



Positive associations between respiratory outcomes and fungal index in rural inhabitants of a representative sample of French dwellings

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ABSTRACT

Our study aims at estimating exposure to molds at home, based on microbial Volatile Organic Compounds (MVOCs) assessment, and evaluating its effect on respiratory diseases in a representative sample of dwellings. In the framework of a national campaign, indoor pollution was monitored in a sample of the 24 million dwellings of metropolitan France ($n=567$). 727 subjects answered to a standardized questionnaire on respiratory diseases and had MVOCs sampled in their bedrooms and a fungal index (FI) defined. Among the 431 dwellings with complete data, one out of three was contaminated by molds as assessed by a positive FI: 27.0% in urban, 38.2% in periurban and 34.9% in rural dwellings respectively. Positive associations were observed between fungal index and current asthma (8.6%) and chronic bronchitis-like symptoms (8.4%), especially in rural areas (OR = 2.95, 95%CI (1.10; 7.95) and 3.35, 95%CI (1.33; 8.48) respectively). Our study, based on objective assessments of fungal contamination, is in agreement with previous results suggesting mold-related respiratory effects. Moreover associations found among rural population could indicate specific pollution and impact in this environment.

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Introduction

Up to 50% of buildings are known to have dampness and mold problems. A large number of studies report associations between exposure to dampness or visible molds and respiratory symptoms (Mendell et al., 2011). It has been estimated that, out of the 21.8 million people reported to have asthma in the USA, approximately 4.6 (2.7–6.3) million cases are estimated to be attributable to dampness and mold exposure in the home (Mudarri and Fisk, 2007). However, in most epidemiological studies focusing on health effects of molds, buildings were considered as contaminated on the basis of visual detection (questionnaire reports and/or technician observations). Using these techniques can lead to a strong underestimation of the proportion of individuals exposed to molds: neither “hidden” contamination (development of molds behind wall paper or in ventilation filters) nor recent contamination, hardly visible, can be

detected. In order to overcome these limits, Moularat et al. (2008b, 2008c) have developed a specific chemical fingerprint for fungal development, based on the detection of a list of pertinent Volatile Organic Compounds (VOCs) characteristic of mold growth (microbial VOCs (MVOCs)). Using this index, Moularat et al. (2008a, 2011) have shown that around 70% of dwellings which present fungal development according to the fungal index (FI) was not considered as contaminated through visual detection. Therefore, in order to obtain a better estimate of the risk associated with molds, the use of objective methods to assess exposure became essential.

The rural environment has been shown to present a higher exposure to biocontaminants in homes and stables than the urban environment, in particular to endotoxins (Lee et al., 2006). Recent results have suggested that compared to urban children, rural children were also exposed to an excess of molds at home (Moularat et al., 2011; Toivola et al., 2004; Wady et al., 2004), particularly children living in farms (Schram et al., 2005; van Strien et al., 2004). In this environment, it has been hypothesized that under particular circumstances early life exposure to an excess of biocontaminants could protect from allergies (Braun-Fahrlander et al., 2002; Eduard et al., 2004; Schram-Bijkerk et al., 2005). It has also been suggested that such exposure could induce non-atopic asthma (Eduard et al., 2004; Hulin et al., submitted for publication) or chronic

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bronchitis in adults (Kiryuchuk et al., 2006; Post et al., 1998; Preller et al., 1995; Vogelzang et al., 1998) through exposure to other molds components than the allergens (MVOCs, mycotoxins). Despite these features, few studies have focused on the effects of indoor air pollution in the rural population.

Our study aims at evaluating exposure of French population to molds, as assessed with an objective index of fungal contamination based on MVOC measurements, as well as its relation with allergic and respiratory diseases in adults. We also investigated health effects of molds in rural, periurban and urban environments, using a representative sample of French dwellings, in order to look for differences according to the type of areas.

Materials and methods

Study sample

A national housing survey was carried out by the French Indoor Air Quality Observatory (OQAI) during the 2003–2005 period in order to assess indoor air quality in French dwellings. This cross-sectional survey collected information about 567 occupied dwellings, randomly selected to be representative of the 24 million principal residences in mainland France, and their 1612 inhabitants distributed in 74 cities in a three-stage clustering (Billionnet et al., 2011; Kirchner et al., 2007). The survey protocol was composed of health and house questionnaires and pollution measurements. All these information have been collected during the same week.

Each dwelling was geo-localized. The sample was divided into three categories regarding residential outdoor environment: rural, periurban and urban according to the land use, given by the European database CORINE Land Cover (EEA, 2007), and the population density around each dwelling (Malherbe and Letinois, 2009). Four criteria were used in this classification:

- The land cover (urban vs. agricultural or forestry) at the dwelling location;
- The major land cover within 500 m of the location;
- The percentage of built-up area (more or less than 30%) within 2 km of the location;
- The population density within 1 km of the location.

The reliability of this method was checked visually using satellite views (data not shown).

Health outcomes

A standardized self-administered questionnaire was filled in by individuals aged 15 years or more who lived permanently in the dwelling, corresponding to a population of 1234 individuals. This was derived from the European Community Respiratory Health Survey (ECRHS) questionnaires (<http://www.ecrhs.org/>) and included items about respiratory health, smoking status and socio-demographic data.

According to replies to the questionnaire, health outcomes were classified and defined as follows:

Current asthma: defined according to the definition of the ECRHS, namely a positive response to any of the three following questions: “Have you had an attack of asthma in the last 12 months?”, “Are you currently taking medicines for asthma (including inhalers, aerosols or tablets)?” and “Have you been woken by an attack of shortness of breath at any time in the last 12 months?” (Sunyer et al., 2004).

Chronic bronchitis: defined according to the definition of the British Medical Research Council, also used by ECRHS, namely the presence of chronic cough (cough during winter, on most days for at least 3 months each year) and chronic phlegm (phlegm from chest,

on most days for at least 3 months each year) were used to define chronic bronchitis (Sunyer et al., 2005). In agreement with a previous work (Bentayeb et al., 2010), chronic bronchitis-like symptoms (at least one of the 2 symptoms: chronic phlegm or chronic cough) were also considered.

Environmental exposure

A detailed questionnaire was filled in during the home visit and included information about the dwelling (building characteristics, environment, visible signs of molds), the occupants (age, education, income), and their daily domestic activities (e.g. place, duration, type of products used, smoking, opening window habits). This questionnaire was divided into two parts: one completed with the inhabitants and the other for the technician's observations.

In order to assess exposure to indoor air pollution, VOCs were measured during one week (7 days), in the main bedroom, by passive diffusion samplers (Radiello, Padua, Italy). Adsorbed VOCs were analyzed using gas chromatography (GC-3800 Gas Chromatograph, Varian, USA) for separation of VOC coupled with flame ionization (FID) for detection and mass spectrometry (MS – ion trap Saturn 2000, Varian, USA) for identification. Aldehyde-hydrazones formed in the cartridge were eluted by acetonitrile solvent and analyzed by liquid chromatography associated with a UV detector. Total VOC concentration was calculated as the sum of the assessed VOCs: 4 aldehydes (acetaldehyde, acrolein, formaldehyde, hexaldehyde), 12 hydrocarbons (benzene, 1,4-dichlorobenzene, ethylbenzene, n-decane, n-undecane, styrene, tetrachloroethylene, toluene, trichloroethylene, 1,2,4-trimethylbenzene, m/p-xylene, o-xylene), and 4 glycol ethers (2-butoxyethanol, 2-butoxyethylacetate, 1-methoxy-2-propanol, 1-methoxy-2-propylacetate).

In order to objectively detect presence of molds, we used the fungal index (FI) developed by the French Scientific and Technical Centre for Building (CSTB). This index aims at determining the presence or the absence of fungal development on the basis of the detection of specific VOCs (Moularat et al., 2008a, 2008b, 2008c). Selection of MVOCs was based on their origin and specificity toward fungal species or materials identified through laboratory experimentation (Moularat et al., 2008c). The fungal contamination index was then developed in order to aggregate all the data supplied regarding the presence or absence of 19 pertinent chemical tracers. The list of these markers can be found elsewhere (Moularat et al., 2008c). This includes two main observations: (1) VOCs emitted during biodegradation are not specific of mold contamination as they can come from other type of degradation (thermal, radiation, etc.); (2) VOCs emitted during metabolism can be specific from one species or one type of materials. Based on all the data supplied regarding the presence or absence of identified tracers in the VOC measurement made in the main bedroom, the FI was constructed from binary responses (presence or absence of specific target compounds) and can conclude to the presence of mold development in a dwelling.

Statistical analyses

Comparisons between groups in the case of population characteristics were tested with Wilcoxon – Mann Whitney two samples tests for continuous variables and Pearson χ^2 test or exact Fisher test for categorical variables. In order to study the buildings characteristics associated with detection of mold contamination, a logistic model was used with the FI as dependent variable, which included all studied characteristics and then selected them using the stepwise procedure to obtain the final model.

Logistic regression analyses using marginal models were used to investigate the associations between exposure to molds and

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