Fourier Transformer Infrared Spectrometer Microspectrometry Revealed Variation of Epithelial Ovarian Cell Lines

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Fourier transformer infrared spectrometer (FTIR) can be used to detect the molecular vibration to analyse the distribution of functional group. The FTIR detections are highly sensitive and are non-destructive to biological sample. Before the infrared experiments, we cultured ovarian cell line, IOSE, and cancer cell lines, A2780, and CP70 with 5-AZA-2'-deoxycytidine (5-aza-dc) known to inhibit DNA methyltransferase (DNMT). 5-aza-dc results in demethylation of CpG islands on DNA of both A2780 and A2780, and then causes upexpression of protiens. (Table.1) Further analysis indicated that the protein amount of IOSE dose not appear significant variation, which proves that 5-aza-dc would not influence normal ovarian epithelial cells.

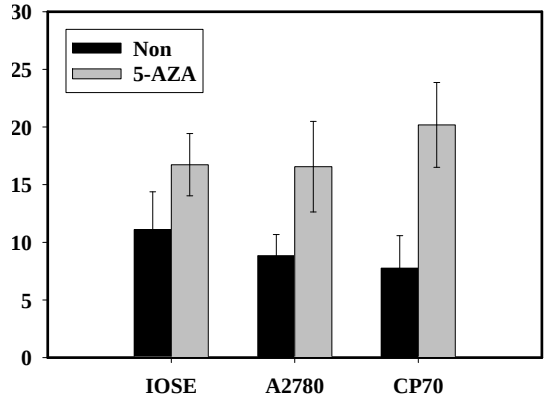

Table 1: Expression of both Amide I and Amide II which stand for protein component in A2780, CP70, and IOSE. After treating 5-aza-dc, the spectra of cells were captured by FTIR. The amount of expression was determined by integration from 1470 cm^{-1} to 1730 cm^{-1}, and then normalized: Log $(1/R_{Protein})$/ Log $(1/R_{Lipid})$.

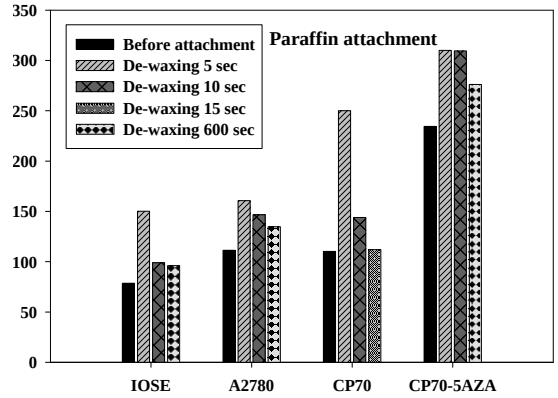

Table 2: Integration from 2800 cm^{-1} to 3000 cm^{-1} of paraffin attachment rates on each ovarian cell line. The results of paraffin attachment of IOSE and A2780 reveal dominantly that remnant exist even if de-waxing after 600 sec, but remaining paraffin on CP70 membrane is rinsed away in 15 sec. As same as IOSE and A2780, CP70 treated with 5-aza-dc presented the result against CP70 without treatment.

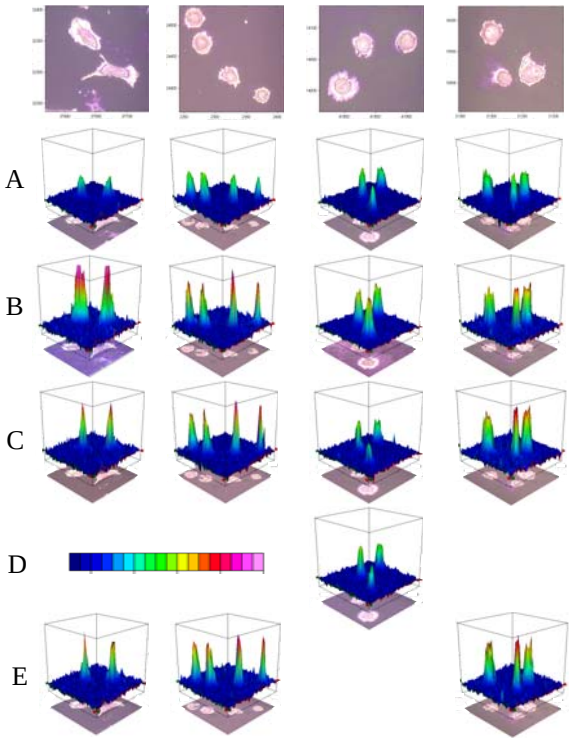

Fig. 1: The paraffin amounts compared among ovarian normal cell line IOSE, cancer cell lines CP70 and A2780 (A) before attachment, at (B) de-waxing 5 seconds, (C) de-waxing 10 seconds, (D) de-waxing 15 seconds, and (E) de-waxing 600 seconds. After wax affixed on membrane, the cells were washed with xylene in each different time interval. The amount of paraffin was determined by integration of Log(1/R)[a] from 2800 cm^{-1} to 3000 cm^{-1}.
[a] Log(1/R) = Log(100/%R), %R = Reflectance value of infrared energy. There is a linear relationship between Log(1/R) and concentration of component.

In the other hand, we use paraffin to attach ovarian cells, de-wax by xylene, and then detect the variety of amounts of paraffin by FTIR. (Table.2, Fig.1) The CP70 cell line reveals that paraffin easily comes off in 15 seconds but other two cell lines remain even in 600 seconds. Against the result of non-treatment CP70, the CP70 cells treated with 5-aza-dc represent the same consequence as IOSE and A2780. These data show that 5-aza-dc affects the expression of CP70, and it is extremely probable on biological constituents of membrane such as transporter.