

Supplementary

# Borneol-Modified Chitosan Coating with Antibacterial Properties via Layer-by-Layer Strategy

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## Methods

### Materials

Methyl iodide and 4-pyridinecarboxaldehyde were purchased from Aladdin Co. (Shanghai, China). Tryptic soy agar (TSA) was bought from Beijing Aubergine Co (Beijing, China). The microbial strains were obtained from the Chinese Industrial Culture Strain Bank.

### Scanning Electron Microscopy (SEM)

The surfaces of GCBP-modified quartz plates were observed using SEM (Hitachi S-4700, Japan).

### Minimum Inhibitory Concentration (MIC)

The MIC test was based on the appropriate procedures outlined in Refs [1, 2]. Serial two-fold dilutions ranging from 128  $\mu\text{g/mL}$  to 0.125  $\mu\text{g/mL}$  were prepared in a 96-well plate (100  $\mu\text{L}$ ), with a bacterial concentration of  $10^6$  CFU/mL (100  $\mu\text{L}$ ), for determining the MIC value in TSA broth. The control consisted solely of inoculated broth and was then incubated for 24 hours at 37°C. The visual turbidity of the wells was observed both before and after incubation to confirm the MIC value.

### Preparation of 4-formyl-1-methylpyridin-1-ium

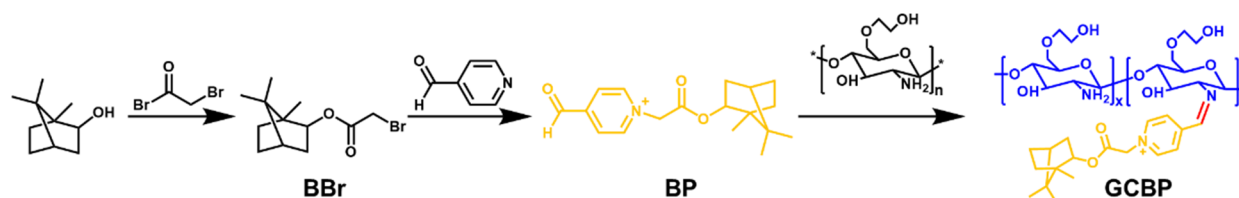
0.4 mL of methyl iodide and 0.4 mL of 4-pyridinecarboxaldehyde were dissolved in 2 mL of toluene, respectively. The mixture was degassed under an  $\text{N}_2$  atmosphere and kept overnight to form a precipitate (4-formyl-1-methylpyridin-1-ium). The product was washed twice with toluene and dried for further use.  $^1\text{H}$  NMR (400 MHz,  $\text{DMSO-d}_6$ )  $\delta$  10.20 (s, 1H), 9.22 (d, 2H), 8.47 (d, 2H), 4.43 (s, 3H).

### Inhibition zone experiment

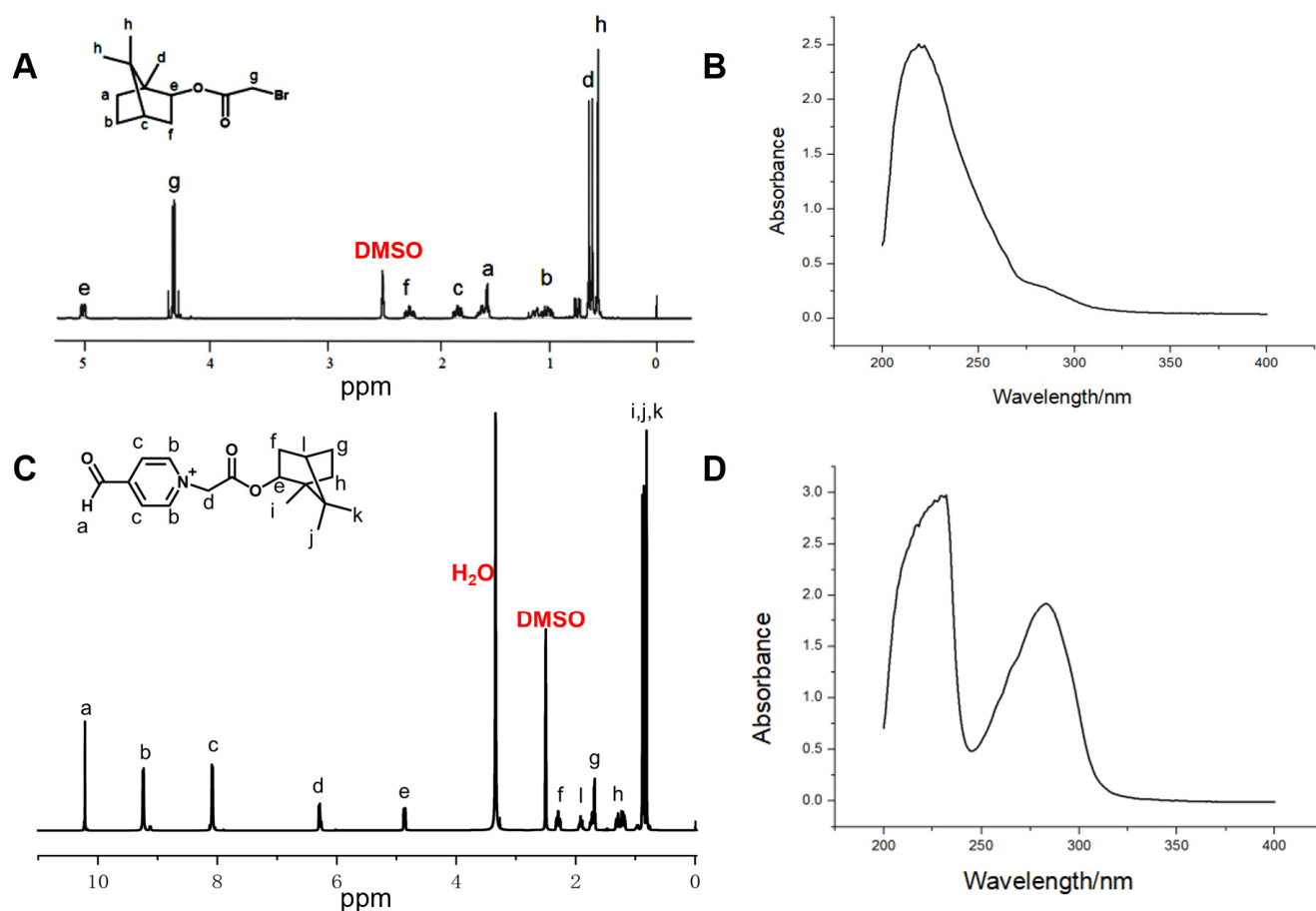
100  $\mu\text{L}$  of bacterial suspension with a concentration of  $10^6$  CFU/mL was added onto TSA agar, then spread evenly with a spreader to ensure the bacterial suspension was uniformly distributed on the agar surface. The GCBP-modified quartz plates were placed onto the agar surface, then incubated at 37°C for 24 hours.

### Verification of the fracture of the Schiff base

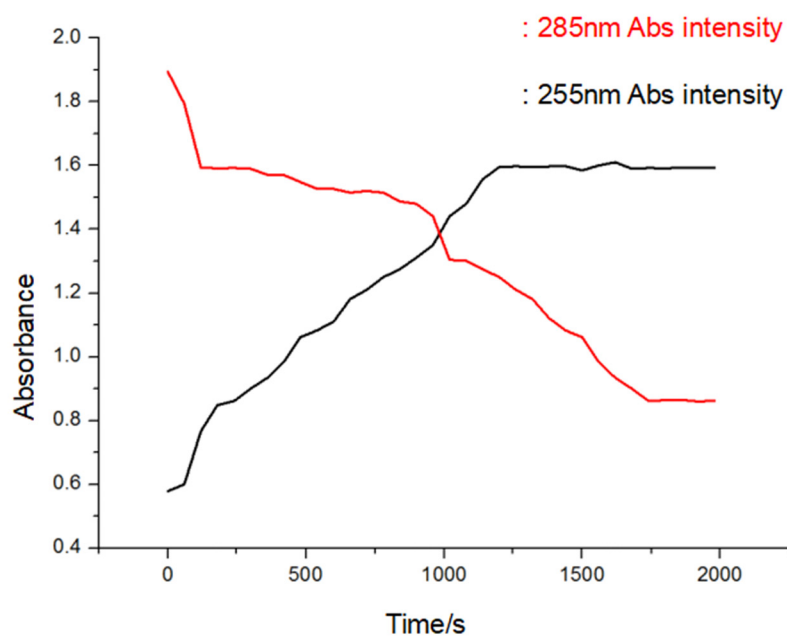
After the inhibition zone experiment, 4-layer GCBP-modified quartz plates were washed twice with water, dried, and subjected to UV-vis testing to observe changes in signal peaks.



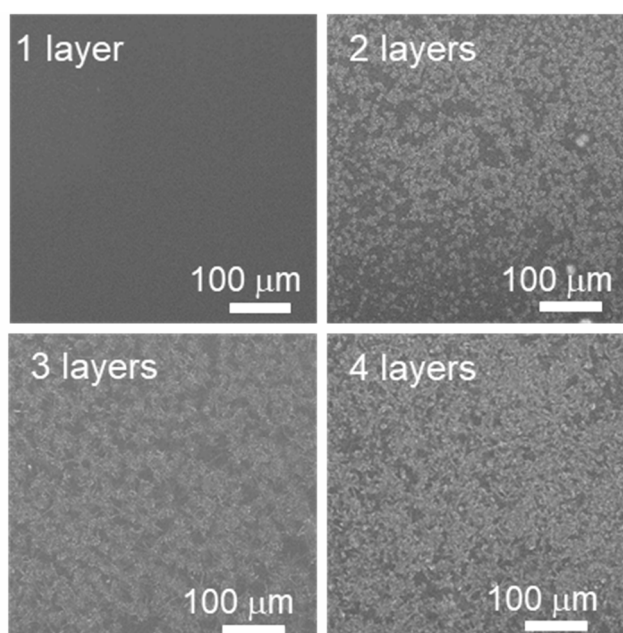
**Figure S1.** Procedures for the preparation of glycol chitosan-pyridylcarboxaldehydeborneol (GCBP).



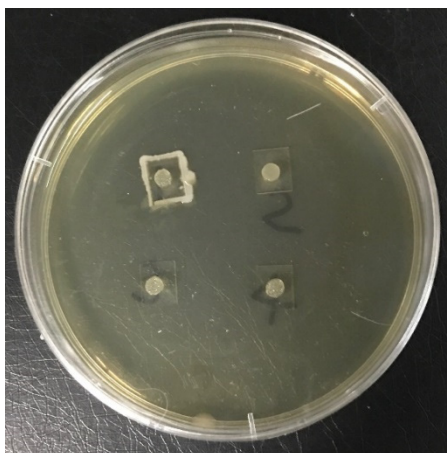
**Figure S2.** (A)  $^1\text{H}$  NMR spectra of BBr, (B) UV-vis spectra of BBr, (C)  $^1\text{H}$  NMR spectra of BP, and (D) UV-vis spectra of 4-pyridinecarboxaldehyde.



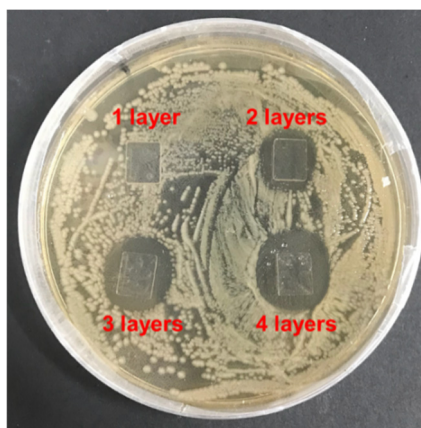
**Figure S3.** Time-dependent UV-vis absorbance changes at 255 nm and 285 nm during the formation of BP.



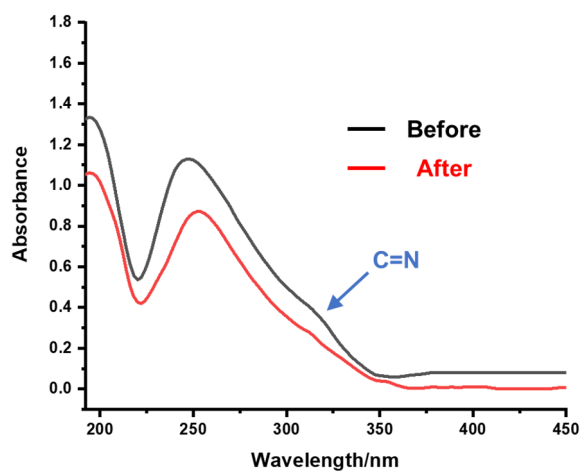
**Figure S4.** Scanning electron microscope (SEM) images of the 1-4 layer GCBP-modified quartz plates.



**Figure S5.** Prison break experiment of GCBP quartz plates against *E. coli*.



**Figure S6.** Inhibition zone experiment of 1-4 layer GCBP-modified quartz plates against *E. coli*.



**Figure S7.** UV-vis spectra of a 4-layer GCBP-modified quartz plate before (black line) and after (red line) the inhibition zone experiment.

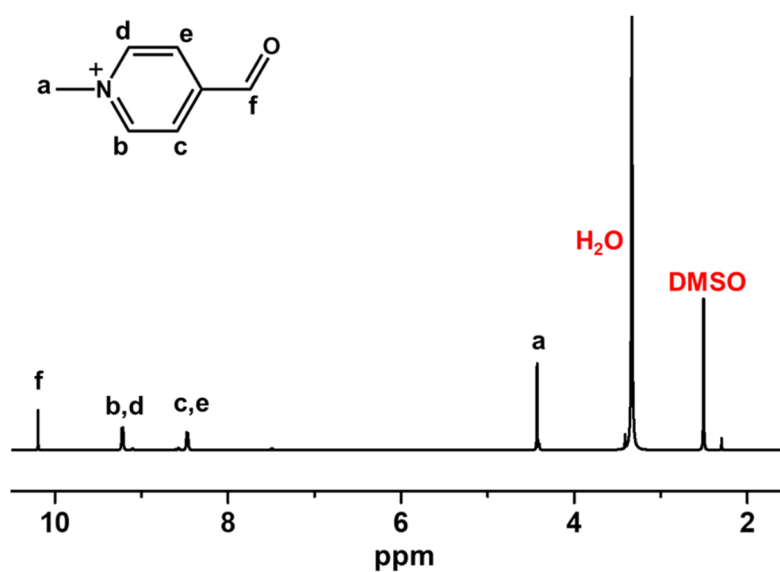


Figure S8.  $^1\text{H}$  NMR spectra of 4-formyl-1-methylpyridin-1-ium.

**Table S1.** MIC results of GC, 4-Pyridinecarboxaldehyde, BP, and 4-formyl-1-methylpyridin-1-ium

Bacteria	MIC ( $\mu\text{g/mL}$ )			
	GC	4-Pyridinecarboxaldehyde	BP	4-formyl-1-methylpyridin-1-ium
<i>E. coli</i>	64	128	32	128

## References

1. Mueller, J.H. and J. Hinton, A protein-free medium for primary isolation of the Gonococcus and Meningococcus, *Proc. Soc. Exp. Biol. Med.* **1941**, 48(1), 330-333.
2. CLSI. Performance Standards for Antimicrobial Susceptibility Testing, 25th Edition; Clinical and Laboratory Standards Institute (CLSI), Wayne, PA, USA 2015; pp 158-176.