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EMERGING RISKS AND CHALLENGES ON ENVIRONMENT, HEALTH AND SAFETY

Allied Health Sciences School of Polytechnic Institute of Porto, Portugal

Health risks following wheat dust exposure during agricultural work - Focus on *Fusarium* spp

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INTRODUCTION:

Although inflammatory and innate responses are essential for protective immunity against infection, persistent inflammation in the lungs promotes respiratory diseases. Chronic exposure to high concentration of organic dust can promote such respiratory pathologies. One of the most concerned occupational populations is that of agricultural workers, in particular grain workers who are highly exposed to organic dust during their activities. This population is known to develop complex respiratory diseases due to exposure to grain dust, a complex mixture of fungal particles, bacteria, insect compounds, animal wastes, inorganic compounds(silicates), chemicals, gases and fumes (Linaker & Smedley, 2002).The grain dust components responsible of these pathologies are still unknown. However, *Fusarium* spp., particularly *Fusariumculmorum* is one of the most predominant fungal species present in grain dust. *Fusarium* spp. are responsible for infectious, allergic and toxic effects in humans. Yet, the innate immune response to these species is poorly characterized.

Innate immune cells such as alveolar macrophages lining the respiratory tract and dendritic cells (DCs) located within the epithelium and interstitium of the lung, form the primary line of defense against invading microorganisms. These cells sense microorganisms via pattern recognition receptors (PRRs), which are specialized in the recognition of conserved microbial structures called microbial associated molecular patterns (MAMPs). Upon microbial sensing, innate immune cells produce a panel of cytokines and chemokines that play a key role in the initiation, amplification and regulation of the inflammatory and innate and adaptive immune responses.

OBJECTIVES:

Our objective is to characterize the response of innate immune cells to *Fusariumculmorum* and to identify the PRRS involved in host-fungus interaction.

MATERIALS AND METHODS:

Fusariumculmorum 2156 (thereafter called *Fc*2156) was isolated by Agroscope from Swiss wheat samples and characterised in our lab.*Fc*2156 was grown on potato dextrose agar plates. Spores were collected and stored for 18 hours in PBS at 4°C or 22°C before usage. *Fc*2156was used as total preparation or centrifuged to isolate spores and supernatant. Bone marrow precursors were cultured for 7 days with granulocyte-macrophage colony-stimulating factor (GM-CSF) to obtain bone marrow-derived dendritic cells(BMDC). BMDCs ($2.5.10^6$cells/ml in96-well plates) were exposed for 24 hours to lipopolysaccharide (LPS, 10 ng/ml) as control and*Fc*2156preparations (total preparation, spores and supernatant) at an equivalent multiplicity of infection (MOI, *i.e.* a cell/spore ratio) of 0.01, 0.1 and 1. The concentrations of tumor necrosis factor (TNF), interleukin (IL)-6, IL-12p40, macrophage inflammatory protein (MIP)-1α and MIP-1βin cell culture supernatants were quantified by Luminex.

RESULTS AND DISCUSSION:

Total preparation and spores, but not supernatant, of *Fc*2156 induced vigorous production of TNF, IL-12p40, MIP-1α, MIP-1β and, to a lesser extent, IL-6 (Table 1 and data not shown). The stimulatory effect followed a bell-shaped response curve, with the highest concentrations of cytokines and chemokines obtained using

*Fc*2156at MOI 0.1. The lowest concentrations of mediators were measured using *Fc*2156 at MOI 1, due to cytotoxic effects.The differential effects of *Fc*2156 preparations were also observed using primary macrophages (data not shown).

Table 1 - Cytokine and chemokine release (in pg/ml) by BMDCsexposed to *Fusarium* spores

	TNF	IL-6	IL-12p40	MIP-1α	MIP-1β
Medium	7	3	19	4	23
LPS 10 ng/ml	52	200	975	323	910
***F.c* 2156MOI 0.01**	1188	81	857	946	3115
***F.c*2156 MOI 0.1**	5026	206	1478	7105	6594
***F.c*2156 MOI 1**	213	10	323	770	976

CONCLUSION:

F. culmorum spores strongly activate innate immune cells causing them to release cytokines and chemokines. It is well known that *Fusarium* spp. produce toxic secondary metabolites such as mycotoxins when exposed to stress conditions. This may account for the observed cytotoxicity and reduced production of inflammatory mediators (Stockmann-Juvala, Alenius, & Savolainen, 2008)when using high loads of *F. culmorum*. Work is in progress to identify the PRRs implicated in the sensing of *F. culmorum spores*.

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REFERENCES:

1. Linaker, C., & Smedley, J. (2002). Respiratory illness in agricultural workers. [Review]. Occupational medicine, 52(8), 451-459.
2. Stockmann-Juvala, H., Alenius, H., & Savolainen, K. (2008). Effects of fumonisin B(1) on the expression of cytokines and chemokines in human dendritic cells. [In Vitro Research Support, Non-U.S. Gov't]. Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association, 46(5), 1444-1451.