## Supplementary Information

## Hollow Mesoporous Carbon Nanocarriers for Vancomycin Delivery:

## Understanding the Structure-Release Relationship for Prolonged

## **Antibacterial Performance**

Yusilawati Ahmad Nor,<sup>a</sup> Hongwei Zhang,<sup>a</sup> Swasmi Purwajanti,<sup>a</sup> Hao Song,<sup>a</sup> Anand Kumar,<sup>a</sup> Yue

Wang,<sup>a</sup> Neena Mitter,<sup>b</sup> Donna Mahony,<sup>b</sup> Chengzhong Yu\*a

<sup>a</sup>Australian Institute for Bioengineering and Nanotechnology, The University of Queensland,

Brisbane, QLD 4072, Australia

<sup>b</sup>Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, Brisbane, QLD 4072, Australia



Figure S1 TEM image of microporous hollow carbon (MicroHC) nanospheres.



Figure S2 Nitrogen absorption-desorption isotherm profiles of MicroHC nanospheres before and after loading of Van.



Figure S3 UV adsorption measurement of the remaining concentration of Van in the solution after absorption by hollow carbon nanoparticles. The absorbance at 281 nm was used to calculate the amount of Van in the supernatant before and after absorption.



Figure S4 Van uptake during 24 h adsorption with initial Van concentration of 1 mg/ml (a) Van adsorption isotherm on MHCs.



Figure S5: OD growth curves of *S. epidermidis* (a) and *E. coli* (b) after treatment at 24 h with the MHC6b (100  $\mu$ g ml<sup>-1</sup>) in LB broth and PBS at 37 °C.



Figure S6 Colloidal stability of MHC6b-Van in PBS and DMEM media with 10% serum for 3 days. The particle sizes of MHC6b-Van were measured as a function of time by DLS analysis.



Figure S7 Cell viability of HEK 293T cells treated with different concentrations of MHC6b.

Human embryonic kidney (HEK 293T) cells were maintained in Dulbecco's Modified Eagle Medium (DMEM) supplemented with fetal calf serum (10%), L-glutamine (2%), penicillin (1%), streptomycin (1%) in 5% CO<sub>2</sub> at 37 °C. The medium was routinely changed every 2 days. The cytotoxicity of MHC6b in HEK 293T cells was tested by the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) method. HEK 293T cells were seeded in a 96-well cell culture plate with a density of  $6 \times 10^3$  cells per well. After incubation for 24 h, MHC6b were added into the cells with different concentrations (10-200 µg/mL). After 48 h, 20 µL of MTT solution (5 mg/ml) was added into each well with 200 µL of medium. The plate was then incubated in the culture oven for 4 h. After removing medium, 100 µL of DMSO was added into each well, followed by shaking for 15 min. To avoid the interference from carbon nanoparticles in absorbance reading, the plate was centrifuged and the supernatant was transferred into a new 96-well plate. Then absorbance readings were measured at a wavelength of 540 nm using a Synergy HT microplate reader. The cells incubated in the absence of particles were used as the control. All the experiments were performed in triplicates for each group. The statistical data were shown as mean± (SD).



Figure S8 UV adsorption curve measuring the concentration of FITC in the solution before and after absorption by the MHC6b nanoparticles (a) and FITC-MHC6b attachment on the *E.coli* surface after 2 h culture. Inset image showing high density of nanoparticles attached on the bacterial surface.

Sample ID	Average size (nm)		PDI <sup>a</sup>
	TEM	DLS	
MHC6a	293	340	0.26
MHC13	219	280	0.14
MHC6b	218	236	0.24
MicroHC	197	240	0.30

Table S1: Particle size of the nanoparticles measured by TEM and DLS

<sup>a</sup>Polydispersity index

Table S2: MIC of the Van against gram positive and gram negative bacteria in LB broth after 24 h incubation at 37  $^{\circ}$ C

Bacteria strains	S. Epidermidis	E .coli
MIC (µg/mL)	2	300