Supplementary Information

Control of cell migration using a conducting polymer device

Abdurrahman Gumus, Joseph P. Califano, Alwin M.D. Wan, John Huynh, Cynthia A. Reinhart-King and George G. Malliaras

Cell culture: Bovine aortic endothelial cells (ECs) were maintained at 37˚C and 5% CO_2 in Medium 199 (Invitrogen, Carlsbad, CA) supplemented with 10% Fetal Clone III (HyClone, Logan, UT), and 1% each of penicillin-streptomycin, MEM vitamins (Mediatech, Manassas, VA) and MEM amino acids (Invitrogen, Carlsbad, CA).

Preparation of the PEDOT:TOS films: ITO patterned glass slides were cleaned carefully with liquid laboratory detergent followed by extensive rinsing with DI water and drying with nitrogen. The ITO stripe was 3 mm wide and 2.5 cm long. A special polymerization tool provided by ReynoldsTech (Syracuse, NY) was used for the deposition of PEDOT:TOS, according to a method adapted from literature*[28]*. A solution consisting of 0.785 grams of Fe(III)-tosylate in 5mL of isopropanol was combined with 32.1 μL of pyridine. The solution was filtered through a 0.45 μm PTFE filter and was spin coated on a cleaned ITO patterned glass slide. The latter was baked for 2 min at 80^0C on a hot plate and was introduced in the ReynoldsTech vapor phase polymerization chamber. 100 μL EDOT were added in a crucible that was held at 80^0C , while the substrate was held at 35^0C. After polymerization for 15-20 minutes, the devices were baked at 50^0C for 30 min followed by rinsing with ethanol (2x5min) to remove the iron salts.

Statistical Analysis of Cell Speed and Persistency Measurements: Images were acquired in five locations ("pixels") across the PEDOT:TOS stripe that were centered at -0.9, -0.5, 0, 0.5, and 0.9V. As the length of these pixels along the direction of the applied bias was 1.2 mm, the length of the PEDOT:TOS stripe

inside the reservoir was 12 mm, and the potential difference inside the PDMS reservoir was 2 V, the variation in the bias within a pixel was ±0.1 V. Six cells were monitored within each pixel. The cell centroids were determined with ImageJ (National Institutes of Health) and coordinates were converted to micron displacements to calculate the mean-square displacement. The speed and direction persistence time were determined from Equation (1) using nonlinear least squares regression analysis*[18]*. Matlab was used to perform migration analysis. Cell speed and direction persistence time measurements were normally distributed and had equal variances. Data were compared with analysis of variance and Tukey's Honestly Significantly Different test in JMP software. All data were reported as mean ± standard error of the mean (SEM).

Control experiment: An unbiased PEDOT:Tosylate film was used for a control experiment. The trajectories of 10 cells were monitored, and the average speed and directional persistence time were found to be 11.5±1.6 μm/h and 53.3±5.3 min, respectively. A T-test revealed that these results were statistically similar to results from the 0V location and statistically different to results from the +0.9V location.

Fibronectin Immunostaining: After electrical biasing for one hour, the devices were rinsed 3 times with warm phosphate buffered saline (PBS). Fibronectin was fixed in 3.7% Formaldehyde (Mallinckrodt Baker, Phillipsburg, NJ) for 30 min at room temperature, then washed with PBS. The device was incubated with 1% Triton in PBS and 0.02% Tween (Mallinckrodt Baker, Phillipsburg, NJ)/1% Bovine Serum Albumin (BSA, Sigma-Aldrich, St. Louis, MO) in PBS for 1 hour as a blocking step. The device was incubated 1:50 with a mouse monoclonal fibronectin primary antibody (Santa Cruz Biotechnology, Santa Cruz, CA) in PBS/3% BSA in a humidified chamber at 4^0 for 3 hours. A secondary antibody (1:200 fluorescein isothiocyanate (FITC)-conjugated goat anti-mouse, (Santa Cruz Biotechnology, Santa Cruz, CA)) in PBS/3%BSA was applied to the device for 1 hour at room temperature. Fibronectin localization was

visualized with a Zeiss Axio Observer.Z1m with a Hamamatsu ORCA-ER camera. Nine pictures were taken along the PEDOT:TOS stripe. Images were pseudo-colored with Axiovision software v. 4.6 and stitched with Canon PhotoStitch software (version 3.1, Canon INC, NY).