## **Supporting Information**

## Hierarchical microspheres with macropores fabricated from chitin as 3D cell culture

Xiaojuan Su<sup>1, 3#</sup>, Mengtian Tan<sup>2#</sup>, Bo Duan<sup>1, 3</sup>, Jie Cai<sup>1, 3\*</sup>, Wei Jiang<sup>2, \*</sup>, Lina Zhang<sup>1, 3\*</sup>

<sup>1</sup> College of Chemistry and Molecular Sciences, Wuhan University, Wuhan 430072,

China

<sup>2</sup> Department of Biological Repositories, Zhongnan Hospital of Wuhan University;

Medical Research Institute, School of Medicine, Wuhan University, Wuhan 430072,

China

<sup>3</sup> Hubei Engineering Center of Natural Polymers-based Medical Materials, Wuhan University, Wuhan 430072, China

Correspondence to:

zhangln@whu.edu.cn (L. Zhang), caijie@whu.edu.cn (J. Cai), jiangw.mri@whu.edu.cn (W. Jiang)



**Figure S1.** SEM images of chitosan microspheres at low magnification (a) and high magnification(b).



**Figure S2.** SEM images of chitin microspheres with chitosan template microspheres of different diameters at low magnification (a1, b1, c1) and high magnification (a2, b2, c2).



Figure S3. SEM images of chitin microspheres at low magnification (a) and high

magnification(b).



**Figure S4.** Schematic diagram of compressive test of the chitin microspheres and PCMS with the FTIR Infrared compression mold (a); Typical compressive stress – strain curves of chitin microspheres and PCMS with different pore size (b).



**Figure S5.** Nitrogen adsorption and desorption and Barrett–Joyner–Halendar (BJH) pore size distribution (b) of the chitin microspheres (a, b) and PCMS (c, d).



Figure S6. Growth evaluation of hESCs cultured on chitin and chitosan.



**Figure S7.** Bright field and live/dead assay fluorescent image of hESCs cultured on the PCMS with different pore diameters (130-140  $\mu$ m(a), 90-100  $\mu$ m(b), 60-70  $\mu$ m(c)).



**Figure S8.** Bright field and DAPI staining image of hESCs cultured on the PCMS for 7 day.