

Supporting Information for

Silver@Silica nanopollens modified membranes for wastewater treatment in membrane bioreactors: Limited adverse effects on microorganisms and compelling antifouling properties

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This supporting information has ten pages, containing three associated text sections (namely Section S1, Section S2 and Section S3), four figures (Fig. S1, Fig. S2, Fig. S3 and Fig. S4) and one table (Table S1).

Section S1 Synthesis and characterization of Ag@silica nanopollens.

Silica nanopollens (SNPs) were synthesized according to Stöber method [1]. Briefly, resorcinol (0.15 g) and formaldehyde (37 wt%, 0.21 mL) were dissolved in the mixture of ammonia aqueous solution (28 wt%, 3.0 mL), deionized water (10 mL) and ethanol (70 mL). The mixture was stirred for 6 h at room temperature, and then 0.6 mL of TEOS was added into the solution and stirred for 8 min, followed by addition of resorcinol (0.4 g) and formaldehyde (37 wt%, 0.56 mL) and then 2 h stirring. The products were collected by centrifugation, ethanol-washing and drying at 50°C. Finally, SNPs were obtained after calcination at 550 °C for 5 h in air. Ag-SiO₂ synthesis method was used to fabricate Ag@silica nanopollens [2]. In brief, silver nitrate (8.83 mmol) was added into silica nanopollens slurry (50 mmol in 200 mL water) with ammonia solution (105 mmol) as a catalyst, and then the mixture was stirred for 6 h at room temperature. The harvested products were purified with deionized water and then dried at room temperature for 2 h.

The microstructures of SNP and ASNP samples were observed using field emission transmission electron microscopy (TEM) (JEOL, JEM- 2100F, Japan). The surface areas of the powders were determined using the Brunauer-Emmett-Teller (BET) (Micromeritics ASAP 2460, USA) with N₂ as adsorbate gas.

Section S2 Information about the pilot-scale MBR that the sludge inoculums were collected

In this study, the sludge inoculums were collected is an anoxic/oxic (A/O) MBR fed with municipal wastewater in Quyang municipal wastewater treatment plant (WWTP) of Shanghai, China. The reactor had an oxic zone with the effective volume of the oxic zone and anoxic zone were 30 L and 22 L, respectively. Four flat-sheet membranes with 0.63 m² total effective filtration (Zizheng Environment Inc., China, 0.2 µm of pore size) were immersed in the oxic zone. Membrane flux was maintained at 15 (L/m²·h). The hydraulic time (HRT) and sludge retention time (SRT) were 6.6 h and 60 d, respectively.

Section S3 DNA extraction, PCR amplification and 16S rRNA gene pyrosequencing.

DNA of all the samples were extracted using E.Z.N.A.® Soil DNA kit (Omega Bio-Tek, Inc., Norcross, GA, U.S.) and the quality of DNA was assessed using a 1.0 % (w/v) agarose gel electrophoresis. PCR amplifications of 16S rRNA were conducted by the following primers: 338F (5'-ACTCCTACGGGAGGCAGCA-3') and 806R (5'-GGACTACHVGGGTWTCTAAT-3').

PCR mixture consist of 20 µL reaction volume containing 4 µL of 5 × FastPfu Buffer, 2 µL of 2.5 mM dNTPs, 0.8 µL of each primer (5 µM), 10 ng of template DNA and 0.4 µL of FastPfu Polymerase (TransGen AP221-02, Beijing, China). Reactions were carried out on a ABI GeneAmp® System 9700 (Perkin-Elmer Applied Biosystems, Foster City, CA, U.S.) under the following thermocycling steps: 95°C for

2 min, followed by 25 cycles at 95°C for 30 s, 55°C for 30 s and 72°C for 30 s, and a final extension at 72°C for 5 min and 10°C until halted by user. PCR products of three replicates were combined and purified using AxyPrep DNA gel extraction kit and Tris_HCl for each sample. Amplicons from the two samples were mixed at the equal concentrations.

FLASH was employed to overlap the paired-end reads followed by quality filtering conducted on Trimmomatic pipeline using the Sliding Window approach [3]. Sequences were clustered into different operational taxonomic units (OTUs) by threshold of 0.97 using UPARSE pipeline (version 7.1, <http://drive5.com/uparse/>) [4] and chimeric sequences were identified and removed using UCHIME. Rarefaction curves, Chao1 (<http://www.mothur.org/wiki/Chao>), Shannon index (<http://www.mothur.org/wiki/Shannon>) and the Good's Coverage (<http://www.mothur.org/wiki/Coverage>) were calculated by MOTHUR (version v.1.30.1 http://www.mothur.org/wiki/Schloss_SOP#Alpha_diversity) according to the standard procedures [5]. The taxonomy of each 16S rRNA gene sequence from OTUs was analyzed using RDP Classifier (<http://rdp.cme.msu.edu/>) and unite pipeline respectively with a set confidence threshold of 80% [6].

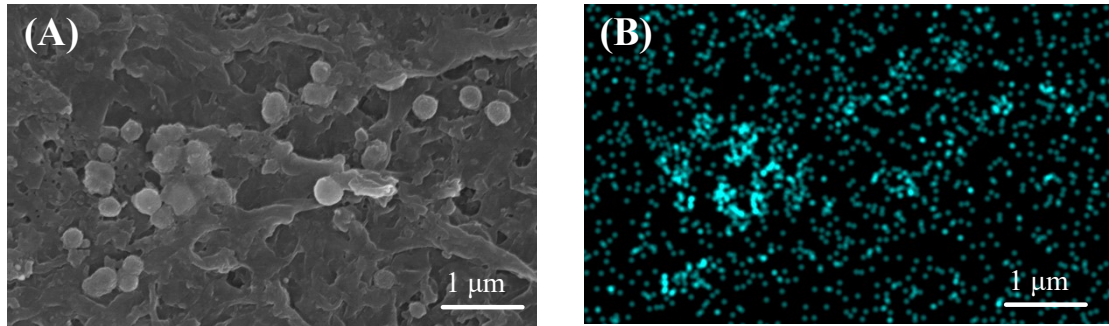


Fig. S1. (A) SEM image of MA and (B) its EDX mapping of silica

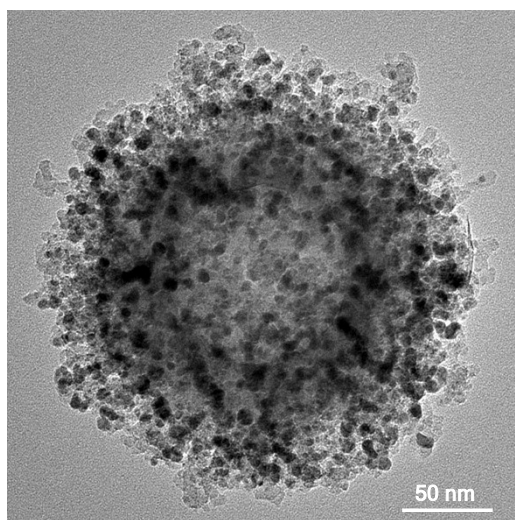


Fig. S2. TEM image of Ag@silica nanopollens

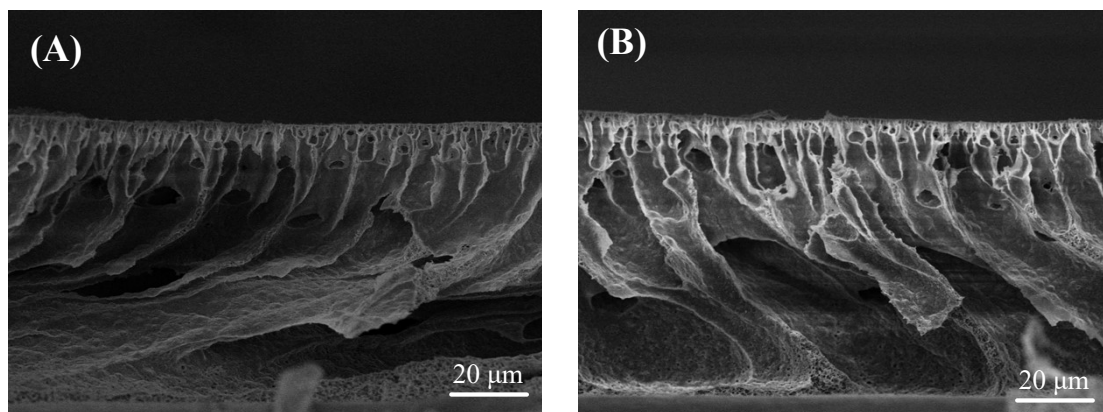


Fig. S3. Cross-section morphologies of (A) M0 and (B) MA membrane.

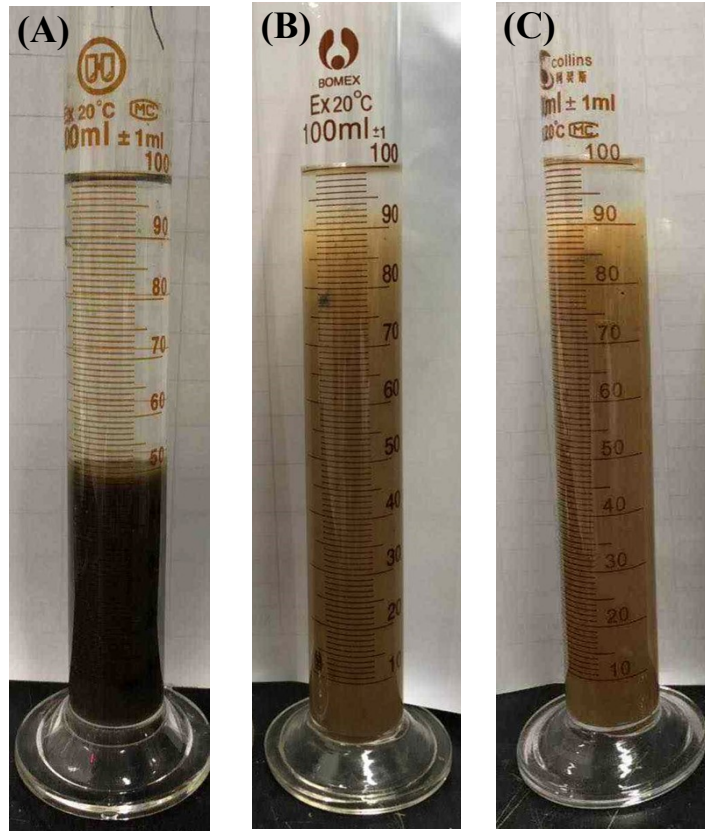


Fig. S4. Images on the sedimentation performance of sludge in MBR system. (A) inoculated sludge and (B) sample from R1 and (C) R2 at 75 d under 30 min settling time

Table S1 The composition of synthetic wastewater

Substance	Concentration (mg/L)
CH ₃ COONa	450
NH ₄ Cl	150
K ₂ HPO ₄ •3H ₂ O	52
CaCl ₂	11.5
MgSO ₄	12
NaHCO ₃	12
FeSO ₄ •7H ₂ O	10
NaCl	800

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