Electronic Supplementary Material (ESI) for Analyst.

Supplementary Information:

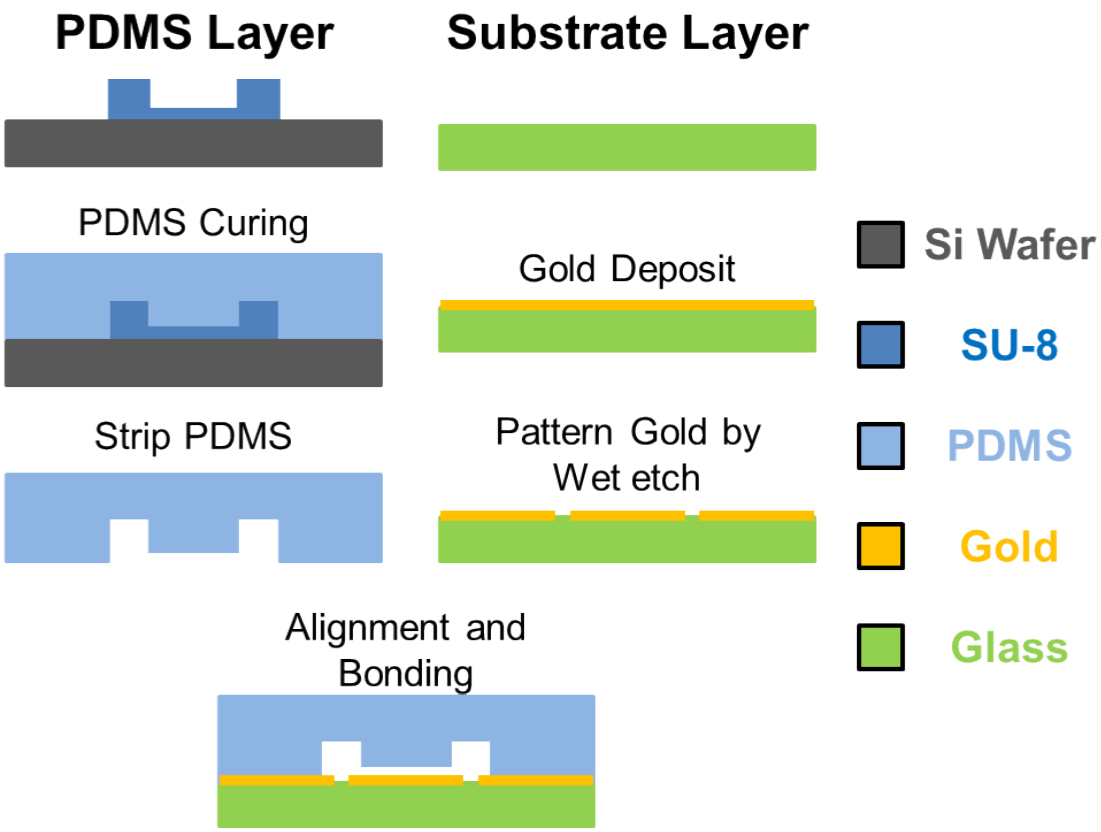

Fig. S1. Fabrication process flows of the presented electrolytic isolation chip: Two masks were used to fabricate an SU8 master. One was used to make shallower (10 µm) interaction bridges and the other to fabricate capture sites and deeper (40 µm) channels. The patterned PDMS (PDMS, Sylgard 184, Dow Corning) was formed by the standard master fabrication process. The Au/Cr (400 nm/10 nm) layer was deposited on the glass wafer by evaporation (EnerJet Evaporator), and patterned by gold etchant (Transene GE-8148 Gold Etch). To seal the device, both the PDMS and the glass substrate were treated by oxygen plasma and then aligned and bonded together.[1]

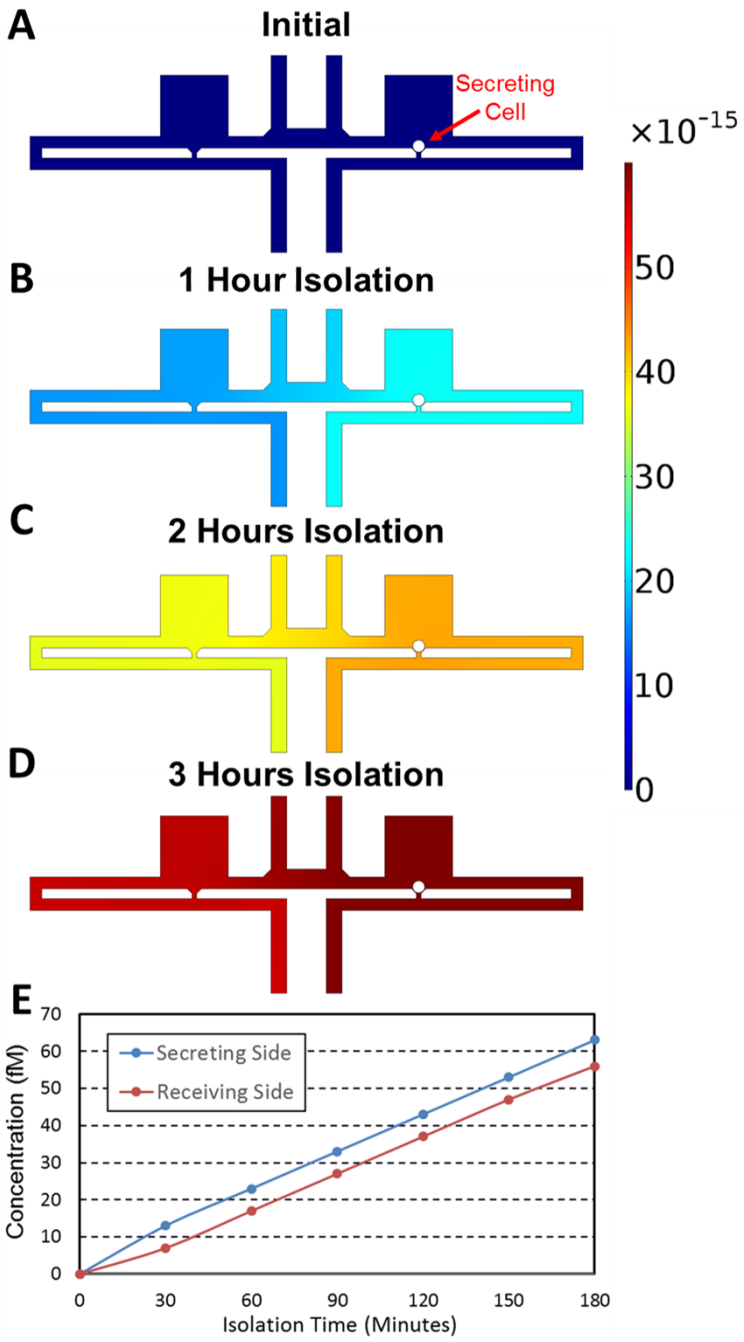

Fig. S2. Simulations of diffusion for signaling proteins through a narrow bridge channel by COMSOL 4.3. We assume that single cell secretes 1^{-20} mole of proteins per second and its diffusion coefficient is 1^{-10} m^2/s.[2-4] We assume that the secreting cell is captured in the right chamber. We simulate the diffusion of signaling proteins from the right (secreting) chamber to the left (receiving) chamber for 3-hour isolation time. (A) Initial condition. Both chambers (left and right) have zero concentrations. (B) Concentration distribution after 1 hour isolation. (C) Concentration distribution after 2 hours isolation. (D) Concentration distribution after 3 hours isolation. (E) Concentration change of signaling proteins over time. Concentration of the receiving chamber (left) closely follows that of the secreting chamber (right). After 3 hour isolation, the concentration difference between two chamber is only 11%, confirming that the diffusion through a bridge channel is sufficient for adequate the cell-cell interaction.

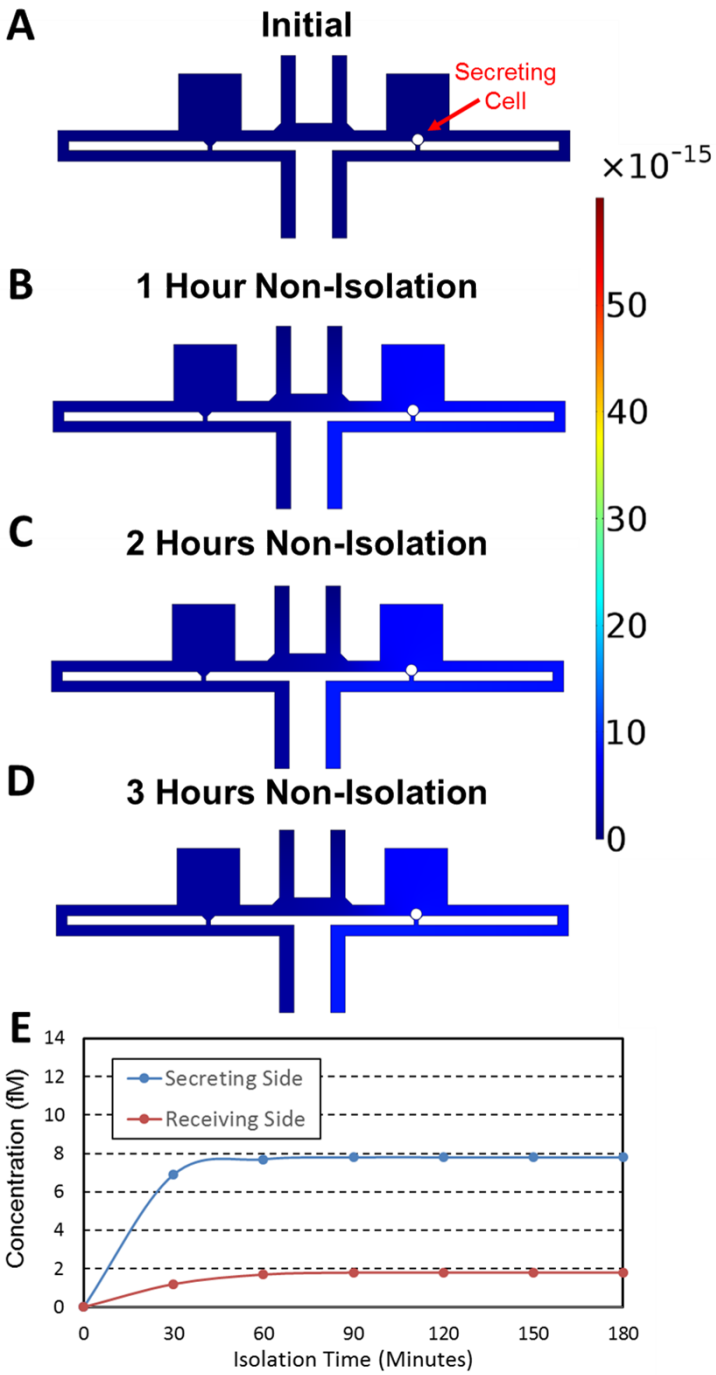

Fig. S3. Simulations of diffusion for signaling proteins when washed through perfusion flow by COMSOL 4.3. We assume that single cell secretes 1^{-20} mole of proteins per second and its diffusion coefficient is 1^{-10} m^2/s.[2-4] As the chambers are not isolated, the secreted proteins are washed by perfusion flow of the media flowing from upstream to downstream. 50 Pa was assumed as a pressure difference between the upstream and downstream. Initially, the concentration is assumed zero and the secreting cell is captured in the right chamber. (A) Initial concentration distribution. (B) Concentration distribution after 1 hour. (C) Concentration distribution after 2 hours. (D) Concentration distribution after 3 hours. (E) Concentration change of signaling proteins over time. As the secreted proteins are washed away by perfusion flow, the protein concentration in the receiving chamber (left) is only 3.3% of that of the isolated case after 3 hours, confirming that cell-cell interaction is negligible.

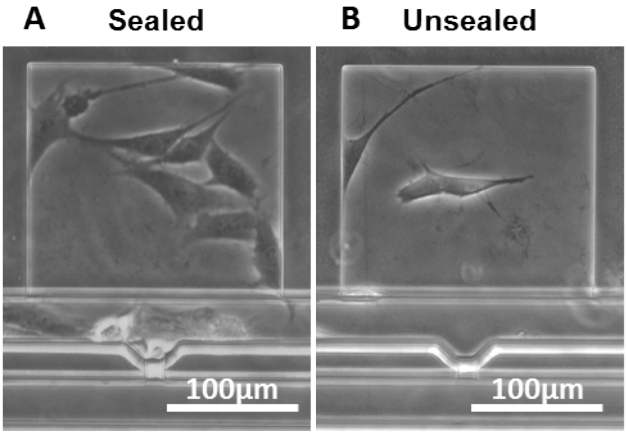

Fig. S4. Cell-cell interaction between C2C12 and PC3 cells: (A) C2C12 cell chambers after 3 day co-culture with five PC3 cells with electrolytic bubble isolation and (B) without electrolytic bubble isolation.

Reference

1. P. Ingram, Y. J. Kim, T. Bersano-Begey, X. Lou, A. Asakura, and E. Yoon, *Proceedings of MicroTAS*, Groningen, 2010, 277-279.
2. M. P. Raphael, J. A. Christodoulides, J. B. Delehanty, J. P. Long, and J. M. Byers, *Biophysical Journal*, 2013, 105, 602–608.
3. D. Brune and S. Kim, *PNAS*, 1993, 90 (9), 3835-3839.
4. M.T. Tyn, T.W. Gusek, *Biotechnol Bioeng*. 1990, 35(4), 327-38.