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The impact of dust in filter materials of respiratory protective devices on the microorganisms viability

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ABSTRACT

Plant biomass processed in a heat and power plant is a source of airborne dust present in the working environment. The aim of this study was to assess the influence of various levels of organic dust content in the filter material on the viability of microorganisms in model conditions, taking into consideration the workplace environment and the physico-chemical as well as the microbiological characteristics of the dust particles. The possibility of a biofilm developing on the reusable filtering facepiece respirators (FFR) in standard use by heat and power plant workers for respiratory tract protection was also investigated.

Standard reusable filtering half masks FFP3R and high-performance melt-blown fabrics were used in the study. The biomass dust characteristics were determined using an elementary analyser and a condensation particle counter with an electrostatic classifier. For quantitative analysis of microbiological pollution we used the plate count method with the microorganisms identified either using the API biochemical tests (for bacteria and yeasts) and the taxonomic key (for moulds). Biofilm development on the filtering half masks was assessed using SEM scanning microscopy. The viability of microorganisms (namely *Escherichia coli* ATCC 10536, *Staphylococcus aureus* ATCC 6538, *Candida albicans* ATCC 10231, *Aspergillus niger* ATCC 16404; *Bacillus subtilis* NCAIM 01644) on the filter materials with different dust content was measured using the quantitative method AATCC 100-2004 (2008).

In the plant biomass processed at the heat and power plant, a high concentration of bacteria (2.30×10^7 CFU g^{-1}) and a lower concentration of fungi (4.46×10^5 CFU g^{-1}) were detected. It was determined to be a good environment for the growth of microorganisms (the carbon to nitrogen ratio was 48:1). The presence of dust and the development of microbiological biofilms on reusable FFR, which is used by the heat and power plant workers for processing the plant biomass in increased humidity conditions, were also confirmed.

It was found that the viability of microorganisms on a filter material depends on the kind of microorganism and the dust content on the material. Model research showed that biomass dust in filter materials stimulates the growth of *E. coli*. Moreover, dust concentration above 21% inhibited the growth of *S. aureus* and *C. albicans* but had no significant influence on the growth of *B. subtilis* and *A. niger*.

1. Introduction

Traditional energy sources, such as coal, oil and natural gas, are constantly being depleted, which is why there is an increasing need for energy from renewable sources. An important fact contributing to intensifying the use of renewable energy sources was the necessity of implementing the provisions of the United Nations Framework Convention on Climate Change (1992) and the Kyoto Protocol (1998) on carbon dioxide reduction. In the European Union the regulations for these provisions are contained in Directive 2009/28/EC (2009), according to which renewable energy sources include: wind, solar power, geothermal energy, the energy of tides and sea waves, hydropower, gas obtained from landfills and waste, biogas and biomass. Biomass consists of easily biodegradable solid or liquid substances of plant or animal origin, mainly from agricultural or forest production.

Depending on the kind of biomass, various technologies are used for processing it however in all those processes the workers are exposed to harmful effects of organic dust. This threat affects

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workers involved in various tasks especially involved in boiler room and comminution devices operation, unloading and pouring of plant biomass (Lawniczek-Walczyk et al., 2012).

The organic dust accompanying plant biomass processing contains mineral substances, organic plant fragments, macro- and microorganisms with the substances they release (including bacterial endotoxins, glucans, mycotoxins, volatile organic compounds) making it a major source of harmful biological agents. Organic dust particles entering through the respiratory system of exposed workers can have toxic, irritating, allergenic, carcinogenic or fibrogenic effects leading to: chronic obstructive lung disease, asthma, chronic bronchitis, bronchial hyper responsiveness, organic dust toxic syndrome and irritation to mucous membranes, eyes and skin (Lacey and Dutkiewicz, 1994; Demers et al., 1997; Rusca et al., 2008).

The degree of hazard due to the effects of harmful biological agents contained in organic dust depends largely on the kind of raw material used and its transport and storage conditions (Lawniczek-Walczyk et al., 2012). Plant biomass stored for a long time in piles that are unsecured from the effects of atmospheric conditions easily absorbs humidity contributing to microorganism development. In humid biomass, mesophiles, actinomycetes and moulds (including the pathogen *Aspergillus fumigatus*) multiply with a particular intensity, for e.g. in woodchips reaching the concentration above 10^2 CFU g^{-1} (Sebastian et al., 2006). One of the solutions for reducing the workers' exposure to the effects of organic dust is the use of Filtering Respiratory Protective Devices (FRPD). Currently the FRPD, including filters and filtering half facepiece respirators (FFRs), in accordance with the European Union norms (Directive 89/686/EEC, 2000) are both disposable or reusable. During FFR use, water vapour and sweat are released, increasing humidity of the material, which in conjunction with proper temperature, provides favourable conditions for growth of microorganisms (Pasanen et al., 1994; Brosseau et al., 1997a, 1997b; Maus et al., 1997; Maus et al., 2001; Reponen et al., 1999; Wang et al., 1999; Jankowska et al., 2000; Majchrzycka et al., 2010). Viability of microorganisms on filter materials may also be affected by the presence of nutritional substances providing carbon and minerals present in plant biomass originating from organic and inorganic particles that were blocked during air filtering. The basic form in which microorganisms live in the air are bioaerosols, while on surface they grow as biofilms (Donlan, 2002). Due to the fact that the pathogenous microorganisms constituting a biofilm are able to release toxins and mycotoxins, their presence in the filter material of FFR may be a serious hazard to the workers using reusable FFR. So far, there are no studies on the influence of organic dust from plant biomass on the viability of microorganisms on FFR and their ability to develop biofilms during long-term usage and storage of reusable FFR in increased humidity conditions.

The aim of this study was to assess the impact of various organic dust level contents on the filter material on microorganism viability in model conditions, taking into account the factors present at the workplace and the physico-chemical and microbiological characteristics of dust. The possibility of biofilm development on the reusable FFRs in standard use by heat and power plant workers for respiratory tract protection was also assessed.

2. Materials and methods

2.1. Plant biomass

Plant biomass processed in a heat and power plant with maximum total heat power of 557 MW was used in the study. Three kinds of biomass were tested: willow wood chips, forest wood chips and sunflower pellets. For microbiological analyses, untreated specimens of 250 g mass were used. For physico-chemical tests, plant biomass in the form of homogeneous organic dust was used. For this purpose, the biomass of each kind was dried three times for 24 h at 70 °C, in decreased pressure conditions (100 mbar) in a vacuum drying chamber (VD 53, Binder, Germany) and grounded afterwards. An averaged specimen consisting of dust mixed at equal ratios from each kind of biomass was also prepared. Organic dust for model tests was prepared by subsequent sterilisation for 15 min at 115 °C and drying for 24 h at 70 °C under reduced pressure of 100 mbar in drying chamber (VD 53, Binder, Germany).

2.2. Tested FFR and filter material

For our study, standard FFRs were used. These were approved for workplace use in the EU in accordance with the provisions of Directive 89/686/EEC (2000), which evaluates the compliance of personal protective equipment (PPE) with basic safety and ergonomics requirements. FFR effectiveness was 99% at a flow rate of 95 l/min against an aerosol of solid particles (NaCl) and paraffin oil mist. FFRs were rated as protective class 3 (FFP3) and were reusable (R) in accordance with the requirements of the European Standard EN 149 (2009). Fig. 1 shows a construction scheme of the FFR used for the tests.

The tests for microbiological pollution measurements were conducted for 2 FFRs used during biomass processing by workers of the heat and power plant for 3 days during 8-h work shifts.

For microbiological evaluation using the plate count method, the layers a1-a4 and b1-b4 were used; the possibility of biofilm development was investigated microscopically for the layers: a1, b1 and b2 (Fig. 1).

For the model studies on microorganism viability, specimens of high-performance polypropylene melt-blown nonwovens were used. The fabrics were produced commercially for use as filter layers in FFR production. The average surface mass of the fabric specimens was (30 ± 3) g/m^3.

2.3. Microorganisms

Four strains from pure culture collections, namely the American Type Culture Collection (ATCC): *Escherichia coli* 10536, *Staphylococcus aureus* 6538, *Candida albicans* 10231, *Aspergillus niger* 16404; and *Bacillus subtilis* 01644 from the National Collection of Agricultural and Industrial Microorganisms (NCAIM), were tested in model studies for the survival of microorganisms on filter material. The strains were selected based on the taxonomic variety (gram-positive cocci, gram-negative rods, gram-positive bacilli, yeast, and mould). Selected strains were also characterised by their varying ability to survive in the environment based on the production of either endospores (*B. subtilis*), spores (*A. niger)* or just vegetative cells (*E. coli, S. aureus, C. albicans*). Moreover, the tested species, in accordance with the Directive 2000/54/EC (2000), were classified in group 2 of health hazards and can be potentially dangerous to people in the working environment.

The average density of the suspension of microorganisms (inoculum which was loaded on the filter material) ranged from 5.0×10^6 to 5.9×10^9 CFU ml^{-1} depending on the strains.

2.4. Physical and chemical characteristics of the plant biomass dust

Particle size distribution of an averaged biomass dust specimen was determined using a condensation particle counter (model 3775, TSI, USA) connected to an electrostatic classifier (model 3080, TSI, USA). The elemental composition of the biomass, taking into account carbon, nitrogen and phosphorus contents, was determined using an elementary analyser (type NE 2500, CE