Analysing biological activity of dust from moisture damaged buildings: Relation of microbial components with toxicity

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Moisture damages in buildings occur frequently all around the world. Exposure to a moisture damaged environment has been associated with increased occurence of respiratory infections and upper respiratory tract symptoms, cough, wheeze and dyspnoea (WHO 2009). Available moisture in building materials enables the growth of micro-organisms, resulting in potential exposure to spores, spore fragments, secondary metabolites and cellular components of microbes.

Collecting settled dust is a way to get an integrated sample of particles that once were airborne. The sampling time extends typically over several weeks, which improves the representativeness of the sample compared to short time sampling. However, settled dust lacks both the gaseous phase and the smallest, non-settling particles present in indoor air. The aim of this study was to compare the concentration of microbial components in settled dust samples with toxicity and inflammatory responses caused by exposure to the dust in a cell culture model.

Methods

Dust samples were collected from moisture- and mold damaged schools (n=14) and reference schools (n = 11) in Spain, The Netherlands and Finland. Settled dust was collected to cardboard boxes for 8 weeks, the sample was vacuumed on a filter, suspended in a dilution buffer, and stored frozen. Before exposure experiments, the samples from each school were pooled together, filtered to remove the largest particles, aliquotted and frozen again. The concentrations of ergosterol and endotoxin were measured with GC-MS-MS (Lappalainen *et al* 2008) and Limulus assay (Jacobs *et al* 2012), respectively.

A mouse macrophage cell line (RAW264.7) was exposed to four doses of dust suspension for 24 hours. The viability of the exposed cells was analysed by measuring the function of the mitochondria [3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT)-test] and permeability of the cell membrane [propidiumiodide (PI) –staining] (Markkanen *et al* 2008). The concentrations of proinflammatory mediators [nitric oxide (NO), tumor necrosis factor (TNF)α, interleukin (IL)-6 and macrophage inflammatory protein (MIP)2] were analysed from the cell culture medium (Huttunen *et al* 2003).

The concentration of microbial components in dust correlated significantly with both toxicity and the ability to induce inflammatory responses (Table 1). Interestingly, the correlation between toxicity and ergosterol was strongest among the samples collected from damaged schools, whereas the correlation between toxicity and endotoxin was strongest within the reference schools. However, associations between microbial components and dust levels should be considered simultaneously in further analyses.

Table 1. Spearman correlation between concentrations of microbial components and biological activity of the settled dust samples collected form moisture damaged and reference schools in three European countries.

Toxicity	Endotoxin	Ergosterol
MTT-test	-0,836*	-0,724*
PI-exclusion	0,808*	0,734*
Inflammatory response		
NO	0,841*	0,731*
IL-6	0,857*	0,792*
TNF	0,848*	0,788*
MIP2	0,833*	0,768*

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