ACCSCIENCE PUBLISHING

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RESEARCH ARTICLE

Microstructure control during cryogenic 3D printing to obtain biomimetic porous and tough cross-scale mineralized collagen bone scaffold

Supplementary File

Table S1. Comparison of compression modulus and porosity of different scaffolds

Compression modulus (kPa)	Porosity (%)	Fabrication method	Reference
60	65	Extrusion 3D printing	[1]
100	65	Extrusion 3D printing + UV	[2]
5	73	Extrusion 3D printing + UV	[3]
400	40	Extrusion 3D printing + freeze-drying	[4]
300	90	Cryogenic 3D printing	[5]
40	89	Cryogenic 3D printing	[6]
254	85	Extrusion 3D printing	[7]
130	77	Freeze-casting	[8]
177	75	Freeze-casting	[9]
230	70	Freeze-casting	[10]
50	94	Freeze-casting	[11]

Abbreviation: UV, ultraviolet light irradiation.

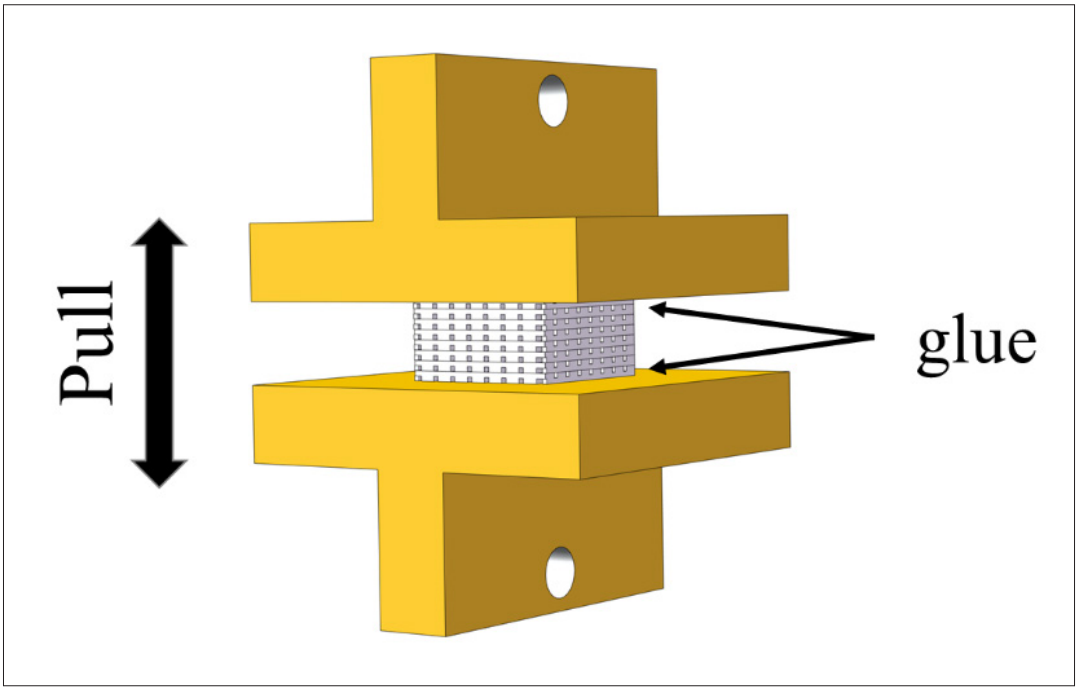

Figure S1. Scaffold stretching model. The top and bottom surfaces of the scaffold were adhered to the ends of the stretching mound. The stretching process was initiated at a rate of 0.5 mm/min and continued until the material failed.

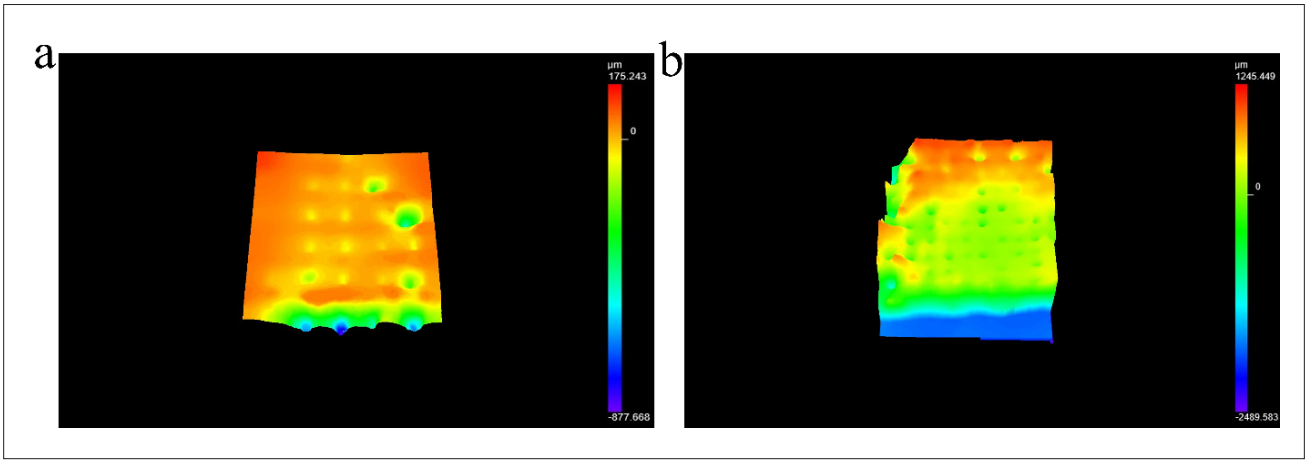

Figure S2. Comparison of crater maps using super depth of field camera: (a) original state; (b) after the falling-ball experiment.

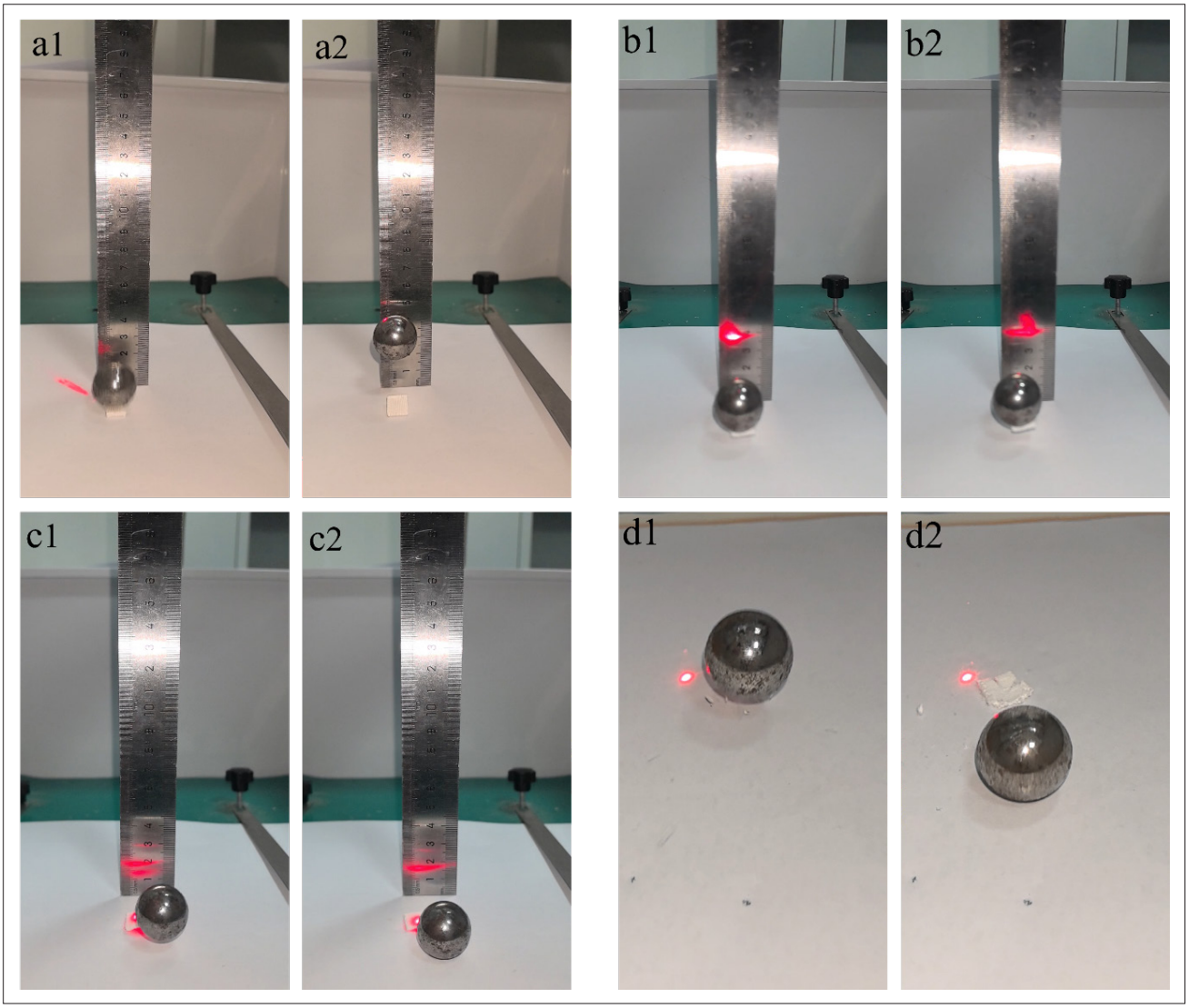

Figure S3. Falling-ball impact experimental process. The drop heights of Figures a and b are 0.3 m. a2 and b2 capture the maximum height of the ball rebound before and after mineralization, a2 before mineralization, and b2 after mineralization. The drop height of Figure c is 0.7 m, which is the maximum height at which the unmineralized stent can absorb the impact energy, and the drop height of Figure d is 0.5 m, which is the maximum height at which the mineralized stent can absorb the impact energy. c2, d2 captures the morphology of the stent after energy absorption. a1–d1 capture the moment when the ball just touches the stent in the different conditions. The position of the red light in the picture is the position of the ball's landing point, as determined by the machine used for the impact experiment.

Supplementary videos

Video S1. The falling ball experiment of cryogenic-printed scaffold before mineralization.

Video S2. The falling ball experiment of cryogenic-printed scaffold after mineralization.

References

1. Yang X, Lu Z, Wu H, Li W, Zheng L, Zhao J. Collagen-alginate as bioink for three-dimensional (3D) cell printing based cartilage tissue engineering. *Mater Sci Eng C*. 2018;83:195-201.
doi: 10.1016/j.msec.2017.09.002
2. Wang Z, Yang Y, Gao Y, Xu Z, Yang S, Jin M. Establishing a novel 3D printing bioinks system with recombinant human collagen. *Int J Biol Macromol*. 2022;211:400-409.
doi: 10.1016/j.ijbiomac.2022.05.088
3. Kajave NS, Schmitt T, Nguyen T-U, Gaharwar AK, Kishore V. Bioglass incorporated methacrylated collagen bioactive ink for 3D printing of bone tissue. *Biomed Mater*. 2021;16(3):10.1088/1748-605X/abc744.
doi: 10.1088/1748-605X/abc744
4. Song Y, Wu H, Gao Y, et al. Zinc silicate/nano-hydroxyapatite/collagen scaffolds promote angiogenesis and bone regeneration via the p38 MAPK pathway in activated monocytes. *Acs Appl Mater Interf*. 2020;12(14):16058-16075.
doi: 10.1021/acsami.0c00470
5. Lee H, Yang GH, Kim M, Lee J, Huh J, Kim G. Fabrication of micro/nanoporous collagen/dECM/silk-fibroin biocomposite scaffolds using a low temperature 3D printing process for bone tissue regeneration. *Mater Sci Eng C*. 2018;84:140-147.
doi: 10.1016/j.msec.2017.11.013
6. Liu H, Wang C, Sun X, et al. Silk fibroin/collagen/hydroxyapatite scaffolds obtained by 3D printing technology and loaded with recombinant human erythropoietin in the reconstruction of alveolar bone defects. *Acs Biomater Sci Eng*. 2022;8(12):5245-5256.
doi: 10.1021/acsbiomaterials.2c00690
7. Hwangbo H, Lee H, Roh EJ, et al. Bone tissue engineering via application of a collagen/hydroxyapatite 4D-printed biomimetic scaffold for spinal fusion. *Appl Phys Rev*. 2021;8(2):021403.
doi: 10.1063/5.0035601
8. Liu Y, Gu J, Fan D. Fabrication of high-strength and porous hybrid scaffolds based on nano-hydroxyapatite and human-like collagen for bone tissue regeneration. *Polymers (Basel)*. 2020;12(1):61.
doi: 10.3390/polym12010061
9. Chacon EL, Bertolo MRV, de Guzzi Plepis AM, et al. Collagen-chitosan-hydroxyapatite composite scaffolds for bone repair in ovariectomized rats. *Sci Rep*. 2023;13(1):28.
doi: 10.1038/s41598-022-24424-x
10. Minardi S, Taraballi F, Cabrera FJ, et al. Biomimetic hydroxyapatite/collagen composite drives bone niche recapitulation in a rabbit orthotopic model. *Mater Today Bio*. 2019;2:100005.
doi: 10.1016/j.mtbio.2019.100005
11. Rahman MS, Rana MM, Spitzhorn LS, et al. Fabrication of biocompatible porous scaffolds based on hydroxyapatite/collagen/chitosan composite for restoration of defected maxillofacial mandible bone. *Prog Biomater*. 2019;8(3):137-154.
doi: 10.1007/s40204-019-0113-x