

## Supporting Information

# New BMSC-laden gelatin hydrogel *in situ* forming by dual-enzymatic cross-linking accelerates dermal wound healing

Minghao Yao,<sup>†§\*</sup> Junni Zhang,<sup>†§</sup> Feng Gao,<sup>†</sup> Yihao Chen,<sup>†</sup> Shanshan Ma,<sup>†</sup> Kun Zhang,<sup>†</sup>

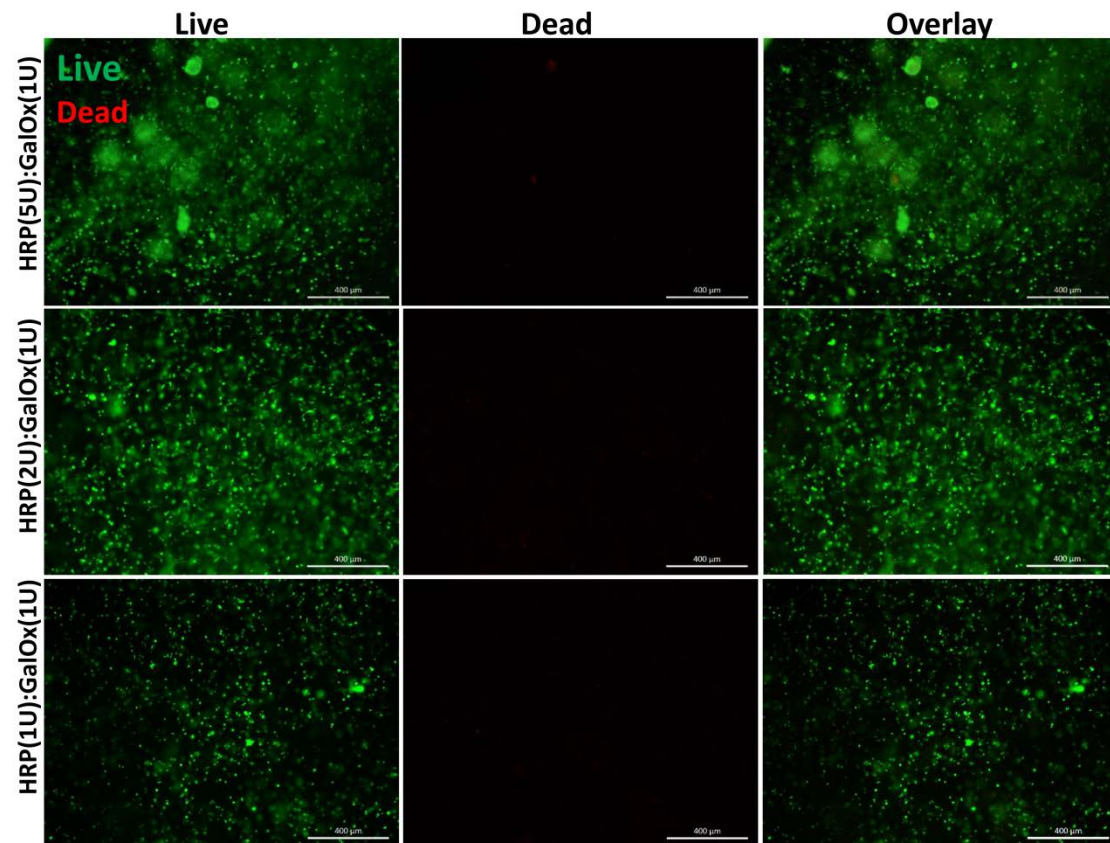
Hongtao Liu,<sup>†</sup> and Fangxia Guan<sup>†\*</sup>

<sup>†</sup>School of Life Science, Zhengzhou University, 100 Science Road, Zhengzhou 450001, P. R. China

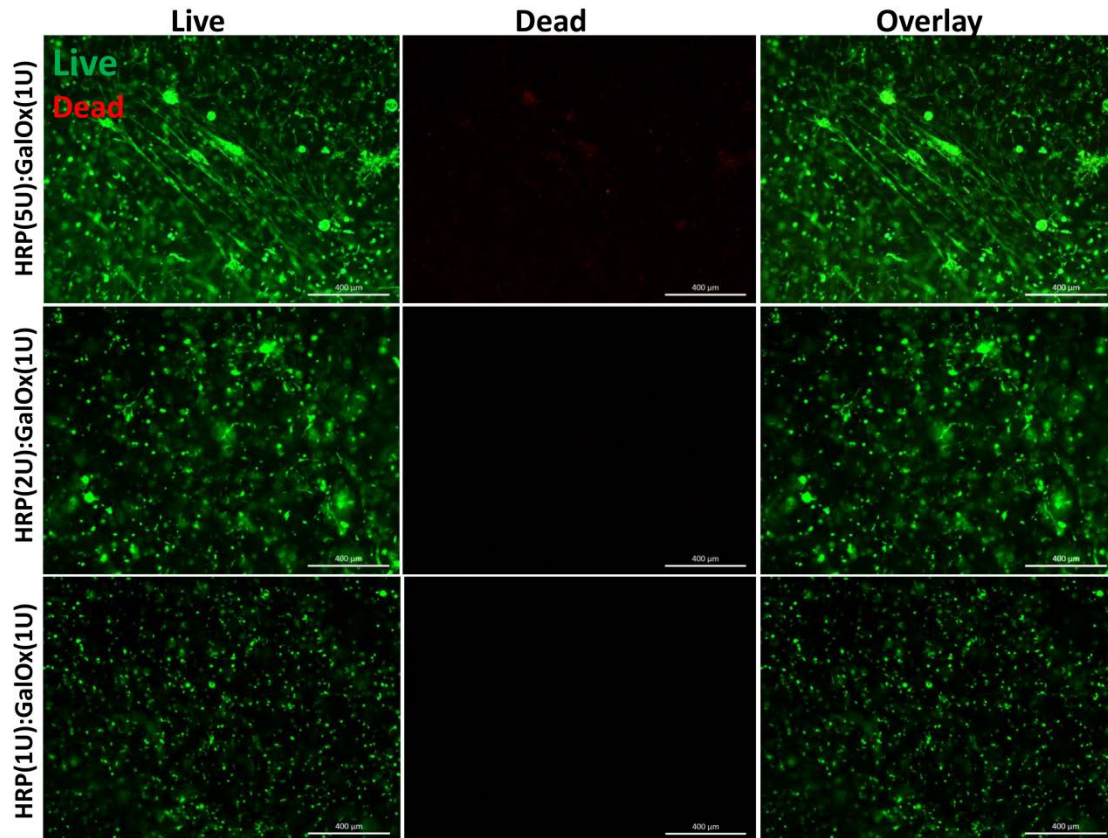
### 3D culture of BMSC within GH hydrogel

BMSC were suspended in the 8% GH solution containing HRP (1, 2, 5 U/ml) and d-galactose (50 mM) at a concentration of  $1 \times 10^6$  cells per ml. Then GalOx (1 U/ml) were added to the GH/HRP/d-galactose/BMSC solution to induce gelation at 37 °C for 10 min. The BMSC-loaded GH hydrogels were cultured using fresh DMEM/F12 complete medium (with 10% fetal bovine serum) at 37 °C in a humidified atmosphere containing 95% air and 5% CO<sub>2</sub>.

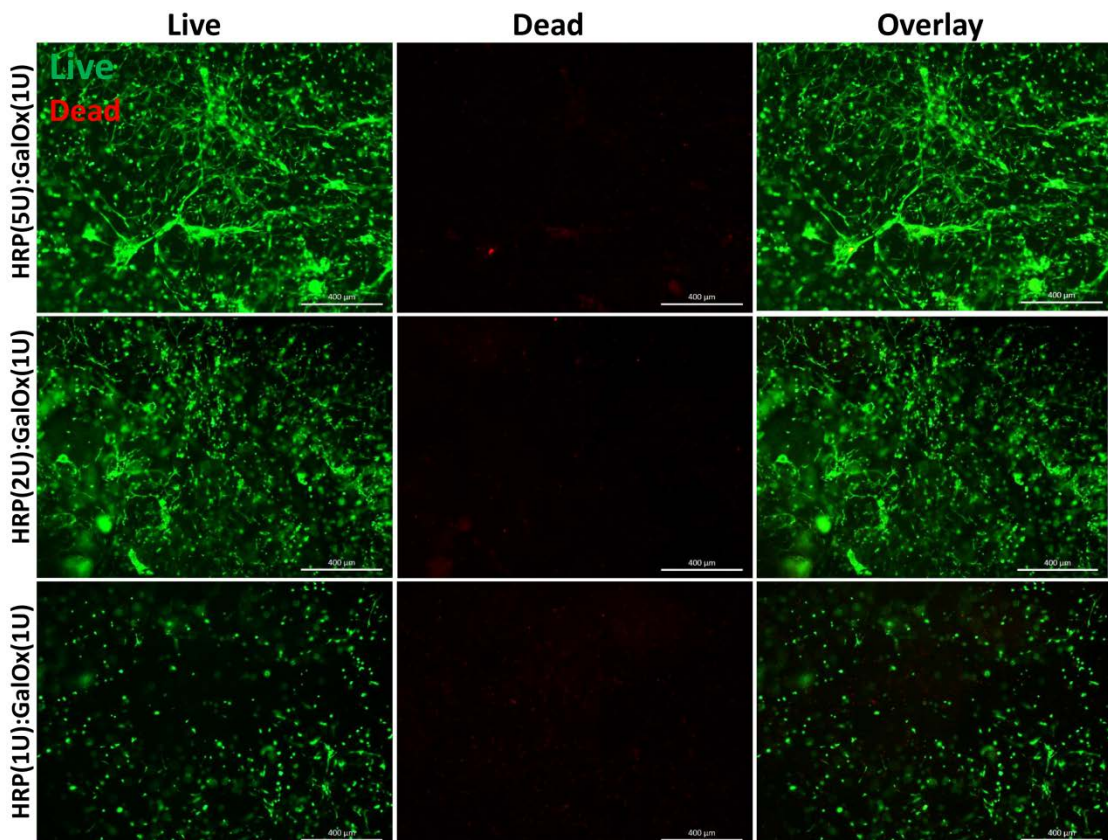
After culturing for 1, 3 and 7 days, the BMSC-loaded hydrogels were stained with cell Live-Dead kit (Calcein-AM/PI) at 37 °C for 10 min, and then observed using fluorescence microscopy (Leica DFC7000T, Germany).



**Figure S1** Live/Dead staining and viability ratio results of BMSC loaded in the hydrogels of HRP(5U): GalOx(1U), HRP(2U): GalOx(1U), and HRP(1U): GalOx(1U) after culturing for 1 day. Scale bar is 400 μm.



**Figure S2** Live/Dead staining and viability ratio results of BMSC loaded in the hydrogels of HRP(5U): GalOx(1U), HRP(2U): GalOx(1U), and HRP(1U): GalOx(1U) after culturing for 3 days. Scale bar is 400  $\mu\text{m}$ .



**Figure S3** Live/Dead staining and viability ratio results of BMSC loaded in the hydrogels of HRP(5U): GalOx(1U), HRP(2U): GalOx(1U), and HRP(1U): GalOx(1U) after culturing for 7 days. Scale bar is 400  $\mu\text{m}$ .

### Notes and references

1. Lee, S. H.; Lee, Y.; Chun, Y. W.; Crowder, S. W.; Young, P. P.; Park, K. D.; Sung, H. J. In Situ Crosslinkable Gelatin Hydrogels for Vasculogenic Induction and Delivery of Mesenchymal Stem Cells. *Adv Funct Mater* **2014**, *24* (43), 6771.
2. Lee, Y.; Bae, J. W.; Oh, D. H.; Park, K. M.; Chun, Y. W.; Sung, H.-J.; Park, K. D. In situ forming gelatin-based tissue adhesives and their phenolic content-driven properties. *Journal of Materials Chemistry B* **2013**, *1* (18), 2407.
3. Lee, Y.; Son, J. Y.; Kang, J. I.; Park, K. M.; Park, K. D. Hydrogen Peroxide-Releasing Hydrogels for Enhanced Endothelial Cell Activities and Neovascularization. *ACS Appl Mater Interfaces* **2018**, *10* (21), 18372.