Electronic Supplementary Material (ESI) for Lab on a Chip.

Multifunctional cardiac microphysiological system based on transparent ITO electrodes for simultaneous optical measurement and electrical signal monitoring

Zhangjie Li,[#a] Kai Niu,[#a] Chenyang Zhou,[a] Feifan Wang,[a] Kangyi Lu,[a] Yijun Liu,[a] Lian Xuan[b] and Xiaolin Wang[*a,b,c,d]

[a] Department of Micro/Nano Electronics, School of Electronic Information and Electrical Engineering, Shanghai Jiao Tong University, Shanghai, 200240, China. E-mail: xlwang83@sjtu.edu.cn
[b] Institute of Medical Robotics, Shanghai Jiao Tong University, Shanghai, 200240, China.
[c] National Key Laboratory of Advanced Micro and Nano Manufacture Technology, Shanghai Jiao Tong University, Shanghai, 200240, China.
[d] National Center for Translational Medicine (Shanghai) SHU Branch, Shanghai University, Shanghai， 200444, China.

Corresponding author:

Xiaolin Wang
Email: xlwang83@sjtu.edu.cn; Tel: +86 (021) 3420-6683

Supplemental Movie S1: The original cardiomyocyte beating video and corresponding impedance signal on day 7.

Supplemental Movie S2: The comparison of impedance data with the cardiomyocyte motion recognized by the OHW software on the same time axis, along with a dynamic vector map and dynamic heat map of cardiomyocyte motion at the corresponding time.

Supplemental Movie S3: Under electrical stimulation at various frequencies, the cardiomyocyte motions recognized by OHW software, along with dynamic vector maps and dynamic heat maps of the cardiomyocyte motions.

Supplemental Movie S4: Field potential signals in both cardiomyocyte beating and resting states and corresponding dynamic heat maps.

Supplemental Movie S5: Original cardiomyocyte beating videos at different time points post-ISO addition, accompanied by corresponding dynamic vector and heat maps.

Supplemental Movie S6: Original cardiomyocyte beating videos at different time points post-NE addition, accompanied by corresponding dynamic vector and heat maps.

Supplemental Movie S7: Original cardiomyocyte beating videos at different time points post-VP addition, accompanied by corresponding dynamic vector and heat maps.

Supplemental Movie S8: Original cardiomyocyte beating videos at different time points post-QD addition, accompanied by corresponding dynamic vector and heat maps.

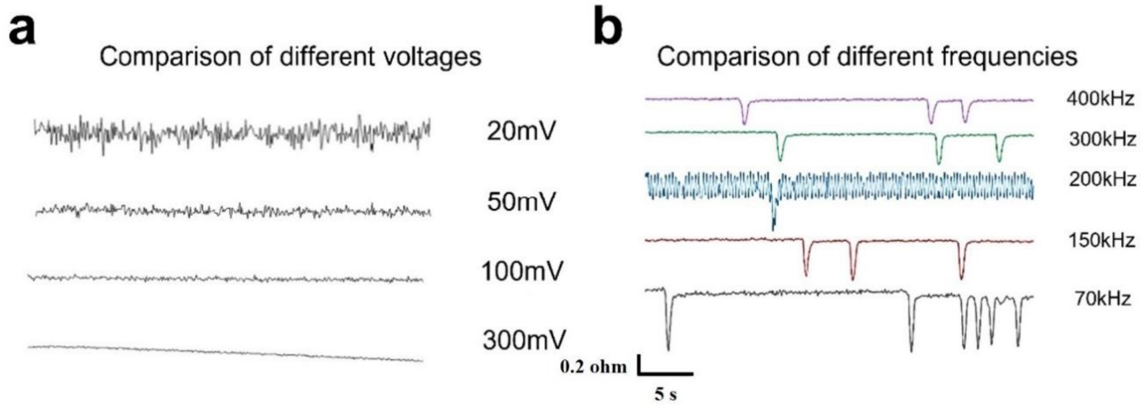

Fig. S1 (a) Impedance obtained at different voltages. (b) Impedance obtained at different frequencies.

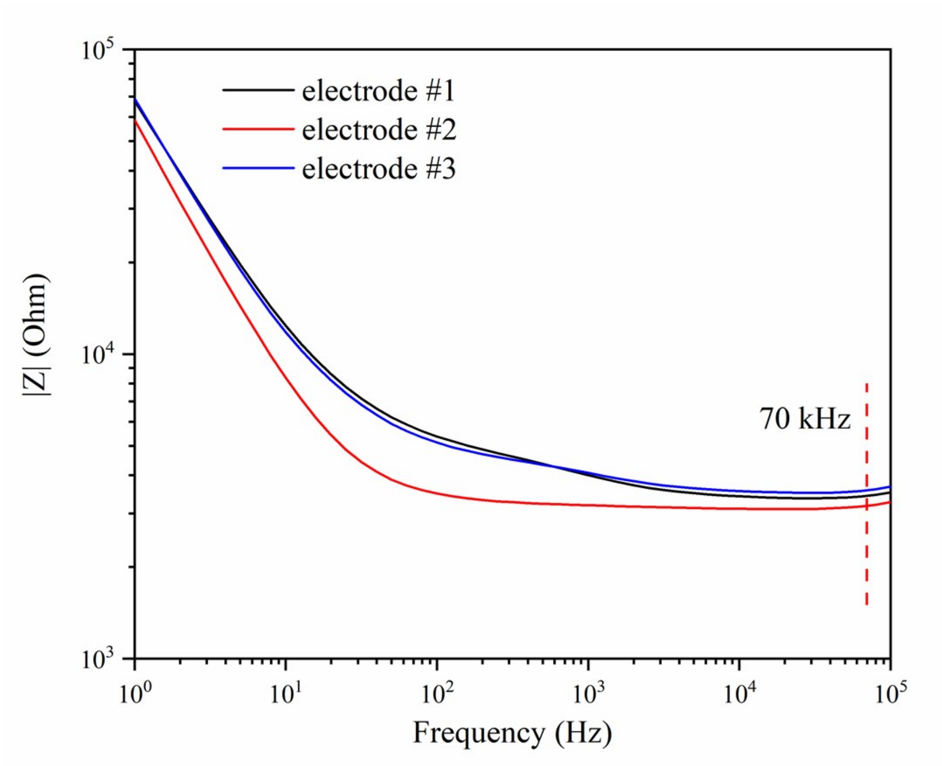

Fig. S2 EIS measured with electrode #1, #2, and #3 in PBS solution.

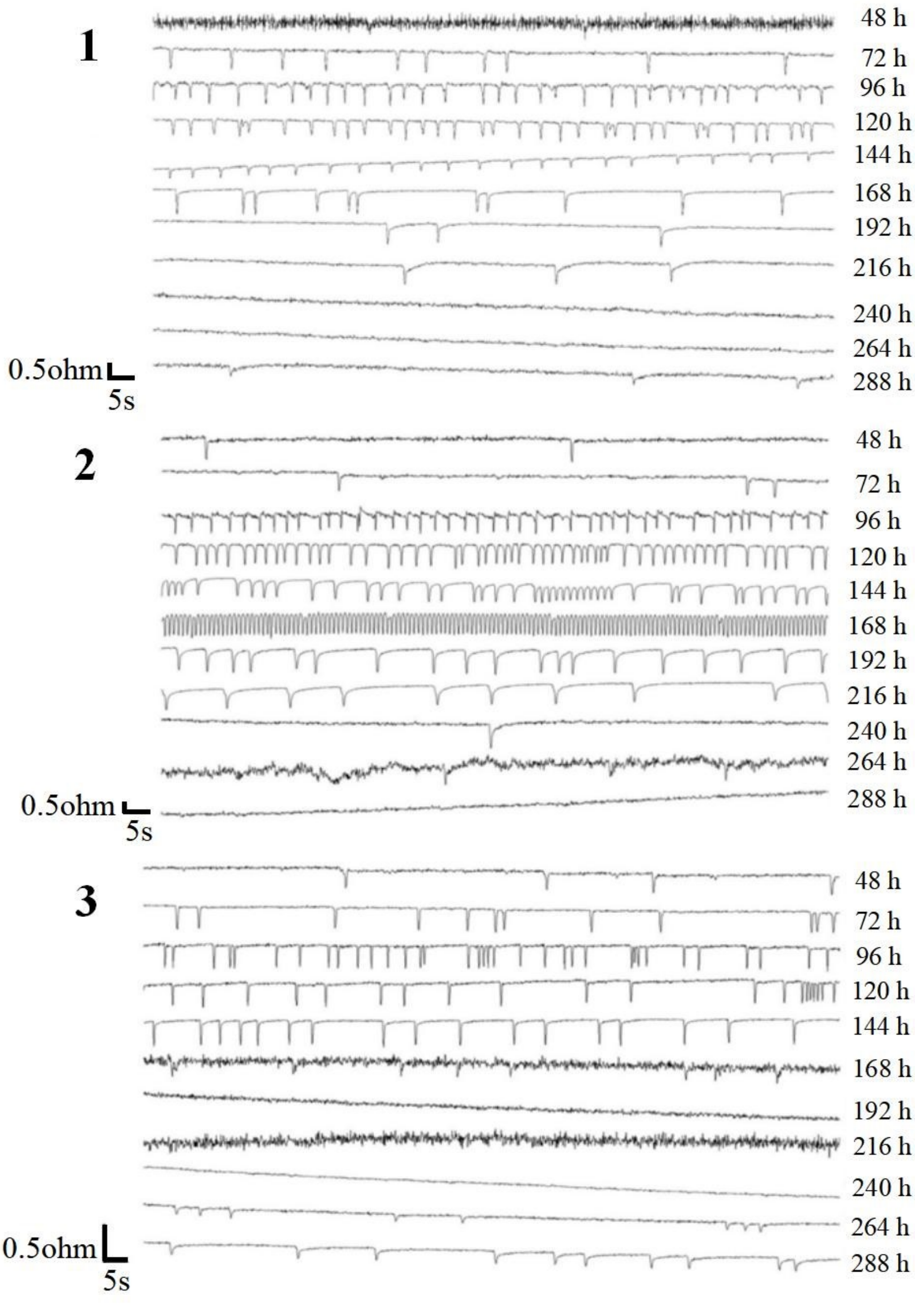

Fig. S3 The impedance changes recorded when culturing cardiomyocytes on the surfaces of three impedance measurement electrodes with differing structures.

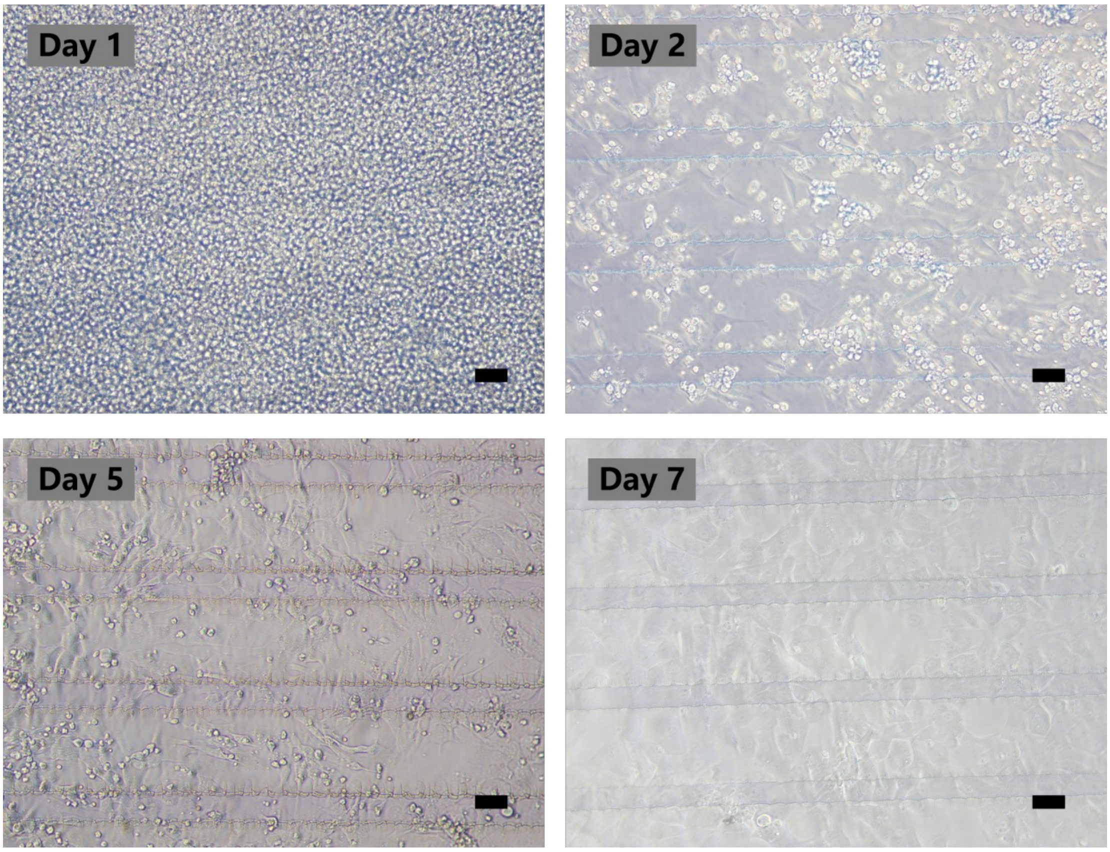

Fig. S4 Growth status of cardiomyocyte on the chip at different days (Scale bar:80μm)

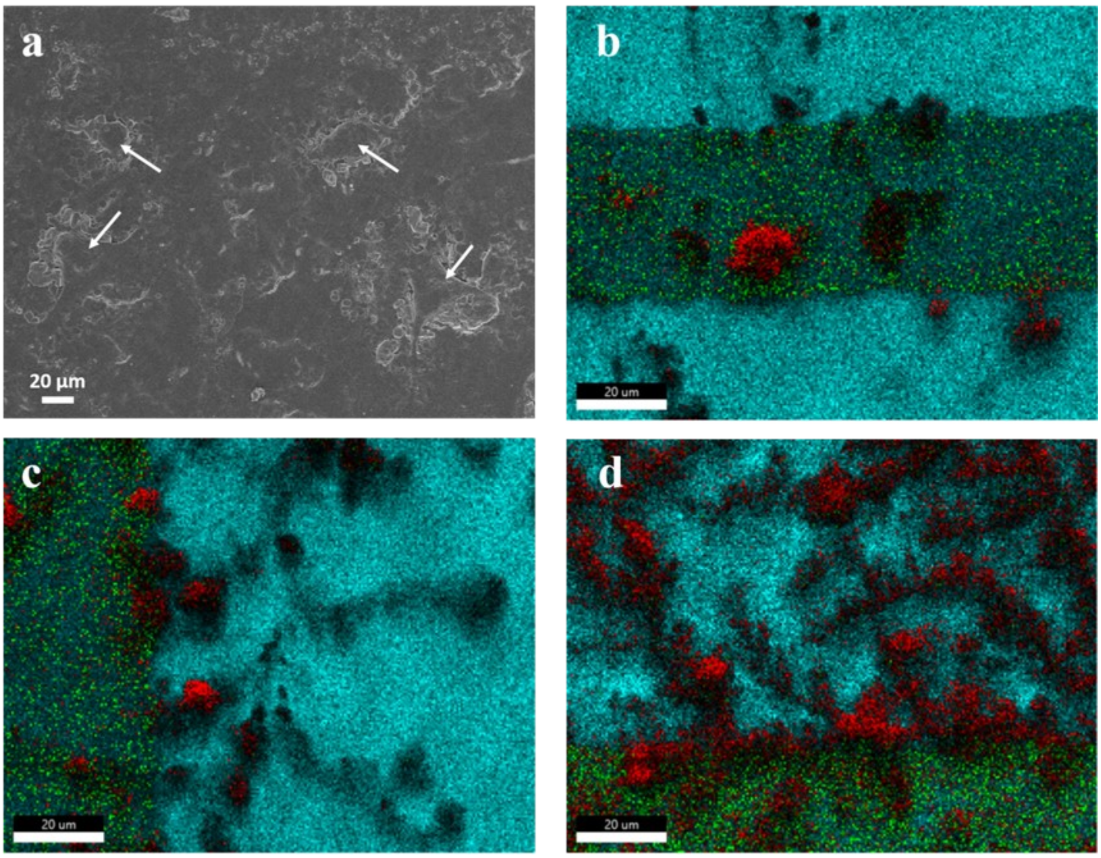

Fig. S5 (a) SEM image of cardiomyocyte on IDE. White arrows indicate cells. EDS mapping of IDE under different cardiomyocyte seeding densities: (b) 5×10^4 cells/mL, (c) 1.5×10^5 cells/mL, (d) 4.5×10^5 cells/mL. In the EDS mapping images, red represents C, indicative of cardiomyocytes; green represents Sn, indicative of ITO; and cyan represents Si, indicative of the glass substrate.

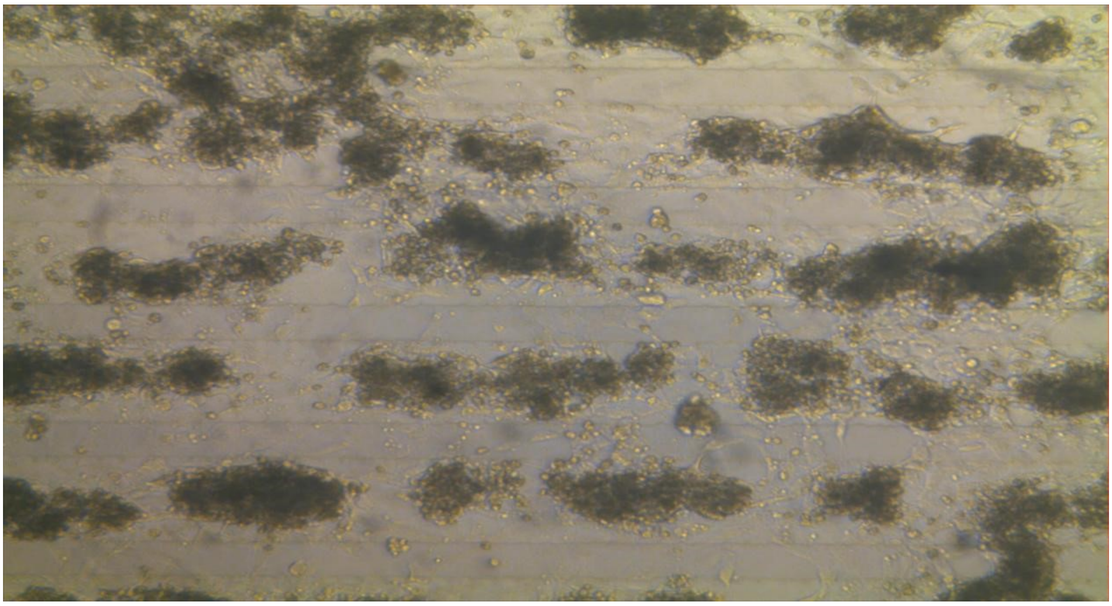

Fig. S6 Cardiomyocyte arrangement along the electrode orientation in the X-axis direction

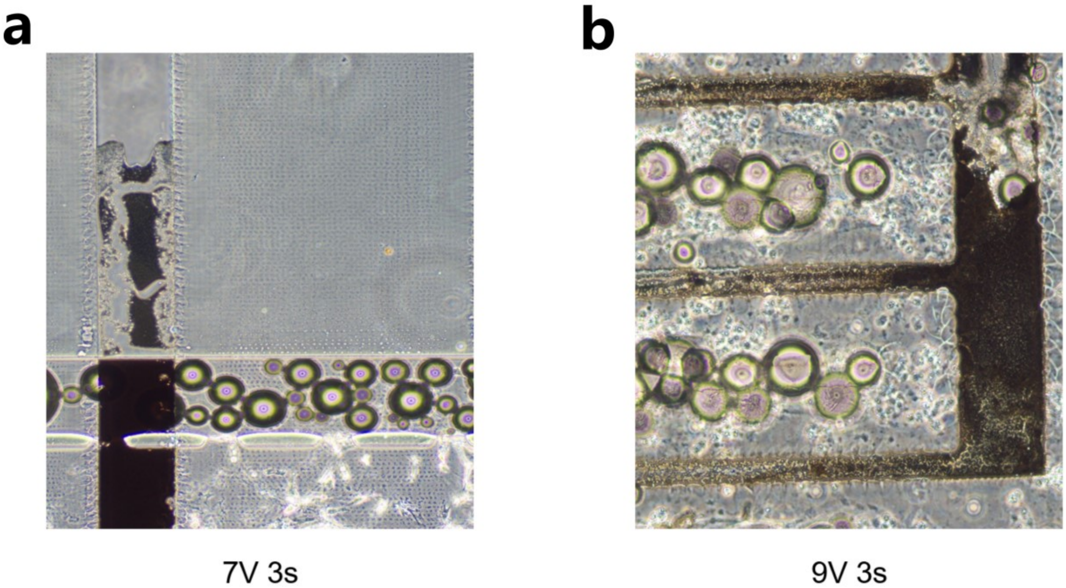

Fig. S7 At high voltages, ITO electrodes decomposed and DMEM medium underwent electrolysis.

Table S1. The parameters of the three IDE geometries

IDE structure	Number of fingers	Average finger length, μm	Average overlay length, μm	Finger width, μm	Interspace, μm	Electrode area, cm^2	Calculated K
#1	108	677.5	538.5	80	80	0.1629	0.276
#2	62	1329.5	1165.5	80	100	0.1397	0.266
#3	58	600.5	475.5	80	60	0.1571	0.531

Table S2. Parameters extracted from fitting EIS data with the equivalent circuit model

IDE structure	R_s, Ohm	C_p, μF	R_{trace}, Ohm	CPE, μF
#1	1042	0.172	3401	3.73
#2	983	56.24	3162	3.43
#3	830.6	0.174	3531	3.54

In addition to conducting EIS measurements on the three electrodes with different structures, we also performed theoretical calculations on the performance of IDE as a supplement to the main text. The IDE can be characterized using the cell constant (K) in, and K can be related to the solution resistance (R_s) and the capacitance (C_p) of the trace and IDE as follows:

$$K = R_s\sigma = \frac{\varepsilon_0\varepsilon_r}{C_p} \quad \text{(1)}$$

Where σ represents the conductivity of the PBS solution, ε_0 represents the vacuum permittivity, and ε_r represents the relative permittivity of the PBS solution[1]. It can be seen that K is inversely proportional to C_p. As C_p increases, the overall impedance decreases. Therefore, an IDE with a smaller K value is more in line with the requirement of minimizing IDE impedance.

For basic IDE geometries, K can be calculated from geometry according to the following equation:

$$K = 2\frac{\sqrt[3]{S/W}}{L(N-1)} \quad \text{(2)}$$

Where S is inter-trace distance, W and L are finger width and length and N is number of fingers in IDE[2]. Although, in this study, the three electrodes used for screening impedance electrode structures are not the basic IDE geometry and do not fully meet the applicable conditions of Formula 2. However, the K value calculated by substituting the parameters of the three electrodes into Formula 2 is still used as a reference for evaluating the performance of the three electrodes. The parameters of the three electrodes along with their corresponding K values are presented in Table S1. As can be seen from Table S1, electrode #2 has the lowest K value. As mentioned earlier, a smaller K value corresponds to a lower impedance which is corroborated by the blank impedance results of the IDE (Fig. S2). The lowest K value of electrode #2 can be attributed to its longest average overlay length. According to Formula 2, the K value and overlay length exhibit an inverse relationship.

To calculate the signal-to-noise ratio of the field potential signals, we choose the signals during cardiomyocyte resting as the noise, and the signal-to-noise ratio can be calculated using the following formula:

$$SNR = 10\log_{10}\frac{\overline{S^2}-\overline{N^2}}{\overline{N^2}} \quad \cdots\cdots(3)$$

where S represents the signal, N represents the noise, and given that $\overline{S^2} = 118$ and $\overline{N^2} = 17.6$, the calculation reveals that the Signal-to-Noise Ratio (SNR) is 7.56.

Reference

1. W. Olthuis, W. Streekstra and P. Bergveld, Sens. Actuators, B, 1995, 24, 252-256.
2. F. Qian, C. Huang, Y.-D. Lin, A. N. Ivanovskaya, T. J. O'Hara, R. H. Booth, C. J. Creek, H. A. Enright, D. A. Soscia and A. M. Belle, Lab Chip, 2017, 17, 1732-1739.