**Supplementary Figures**

**Figure S1.** Design and fabrication of nanopatterned cardiosphere-derived cell patch. (A) Replica molding procedure for large area (centimeters) fabrication of nanopatterned PEG scaffolds. (B) Cross-sectional SEM image of the ANFS. (C) SEM images of CDCs cultured on the ANFS for 7 days illustrating the confluent cell alignment, and formation of tight intercellular junctional structures. Scale bar: 2 µm in (B); 50 µm in (C).

**Figure S2.** Cardiomyogenesis of CDCs promoted by ANFS. Cardiomyogenesis of CDCs on (A, B) ANFS vs. (C) the flat substratum. (A) and (B) correspond to the experiment shown in Fig. 1E, at higher magnification and showing the

corresponding phase contrast image revealing the underlying nano-structured substratum; (C) The control experiment, with CDCs cultured on the flat substratum for 14 days. Scale bar: 20 µm in (A) and (B); 50 µm in (C).

**Figure S3.** Co-culture of CDCs with neo-natal rat ventricular myocytes on ANFS promote cardiomyogenesis. NCX-GFP expression measured using flow cytometry in DiI labeled CDCs following 14 days co-culture with NRVMs (also see Fig. 3E). (A) Representative example of NCX-GFP expression in DiI labeled CDCs after 14 days of co-culture with NRVMs. CDCs were lentivirally transduced with NCX-GFP, and labeled with DiI and co-cultured with NRVMs on the flat substratum and ANFS in a 1:10 ratio. DiI positive CDCs were gated using APC filter (x-axis) and NCX-GFP expression was measured using FITC filter (y-axis). Results were normalized against NCX-GFP transduced CDCs on day 1. In this analysis the ‘Probability distribution’ mode was used to evaluate the fraction of NCX-GFP expressing CDCs, labeled with DiI on the flat substratum in co-culture with NRVMs on (B) day 1 and (C) day 14, and (D) ANFS in co-culture with NRVMs on day 14. Horizontal line represents normalization with GFP expression at day1. Y-axis represents NCX-GFP expression level, and X-axis represents DiI staining intensity.

**Figure S4.** Statistical analysis of co-culture of CDCs with neo-natal rat ventricular myocytes on ANFS (see Fig. S3).

**Supplementary Movie**

**Movie S1.** Live cell movie showing polarization and directional migration of individual CDCs along the orientation of the ANFS.

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