Supplementary Information

Label-free purification and characterization of optogenetically engineered cells using optically-induced dielectrophoresis

Jia Yang,^{a,b,c†} Yanyu Gu, ^{a,b,d†} Chuang Zhang,^{a,b} Yuzhao Zhang,^{a,b,c} Wenfeng Liang,^e Lina Hao,^{d,*} Ying Zhao,^{f,*} Lianqing Liu,^{a,b} Wenxue Wang^{a,b,*}

- ^a State Key Laboratory of Robotics, Shenyang Institute of Automation, Chinese Academy of Sciences, Shenyang 110016, China
- ^b Institutes for Robotics and Intelligent Manufacturing, Chinese Academy of Sciences, Shenyang 110169, China
- ° University of Chinese Academy of Sciences, Beijing 100049, China
- ^d School of Mechanical Engineering and Automation, Northeastern University, Shenyang 110819, China
- ^e School of Mechanical Engineering, Shenyang Jianzhu University, Shenyang 110168, China
- ^fDepartment of General Surgery, Shengjing Hospital of China Medical University, Shenyang, 110004, China.
- †These authors contributed equally to this work and should be considered co-first authors.

*Corresponding authors: wangwenxue@sia.cn, haolina@me.neu.edu.cn, and zhaoy3@sj-hospital.org.

Section 1: Supplementary figures



Figure S1. The solution conductivity rising linearly with the increasing BSA mass concentration.



Figure S2. Theoretical simulation results of the electric field with a normal light pattern intensity

and those with an excessive light pattern intensity. The conductivity setting of a-Si:H along the X-axis direction and the sectional view along the diameter of the circular light pattern in the X-axis direction. (a– c) corresponds to the normal light pattern intensity, and (d–f) corresponds to the excessive one.



Figure S3. Validation of the successful expression of ChR2 in HEK293 cells. (a-d) Fluorescence verification of ChR2-expressing cells. The upper row is the bright-field images and the lower one is the fluorescence images. The left column is HEK293 cells after transient transfection and the right column is HEK293 cells without transfection. (e) Blue light-induced photocurrent verification of the HEK293-ChR2 cell and the HEK293 cell. The blue light irradiation is indicated by the blue shadow.



Transfection rate: ~ 61% Transfection rate: ~ 37%

Transfection rate: ~ 20%

Figure S4. Micrographs of transiently transfected HEK293 cell populations with three transfection rates. The upper row is the bright-field images and the lower one is the fluorescence images. The images from left to right correspond to the transfection rates of 61%, 37%, and 20%, respectively.



Figure S5. Physical map of the ODEP system and chip. (a) Overall system diagram. (b) Locally enlarged diagram showing the position of the chip in the system. (c) Physical map of the ODEP chip.



Figure S6. Schematic diagram of the cell separation process. (a) Selecting an area as the cell transportation destination that is emptied by injecting the isotonic solution with a hose. (b) Moving the circular light pattern to another area full of cells. (c) Setting the AC voltage frequency at 50 kHz to attract HEK293-ChR2 cells and repel the HEK293 cells. (d) Moving the circular light pattern followed by the attracted cells back to the destination area. (e) The attracted cells reaching the inlet of the hose. (f) Extracting and collecting the cells by the hose.



Figure S7. The mean cell radius of the cell population dropping linearly with an increasing ratio of engineered cells in the cell population.



Figure S8. Micrographs of cells cultured on the graphene transistors. The left column is the brightfield images and the right one is the fluorescence images. The lower row is the cells separated from the transiently transfected cell population, and such a cell population was used as the control and shown in the upper row.



Figure S9. The target imaged by the bio-syncretic graphene transistors based on the separated cells or the transiently transfected cell population.

Section 2: Supplementary text

The conductivity of a-Si:H varies with the light intensity. To interpret the influence of excessive light pattern intensity on the electric field and further the cell separation, assuming that the conductivity of a-Si:H along the Y-axis is constant, the conductivity of a-Si:H along the X-axis is set as piecewise functions. For the light pattern with normal light intensity, the conductivity of a-Si:H is set as shown in **Fig. S2a–b**, where the conductivities of the illuminated area and other areas are different constants, and there is no transition between them. For the light pattern with excessive light intensity, the adjacent area of the light pattern is also illuminated to a certain extent, so the conductivity of a-Si:H at the critical regions between the illuminated area and other areas are set to change linearly (**Fig. S2d–e**). The results show that the excessive light intensity leads to weaker non-uniformity of the electric field and a larger attraction area, and thus target cells cannot be effectively concentrated to the light pattern (**Fig. S2c and f**).