Electronic Supplementary Information

Cell Adhesion and Proliferation in Chiral Pores Triggered by Polyoxometalates

Yingyi Ma,[a,b] Lei Shi,[a] Mengcheng Zhou,[a] Bao Li,[a] Zhijun Chen[a,] and LixinWu[a,]**

[a]State Key Laboratory of Supramolecular Structure and Materials, College of Chemistry, Jilin University, Changchun 130012, China

[b]College of Material Science & Engineering, Harbin University of Science and Technology, Harbin 150040, China

To whom correspondence should be addressed. Email:wulx@jlu.edu.cn

Experimental Section:

Materials. Polystyrene (PS, Mw: 349 kg mol^{-1}) was purchased from Acros Organics. Didodecyldimethylammonium bromide (DDDA·Br) and Propidium Iodide (3,8-Diamino-5-[3-(diethylmethylammonio)propyl]-6-phenylphenanthridinium diiodide, PI) were the product of Aladdin and used as received. Branched poly(ethylenimine) (PEI, Mw: 750 kDa, 50%wt in water) was received from Sigma Aldrich. Bovine serum albumin (BSA) was purchased from Beijing Biosynthesis Biotechnology Company. $[(CH_3)_2NH_2]_{15}\{[\alpha\text{-}P_2W_{15}O_{55}(H_2O)]Zr_3(\mu_3\text{-}O)(H_2O)(L\text{-}tartH)[\alpha\text{-}P_2W_{16}O_{59}]\}$ (L-Zr-P_2W_{15}), $[(CH_3)_2NH_2]_{15}\{[\alpha\text{-}P_2W_{15}O_{55}(H_2O)]Zr_3(\mu_3\text{-}O)(H_2O)(D\text{-}tartH)[\alpha\text{-}P_2W_{16}O_{59}]\}$ (D-Zr-P_2W_{15}) and $Na_{16}P_4W_{30}Zn_4(H_2O)_2O_{112}$ ($P_4W_{30}Zn_4$) were freshly prepared according to the literature procedures, respectively.[1,2] Distilled water (Milli-Pore 18.2 MΩ/cm) and dichloromethane of analytical grade (Beijing Chemical Works) are used in the experiments. Tryptone (Oxoidltd Basingstoke Hampshire England) and resin (Shanghai Yiyang Instrument Inc) are used in the experiments.

Preparation of DDDA decorated honeycomb-pattered film. 30 mg of PS and 0.5 mg of DDDA·Br were dissolved in 5 mL of dichloromethane. Then, the prepared organic solution was mixed with distilled

water in a controlled volume ratio of 20:1. The mixed solution was sonicated 10 min to disperse distilled water in organic phase, obtaining a translucent white microemulsion. After filtering with 220 nm filter, 20 μL of the filtrate was cast onto a clean glass under ambient environment to get the desired DDDA modified honeycomb patterned film.

Fabrication of chiral and racemic pores. Negatively charged polyoxometalate (POM) were assembled onto the inner surface of the pores through electrostatic interaction using positively charged PEI as the connecting layer. Briefly, L-Zr-P_2W_{15}, D-Zr-P_2W_{15}, racemic POM (L-Zr-P_2W_{15}:D-Zr-P_2W_{15}=1:1) and PEI were dissolved in distilled water to obtained 0.5 mg mL^{-1} solution, respectively. And each bilayer was assembled by contacting with POM solution for 30 min, rinsing three times with water for 5 min, drying in the oven at 40°C for 20 min, then contacting with PEI solution for 30 min, and again rinsing with distilled water and drying. This cycle was repeated six and a half times to prepare the chiral and racemic pores. In addition, the film was also assembled with $P_4W_{30}Zn_4$ and PEI to construct achiral porous film as a control.

Cell adhesion and culture in chiral and racemic pores. Cell adhesion studies were performed by dropping the stable-phase *E.coli-BL21* cells (4 mL) onto POM modified film at 30°C. After 20 min of incubation, the film was rinsed four times with phosphate buffered saline (PBS) buffer to remove unspecific binding of the cells to the film. Then the film containing adherent cell were incubated in 4 ml of fresh YPD media for 16 h at 30°C. The sample was fixed with 4% formaldehyde before the scanning electron microscope (SEM) characterization. The film was rinsed with PBS for three times followed by fixation with 4% formaldehyde and stained with PI, respectively for the confocal laser scanning microscopy (CLSM) characterization.

Measurements. The optical photographs were taken with an olympus BX-51 optical microscope (OM). Scanning electron microscopy (SEM) images and X-ray energy-dispersive spectroscopic (EDX) analysis were acquired on a JEOL FESEM 6700F electron microscope. Acquisition of confocal laser scanning microscopy (CLSM) images was obtained on an OLYMPUS FLUOVIEW FV1000. Data analysis was carried out with the software FV10-ASW Version. Quartz crystal microbalance (QCM) measurements were taken with a KSV QCM-Z500 using quartz resonators with both sides coated with Ag (f_0= 9 MHz). A MicroCal ITC_{200} (GE) was used to measure the ITC curves at room temperature. The optical density (OD) at 600 nm was measured using the Eppendorf Biophotometer Plus.

Characterization Data:

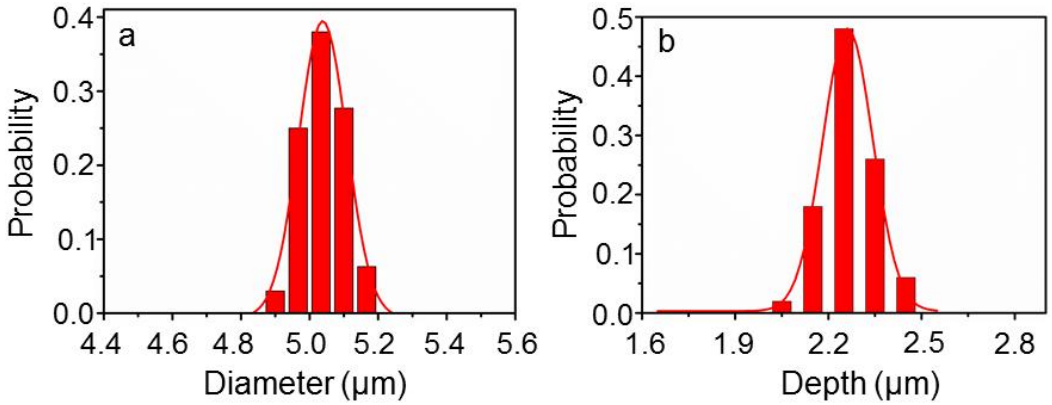

Fig. S1 Histograms and Gaussian fit curves of the pore size: (a) diameter and (b) depth (in total, 300 pores were counted) in Figure 1a.

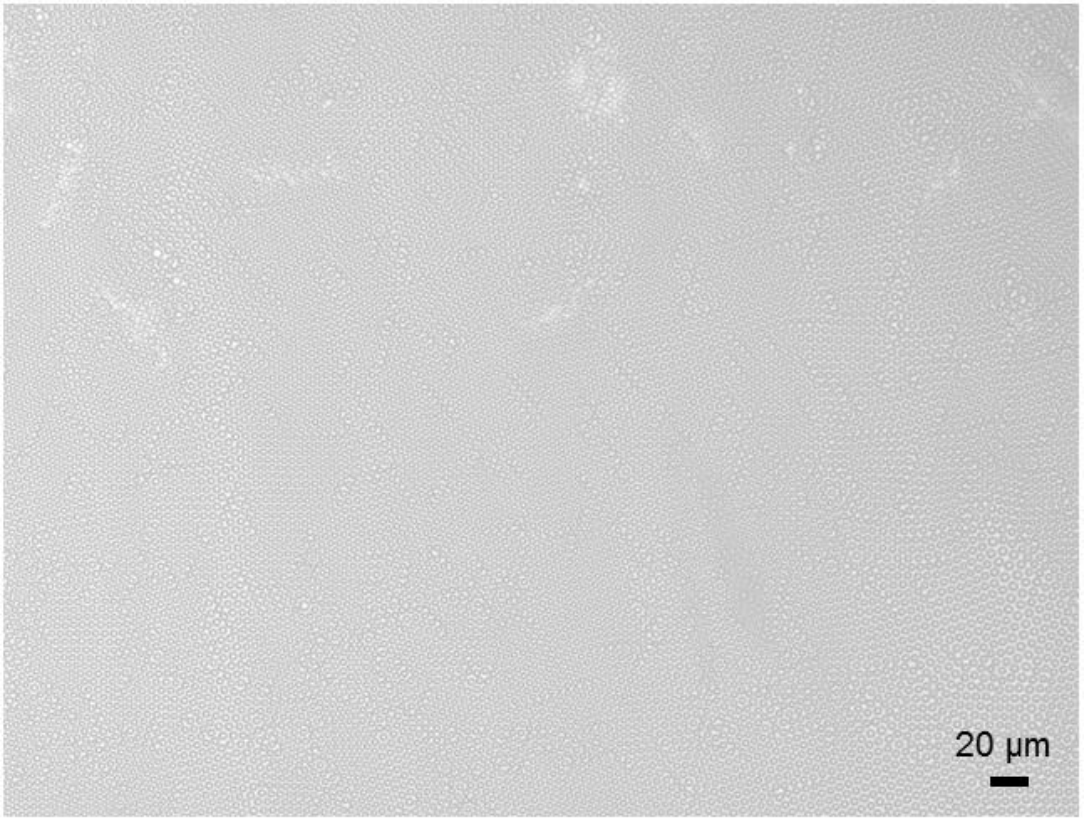

Fig. S2 Optical image of the DDDA decorated honeycomb-patterned film.

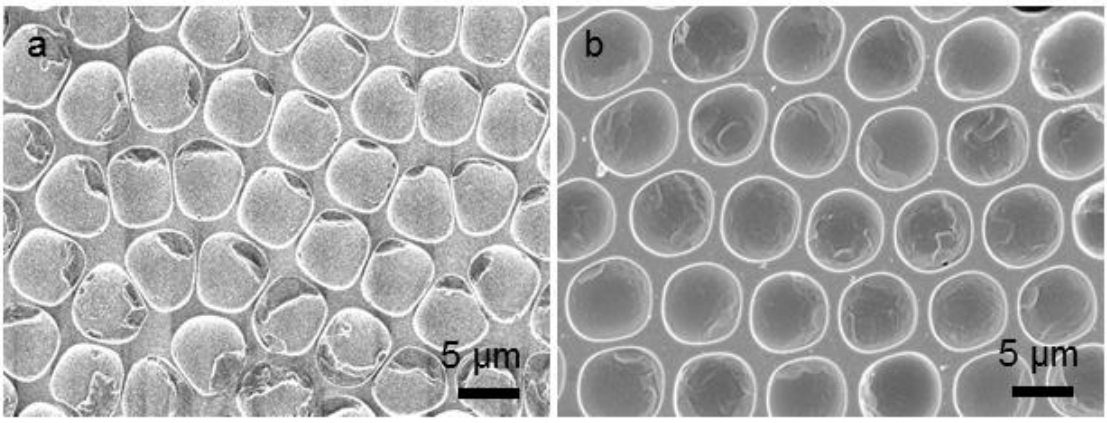

Fig. S3 SEM images of *E.coli* cells adsorbed on an achiral POM ($P_4W_{30}Zn_4$) modified porous film (a) before and (b) after proliferation for 16 h.

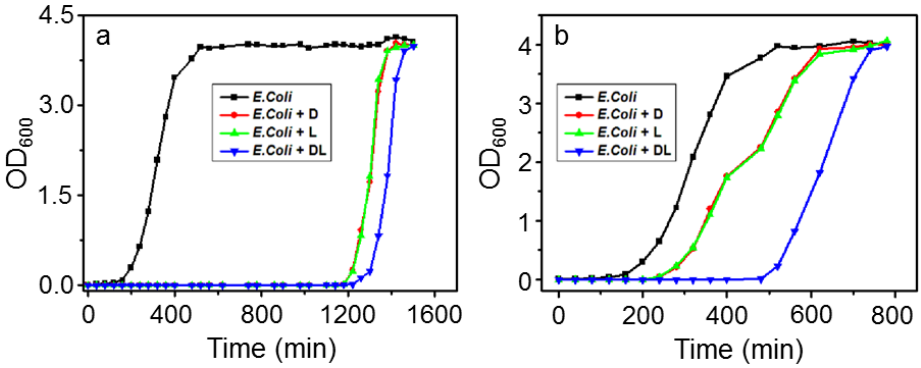

Fig. S4 Growth curves based on the OD_{600} at 600 nm for *E.coli* cells cultured in D-, L- and racemic porous film for (a) 15 min and (b) 16 h, the measurements were initiated immediately after cells released from the porous film.

Table S1. Thermodynamic parameters for the binding of BSA to D-, L- and racemic POM according to ITC measurements.

Sample	n	K (M^{-1})	ΔH (kcal mol)	ΔG (kcal mol)	ΔS (kcal mol)
D	0.363±0.010	$(4.13±1.24)×10^6$	−49.29±2.54	−8.84	−0.138
L	0.368±0.008	$(4.92±1.34)×10^6$	−47.08±1.99	−8.97	−0.130
D,L	0.372±0.012	$(2.91±0.84)×10^6$	−51.29±2.95	−8.78	−0.145

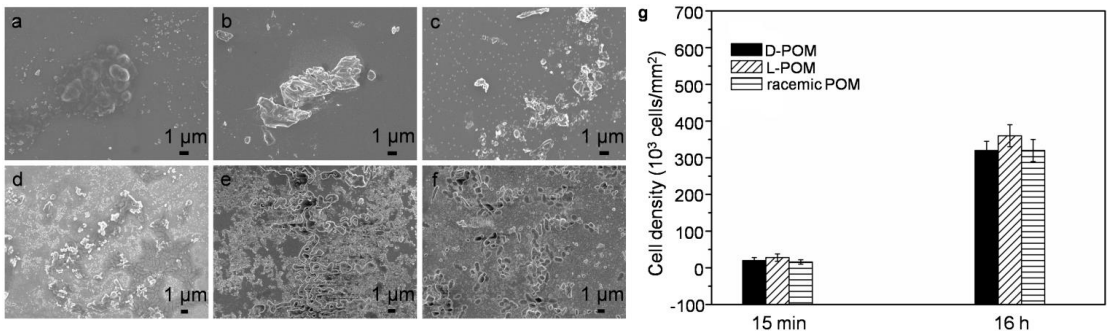

Fig. S5 SEM images of *E.coli* cells before (a, b, c) and after (d, e, f) proliferation on D- (a, d), L- (b, e) and racemic (c, f) POM assembled flat films, and (g) the number of *E.coli* cells adhered to different POM assembled flat films.

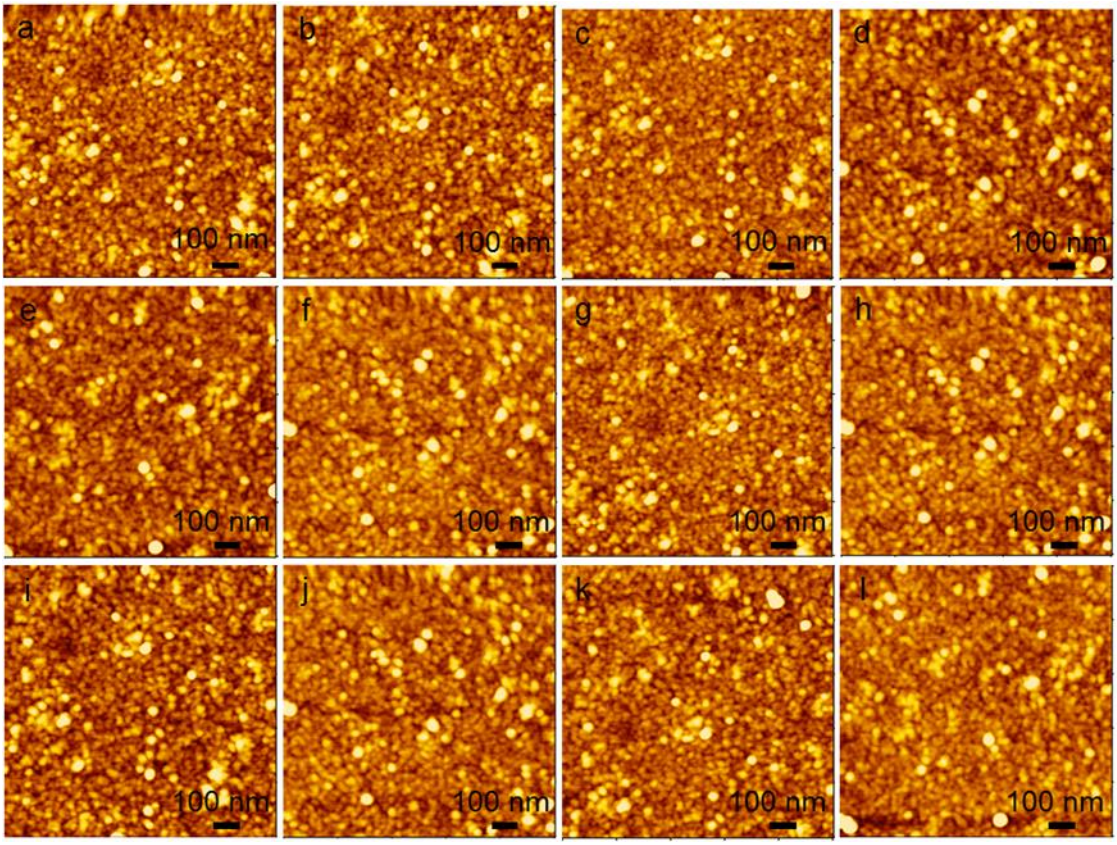

Fig. S6. AFM images of D- (a, d, g, j), L- (b, e, h, k) and racemic (c, f, i, l) POM assembled film with different deposition cycles: (a, b, c) one, (d, e, f) two, (g, h, i) four, and (j, k, l) six.

Table S2. Ra and Rz values for the D-, L- and racemic POM assembled film with different deposition cycles.

Sample (deposition cycle)	Ra[a]	Rz[b]
D (one)	0.3537	6.473
L (one)	0.3659	6.582
DL (one)	0.3546	6.542
D (two)	0.3665	6.644
L (two)	0.3511	6.410
DL (two)	0.3677	6.709
D (four)	0.3543	6.638
L (four)	0.3910	6.445
DL (four)	0.3511	6.443
D (six)	0.3543	6.561
L (six)	0.3721	6.498
DL (six)	0.3734	6.808

a: Ra is the arithmetic average ruoughness;

b: Rz is the mean roughness depth.

References:

1. X. K. Fang, T. M. Anderson, C. L. Hill, *Angew. Chem. Int. Ed*. **2005**, *44*, 3540; *Angew. Chem.* **2005**, *117*, 3606.

2. R. G. Finke, M. W. Droege, P. J. Domaille, *J. Am. Chem. Soc*. **1984**, *106*, 2737.