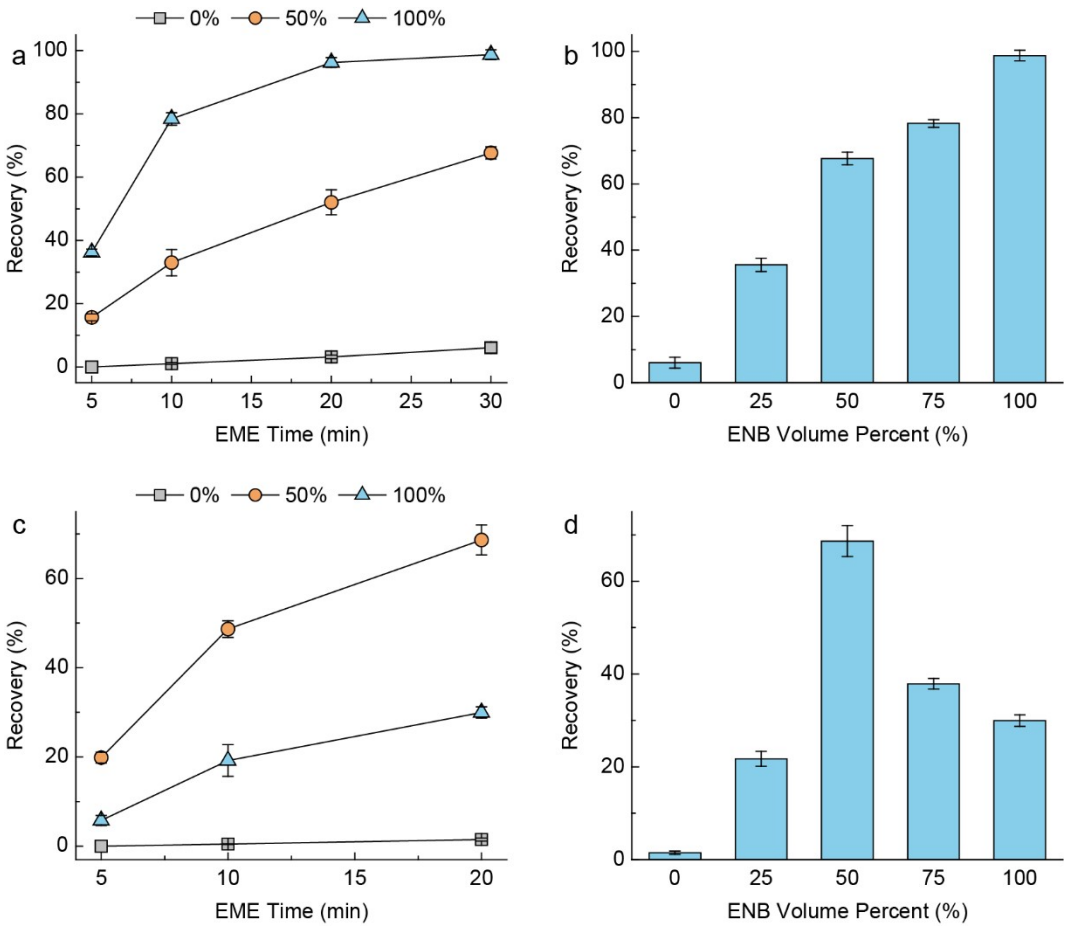

Fig. S6 Recoveries of methcathinone separated from water (a, b) and blood (c, d) using the liquid-infused membranes. The infused liquid includes low polar solvent (C11-OH), specific solvent (ENB), and their mixture, in which the ENB volume percents are adjusted from 0 to 100%.

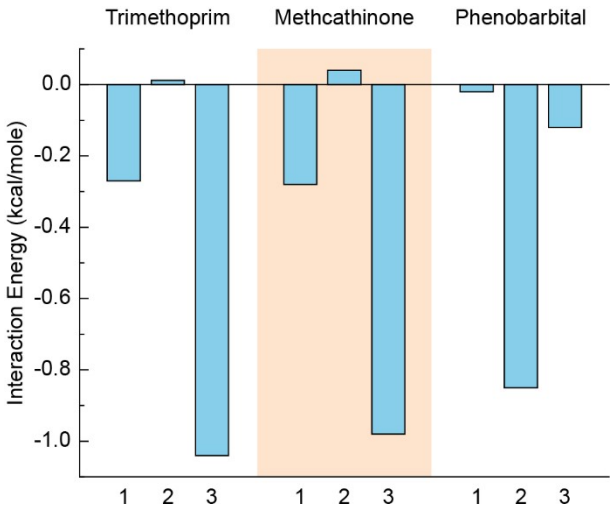

Fig. S7 Interaction energy of the infused liquid molecules including C11-OH (1), TBP (2), and ENB (3) with the model drug molecules such as trimethoprim, methcathinone, and phenobarbital via the theoretical calculation.

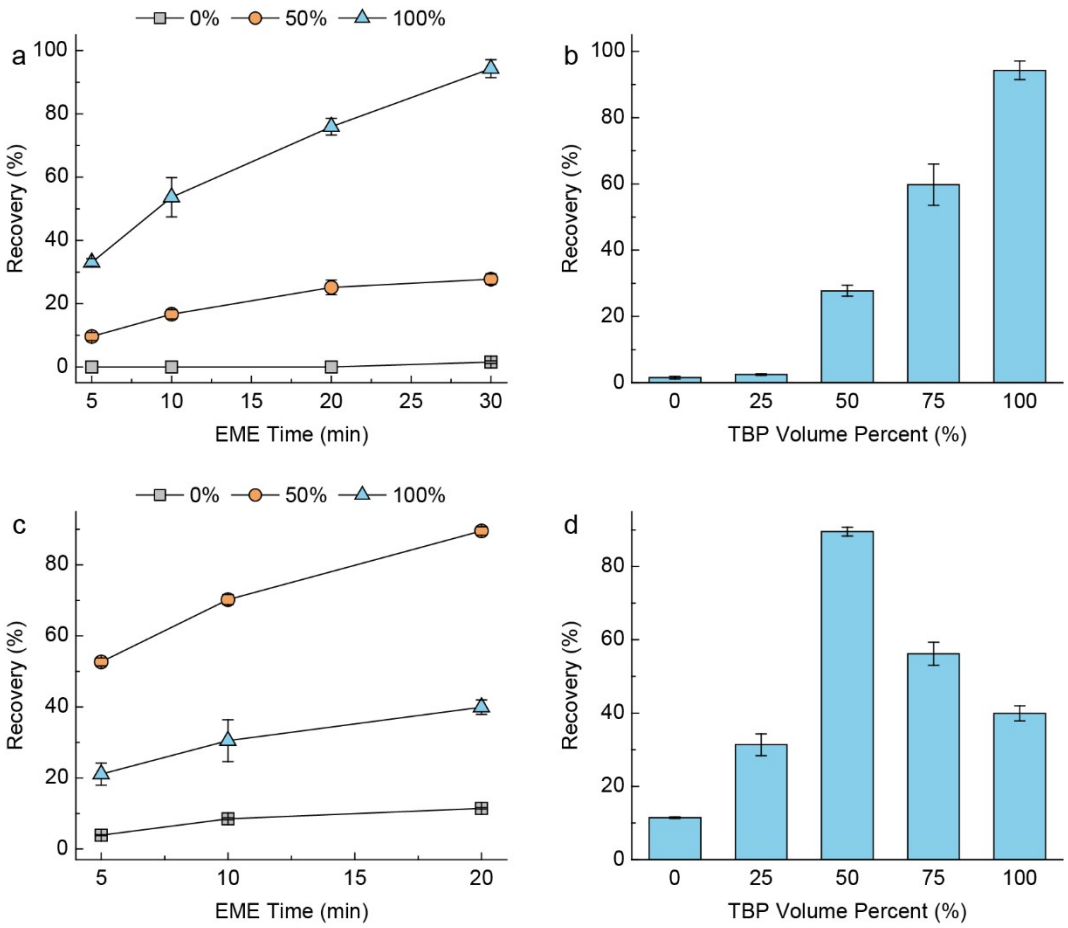

Fig. S8 Recoveries of phenolbarbital separated from water (a, b) and blood (c, d) using the liquid-infused membranes. The infused liquid includes low polar solvent (C11-OH), specific solvent (TBP), and their mixture, in which the TBP volume percents are adjusted from 0 to 100%.

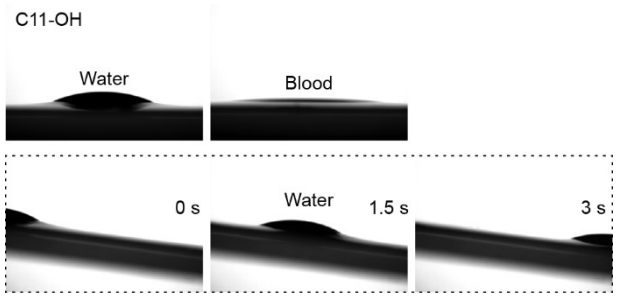

Fig. S9 (Upper) Water and blood contact angles of the C11-OH-infused membrane. (Below) Time-sequence photographs of slippage of a water droplet (about 10 μL) on the C11-OH-infused membrane under a tilted angle (about 10°).

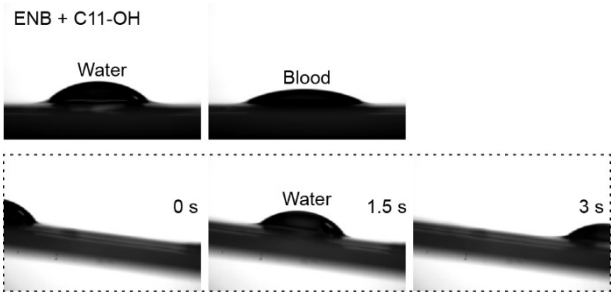

Fig. S10 (Upper) Water and blood contact angles of the membrane infused with C11-OH and ENB. (Below) Time-sequence photographs of slippage of a water droplet (about 10 μL) on the membrane infused with C11-OH and ENB under a tilted angle (about 10°).

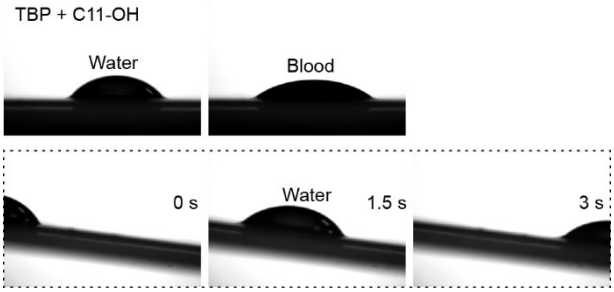

Fig. S11 (Upper) Water and blood contact angles of the membrane infused with C11-OH and TBP. (Below) Time-sequence photographs of slippage of a water droplet (about 10 μL) on the membrane infused with C11-OH and TBP under a tilted angle (about 10°).

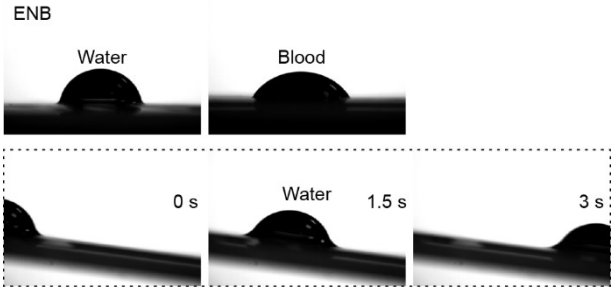

Fig. S12 (Upper) Water and blood contact angles of the ENB-infused membrane. (Below) Time-sequence photographs of slippage of a water droplet (about 10 μL) on the ENB-infused membrane under a tilted angle (about 10°).

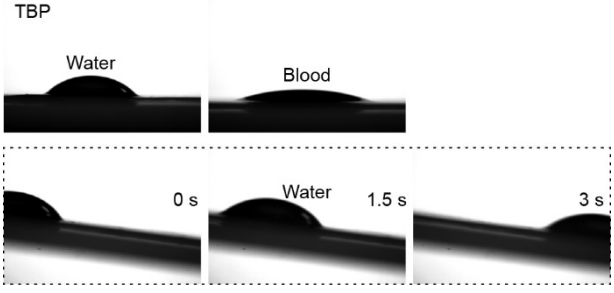

Fig. S13 (Upper) Water and blood contact angles of the TBP-infused membrane. (Below) Time-sequence photographs of slippage of a water droplet (about 10 μL) on the TBP-infused membrane under a tilted angle (about 10°).

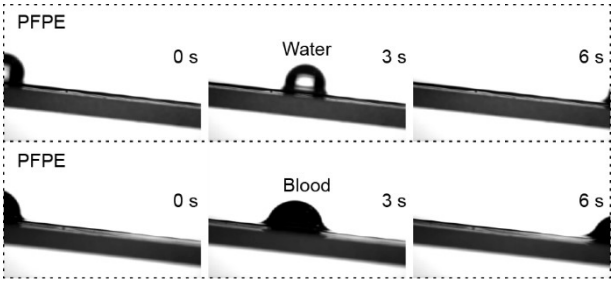

Fig. S14 Time-sequence photographs of slippage of water droplet or blood droplet (about 10 μL) on the PFPE-infused membrane under a tilted angle (about 10°).

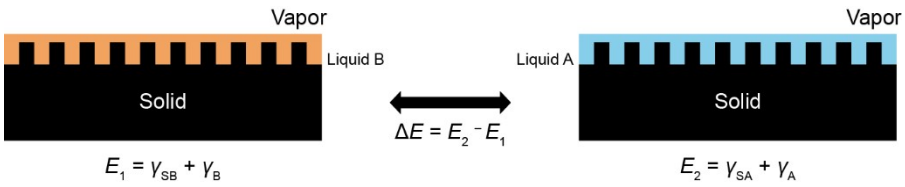

Fig. S15 Surface energy change after the fouling of liquid-infused membranes.

At the initial state, the total interfacial energy (E_1) per unit area of liquid-infused membranes can be expressed by the following equation: $E_1 = \gamma_{SB} + \gamma_B$, where γ_{SB} and γ_B represent the surface energies of solid-Liquid B interface and Liquid B-vapor interface, respectively. After fouling, the total interfacial energy (E_2) per unit area of liquid-infused membranes can be expressed by the following equation: $E_2 = \gamma_{SA} + \gamma_A$, where γ_{SA} and γ_A represent the surface energies of solid-Liquid A interface and Liquid A-vapor interface, respectively. Thus, the surface energy change $\Delta E = (\gamma_{SA} + \gamma_A) - (\gamma_{SB} + \gamma_B) = (\gamma_{SA} - \gamma_{SB}) + (\gamma_A - \gamma_B)$, which can be evolved to measurable values with use of the Young equation.

We have,

$$\Delta E = [(\gamma_{SV} - \gamma_A \cos\theta_A) - (\gamma_{SV} - \gamma_B \cos\theta_B)] + (\gamma_A - \gamma_B)$$

where γ_{SV}, θ_A, and θ_B are the surface energy of the solid-vapor interface, and the intrinsic contact angles of the flat solid surface for Liquid A and Liquid B, respectively.

After futher simplification, we have,

$$\Delta E = (\gamma_B \cos\theta_B - \gamma_A \cos\theta_A) + (\gamma_A - \gamma_B) = \gamma_B(\cos\theta_B - 1) + \gamma_A(1 - \cos\theta_A)$$

If the liquid-infused membranes have high antifouling ability, the State 2 always has a higher total surface energy than the State 1, that is, $\Delta E > 0$.

Thus, the infused liquid with lower surface tension (γ_B) can enhance the antifouling ability, such as PFPE and C11-OH. The repelled liquid with lower surface tension (γ_A) is easier to foul the membrane substrates, such as blood.

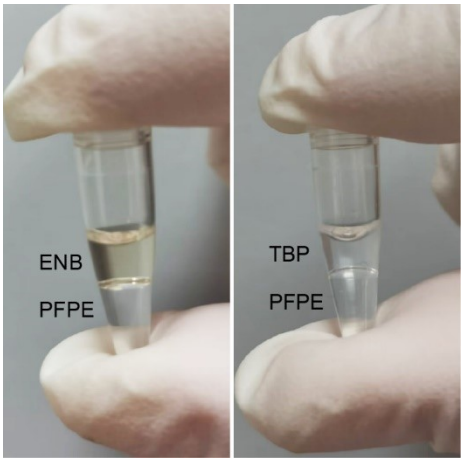

Fig. S16 Photographs of PFPE mixed with ENB or TBP at the same volume.

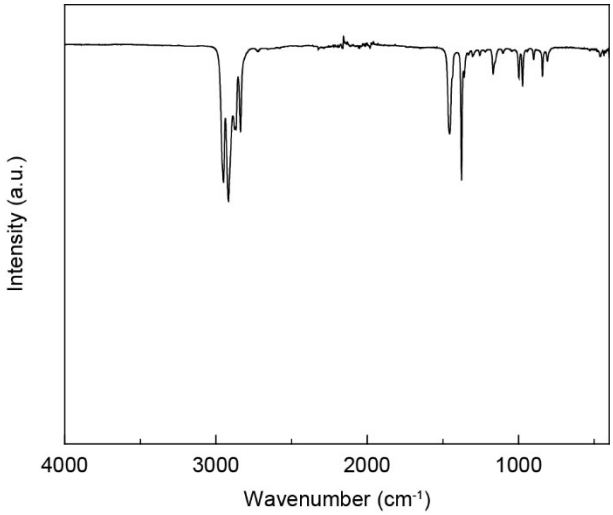

Fig. S17 IR spectrum of the original PP membrane.

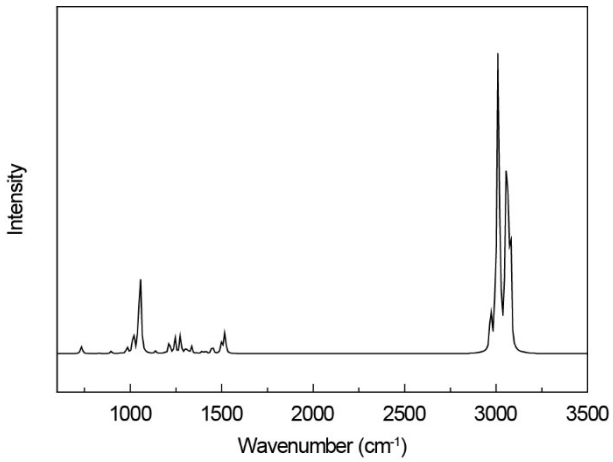

Fig. S18 IR spectrum of C11-OH via the theoretical calculation. The peak at 1057 cm^{-1} is attributed to the C-O stretching vibration of C11-OH.

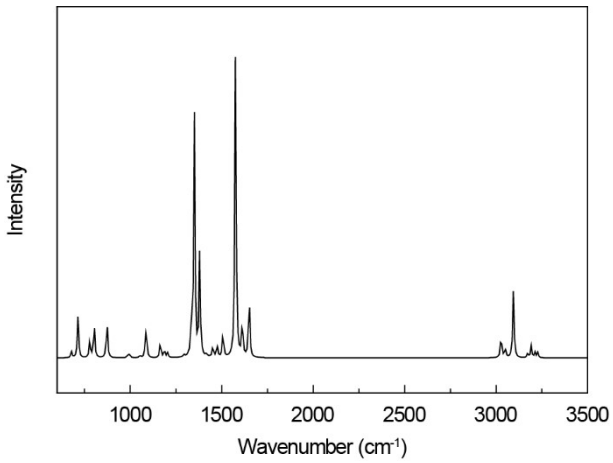

Fig. S19 IR spectrum of ENB via the theoretical calculation. The peak at 1530 cm^{-1} is attributed to the aromatic skeletal vibration of ENB.

Table S1. Physical properties of the used solvents.

Solvent	Viscosity (mPa.s)	Dielectric constant	Dipole moment	Surface tension (mN/m)
PFPE	-	-	-	17.1[a]
Undecanol	10.9	5.98	1.71	24.6[a]
Tributyl phosphate	11.1	8.34	3.07	31.7[b]
1-Ethyl-2-nitrobenzene	2.37	21.9	3.72	41.0[b]

[a]The data are obtained from Ref. S1. [b]The data are obtained from the Lange's Handbook of Chemistry.

References

S1 T.-S. Wong, S. H. Kang, S. K. Y. Tang, E. J. Smythe, B. D. Hatton, A. Grinthal and J. Aizenberg, *Nature*, 2011, **477**, 443.

Supplementary Movies

Movie S1 In n-hexane, dynamic adhesive behavior of a water droplet on the PP membrane at the vertical direction.

Movie S2 In n-hexane, dynamic adhesive behavior of a water droplet on the PP membrane at the horizontal direction.

Movie S3 In n-hexane, dynamic adhesive behavior of a blood droplet on the PP membrane at the vertical direction.

Movie S4 In n-hexane, dynamic adhesive behavior of a blood droplet on the PP membrane at the horizontal direction.

Movie S5 Slippage of a water droplet (about 10 μL) on the C11-OH-infused membrane under a tilted angle (about 10°).

Movie S6 Slippage of a water droplet (about 10 μL) on the membrane infused with C11-OH and ENB under a tilted angle (about 10°).

Movie S7 Slippage of a water droplet (about 10 μL) on the membrane infused with C11-OH and TBP under a tilted angle (about 10°).

Movie S8 Slippage of a water droplet (about 10 μL) on the ENB-infused membrane under a tilted angle (about 10°).

Movie S9 Slippage of a water droplet (about 10 μL) on the TBP-infused membrane under a tilted angle (about 10°).

Movie S10 Slippage of a blood droplet (about 10 μL) on the C11-OH-infused membrane under a tilted angle (about 10°).

Movie S11 Slippage of a blood droplet (about 10 μL) on the membrane infused with C11-OH and ENB under a tilted angle (about 10°).

Movie S12 Slippage of a blood droplet (about 10 μL) on the membrane infused with C11-OH and TBP under a tilted angle (about 10°).