| 研究区分 | 教員特別研究推進　教育推進 |
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| 研　究<br>テーマ | The effect of diesel exhaust particles on lung epithelial tight junction proteins | | | | |
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**講演題目**

Establishing the electrical parameters and culture conditions for A549 cells

**研究の目的、成果及び今後の展望**

Air pollution is responsible for the morbidity and mortality of millions of people each year. It is important to understand how air pollutants, such as diesel exhaust particles affect the cells of the airway and contribute to illness. The cells of the airway are composed of epithelial cells as well as undifferentiated cells comprising the interface separating the outer compartment (the airway lumen) from the inner compartment of the body. Bordering epithelial cells have complexes of proteins called the tight junction that both act as a barrier as well as regulating the passage of small molecules between the cells. When the tight junctions are disrupted, it may compromise the epithelial barrier and affect the polarization of the epithelia. For this reason, the aim of this research was to investigate the effect of carbon black nanoparticles (found in diesel exhaust) on the epithelial integrity of monolayers of human airway epithelial cells (A549 cells). As a first step, it was necessary to establish culture conditions. A549 cells were cultured on inserts in 12 well plates for 1 week, 2 weeks, 3 week, or 4 weeks in Ham's/F12 medium supplemented with 10% FBS, and 1% penicillin/streptomycin. Transepithelial conductance, equivalent short-circuit current, and dilution potential were measured at each time point to establish baseline parameters for these cells. Compared to 1 week of culture, 4-week cultured A549 cells had significantly lower conductance, showing that the barrier function increased with culture time. In addition, while the relative permeability of NaCl did not change overall, the permeability of $Na^+$ and $Cl^-$ decreased with longer culture time. From this data, it can be concluded that 1 week of culture is likely not sufficient as the electrical parameters of the tight junctions change over time. As a next step, using the established culture conditions, the cells will be treated with carbon black particles and the effect on the barrier will be examined by Ussing chambers. In addition, the effect of the treatment on the tight junction proteins (such as occludin) will be investigated by fluorescence microscopy.