

Supporting Information

A bioactive calcium silicate nanowires-containing hydrogel for organoid formation and functionalization

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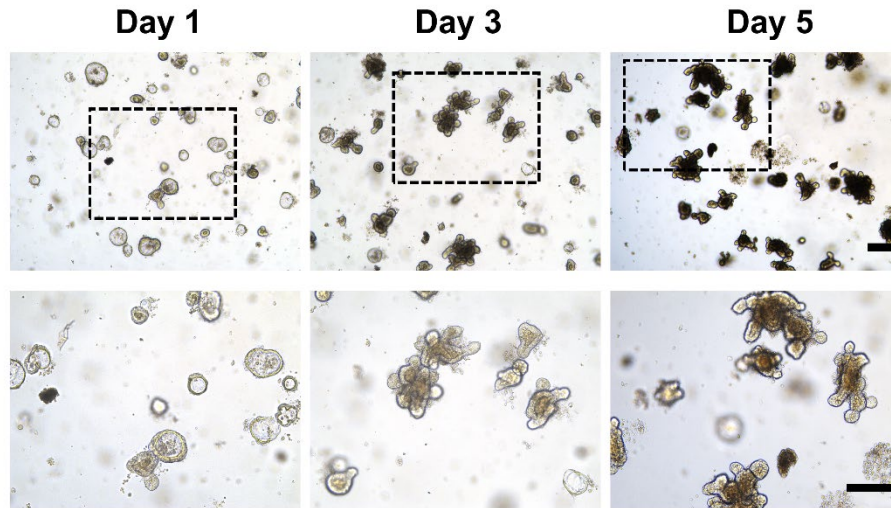


Fig. S1 The developmental process of the intestinal organoids at different times. Scale bars: 250 μ m.

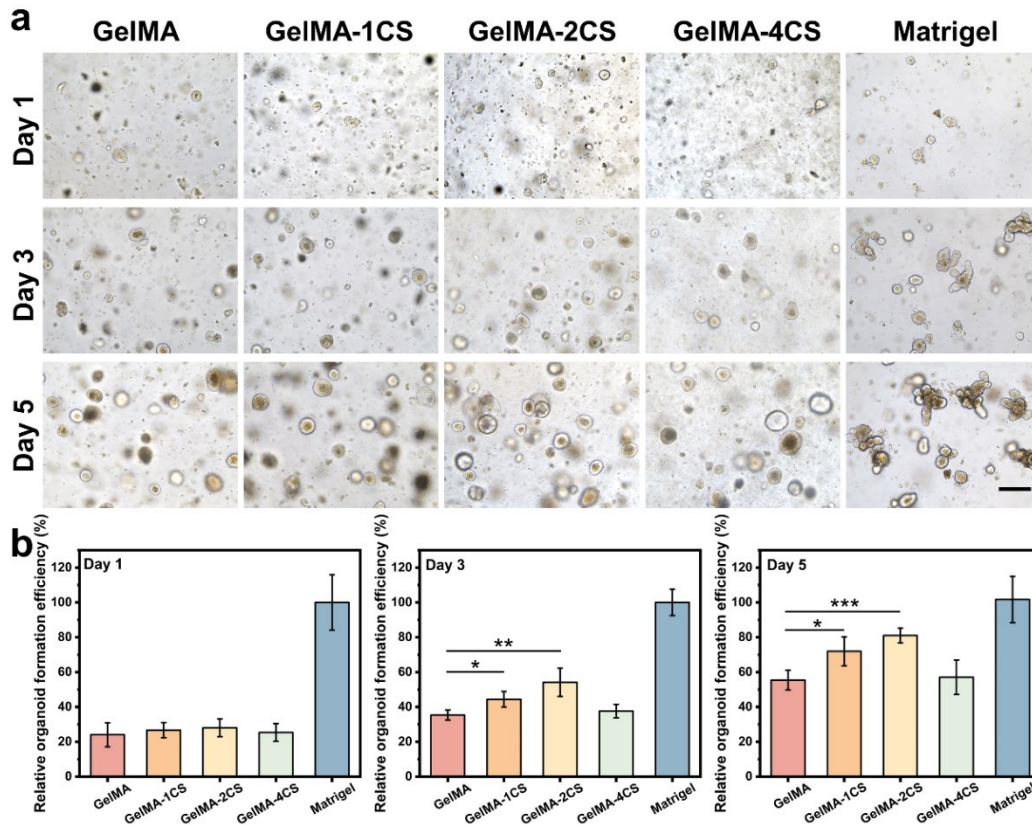


Fig. S2 (a) The optical microphotographs of the intestinal organoids grown in the CS/GelMA hydrogel with different content of CS nanowires for 1, 3, and 5 days. Scale bars: 250 μ m. (b) Quantitative analysis of colony formation efficiency cultured in different CS/GelMA composite hydrogel for 1, 3, and 5 days (n=4). Data are presented as means \pm SD. * p < 0.05, ** p < 0.01, *** p < 0.001.

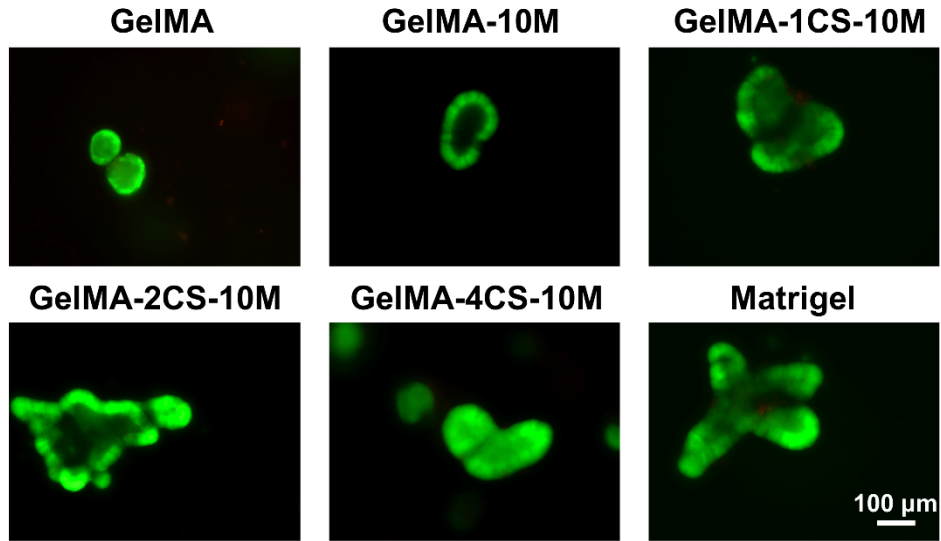


Fig. S3 The live/dead staining of intestinal organoids grown in different hydrogels.

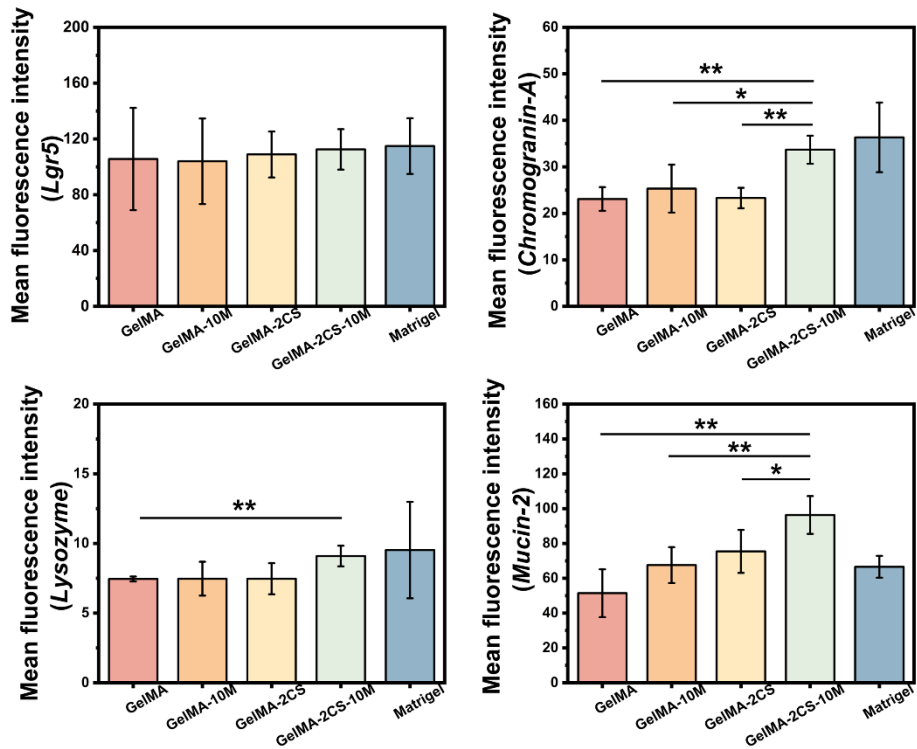


Fig. S4 The mean fluorescence intensity of four differentiated cell type markers in intestinal organoids grown in different hydrogels. Data are presented as means \pm SD. * $p < 0.05$, ** $p < 0.01$.

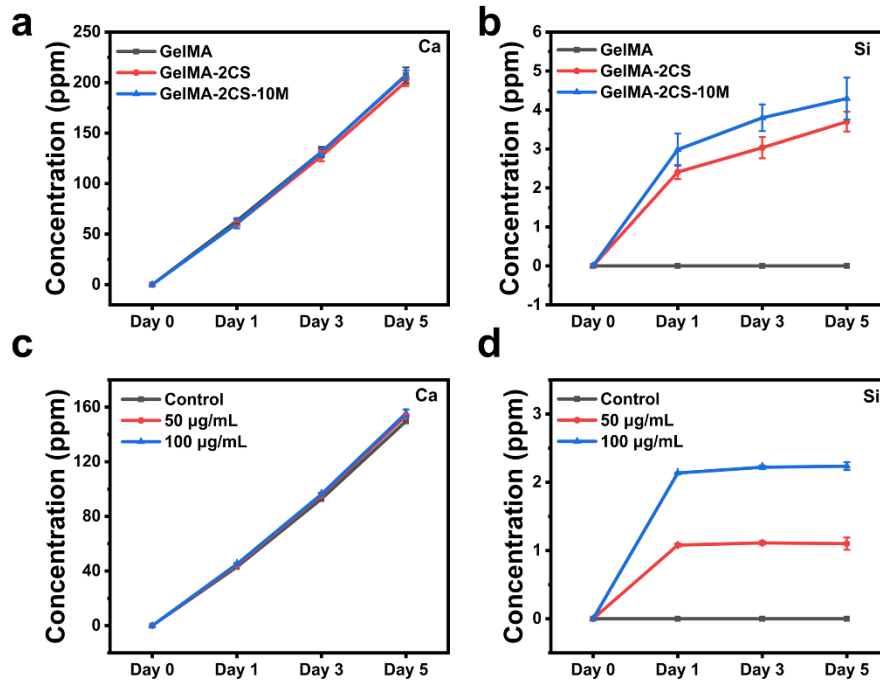


Fig. S5 The ions release of CS/GelMA hydrogel (a, b) and CS nanowires (c, d) soaked in DMEM/F12 medium for 1, 3, and 5 days, respectively.

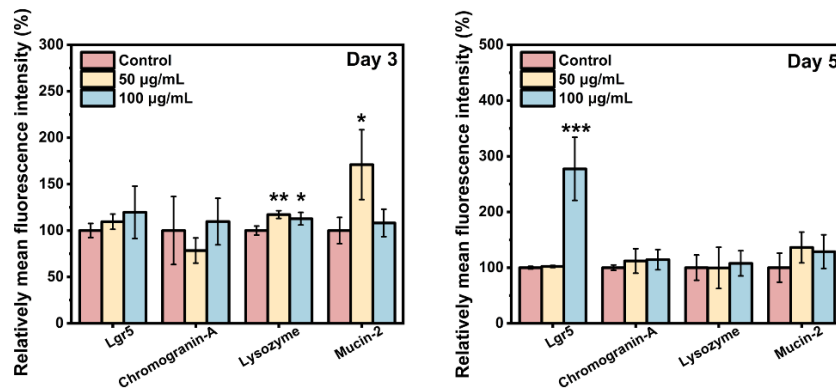


Fig. S6 The relatively mean fluorescence intensity of four differentiated cell types markers in intestinal organoids treated with 0, 50 µg/mL, and 100 µg/mL CS nanowires for 3 and 5 days. Data are presented as means ± SD. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

Table S1 Primer sequences for RT-qPCR.

Gene	Primer sequence
Lgr5 F	ATTCGGTGCATTAGCTTGG
Lgr5 R	CGAACACCTGCGTGAATATG
Chromogranin-A F	AAGGTGATGAAGTGCCTCCT
Chromogranin-A R	GGTGTTCGCAGGATAGAGAGG

Lysozyme F	GTCACACTTCCTCGCTTTCC
Lysozyme R	TGGCTTTGCTGACTGACAAG
Mucin-2 F	CCGACTTCAACCCAAGTGAT
Mucin-2 R	GAGCAAGGGACTCTGGTCTG
GAPDH F	GATTTGGTCGTATTGGGCG
GAPDH R	CTGGAAGATGGTGATGG
ALB F	GCATTGGTCTCATCTGTCCG
ALB R	ATTGGGGAATGTCTGGCTCA
KRT19 F	TGACCTGGAGATGCAGATTG
KRT19 R	CCTCAGGGCAGTAATTCCTC
CYP3A11 F	GGATGAGATCGATGAGGCTCTG
CYP3A11 R	CAGGTATTCCATCTCCATCACAGT
SOX9 F	AGGAAGCTGGCAGACCAGTA
SOX9 R	CGTTCTTCACCGACTTCCTC
HNF4a F	AGCCGACAATGTGTGGTAGA
HNF4a R	CTTCCTTCTTCATGCCAGCC
GAPDH F	CACTGCCACCCAGAAGACTGT
GAPDH R	GGAAGGCCATGCCAGTGA

Table S2 Primer sequences for RT-qPCR.

Gene	Primer sequence
Wnt3 F	CTCGCTGGCTACCCAATTTG
Wnt3 R	CTTCACACCTTCTGCTACGCT
LRP6 F	GCGAAGACCACAGCGATGAA
LRP6 R	TTGTCCTGGCAGTTCGCATC
R-spondin1 F	TGTGAAATGAGCGAGTGGTCC
R-spondin1 R	TCTCCCAGATGCTCCAGTTCT
R-spondin2 F	TTGCATAGAGGCCGCTGCTTT
R-spondin2 R	CTGGTCAGAGGATCAGGAATG
R-spondin3 F	GTACACTGTGAGGCCAGTGAA
R-spondin3 R	ATGGCTAGAACACCTGTCCTG
GAPDH F	ATGGTGAAGGTTCGGTGTGAA
GAPDH R	TGGAAGATGGTGATGGGCTT