Electronic Supplementary Material (ESI) for Lab on a Chip

Support information

A vertically aligned carbon nanotube-based impedance sensing biosensor for rapid and high sensitive detection of cancer cells

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Investigation about the mechanism of cancer cell entrapment on CNT array

As entrapment of cancer cells on CNT arrays occurs in a short time, no sufficient time is assumed for adhesive protein secreting from cells membrane and in contrary, CNT nano size tips act as adhesive sites to entrap cancer cells. On the other hand high metastatic cancer cells (like SW48) have a deformable cytoskeletal structures[1-4] which could have a key role in the entrapment on CNT structures.

In other words the CNT elastic beams, as stopping agents, were deflected during the cells motility which may result in the entrapment of cells with deformable structure (such as cancer cells) on these nano arrays. To investigate the validity of this suggested mechanism, one must study the entrapment of stiffer cells on CNT arrays. For this purpose, the SW48 cells were fixed by glutardhyde. Fixation of the cells results in an increment of their stiffness which leads to a dramatic increase in the cytoskeletal Young's modulus[5-6]. After the fixation process, the cells were flown on the surface of vertically aligned CNT arrays with the speed

of 0.5 ml/sec. This process was repeated for live SW48 cells with the same concentration. Figure 1S presented the SEM image of entrapped fixed (1S-A) and live (1S-B) colon cancer cells. The fraction of live entrapped cells is about 15 times more than fixed ones which may confirm the critical role of cell deformability in their entrapment on CNT arrays. The high speed of cells flowing on CNT arrays resulted in the escape of rigid cells from the CNT arrays whereas more deformable cells such as live cancer cells were entrapped with higher efficiency.

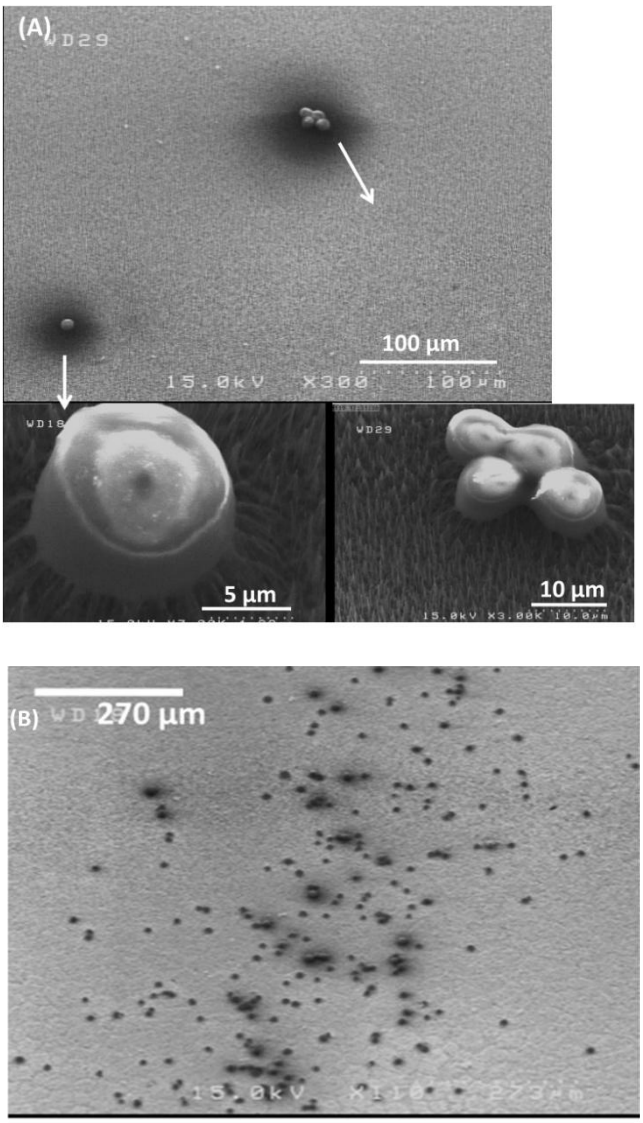

Figure 1S. SEM image from the entrapment of (A) fixed and (B) live SW48 cancer cells with the same concentration on MWCNT arrays. The fraction of entrapped fixed cells is observably lower than live ones.

An interesting point is that vertically aligned MWCNTs have hydrophobic surface[7]. But all of eyukairotic cells (such as cells we investigated on) have hydrophilic membrane[8]. So, cells and CNTs might not have any hydro chemical attraction for adherence to each other in such a rapid time. In addition, for better investigation on the effect of chemical bonding in cell entrapment we investigated the effect of a chemical anti-adhesive material, BSA (bovine serum albumin), on the fraction of cancer cells entrapment. For this aim, two separate cell solutions with the same cell concentration were flown on two separate CNT surfaces in a manner which BSA was added in just one of the cell solutions. The fraction of cells entrapment on CNT arrays was the same for both of them which confirm the non-crucial effect of chemical adhesive bonding in cells entrapment on CNT arrays. As a result chemical processes might not have a key role in cells attachment to CNT surface. On the other hand mechanical interaction of such deformable cancer cells by elastic and deflectable CNTs might be the important parameters in their attachment.

Effect of Ni catalyst on cell trapping

The presence of nickel in the top side of the CNTs is due to the tip-growth mechanism in our PECVD unit. Although Ni acts as a catalyst during the growth, it is usually coated with a few layers of carbon and hence it is not directly exposed to cells. We believe that the round and smooth shapes of the tips could prevent the cell membranes to be torn by the sharp side walls of nanotubes. In addition, it might have a favorable contribution in the electrical interaction between CNTs and cell membrane. The hemispherical shape of Ni catalyst (as shown in figure 2S-A) would lead to a smooth penetration into the cell membrane (as tested by LDH assay) and extract signal from inside the cell.

Contrary to Ni/MWCNTs which have shown to be biocompatibile[10], the Ni-removed CNT show some low level of cytotoxicity[9]. We have also observed that the removal of the nickel

from the top side of the CNTs would lead to smaller electrical signals which could be due to the lack of electrical contacts between the CNT tips and the cells. Finally we could result that Ni catalysts have constructive role in mechanical, biological, and electrical parameters of CNT-ECIS.

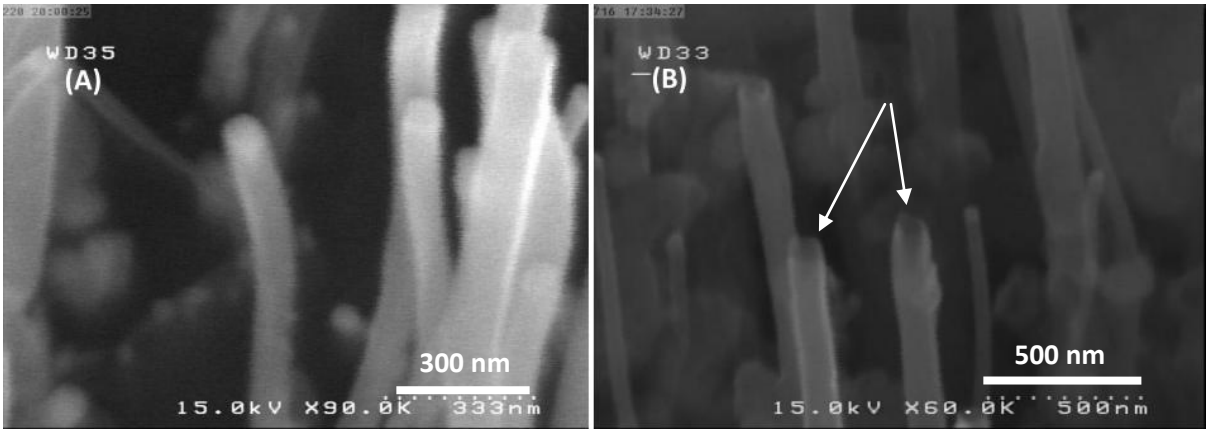

Figure 2S. High magnification SEM images of, (A) Ni/MWCNT in which the Ni has a dome shape. (B) The sharp side walls of Ni removed MWCNT.

Single-cell deformability has recently been recognized as a unique label-free biomarker for cell phenotype with implications for assessment of cancer invasiveness[11].The presented device has practical potential for high-throughput deformability based diagnosis of cancer cells from healthy ones to obtain viable target cells of interest in cancer research, immunology, and regenerative medicines. We can divide the application of CNT-ECIS in two categories which we are being developed. As an immediate application, we are trying to use these devices to distinguish between various metastatic stages in cancer transformation and hopefully detect cancer cells at early stages of disease. The current paper describes a method

to observe (study) the electrical signals and correlate them with different cancer cell stages based on CNT-ECIS devices.

As a second category, the level of cell entrapment with respect to the metastatic grade as well as the type of the cancerous cells could be investigated by the device. We have observed that higher metastatic cells are attached to the CNTs in a more remarkable fashion and hence they can be counted to have an estimation of the grade of the metastasis of the cells. Also this device could be applicable in detection of rare cells such as Circulating Tumor Cells (CTC) in blood samples. Finally, we believe that the rapid response of such sensors could be applicable for monitoring some fast necrosis processes of the cells as well as on-line monitoring of drug effect on the viability of cancer cells. Further study about the cancer cell entrapment on vertically aligned CNT arrays as well as new generations of CNT-ECIS is being pursued.

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