Supplemental Information:

Contractile response: Constructs sampled on culture day 11 were analyzed with respect to excitation threshold (ET), maximum capture rate (MCR) and amplitude of contraction (fractional area change) as previously described [19]. A total of 9 constructs were analyzed in the CM and CF+ CM groups, and 10 constructs in the US group. In brief, constructs were placed in a 60 mm Petri dish containing 120 mL Tyrode's solution (140 mM NaCl, 5.4 mM KCl, 0.33 mM NaH_2PO_4, 1.8 mM $CaCl_2$, 1 mM $MgCl_2$, 5 mM HEPES, 5.5 mM D-glucose, pH 7.4) between two carbon rods (Ladd Research Instruments) that were connected to a cardiac stimulator (Nihon Kohden). The measurements were done at room temperature. Electrical stimuli (square pulses, 2 ms duration) were applied at a rate of 1 Hz and a voltage of 1 V, and the voltage was gradually increased until the entire construct was observed to beat synchronously, as assessed by videomicroscopy at 10X magnification. The corresponding voltage was defined as the ET. Maximum capture rate (MCR) was determined at 200% of ET by increasing the stimulation frequency until the paced contractions ceased or became irregular. Contractile amplitude was determined for constructs paced at 1 Hz and 200% of ET by obtaining digital video recordings (30 frames/s) and assessing the change in crosssectional area. In particular, average amplitude of contraction (i.e. average fractional area change measured during a 1-to-5 min. long contractile sequence) and contraction profile (i.e. time history of fractional area change obtained measured during a single, 800 ms contractile cycle) were determined using image analysis software (Scion Image).

Histology and Immunofluorescence: Histological analyses were done on constructs (n = 4-5 per group) sampled on culture day 11 from all four groups and on culture day 5 for the CF group (CF-5). Constructs were rinsed in PBS, fixed in 10% neutral buffered formalin (Sigma-Aldrich) for 12 h, embedded in paraffin, and sectioned within the first 100 μm from the surface at a thickness of 5 μm. Sections were stained with hematoxylin and eosin for general evaluation and Masson's Trichrome for collagen. Sections were immunofluorescently stained for markers of CM (troponin I, sarcomeric α-actin, connexin-43) [19] and markers of CF (vimentin and prolyl-4-hydroxylase) [17]. For immunofluorescence, sections were deparaffinized and antigen was retrieved by heat treatment for 20 min at 95°C in de-cloaking chamber (Biocare Medical), blocked with 10% horse serum (Vector Laboratories) for 40 minutes at RT, then incubated for 1 hour at 37°C with the following primary antibodies diluted in PBS containing 0.5% Tween 20 and 1.5% horse serum: (i) polyclonal rabbit troponin I (Chemicon 1:200), (ii) polyclonal rabbit anti-Connexin-43 (Chemicon, 1:50), (iii) mouse anti-prolyl-4-hydroxylase β subunit (Clone 6-9H6, Chemicon 1:50), (iv) mouse anti-sarcomeric α-actin (clone5C5, 1;100, Sigma) and (v) Cy3 conjugated mouse anti-vimentin (clone V9, Sigma, 1:100). Subsequently, the slides were rinsed in PBS and incubated with the appropriate secondary antibodies (all from Vector Laboratories) for 30 minutes at 37°C: fluorescein conjugated goat anti-rabbit IgG (1:200) for Cx-43 and troponin visualization and fluorescein conjugated horse anti-mouse IgG (1:200) for prolyl-4-hydroxylase and sarcomeric α-actin visualization. The sections were counterstained with DAPI and coverslipped (Vectorshield mounting medium with DAPI). Neonatal rat heart tissue and bovine articulate cartilage served as positive and negative control, respectively.

Implantation studies:

All surgeries were according to an approved Columbia University Animal Care Protocol. Adult nude athymic rats were anesthetized with inhaled isoflurane (5.0%) and rapidly intubated and

placed on a rodent ventilator at a rate of 90 and a tidal volume of 2.5cc. After initiation of mechanical ventilation, anesthesia was maintained by inhalation of 2.0% isoflurane. A 1.5cm left thoracotomy incision was made with a #10 blade over the point of maximal impulse. The lung was gently retracted and the pericardium opened exposing the left ventricle and left anterior descending artery (LAD). A 7-0 prolene suture was passed around the LAD and tied producing an anterior wall myocardial infarction. The scaffold was then placed over the infarct bed and secured to the myocardium with 4 interupted 7-0 prolene sutures. A warm water recalculating pad was used during the surgery for thermoregulation and intra-muscular buprenorphone (0.05mg/mkg) was applied as a pre-emptive anitbiotic. The rats were sacrificed after 2 weeks, the hearts were explanted, formaledyde fixed and paraffin embedded.