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Indoor air quality in mass catering plants: Occurrence of airborne eumycetes in a university canteen

Andrea Osimani, Cristiana Garofalo*, Vesna Milanović, Manuela Taccari, Lucia Aquilanti, Serena Polverigiani, Francesca Clementi

Dipartimento di Scienze Agrarie, Alimentari ed Ambientali, Università Politecnica delle Marche, via Brecce Bianche, 60131, Ancona, Italy

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ABSTRACT

The present study aimed to investigate the viable eumycetes population of indoor air sampled in a mass catering plant that produces up to 1200 meals per day. The combination of a culture-dependent and -independent approach applied in the food-processing environment was efficient for the identification of airborne eumycetes. *Penicillum, Cladosporium* and *Alternaria* were the dominant genera, while *Thysanophora, Aspergillus, Rhizopus, Annulohypoxylon, Arthrinium, Aureobasidium* and *Trametes* were found to be in minority. The size and composition of the eumycetes population found in this study do not raise specific concerns. Nevertheless, a quantitative and qualitative monitoring of these bioaerosol should be included in the progress and criticality reports of the HACCP system in the catering services where eumycetes can pose different risks, such as worsening of the overall quality of meals and/or of the work environment. Indeed, these reports are a useful tool in the preventive HACCP approach, provided that an adequate frequency of sampling is planned in order to quicken the pre-warning process.

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1. Introduction

The recent and dramatic changes in the lifestyle of families and individuals have led to a growing number of people who need to consume at least one meal away from the home using public catering (schools, hospitals, corporates). In the foodservice industry, therefore, the assurance of the quality of meals is fundamental in terms of both sensory and nutritional quality, as well as that of safety (Osimani and Clementi, 2016a,b). The quality of food depends primarily on the raw materials (Petruzzelli et al., 2014b) and on the storage and process operations and the environment; the latter is in fact responsible for risks arising from the potential chemical and biological contamination of work surfaces and air, as pointed out by several authors (Osimani et al., 2011, 2013, 2014; Wray, 2011; Brown and Wray, 2014; Curiel and Lelieveld, 2014; Osimani et al., 2015a,b). Risks related to air contamination are linked to the composition of bioaerosol which is composed of airborne biological particles including viruses and viable organisms like bacteria and fungi, fungal spores, pollen and by-products such as endotoxins (Stetzenbach et al., 2004). The most common microbiological parameters usually evaluated in bioaerosol are the following: total mesophilic bacterial count as an indicator of contamination of human and animal origin, and total eumycetes (moulds and yeasts) as an environmental indicator often related to the presence of high humidity and dust, reduced ventilation and poor air quality. The typical bacterial indoor air contamination is predominantly represented by members ascribed to the genera *Aeromonas*, *Bacillus*, *Micrococcus*/*Kocuria*, *Nocardia*, *Pseudomonas*, and *Staphylococcus* (Di Giulio et al., 2010; Xu and Yao, 2013), while the most common airborne eumycetes identified in indoor air usually belong to the genera *Alternaria*, *Aspergillus*, *Chaetomium*, *Cladosporium*, *Mucor*, *Penicillium*, and *Stachybotris* (Orman et al., 2005; Ayanbimpe et al., 2010; Di Giulio et al., 2010; Xu and Yao, 2013). Among the above-mentioned microrganisms, potential pathogens and spoilage species can be found; it is worth noting that some moulds can also be responsible for infectious diseases, hypersensitivity reactions, allergic reactions and intoxications. In addition, the presence of fungi in indoor air can result in the sick building syndrome (SBS) which encompasses a variety of symptoms such as nasal drainage, watering eyes, headaches, nausea, generalized malaise and airway infections (Cabral, 2010).

In order to avoid health risks for the consumer, the European food industry, must comply with specific regulations issued in the field of food safety. One of them, Regulation (EC) No. 852/2004,

* Corresponding author.
E-mail address: c.garofalo@univpm.it (C. Garofalo).

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requires that every food processing plant must be managed in accordance with the Hazard Analysis and Critical Control Points (HACCP) system principles. Hence, the microbiological monitoring of indoor air can play a key role for obtaining good quality food. In fact, air monitoring can provide information on the quality of the process environment during food manufacturing and allows for the study of microbiological air quality trends. The monitoring of airborne microorganisms can be active (mostly performed with calibrated impaction samplers) or passive and be quantitative rather than qualitative. A quantitative analysis can provide an estimation of the total amount of microorganisms present in known volumes of air, while a qualitative investigation can be useful for the study of the microbial ecology of indoor air. In both types of monitoring, the microorganisms present in representative volumes of air can be collected on suitable culture media and incubated under appropriate conditions in order to allow the formation of colonies. Since each colony is theoretically attributable to a single original microorganism, the number of colonies grown on agar media (indicated as colony forming units, CFU) is summarily related to the number of viable microorganisms present in the air sample; the results are usually then expressed as CFU/m^3 of air.

The activities of mass catering are characterized by a continuous supply of a wide range of raw materials. In addition, the procedures for food preparation can be complex consisting of several steps. As such, indoor air can be contaminated by microorganisms which could be introduced or transported within the different preparation areas and, in turn, could enter the finished product.

Many reviews have been published on the evaluation of the risks related to the presence of potentially harmful airborne microorganisms in indoor environments (Eames et al., 2009; D'Arcy et al., 2012; Eduard et al., 2012), thus highlighting the ever constant attention on this topic; notwithstanding, the dynamics that are responsible for the definition of the pool of microorganisms detected in different environments are still under study (Gandolfi et al., 2013).

The aim of the present study was to investigate the occurrence of viable eumycetes in indoor air samples collected in a university canteen through a culture-dependent method and Polymerase Chain Reaction-Denaturing Gradient Gel Electrophoresis (PCR-DGGE). To this aim, total eumycetes were counted on an appropriate agar media during winter and summer; the bulk cells were collected from agar plates and then analysed through PCR-DGGE.

2. Materials and methods

2.1. Description of the university canteen and activities

The study was carried out in a university canteen, located in central Italy (43°36′57″latitude, 13°31′07″longitude, and altitude of approximately 16 m a.s.l.), producing up to 1200 meals a day. The canteen is located in the city centre, surrounded by buildings and a small park composed of plants belonging to the typical Mediterranean vegetation. The facility is built on two floors: the ground floor and basement (Fig. 1). The basement hosts the rooms which are mainly involved in receiving raw materials, storage and processing, such as: the receiving area, the primary food preparation room (A), the fresh meat cold room (B), the cheese and fermented sausage cold room (C), the fruit and vegetable cold room (D), the frozen food cold room, the locker rooms for the staff, the warehouse, the disposal of wastes, and utensil and pan storage room. The ground floor houses the administrative offices, the dining room, the food and beverage distribution area, and the kitchen (E). A centralized heating, ventilation, and air conditioning system (HVAC) guarantees the air ventilation and heating of the canteen. The HVAC system (Thermac, Vigasio, Italy) is designed for an air capacity of 1500 m^3/h, and it is powered by a 0.5 HP engine. The system is

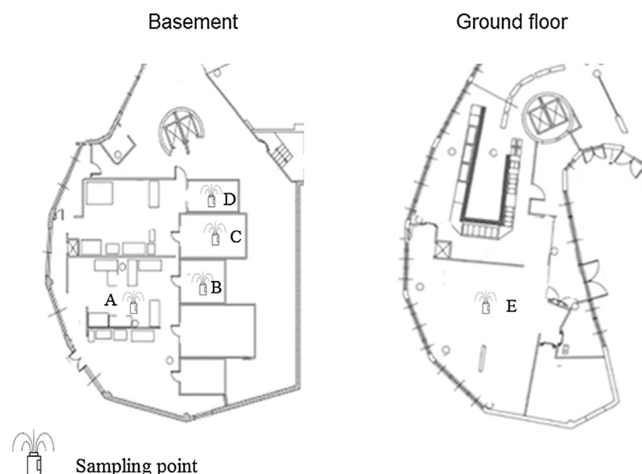

Fig. 1. University canteen map and air sampling points.
A – primary food preparation room, B – meat cold room, C – cheese and fermented sausage cold room, D – fruit and vegetable cold room, E – kitchen.

equipped with throwaway air fiberglass filters that are laid over each other to form the filter media which are reinforced with a metal grating. The filters are replaced with new ones every year.

The raw materials are quickly stored in the appropriate cold room or in the warehouse as they arrive. Goods are subsequently transferred to the primary food preparation room for processing. The primary food preparation room is divided into separate preparation areas which are used for the manipulation of uncooked red meats, white meats, fish, fruits and vegetables, cheeses, and fermented sausages in order to avoid the risk of microbial cross-contamination. All the preparation areas are delimited by partial walls of 3 m height which do not reach the ceiling, hence, the separation of the areas regards just the tableware but not the indoor air. The semi-processed foods are quickly transferred from the primary food preparation room to the kitchen where they are cooked, stored in stainless steel boxes, and administered directly from the retail display unit located in the food distribution area.

2.2. Sampling

In order to estimate airborne eumycetes contamination of indoor air, the air was sampled by using a calibrated impaction sampler (SAS Super 90, International-Pbi, Milan) placed in the centre of each room, approximately 1 m from the floor. The impaction sampler allows the viable microbial particles to impact on the surface of a culture medium where they grow into colonies after incubation. Knowing the aspirated air volume together with the number of colonies on the plate, it is possible to calculate the load of microorganisms in a given air volume (1 m^3). The three cold rooms of about 30 m^3 of volume each, the kitchen, and the primary food preparation room were subjected to air sampling (Fig. 1). Temperature (T°) and relative humidity (R.H.%) were measured with a Wet and Dry Bulb Hygrometer (G.H. Zeal, London, UK). Air samples were collected during winter and summer; for each season, three samplings were carried out as follows: Sampling 1 (S1 – November), Sampling 2 (S2 – December), Sampling 3 (S3 – January) in winter, and Sampling 4 (S4 – June), Sampling 5 (S5 – July), Sampling 6 (S6 – August) in summer. During each sampling, two samples were collected for each of the five rooms, for a total of 30 samples in winter and 30 samples in summer. Microbiological analyses were conducted by sampling 500 L of air in each area. Prior to sampling, all surfaces of the sampler were wiped with isopropyl alcohol and dried with sterile gauze, as prescribed by the manufacturer. All sampling procedures were performed in the presence of the staff and during food

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