



**INVESTIGATIONS INTO THE ROLE OF  
ZINC IN NORMAL AND ALLERGIC  
RESPIRATORY EPITHELIAL CELLS AND  
TISSUES**

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# ABSTRACT

## ABSTRACT

This thesis describes an investigation of the physiological role of Zn within the airway epithelium. Zn is an essential biometal required for the healthy functioning of the body. Damage and shedding of airway epithelial cells (AEC) is a distinct feature of asthma hence, protection of this tissue is likely to have beneficial implications for the management of this disease. Several studies report a hypozincaemia in asthmatic patients, suggesting that decreased Zn levels may contribute to increased oxidative stress, chronic inflammation and decreased protection of the airway epithelium.

The distribution of labile intracellular Zn in the respiratory epithelium was measured by Zinquin, a novel Zn specific fluorophore. The pseudostratified ciliated epithelium was found to be continuously lined at the luminal surface by an intense region of Zinquin fluorescence, extending from the trachea to the bronchioles, but fluorescence was lower in the alveolar tissue. The positioning of intracellular Zn at a region which is most exposed to oxyradicals and other cytotoxins would allow Zn to act as a cytoprotectant in these cells.

Zn was found to be essential for respiratory epithelial cell (REC) survival since a decrease in intracellular Zn levels, mediated by the Zn chelator TPEN, resulted in the activation of caspase-3 activity and enhanced cell susceptibility to oxyradical-induced apoptosis. Increased intracellular Zn levels, via the Zn ionophore sodium pyrithione (NaPYR) and exogenous ZnSO<sub>4</sub>, inhibited caspase-3 activation and apoptosis.

A murine model of allergic airway inflammation using BALB/c mice treated with ovalbumin (OVA) was established and used to determine the effects of mild nutritional Zn deficiency on airway inflammation and airway hyper-responsiveness (AHR). Mice were given Zn normal (ZN) or Zn limited (ZL) diets before and during induction of inflammation. ZL mice had increased systemic and local eosinophilia, mucus cell hyperplasia and AHR compared to the ZN mice. Eosinophilia was pronounced in the allergic ZL mice suggesting that Zn deprivation negatively influenced the pathogenesis of allergic airway inflammation. Increased levels of AHR in the ZL mice were reversed by 14 days of Zn repletion.

Airway inflammation significantly decreased Zinquin fluorescence and increased pro-caspase-3 protein in AEC of OVA-treated mice. These mice also had increased levels of apoptosis as detected by enhanced levels of active caspase-3 (AC3) and increased cleavage of cytokeratin 18 (CK18), a specific substrate of caspase-3. Interestingly, ZL OVA-treated mice had a more pronounced increase in AC3 protein, cleavage of CK18 and the presence of apoptotic bodies within the airway epithelium.

This thesis provides new data on the role of Zn in the respiratory system which should lead to a greater understanding of the association between Zn deficiency and airway disease. Clinical implications of these studies suggest the monitoring of Zn levels in subjects with varying severity of atopy and asthma and the potential for Zn supplementation as a possible therapeutic in the treatment of allergic airways disease.