Supporting information for:

Induction and differentiation of human induced pluripotent stem cells into functional cardiomyocytes on a compartmented monolayer of gelatin nanofibers

Yadong Tang[a,b]，Li Liu[b]，Junjun Li[b]，Leqian Yu[b], Li Wang[a]，Jian Shi[a]， Yong Chen[a,b,c]*

[a] *Ecole Normale Supérieure-PSL Research University, Département de Chimie, Sorbonne Universités - UPMC Univ Paris 06, CNRS UMR 8640 PASTEUR, 24, rue Lhomond, 75005 Paris, France*

[b] *Institute for Integrated Cell-Material Science, Kyoto University, Kyoto 606-8507, Japan*

[c] *Institute for Interdisciplinary Research, Jianghan University, 430056 Wuhan, China*

*Corresponding author: E-mail: yong.chen@ens.fr

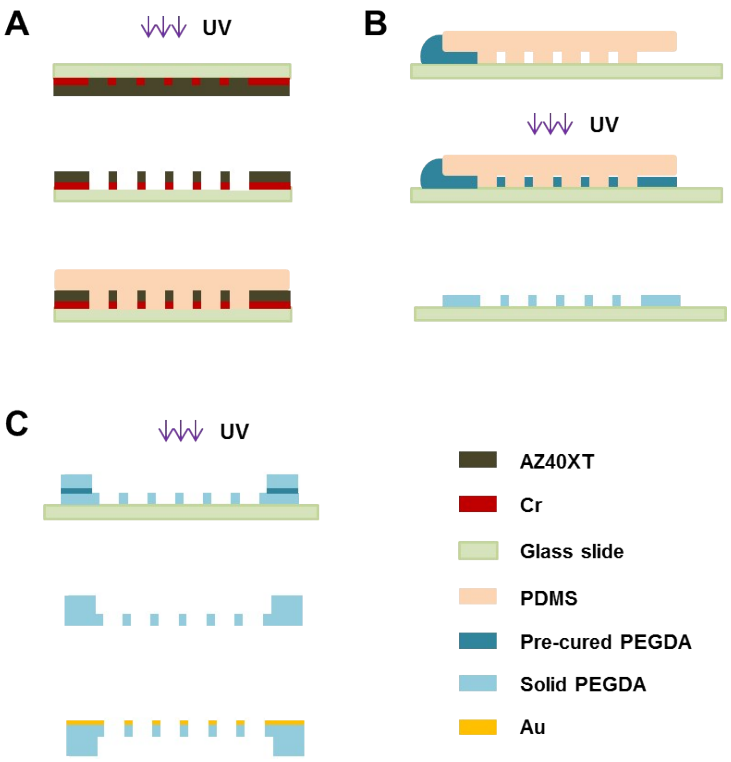

Fig. S1 Fabrication sequence of PEGDA honeycomb frame. (A) Fabrication of PDMS mold by backside UV lithography of a AZ 40XT resist layer spin-coated on a patterned Chromium mask and PDMS casting; (B) Fabrication of PEGDA honeycomb microframe by aspiration-assisted molding. (C) Binding of the PEGDA ring on the honeycomb frame and backside Au deposition.

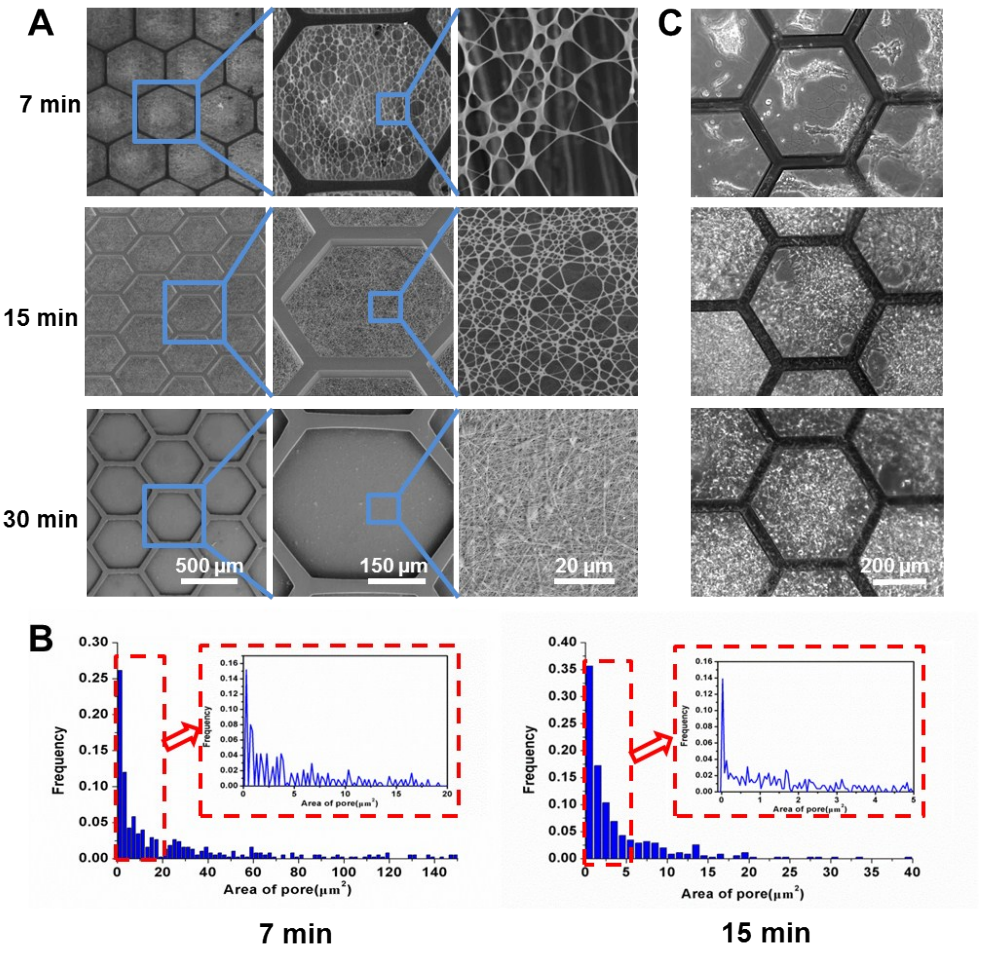

Fig. S2 Electrospinning time dependent patch structure and cell coverage of the patch. (A) SEM images of crosslinked gelatin nanofibers obtained by 7, 15 and 30 min electrospinning. (B) Pore size distribution of 7 and 15 min electrospun samples. (C) Bright field images of hiPSCs on the patch with the same seeding density after 24 h culture.

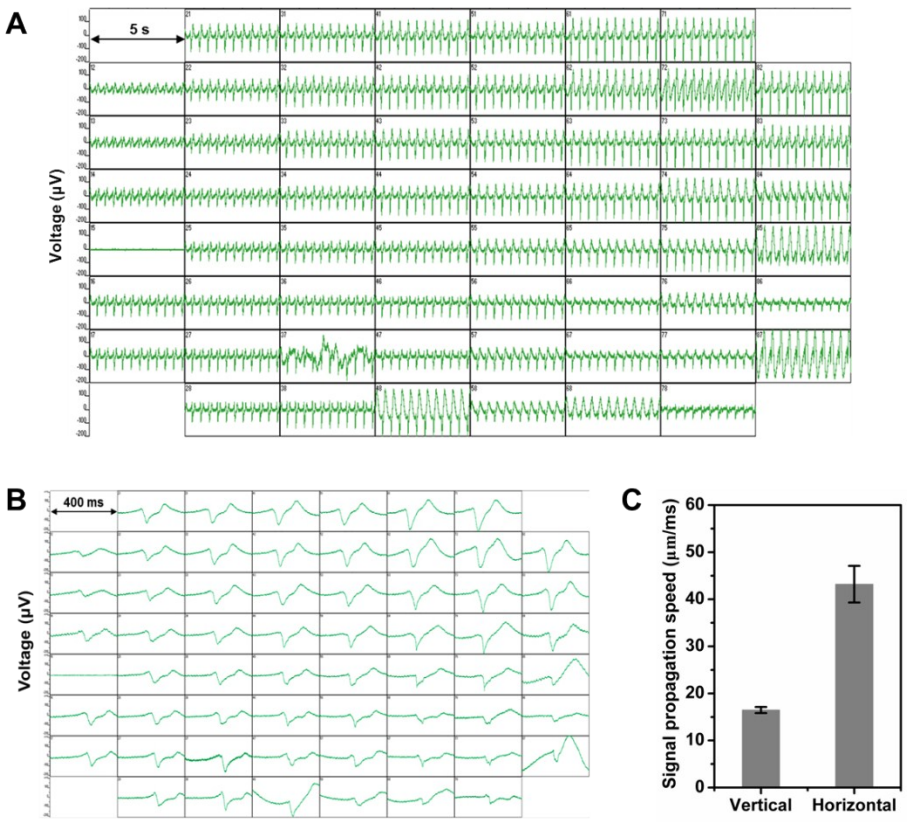

Fig. S3 Overall view (A: 5s interval; B: 400ms interval) of 60-channel representative field potential waves of hiPSC differentiated cardiomyocytes on a patch recorded using a commercial MEA device, showing rather uniform and regular cardiac beatings. (C) Cardiac signal propagation speed in vertical and horizontal directions.

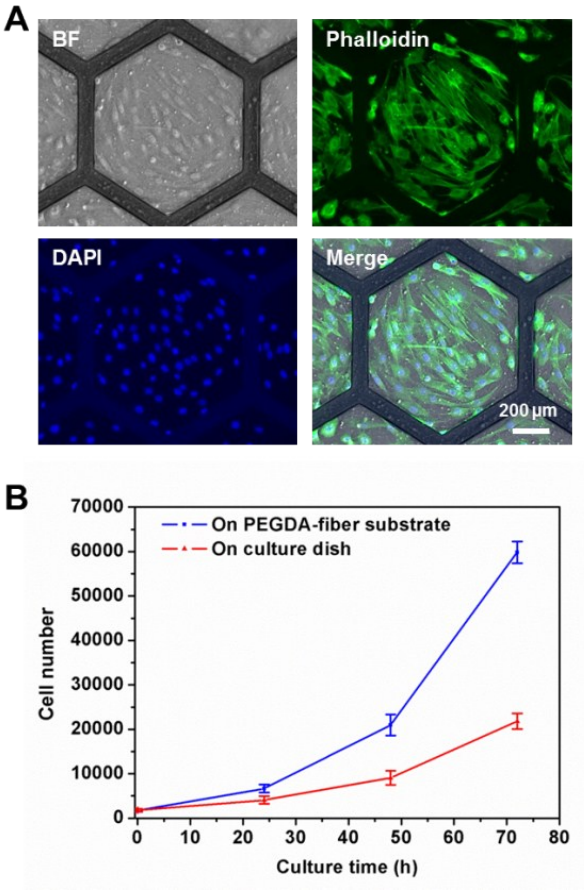

Fig. S4 (A) Bright field and immunostaining fluorescence images of NIH-3T3 cells: Green, Phalloidin for cytoskeleton; Blue, DAPI for nuclear. (B) Increase of the NIH-3T3 cell number versus culture time with a PEGDA-nanofiber device and a normal culture dish for 72 h, showing a significant difference of the cell proliferation rate between the two types of culture substrates.

Table S5 Forward and reverse primer sequences used for pluripotency analyze by RT-PCR.

Primers	**Sequence**
OCT4 forward	5′-GACAGGGGGAGGGGAGGAGCTAGG-3′
OCT4 reverse	5′-CTTCCCTCCAACCAGTTGCCCCAAAC-3′
NANOG forward	5′-CAGCCCCGATTCTTCCACCAGTCCC-3′
NANOG reverse	5′-CGGAAGATTCCCAGTCGGGTTCACC-3′
PAX6 forward	5′-ACCCATTATCCAGATGTGTTTGCCCGAG-3’
PAX6 reverse	5′- ATGGTGAAGCTGGGCATAGGCGGCAG-3′
BRACHYURY forward	5′-GCCCTCTCCCTCCCCTCCACGCACAG-3′
BRACHYURY reverse	5′-CGGCGCCGTTGCTCACAGACCACAGG-3′
AFP forward	5′-GAATGCTGCAAACTGACCACGCTGGAAC-3′
AFP reverse	5′-TGGCATTCAAGAGGGTTTTCAGTCTGGA-3′
GAPDH forward	5′-AACAGCCTCAAGATCATCAGC-3′
GAPDH reverse	5′-GGTCTCTCTCTTCCTCTTGTGC-3′

Video S1 Contraction of cardiomyocytes on a patch at day 12, showing homogenous synchronized beating over all the area of observation.

Video S2 Contraction of cardiomyocytes on a glass slide at day 12, showing irregular beating due to inhomogeneous formation of cell clusters.

Video S3 Contraction of cardiomyocytes on a patch placed in a commercial MEA device at day 21, showing again homogenous synchronized beating and the underneath microelectrodes.