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The Occurrence of Bioaerosols in the Food Preparation Areas of HIV/AIDS Hospices in Central South Africa

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1. Introduction

South Africa is amongst countries that are facing serious challenges with regard to HIV/AIDS and institutions of HIV/AIDS hospices have been introduced in many areas of the country to help with the care of patients infected by HIV. As in any domestic setting, the safety and quality of food served in a hospice depend on the kitchen design, storage conditions, and food preparation practices of the food handlers. Micro-organisms in such areas can become airborne when droplets are generated while cleaning, cooking, preparing food, speaking, coughing, sneezing or vomiting. Most residential and hospice kitchens do not normally use air-filtration systems as it is the situation in hospitals. This means that the principal factors governing the levels of airborne particles indoor are: indoor sources, outdoor particle levels, the deposition rate of particles on indoor surfaces, and the air exchange rate (Nazaroff, 2004). Exposure of building occupants to certain micro-organisms, and elevated concentrations of environmental organisms, could result in allergenic reactions, irritant responses, toxicosis, respiratory illness and other ill effects.

This is especially important in a hospice environment that accommodates patients with compromised immune systems due to infection with the human immunodeficiency virus (HIV). Tuberculosis (TB) is an archetypal example of a disease that is transmitted by airborne route. Primary pulmonary TB is caused by the inhalation of droplet nuclei carrying the causative agent, Mycobacterium tuberculosis (MTB). For hardy bacteria such as mycobacterium TB, only a single organism is needed to cause disease (Haas, 2006). TB acts synergistically with HIV and increases the risk of primary TB infection developing into the active disease by a hundred fold (Davies, 1999). The world-wide occurrence of TB is high, with approximately one third of the world’s population thought to be infected with MTB (Miller, 1996). Globally, TB is estimated to cause the deaths of three million people annually, and this figure is predicted to rise to five million by the year 2050 (Davies, 1999). Nosocomial infections similar to TB are a very real problem in healthcare facilities, with approximately one in 10 patients acquiring an infection during a hospital stay (Schulgen, et al., 2000).

Although most nosocomial infections are generally associated with person-to-person contact, evidence is mounting that they are mostly transmitted via aerosol route. Unlike formal healthcare facilities that generally boast air filtration systems, informal healthcare
facilities like hospices have none. With patients suffering from a plethora of diseases associated with a compromised immune system, contaminated air only serves to aggravate the problem. It is therefore essential at these establishments to limit the prevalence of food-borne and airborne causative agents in the food preparation areas and through preventative measures limiting the chances for secondary infections. Therefore, the aim of this study was to quantify the microbial load of indicator microorganisms and associated environmental factors in the air (outside and inside) of a typical hospice environment. Subsequently, the relationship between environmental factors and microbial concentration was established and compared with normal breathable air in the same environment. It is expected that minimising food-borne contaminants associated with aerosols in HIV/AIDS hospices will improve the safety of food, being a considerable contributor to the food-borne contamination levels. It is envisaged that this study will contribute to the scientific knowledge of microbial contaminants associated with HIV/AIDS hospices in South Africa.

2. Materials and methods

2.1 Sample collection
Samples of breathable air were collected at ten registered hospices in two provinces in central South Africa, namely the Eastern Cape and Free State. The sampling campaigns were conducted during the southern-hemisphere winter (dry months, April-September). Samples were collected in duplicate at each hospice in the morning (before and after food preparation) and also at lunchtime (before and after food preparation), both inside and outside the hospice kitchen for both sessions.

2.2 Air sampling procedure
All microbial samples were collected 1.5 metres above the floor on 65 mm RODAC plates by means of impaction on soft agar. The (SAS) Super-90 surface air sampler (PBI International, Milan, Italy) was used for this purpose. The air sampler was calibrated at an airflow rate of 0.03 m³·min⁻¹ and all detachable parts were sterilised with 70% ethanol before use and between sampling runs (Venter et al., 2004). Plate count agar (PCA) was used for the isolation of total viable aerobic count (Merck, SA). Samples were then placed in a cooler box and immediately transported to the laboratory. Subsequent incubation of the plates was done at temperatures of between 25 and 37 °C for periods ranging from 24 to 72 hours. All colonies were enumerated using the positive hole correlation method and expressed as colony-forming units per cubic meter of air sampled. Subsequent replica plating was performed using a replica plating device and sterile velveteen cloth to quantify the following micro-organisms: Staphylococcus aureus, Pseudomonas spp., Bacillus spp., coliform and total coliform.

2.3 Facility design
Assessment of the kitchen design and setting was done by means of visual observations and note taking. Simultaneous observation of safe food handling and storage practices was also conducted. (See table 2.1)

2.4 Quantification of environmental factors
The extrinsic environmental factors capable of influencing the survival of micro-organisms, i.e. temperature (area heat stress monitor, Questemp, SA) and relative humidity (whirling
psychrometer, Airflow Instrumentation, SA), were measured, as were the factors that could influence the distribution of the assessed microbiota (Chang, et al., 2001), namely airflow (airflow anemometer – LCA 6000 VT, Airflow Instrumentation, SA) and airborne particle (dust) concentrations (hand-held aerosol monitor - 1005/1060, PPM Enterprises, Inc) (Venter, et al., 2004). Positive and negative controls were included and all analysis and assays were repeated at least in duplicate according to Venter, et al., (2004).

3. Results and discussion

3.1 Facility design

The results obtained from the technical investigations are presented in Table 1. From this, it is apparent that most hospices with the exception of a few e.g. Ons plek and Bethlehem, complied with the requirements of good preparation and handling practices of food.

<table>
<thead>
<tr>
<th>Kitchen facility design</th>
<th>Occurrence %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ceiling</td>
<td>60</td>
</tr>
<tr>
<td>Ventilation through natural moving air</td>
<td>60</td>
</tr>
<tr>
<td>Air bricks with filters</td>
<td>50</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Food handling practices</th>
<th>Occurrence %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cleanliness/neatness of the food handler</td>
<td>70</td>
</tr>
<tr>
<td>Wearing of suitable protective clothing</td>
<td>50</td>
</tr>
<tr>
<td>Availability of hand washing facilities</td>
<td>60</td>
</tr>
<tr>
<td>Storage space for hygienic storage of food</td>
<td>50</td>
</tr>
<tr>
<td>Availability of easy to clean refuse containers</td>
<td>50</td>
</tr>
<tr>
<td>Regular washing of hands before/after food preparation</td>
<td>60</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Overall cleanliness of the kitchen</th>
<th>Occurrence %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Environment conducive for cooking/preparing food</td>
<td>50</td>
</tr>
</tbody>
</table>

*The percentages and right hand values represent the level of conformance to Regulation 918 amongst all the hospices sampled (n=10).

Table 1. Kitchen facility design and food handling practices in hospices around Central South Africa.

The lack of air filtration systems due to financial constraints, may have contributed to the presence of bio-aerosol indoors. The majority of the investigated kitchens were also not designed to provide required barriers against moisture, temperature, pests, dust and associated microbes. Therefore limited control over the quality and safety of the food stored under the noted conditions would be expected.

3.2 Influence of extrinsic factors

The extrinsic factors that influence the viability and distribution of micro-organisms that prevail in various hospice kitchens are listed in figure 1. From these results, it is evident that
on average, the hospice kitchens provide an environment that would sustain microbial viability concomitant to proliferation, given that a suitable substrate and sufficient time is available. The transport and the ultimate setting of a bio aerosol are affected by its physical properties and the environmental parameters that it encounters. These physical characteristics are size, density, and shape of droplets or particles; the environmental factors include magnitude of a relative humidity and temperature, which determine the capacity to be airborne. Bioaerosols generated from suspensions undergo desiccation, whereas those generated as dusts or powders partially rehydrate. The presence of moulds as an example indicates a problem with water penetration or high humidity indoors (Goel and Goel 2009).

The persistence of micro-organisms, the presence and density of pathogens and the potential spread of microbial contamination from contaminated food in the household Kitchen have been extensively studied and re-examined. These studies indicated that domestic kitchen sites have been found to be repeatedly contaminated with a variety of bacterial contaminants, including *Listeria monocytogenes* (Beumer, et al., 1996), *Escherichia coli* and *Enterobacter cloacae* (Speirs, et al., 1995). It is well known that dampness and other excess moisture accumulation in buildings are closely connected to observation of mould, mildew, or other microbial growth. Microbial growth has also been associated with building characteristics. In residences measures of microbial contamination have been found to be positively correlated with indoor temperature and humidity, age and size of buildings, use of wood stoves and fireplaces and absence of mechanical ventilation (Dharmage, et al., 1999). From this study it became evident that the evaluated kitchens boasted levels of relative humidity of 60 ± inside as compared to the average of 40 ± outside.

The relative humidity (RH) in one of the hospices was 100% and the dominant bacterial specie was *Bacillus cereus*, therefore showing an increase associated with an increase in RH. The average temperatures were below 20 °C outside and 15 °C inside (Figure 2); hence the prevalence of *Pseudomonas aeruginosa* (Fig 1) due to their ability to survive at low temperatures (Forsythe, 2000). The average airflow for the assessed kitchen was 0.4 m.s⁻¹ outside compared to the average of 0.2 m.s⁻¹ inside. It is also interesting to note that the low airflow inside correlated with a high concentration of airborne particulates inside which was 8.04 mg.m⁻³.

### 3.3 Airborne indicator organism presence

The presence of undesirable bio-aerosols is often associated with sick building syndrome (SBS) and building-related illnesses. Sources include furnishing and building materials, fungal contamination within wall and gaps at structural joints (Jay, 2000). Inadequacies in the building design and improper ventilation may contribute to poor indoor air quality. For purposes of this study, micro-organisms indicative of poor food manufacturing practices are defined as indicator organisms. In the formal food industry, the presence of *Escherichia coli* is indicative of faecal contamination, while the presence of *Staphylococcus aureus* points to extensive human handling, and total viable aerobic organisms is a sign of poor process hygiene. The presence of these organisms in the breathable air of the various hospice kitchens could also be attributed to the aforementioned practices, as food processing is the core business of the kitchens in this setting.
The Occurrence of Bioaerosols in the Food Preparation Areas of HIV/AIDS Hospices in Central South Africa

In this scenario, the presence of *E. coli* could be due to faecal contaminants from the hands of food handlers, or from contaminated working surfaces and utensils. Therefore, it is of the utmost importance to observe proper personal hygiene, particularly with regard to hand-washing after visiting the toilet, in order to prevent contamination. However, three of the hospices that were investigated are without hand-washing facilities and are located in rural areas. As noted in figure 1 the average airborne particulates were 7.90 mg.m\(^{-3}\) inside as compared to 8.0 mg.m\(^{-3}\) outside. These results are comparable to those observed by Hargreaves *et al.*, 2003. The presence of *Bacillus spp* was also noted in the presence of these conspicuous airborne particulates which were dominant throughout the sampling period. This could be due to the fact that these are spore formers and bacteria in many cases are attached to larger airborne particles.

According to Moir *et al.*, 2002, these spores can endure a wide range of extreme environmental stresses while retaining the capacity to return to vegetative growth almost immediately once the nutrient returns to the environment. It was therefore assumed that airflow in and outside the hospice kitchens were sufficient to carry dust particles as well as different bacterial population (Figure 1). In general, the average bacterial counts varied between 1 x 10\(^1\) to 1 x 10\(^2\) cfu.m\(^{-3}\). Compared to the literature these counts were fairly low and apart from being possible allergens they are also associated with decreased lung function, increased respiratory symptoms such as cough, shortness of breath and asthma attacks (WHO, 2002). From the observed results it is apparent that the kitchens in question boast a resident bio-aerosol population that is not significantly influenced by the noted environmental parameters. Though the source of the bio-aerosols assayed is not clear, *S. aureus* probably results from aerosols dispersed by the food handlers in the kitchens. It should also be noted that although microbial counts were low, the kitchens provided an environment conducive to microbial survival as aerosolised particles and subsequently as food contaminants. In general the kitchens had a lower temperature, increased RH, and higher airborne particle count.

4. Conclusion

Indoor air pollution is usually caused by the accumulation of contaminants from various indoor sources. The generation of pollutants within the indoor environment may come from...
primary sources such as fuel combustion for cooking, as well as emissions from fireplaces, stoves, cleaning products and chemicals stored in the home. It is therefore safe to assume that since all these factors were present at the study sites, they all contributed to the indoor air pollution discovered. Factors such as heating, ventilation, air-conditioning and household activities, e.g. cooking and cleaning, all play a crucial role in the wellbeing of a building’s occupants. In a setting such as a hospice, where proper food storage and handling is not always possible due to lack of infrastructure, it is essential that special care is taken regarding the type of food stored and the packaging material used. Micro-organisms detected in the indoor air of the hospice kitchens included in this study could be derived from the hospice occupants, but may also emanate from the outside environment. One such example would be *Bacillus cereus*, which is a common air- and dust-borne contaminant that readily multiplies in meat products. These organisms are able to withstand unfavourable conditions such as low temperature and heat due to their ability to form spores (Whyte et al., 2001). A study by Nel et al., 2003, reported rapidly increasing levels of *B. cereus* when a product was exposed to poor handling and processing procedures. A hospice, where patients are provided with accommodation, food and care, would be a typical example of this setting.

From the results it can be concluded that certain food preparation and storage facilities in the hospices studied are, according to the technical data, not suitable for this purpose. However, it appeared that some of the extrinsic factors influencing microbial viability were being governed. Specific attention should be given to the upgrading of the kitchen infrastructure – for example, it would be ideal to have separate rooms for the preparation of raw and cooked food, as well as a room used only for food storage purposes. The results presented in this chapter further identified the hospice occupants as possible sources of the organisms found in the hospice kitchens and surrounding environments, since the occupants were in some cases also responsible for preparing the food, while moving continuously between the kitchen, bathroom and bedroom. In terms of residency, it would thus be ideal to separate the patients from the food preparation facilities, including the kitchen.

Companies that regularly donate food to the hospices are further cautioned to avoid donating foodstuffs that are past their sell-by date, as the inability of the assessed kitchens to control humidity fluctuations only exacerbates the problem with regard to food safety and proliferation of microbial load. It was further noted that in South Africa, unlike in more humid countries, the facility design and geographical localisation have a limited effect on the resident bio-aerosol profiles. In order to address and verify the concerns raised in this chapter, it is recommended that the ability of micro-organisms to proliferate on the foods (high-risk) provided to the hospices should be assessed and the menus adjusted in accordance to season-associated changes in the extrinsic factors that would influence microbial viability and growth. It should further be noted that the occupants of the hospices concerned have very little knowledge of proper food storage and handling practices and are to a large extent not aware of the threats posed to them by the resident microbiota. This problem could be alleviated by providing the patients and food handlers with educational training in respect of proper food safety (storage and handling).

5. Future research

Due to a lack of empirical data, aerosol transmission of influenza is often questioned and its relationship with other airborne microbiota such as bacteria and fungi still requires further research to establish their impact on food and people indoors (Blachere et al., 2007).
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The book describes the effects of air pollutants, from the indoor and outdoor spaces, on the human physiology. Air pollutants can influence inflammation biomarkers, can influence the pathogenesis of chronic cough, can influence reactive oxygen species (ROS) and can induce autonomic nervous system interactions that modulate cardiac oxidative stress and cardiac electrophysiological changes, can participate in the onset and exacerbation of upper respiratory and cardio-vascular diseases, can lead to the exacerbation of asthma and allergic diseases. The book also presents how the urban environment can influence and modify the impact of various pollutants on human health.

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