
Poster: free communications

A simple and inexpensive *in vitro* model to study the effects of hypoxia on pulmonary epithelial and microvascular endothelial cells

Sara Signorelli⁰, Paul Jennings⁰, Martin Leonard¹, Walter Pfaller⁰

⁰ Innsbruck Medical University (Innsbruck) (AT); ¹ University College Dublin (Dublin) (IR)

e-mail: sara.signorelli@i-med.ac.at

Hypoxia is associated with a number of chronic lung diseases, a major cause of mortality in western populations. Exposure to chronic hypoxia results in pulmonary hypertension and changes in the structure of pulmonary arteries in animal models. *In vivo* test methods are expensive, animal intensive and time consuming. In addition, animal models may not be readily suited to detailed investigations at a cellular and molecular level. Therefore, we have developed a simple and inexpensive *in vitro* system which can mimic the lung environment and allow us to investigate the responses of alveolar epithelial cells (A549) and microvascular endothelial cells (HMEC1) exposed to hypoxia, as these cell types are a major contributors to chronic pulmonary diseases. A549 cells and HMEC-1 cells were exposed to 15%, 7% and 1% oxygen for 24 hours and the effects of hypoxia on gene expression, protein secretion and transcription factor activation were investigated. Specific gene expression was assayed by real time PCR. Transcription factor activation and protein secretion were determined by Enzyme Immuno Assays (EIA). Hypoxia resulted in an induction of cAMP Response Element Binding Protein (CREB) in both cell types. CREB responsive genes were induced under hypoxia conditions in HMEC-1 cells but not in A549 cells. Both cell types demonstrated hypoxia induced secretion of Endothelin-1 (ET-1), VEGF and IL-6. These results demonstrate that both epithelial and endothelial cells can contribute to hypoxia pulmonary hypertension, vascular remodeling and inflammation. Furthermore, these findings suggest a key role of CREB in hypoxia cell specific responses and provide important information for the further elucidation of the molecular mechanisms involved in hypoxia-associated pulmonary diseases. In conclusion, the developed system allows us to study *in vitro* the effect of different oxygen concentrations on the regulation of intracellular signaling pathway in specific cell types.

Keywords: hypoxia, pulmonary hypertension, CREB, ET-1, VEGF, IL-6