## **Supporting information**

# Scanning ion conductance microscopy study reveals the disruption of the integrity of human cell membrane structure by oxidative DNA damage

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Yuan Liu, Ph.D Email: <u>yualiu@fiu.edu</u> S1. Footprint area analysis of individual treated cells.



**Figure S1.** Effect of chromate treatment on the footprint area of individual attached cells. A) Bright-field optical image (40x) of the HEK293H cells treated with K<sub>2</sub>CrO<sub>4</sub>. No apparent changes in morphologies after 90 min of chromate treatment. B) Analysis of the footprint area: Untreated (2546±650.1  $\mu$ m<sup>2</sup>), K<sub>2</sub>CrO<sub>4</sub> [10  $\mu$ M] (3231±1174  $\mu$ m<sup>2</sup>), K<sub>2</sub>CrO<sub>4</sub> [100  $\mu$ M] (3396 ±1549  $\mu$ m<sup>2</sup>), K<sub>2</sub>CrO<sub>4</sub> [500  $\mu$ M] (2986±840.6). "n.s" P>0.05 with N=18.



### S2. Trypan blue exclusion tests on treated HEK293H cells.

**Figure S2.** Bright-field images for a series of trypan blue exclusion tests of cell viability. The HEK293H cells were treated with 10, 100 and 500  $\mu$ M K<sub>2</sub>CrO<sub>4</sub> for 90 min. The red arrows show dead cells.



#### S3. Additional SICM topography images of the fixed HEK293H cells.

**Figure S3.** SICM topography images of cells fixed with paraformaldehyde 4%. A) Treated HEK293H cell fixed after 90 min of treatment with 10  $\mu$ M K<sub>2</sub>CrO<sub>4</sub>. B) Untreated HEK293H cell. The red arrow shows damages on the cell membrane. Height profiles at the right side are along the dashed line marked on the topography images.





**Figure S4.** SICM images of live HEK293H cells. A) Topography (left), surface potential (middle) and surface potential distribution (right) of the cell membrane of the untreated cell. B) Topography (left), surface potential (middle) and surface potential distribution (right) of the cell membrane of treated cells. The red arrows show damages on the cell membrane. Height profiles (red color) are through the red dashed lines marked on the topography images.

#### S5. Extra data of Single-point time-resolved ionic current measurements of live cells.

Figure S5 A-C show individual current time traces. Figure S5D show the RMS noise of current, which is bigger at the beginning. There is no significant difference of current RMS noise between treated and untreated cells at the same time point.



**Figure S5.** Nanopipette single-point measurements. (A-C) The normalized ionic current time traces on untreated (red color) and untreated (blue color) cells. (D) Analysis of the RMS noise of the ionic current at 3, 10 and 16 min. N=4, "n.s" P>0.05 between treated and untreated groups at the same time point.

#### **S6. Immunoblotting results**



**Figure S6**. The level of  $\beta$ -actin protein detected by immunoblotting. A) Cell lysates of untreated and treated HEK293H cells were subjected to SDS-PAGE. The level of  $\beta$ -actin protein was detected by an antibody against human  $\beta$ -actin protein. The left and right lanes represent the  $\beta$ actin level in untreated and treated cells, respectively. The loading controls (bottom panel) were provided with the Coomassie blue stained protein bands. B) The relative levels of  $\beta$ -actin protein in untreated (left bar) and treated (right bar) cells. There is a statistically significant difference between untreated and treated (P < 0.05, n=3).