

Potential airborne microbial hazards for workers on dairy and beef cattle farms in Egypt

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Summary

This study was conducted to determine the concentration and frequency distribution of certain airborne micro-organisms on cattle farms and their potential health hazards to farm workers. The samples (60 air samples and 240 hand and nasal swabs from cattle farm workers) were collected from ten cattle farms (five dairy barns and five beef sheds) located in the Sharkia Governorate of Egypt. Air samples were collected for microbiological examination in liquid media using an all-glass impinger whereas those for fungal examination were placed on agar plates using slit air samplers (aeroscopes). The results showed that the overall means of total culturable bacterial and fungal counts were lower in the air of dairy cattle barns than in beef cattle sheds. Identification of the isolated bacteria revealed the recovery of the following species (from dairy cattle barns versus beef cattle sheds): *Staphylococcus epidermidis* (26.7% vs 36.7%), *S. saprophyticus* (20% vs 33.3%), *S. aureus* (10% vs 16.7%), *Enterococcus faecalis* (23.3% vs 26.7%), *Enterobacter agglomerans* (23.3 vs 13.3%), *Escherichia coli*, (16.7% vs 26.7%), *Klebsiella oxytoca*, (10% vs 16.7%), *K. pneumoniae* (3.3% vs 0%), *Proteus rettegr* (6.7% vs 13.3%), *P. mirabilis* (10% vs 10%), *P. vulgaris* (3.3% vs 6.7%), *Pseudomonas* species (6.7% vs 16.7%), respectively). Mycological examination of air samples revealed the presence of *Aspergillus fumigatus* (46.7% vs 63.3%), *A. niger* (20% vs 36.7%), *A. flavus* (13.3% vs 26.7%), *Penicillium citrinum* (16.7% vs 23.3%), *P. viridicatum* (13.3% vs 6.7%), *P. capsulatum* (3.3% vs 0%),

Cladosporium spp. (30% vs 56.7%), *Alternaria* spp. (13.3 vs 23.3%), *Mucor* spp. (6.7% vs 16.7%), *Fusarium* spp. (3.3% vs 10%), *Absidia* spp. (6.7% vs 10%), *Curvularia* spp. (10% vs 3.3%), *Rhizopus* spp. (6.7% vs 13.3%), *Scopulariopsis* (3.3% vs 6.7%), *Epicoccum* spp. (0% vs 3.4%) and yeast (13.3% vs 20%), respectively. In addition, microbiological examinations of farm workers revealed heavy contamination of their hands and noses with most of the micro-organisms detected in the air of cattle farms. The results showed that potential airborne microbial risks in beef cattle sheds were greater than in dairies.

Keywords

Air, Bacterium, Beef, Biohazard, Cattle, Dairy, Egypt, Fungi, Public health, Zoonoses.

Rischi potenziali di contaminazione microbica per via aerea in lavoratori di allevamenti di bovini da ingrasso e da latte in Egitto

Riassunto

Questo studio è stato condotto per determinare la concentrazione e la distribuzione della frequenza di alcuni microrganismi aerei nelle fattorie di animali e il potenziale pericolo che rappresentano per la salute dei lavoratori. I campioni (60 campioni d'aria e 240 tamponi prelevati da naso e mani dei lavoratori) sono stati raccolti da dieci fattorie (cinque per produzione casearia e cinque per la macellazione) situate nel Governatorato di Sharkia,

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Egitto. I campioni di aria per l'esame microbiologico sono stati raccolti in mezzo liquido mediante impinger in vetro, mentre quelli per l'esame fungino sono stati posti su piastre di agar mediante campionatori "slit air" (aeroscopi). I risultati dimostrano che le medie complessive delle conte batteriche e fungine coltivabili totali erano inferiori nell'aria delle fattorie casearie rispetto a quelle degli animali da macello. L'identificazione dei batteri isolati ha evidenziato il recupero delle seguenti specie (fattorie casearie vs. fattorie di animali da macello): Staphylococcus epidermidis (26,7% vs. 36,7%), S. saprophyticus (20% vs. 33,3%), S. aureus (10% vs. 16,7%), Enterococcus faecalis (23,3% vs. 26,7%), Enterobacter agglomerans (23,3 vs. 13,3%), Escherichia coli, (16,7% vs. 26,7%), Klebsiella oxytoca, (10% vs. 16,7%), K. pneumoniae (3,3% vs. 0%), Proteus rettgeri (6,7% vs. 13,3%), P. mirabilis (10% vs. 10%), P. vulgaris (3,3% vs. 6,7%), specie Pseudomonas (6,7% vs. 16,7%). L'esame micologico dei campioni di aria ha rivelato la presenza di Aspergillus fumigatus (46,7% vs. 63,3%), A. niger (20% vs. 36,7%), A. flavus (13,3% vs. 26,7%), Penicillium citrinum (16,7% vs. 23,3%), P. viridicatum (13,3% vs. 6,7%), P. capsulatum (3,3% vs. 0%), Cladosporium spp. (30% vs. 56,7%), Alternaria spp. (13,3 vs. 23,3%), Mucor spp. (6,7% vs. 16,7%), Fusarium spp. (3,3% vs. 10%), Absidia spp. (6,7% vs. 10%), Curvularia spp. (10% vs. 3,3%), Rhizopus spp. (6,7% vs. 13,3%), Scopulariopsis (3,3% vs. 6,7%), Epicoccum spp. (0% vs. 3,4%) e lievito (13,3% vs. 20%). Inoltre, l'esame microbiologico dei lavoratori ha rivelato una pesante contaminazione di mani e naso ad opera della maggior parte dei microrganismi rilevati nell'aria delle fattorie. I risultati dimostrano che i rischi microbici aerei potenziali nelle fattorie di animali da macello sono maggiori rispetto a quelli rilevati nelle fattorie casearie.

Parole chiave

Aria, Batteri, Bovini, Carne, Egitto, Funghi, Pericolo biologico, Prodotti caseari, Salute pubblica, Zoonosi.

Introduction

The use of confinement systems for raising animals has brought with it not only large increases in productivity per farm but also has

resulted in the potential physical, chemical and biological health hazards both to farm workers and the confined species (6). The biological health hazard arises from the presence of bioaerosols which constitute a natural phenomenon in barn air. These bioaerosols are a complex mixture of live and dead microorganisms and their products as well as other aeroallergens (37). The primary sources of these bioaerosols are the animals themselves, fodder, litter, dung and humans. Airborne bacteria are a constituent of bioaerosols. Their concentrations in air greatly depend on the construction and technical characteristics of the housing, stocking density and health conditions of animals kept in the barn, mode of keeping, microclimatic conditions, feeding, grooming, milking and other activities (23). Airborne bacteria, not only include pathogenic bacteria but also saprophytic bacteria which are potential or typical commensals in animals. It is likely that, although they do not produce disease, they may be responsible for growth depression (18).

Apart from airborne bacteria, the barn air is also polluted with a vast range of mainly saprophytic fungi (15). People are sometimes affected without becoming infected (24). Fungi are also capable of causing human health problems when the opportunity presents itself (36). Airborne bacteria and fungi can be bound to solid or liquid carriers and may be suspended in air for long periods of time which gives rise to endemicity of microorganisms on the farm. In addition, to establish endemic infection within a facility, bioaerosols vented from the building can spread the infection to other buildings on the site or to other sites, resulting in epizootics (18). The contamination of surfaces and food by bioaerosols can promote the spread of infection. Occupations associated with animal breeding and processing of animal materials are exposed to the risk of inhalation of large quantities of bioaerosols which result in respiratory diseases (32). It is important to trace the circulating bacteria in farm animals, air and workers which reflect the risk associated with exposure to bioaerosols in cattle barns.

This study was performed to determine the concentration and species composition of bioaerosols and to investigate the health hazard in people whose occupations expose them to the inhalation of large quantities of these bioaerosols.

Materials and methods

The study included 10 cattle farms (5 dairy cattle barns and 5 beef cattle sheds) in the Sharkia Governorate of Egypt. A visual assessment of each farm was conducted and a brief questionnaire covered questions concerning the husbandry system, characteristics of the floors, ventilation type and area and the number of animal caretakers. Data on the characteristics of animal sheds are summarised in Table I.

Sampling

Each farm was scheduled for sampling six times, with an interval of 15 days in between. During each visit, two air samples were collected from the centre of the farm; one for airborne bacteria and the other for airborne fungi. In addition, hand and nasal swabs from two animal caretakers at each farm were also collected.

Airborne bacteria

Air samples for airborne bacteria were taken at 150 cm above the floor level which is within the human breathing zone. All samples were collected using a liquid impinger device (National Research Centre, Cairo) containing 50 ml buffered peptone water (BPW) as the collection media (16). The flowing rate of air into the apparatus was 10 l/min and was operated for 10 min. After aerosol sampling, the collection media was placed separately in a sterile plastic bottle, labelled and transferred to the laboratory on ice.

Airborne fungi

A slit air sampler (aeroscope) was used to collect fungal propagules. The sampler was operated at the prescribed flow rate of 28.3 l/min and Petri dishes filled with 35 ml of malt extract agar were used (26).

Hand swabs

The entire surface of the hand was swabbed using sterile swabs moistened in sterile BPW. The swab was then immersed in test tubes containing BPW under aseptic conditions, packed and despatched to the laboratory on ice (33).

Table I
Topographical examination of farms investigated

| Farm No. | Production type | Location | Size | | Floor type | No. of housed animals | Ventilation area |
|----------|-----------------|-------------------|------------|-----------|------------|-----------------------|---------------------------|
| | | | Length (m) | Width (m) | | | |
| 1 | Dairy | Hehia town | 32 | 10.5 | Concrete | 37 | 5 windows, each 1.5x3 m |
| 2 | Dairy | Hehia town | 23 | 11.0 | Concrete | 42 | 3 windows, each 1.2x2.5 m |
| 3 | Dairy | Hehia town | 22 | 11.0 | Muddy | 33 | 3 windows, each 1x2.5 m |
| 4 | Dairy | Kafr Sakr town | 18 | 12.0 | Muddy | 44 | 3 windows each, 1.2x2.5 m |
| 5 | Dairy | Hehia town | 33 | 11.0 | Concrete | 52 | 4 windows each, 1x2.2 m |
| 6 | Beef | Hehia town | 20 | 8.0 | Muddy | 46 | 3 windows each, 1.5x2 m |
| 7 | Beef | Hehia town | 22 | 10.0 | Muddy | 50 | 4 windows each, 1x2.5 m |
| 8 | Beef | Hehia town | 18 | 11.5 | Muddy | 34 | 3 windows each, 1.2x2 m |
| 9 | Beef | El Ebrahimia town | 26 | 9.5 | Muddy | 42 | 4 windows each, 1.5x2 m |
| 10 | Beef | Hehia town | 24 | 11.0 | Concrete | 36 | 4 windows each, 1.2x2.5 m |

Nasal swabs

Nasal swabs were collected by rolling sterile swab moistened in sterile BPW firmly against the nasal mucosa and then treated as for hand swabs.

Laboratory analysis

Laboratory analysis of the samples was performed without delay in the Laboratories of Zoonoses and Veterinary Public Health Departments at the Faculty of Veterinary Medicine, Zagazig University.

Airborne bacteria

The airborne samples were examined for microbial enumeration and speciation.

The number of micro-organisms in the air samples were determined using the dilution plating method (9).

The speciation of micro-organisms was determined as described below.

The air samples prepared in BPW were enriched by incubation at 37°C for 18-24 h. Then 0.1 ml of pre-enriched culture broth was spread on blood agar and eosin methylene blue (EMB) agar (Merck, Germany) and incubated at 37°C for 18-24 h (10). The suspected colonies from each plate were collected (on average three suspected colonies from each plate on the basis of colonial morphology) and were purified on nutrient agar slant for further identification (31). Briefly, Gram stain, Indole, methyl red, Voges-Proskauer, citrate utilisation, haemolysis, motility, H₂S production, gelatin liquefaction and sugar fermentation tests were performed.

Airborne fungi

The cultured MEA plates were incubated at room temperature (23±3°C). Plates were inspected after 4 days and periodically up to 14 days. The number of fungi per cubic metre of air was calculated and the isolated fungi were subcultured on Sabaraud dextrose agar (SDA) and Czapek dox agar for further identification (15). The macroscopic characteristics (morphological criteria of the colonies e.g. growth rapidity, colour, texture, surface and central appearance, pigment, margins, aerial hyphae) and the microscopic examination of the colony by scotch tape

preparations, by pressing the sticky side of clear scotch tape to the surface of the colony and placing it, side down, in a drop of lactophenol cotton blue on a microscope slide. The samples were examined with 10× and 40× magnification for characteristic hyphal and spore arrangements (shape of hyphae, septae, chlamidiospore, conidiophore, conidia).

Nasal and hand swabs

The swab tip was vortexed for 1 min in 10 ml sterile phosphate buffer saline (PBS). A volume of 100 µl was cultured onto duplicate sets of three agar media, namely: blood agar, EMB and SDA. The plates were incubated and colonial subcultures and the identification protocol were similar to that used for the air samples (7).

Results

The results given in Table II show the concentration of airborne micro-organisms measured in five dairy cattle barns and five beef cattle sheds. It was found that the total mean bacterial count per cubic metre was higher in beef (1.85×10^5) cattle air samples than in dairy (7.28×10^4) samples. In addition, the overall mean total fungal count per cubic metre were 2.59×10^3 and 1.5×10^3 in the air of beef and dairy cattle sheds, respectively.

Table III gives the type and frequency distribution of bacteria isolated from the air of cattle sheds. *Staphylococcus aureus* was the most predominant Gram-positive cocci in air samples from beef (36.7%) and dairy (26.7%) sheds. Moreover, *Enterobacter agglomerans* (23.3%) and *Escherichia coli* (26.7%) were the predominant Gram-negative bacteria in dairy and beef cattle samples, respectively.

The data recorded in Table IV show that *Aspergillus fumigatus* is the predominant species in the air samples from both dairy (46.7%) and beef (63.3%) cattle farms.

Table V gives the type and frequency distribution of bacteria and fungi isolated from cattle workers. *Staphylococcus epidermidis* was found to be the most common micro-organism in nasal and hand swabs from beef farm workers with the percentage of 51.7% and 31.7%, respectively.

Table II
Concentration of airborne micro-organisms measured in five dairy cattle barns and five beef cattle sheds

| Farm | Production type | Sample No. | Total bacterial count/m ³ | | Total fungal count/m ³ | |
|---------|-----------------|------------|--------------------------------------|---|-----------------------------------|---|
| | | | Mean | Range | Mean | Range |
| 1 | Dairy | 6 | 2.2 × 10 ³ | 2.1 × 10 ² -4.8 × 10 ³ | 5.4 × 10 ² | 1.6 × 10 ² -7.8 × 10 ² |
| 2 | Dairy | 6 | 5.9 × 10 ³ | 4.9 × 10 ² -11.5 × 10 ³ | 7.9 × 10 ² | 4.2 × 10 ² -11.1 × 10 ² |
| 3 | Dairy | 6 | 2.9 × 10 ⁴ | 3.3 × 10 ⁴ -18.2 × 10 ⁴ | 2.3 × 10 ³ | 1.5 × 10 ³ -2.9 × 10 ³ |
| 4 | Dairy | 6 | 2.4 × 10 ⁵ | 4.0 × 10 ⁴ -5.91 × 10 ⁵ | 2.4 × 10 ³ | 1.9 × 10 ³ -2.84 × 10 ³ |
| 5 | Dairy | 6 | 8.5 × 10 ⁴ | 3.3 × 10 ⁴ -1.5 × 10 ⁵ | 1.5 × 10 ³ | 1.1 × 10 ³ -2.04 × 10 ³ |
| Overall | | 30 | 7.3 × 10 ⁴ | 2.1 × 10 ² -5.91 × 10 ⁵ | 1.5 × 10 ³ | 1.6 × 10 ² -2.9 × 10 ³ |
| 6 | Beef | 6 | 3.7 × 10 ⁵ | 1.3 × 10 ⁵ -7.6 × 10 ⁵ | 3.29 × 10 ³ | 2.3 × 10 ³ -4.4 × 10 ³ |
| 7 | Beef | 6 | 1.5 × 10 ⁵ | 2.4 × 10 ⁴ -3.9 × 10 ⁵ | 3.02 × 10 ³ | 1.9 × 10 ³ -4.1 × 10 ³ |
| 8 | Beef | 6 | 2.9 × 10 ⁵ | 9.0 × 10 ⁴ -4.3 × 10 ⁵ | 2.0 × 10 ³ | 1.1 × 10 ³ -3.7 × 10 ³ |
| 9 | Beef | 6 | 6.4 × 10 ⁴ | 8.3 × 10 ³ -1.2 × 10 ⁵ | 2.92 × 10 ³ | 1.7 × 10 ³ -5.4 × 10 ³ |
| 10 | Beef | 6 | 5.1 × 10 ⁴ | 1.8 × 10 ⁴ -9.5 × 10 ⁴ | 1.72 × 10 ³ | 1.3 × 10 ³ -2.0 × 10 ³ |
| Overall | | 30 | 1.85 × 10 ⁵ | 8.3 × 10 ³ -7.6 × 10 ⁵ | 2.59 × 10 ³ | 1.1 × 10 ³ -5.4 × 10 ³ |

Table III
Type and frequency distribution of bacteria isolated from the air of five cattle barns and five beef cattle sheds in Egypt

| Micro-organism | Dairy cattle barns | | Beef cattle sheds | |
|-------------------------------------|--------------------|------------|-------------------|------------|
| | No. of isolates | Percentage | No. of isolates | Percentage |
| Gram-positive cocci | | | | |
| <i>Staphylococcus epidermidis</i> | 8 | 26.7 | 11 | 36.7 |
| <i>Staphylococcus saprophyticus</i> | 6 | 20.0 | 10 | 33.3 |
| <i>Staphylococcus aureus</i> | 3 | 10.0 | 5 | 16.7 |
| <i>Enterococcus faecalis</i> | 7 | 23.3 | 8 | 26.7 |
| Gram-negative bacilli | | | | |
| <i>Enterobacter agglomerans</i> | 7 | 23.3 | 4 | 13.3 |
| <i>Escherichia coli</i> | 5 | 16.7 | 8 | 26.7 |
| <i>Klebsiella oxytoca</i> | 3 | 10.0 | 5 | 16.7 |
| <i>Klebsiella pneumoniae</i> | 1 | 3.3 | 0 | 0.0 |
| <i>Proteus ettegrri</i> | 2 | 6.7 | 4 | 13.3 |
| <i>Proteus mirabilis</i> | 3 | 10.0 | 3 | 10.0 |
| <i>Proteus vulgaris</i> | 1 | 3.3 | 2 | 6.7 |
| <i>Pseudomonas</i> spp. | 2 | 7.7 | 5 | 16.7 |

Discussion

Table II gives an enumeration of airborne micro-organisms measured in five dairy cattle barns and five beef cattle sheds. It was evident that the overall mean of total bacterial counts in air samples of dairy and beef cattle farms was 7.3×10⁴ and 1.9×10⁵ cfu/m³. A very similar total bacterial count (from 2.8×10⁴ to 7.8×10⁴ cfu/m³) was recorded in air of dairy

cattle barns in Croatia (28). A concentration between some hundreds and several thousands per litre were recorded in livestock buildings (19). On the other hand, in Ohio, the average level for airborne culturable bacteria in dairy barns can reach up to 3.3×10⁸ cfu/m³ (25).

Table II also confirms that the overall mean of the total fungal count in air samples was 1.5×10³ and 2.6×10³ cfu/m³ in dairy and beef

cattle farms, respectively. These results substantiate what has been recorded in India, i.e. that the average concentration of total culturable fungi in the air of indoor cattle sheds ranged from 1.65×10^2 to 2.2×10^3 cfu/m³ (2). Higher concentrations of airborne fungi (up to 10^7 cfu/m³ of dairy farm air) was also reported in Finland (17). However, in Germany, fungi accounted for more than 1% of the total amount of airborne micro-organisms in livestock buildings (19).

The results in Table II show that in air samples of dairy cattle farms, the mean total bacterial and fungal counts were highest (24.2×10^4 and 19×10^2 cfu/m³, respectively) in Farm No. 4 and lowest (21.7×10^2 and 5.4×10^2 cfu/m³, respectively) on Farm No. 1. However, the mean total bacterial and fungal counts in air samples of beef cattle farms were higher on Farm No. 6 (37.1×10^4 and 32.9×10^2 cfu/m³, respectively) than Farm No. 10 (5.1×10^4 and 17.2×10^2 cfu/m³, respectively). Similar variations in the concentrations of airborne micro-organisms among dairy cattle farms have been reported (17). These variations in the level of aerial microbiota of cattle farms could be attributed to the differences in hygiene levels, stocking density and ventilation areas observed on these farms. These results are supported by Lago *et al.* (22) who found a significant relation between the bacterial counts of animal pens and stocking density, ventilation rate and floor and bedding types of pens. Thus, the high concentration of airborne micro-organisms recorded on Farm Nos 4 and 6 could be interpreted by the presence of muddy floors, irregular and infrequent cleaning, low ventilation areas and creating high risk factors for respiratory diseases not only for cattle but also for the workers in cattle sheds (2).

Comparing the values of total airborne bacterial and fungal counts in the farms examined revealed higher counts in air of beef cattle farms than dairy farms. In Germany, the median values of airborne inhalable lipopolysaccharides of Gram-negative bacteria in beef cattle farms significantly exceeds those of dairy farms (35). The higher bacterial and fungal counts observed in this study in the air of beef cattle farms can be attributed to high

stocking densities, muddy floors with no separation of wastes from the pen floor that allows water to accumulate in the soil, and irregular and infrequent cleaning of most beef farms compared to dairy farms (5).

The type and frequency distribution of the bacteria isolated from the air of dairy and beef cattle farms are presented in Table III. The results clarify that the isolated Gram-positive cocci from dairy cattle farms versus beef farms were as follows:

- *S. epidermidis* (26.7% vs 36.7%)
- *S. saprophyticus* (20% vs 33.3%)
- *S. aureus* (10% vs 16.7%)
- *Enterococcus faecalis* (23.3% vs 26.7%).

A similar occurrence of these species has been recorded in Egypt (3). Furthermore, 80% of airborne micro-organisms in livestock buildings were found to be *Staphylococcae* and *Streptococcae* (19). The high incidence of *S. aureus* in the air examined indicates not only air pollution from diseased or even carrier humans and animals but also reveals the serious risk to cattle and occupational workers. *E. faecalis* is of public health significance as it may cause endocarditis, urinary tract infection and sepsis in humans (12) and its frequent isolation from the air of cattle sheds is an indication of poor hygiene (3).

Table III shows also that the Gram-negative bacilli isolated from the air of dairy cattle barns were: *E. agglomerans* (23.3%), *E. coli* (16.7%), *Klebsiella oxytoca* (10%), *K. pneumoniae* (3.3%), *Proteus rettegi* (6.7%), *P. mirabilis* (10%), *P. vulgaris* (3.3%) and *Pseudomonas* spp. (6.7%). On the other hand, the corresponding frequency for the same micro-organisms in air of beef cattle sheds were 13.3%, 26.7%, 16.7%, 0%, 13.3%, 10%, 6.7%, and 16.7%, respectively. Most of the isolated bacterial species were isolated previously from the air in cattle barns although the percentage of isolated species differed (3). Moreover, the results obtained coincided with those who found that within airborne Gram-negative microbiota of cattle sheds, *Enterobacteriaceae* and *Pseudomonadaceae* dominated and that within the family *Enterobacteriaceae*, the species *E. coli* and *E. agglomerans* were predominant (38). The high frequency of these species of Gram-

negative bacteria in air of dairy and beef cattle farms increases the risk of exposure to airborne microflora as most detected species are producers of allergens and/or toxins; many produce a biological active endotoxin (11) and may evoke allergic alveolitis (29).

The results given in Table IV reveal that the type and percentage of fungi isolated from the air of dairy cattle farms compared to beef farms were as follows:

- *A. fumigatus* (46.7% vs 63.3%)
- *A. niger* (20% vs 36.7%)
- *A. flavus* (13.3% vs 26.7%)
- *Penicillium citrinum* (16.7% vs 23.3%)
- *P. viridicatum* (13.3% vs 6.7%)
- *P. capsulatum* (3.3% vs 0%)
- *Cladosporium* spp. (30% vs 56.7%)
- *Alternaria* spp. (13.3% vs 23.3%)
- *Mucor* spp. (6.7% vs 16.7%)
- *Fusarium* spp. (3.3% vs 10%)
- *Absidia* spp. (6.7% vs 10%)
- *Curvularia* spp. (10% vs 3.3%)
- *Rhizopus* spp. (6.7% vs 13.3%)
- *Scopulariopsis* spp. (3.3% vs 6.7%)

- *Epicoccum* spp. (0% vs 3.4%)
- yeast (13.3% vs 20.0%).

Similar findings were previously recorded in air of cattle sheds (13). Lower percentages of the fungal species isolated were also reported in cattle sheds in Egypt (3). The frequent isolation of airborne fungi in the present study may be attributed to the time of sampling (in the autumn), when the level of fungi peaked (4). Moreover, water accumulation in cattle sheds that were so densely stocked and that had very muddy floors provides high moisture for fungi to rapidly multiply in faecal matter and feed materials that are abundant on cattle farms (5). The fungal species isolated were the types that prevailed in cattle shed air in other studies (2, 17). The public health significance of the fungi isolated in the present study arises from the fact that at least ten species or genera were reported as potential agents of allergic and immunotoxic diseases of the respiratory tract. The greatest respiratory risk is posed by *A. fumigatus*, *Penicillium* spp., *Cladosporium*, *Alternaria* and *Mucor* spp. (11).

Table IV
Type and frequency distribution of fungi isolated from air of five dairy cattle barns and five beef cattle sheds in Egypt

| Micro-organism | Dairy cattle barns | | Beef cattle sheds | |
|----------------------------|--------------------|------------|-------------------|------------|
| | No. of isolates | Percentage | No. of isolates | Percentage |
| <i>Aspergillus</i> spp. | | | | |
| <i>A. fumigatus</i> | 14 | 46.7 | 19 | 63.3 |
| <i>A. niger</i> | 6 | 20.0 | 11 | 36.7 |
| <i>A. flavus</i> | 4 | 13.3 | 8 | 26.7 |
| <i>Penicillium</i> spp. | | | | |
| <i>P. citrinum</i> | 5 | 16.7 | 7 | 23.3 |
| <i>P. viridicatum</i> | 4 | 13.3 | 2 | 6.7 |
| <i>P. capsulatum</i> | 1 | 3.3 | – | 0.0 |
| <i>Cladosporium</i> spp. | | | | |
| | 9 | 30.0 | 17 | 56.7 |
| <i>Alternaria</i> spp. | | | | |
| | 4 | 13.3 | 7 | 23.3 |
| <i>Mucor</i> spp. | | | | |
| | 2 | 6.7 | 5 | 16.7 |
| <i>Fusarium</i> spp. | | | | |
| | 1 | 3.3 | 3 | 10.0 |
| <i>Absidia</i> spp. | | | | |
| | 2 | 6.7 | 3 | 10.0 |
| <i>Curvularia</i> spp. | | | | |
| | 3 | 10.0 | 1 | 3.3 |
| <i>Rhizopus</i> spp. | | | | |
| | 2 | 6.7 | 4 | 13.3 |
| <i>Scopulariopsis</i> spp. | | | | |
| | 1 | 3.3 | 2 | 6.7 |
| <i>Epicoccum</i> spp. | | | | |
| | – | 0.0 | 1 | 3.4 |
| Yeast | | | | |
| | 4 | 13.3 | 6 | 20.0 |

Table V shows that the types and frequency distribution of bacterial species isolated from hand swabs of dairy farm workers were as follows:

- *S. epidermidis* (18.3%)
- *S. saprophyticus* (13.3%)
- *S. aureus* (8.3%)
- *E. faecalis* (11.7%)
- *E. agglomerans* (23.3%)
- *E. coli* (15%)
- *K. oxytoca* (5%)
- *P. mirabilis* (10%).

The same species were isolated from hand swabs of beef farm workers as follows:

- *S. epidermidis* (31.7%)
- *S. saprophyticus* (20%)
- *S. aureus* (16.7%)

- *E. faecalis* (21.7%)
- *E. agglomerans* (13.3%)
- *E. coli* (26.7%)
- *K. oxytoca* (6.7%)
- *P. mirabilis* (5%).

Table V also shows that the bacterial species isolated from nasal swabs of dairy versus beef farm workers were as follows:

- *S. epidermidis* (35% vs 51.7%)
- *S. saprophyticus* (26.7% vs 40%)
- *S. aureus* (15% vs 23.3%)
- *E. faecalis* (16.7% vs 28.3%)
- *E. agglomerans* (18.3% vs 20%)
- *E. coli* (11.7% vs 16.7%)
- *K. oxytoca* (6.7% vs 8.3%)
- *K. pneumoniae* (1.7% vs 3.3%)
- *P. mirabilis* (6.7% vs 6.7%).

Table V
Type and frequency distribution of bacteria and fungi isolated from workers of dairy and beef cattle farms in Egypt

| Micro-organism | Dairy farm workers | | | | Beef farm workers | | | |
|-------------------------------------|--------------------|------|-------------|------|-------------------|------|-------------|------|
| | Hand swabs | | Nasal swabs | | Hand swabs | | Nasal swabs | |
| | No. | % | No. | % | No. | % | No. | % |
| Gram-positive cocci | | | | | | | | |
| <i>Staphylococcus epidermidis</i> | 11 | 18.3 | 21 | 35.0 | 19 | 31.7 | 31 | 51.7 |
| <i>Staphylococcus saprophyticus</i> | 8 | 13.3 | 16 | 26.7 | 12 | 20.0 | 24 | 40.0 |
| <i>Staphylococcus aureus</i> | 5 | 8.3 | 9 | 15.0 | 10 | 16.7 | 14 | 23.3 |
| <i>Enterococcus faecalis</i> | 7 | 11.7 | 10 | 16.7 | 13 | 21.7 | 17 | 28.3 |
| Gram-negative bacilli | | | | | | | | |
| <i>Enterobacter agglomerans</i> | 14 | 23.3 | 11 | 18.3 | 8 | 13.3 | 12 | 20.0 |
| <i>Escherichia coli</i> | 9 | 15.0 | 7 | 11.7 | 16 | 26.7 | 10 | 16.7 |
| <i>Klebsiella oxytoca</i> | 3 | 5.0 | 4 | 6.7 | 4 | 6.7 | 5 | 8.3 |
| <i>Klebsiella pneumoniae</i> | – | 0.0 | 1 | 1.7 | – | 0.0 | 2 | 3.3 |
| <i>Proteus mirabilis</i> | 6 | 10.0 | 4 | 6.7 | 3 | 5.0 | 4 | 6.7 |
| Moulds and fungi | | | | | | | | |
| <i>Aspergillus fumigatus</i> | 5 | 8.3 | 4 | 6.7 | 9 | 15.0 | 6 | 10.0 |
| <i>Aspergillus niger</i> | 4 | 6.7 | 3 | 5.0 | 7 | 11.7 | 4 | 6.7 |
| <i>Aspergillus flavus</i> | 6 | 10.0 | 4 | 6.7 | 6 | 10.0 | 5 | 8.3 |
| <i>Penicillium citrinum</i> | 3 | 5.0 | 2 | 3.3 | 5 | 8.3 | 3 | 5.0 |
| <i>Cladosporium</i> spp. | 5 | 8.3 | 3 | 5.0 | 7 | 11.7 | 6 | 10.0 |
| <i>Mucor</i> spp. | 2 | 3.3 | – | 0.0 | 4 | 6.7 | 2 | 3.3 |
| <i>Alternaria</i> spp. | 1 | 1.7 | – | 0.0 | 2 | 3.3 | 1 | 1.7 |
| <i>Absidia</i> spp. | – | 0.0 | – | 0.0 | 2 | 3.3 | – | 0.0 |
| <i>Curvularia</i> spp. | 1 | 1.7 | 1 | 1.7 | 2 | 3.3 | – | 0.0 |
| <i>Epicoccum</i> spp. | 1 | 1.7 | – | 0.0 | – | 0.0 | – | 0.0 |
| <i>Rhizopus</i> spp. | 2 | 3.3 | – | 0.0 | 1 | 1.7 | – | 0.0 |

Most of these organisms were previously isolated from hand swabs and sputum of farm workers and veterinarians in contact with animals; although the percentages of isolation were lower (34). The percentages of the bacterial species isolated in this study were comparable to those published by Mohamed *et al.* (30). Although among the bacterial isolates, some were normal human or animal microbiota or environmental saprophytes, others are known as opportunistic pathogens or allergenic for humans, especially for immuno-compromised individuals. *S. aureus* causes suppurative diseases, pyogenic lesions on the skin, septicaemia and food poisoning (1). *E. coli* is associated with diarrhoea, gastroenteritis, endocarditis, septicaemia, urogenital infection and pneumonia (21). Both *Enterobacter* and *Proteus* species have been implicated in diarrhoea and urinary tract infection in humans (31). *K. pneumoniae* is an important cause of pneumonia and pulmonary infections. Gram-negative bacteria are potent endotoxin producers and their remnants may be inhaled into the bronchi and alveoli of the human lung (27), thereby contributing to chronic respiratory symptoms reported frequently among cattle farm workers in Egypt.

The type and frequency distribution of fungi isolated from workers in cattle farms are illustrated in Table V which shows that the following were isolated from hand swabs of dairy farm workers:

- *A. fumigatus* (8.3%)
- *A. niger* (6.7%)
- *A. flavus* (10%)
- *P. citrinum* (5%)
- *Cladosporium* spp. (8.3%)
- *Mucor* spp. (3.3%)
- *Alternaria* spp. (1.7%)
- *Absidia* spp. (0%)
- *Curvularia* spp. (1.7%)
- *Epicoccum* spp. (1.7%)
- *Rhizopus* spp. (3.3%).

The same species in the same order were isolated from hand swabs of beef farm workers as follows:

- *A. fumigatus* (15%)
- *A. niger* (11.7%)

- *A. flavus* (10%)
- *P. citrinum* (8.3%)
- *Cladosporium* spp. (11.7%)
- *Mucor* spp. (6.7%)
- *Alternaria* spp. (3.3%)
- *Absidia* spp. (3.3%)
- *Curvularia* spp. (3.3%)
- *Epicoccum* spp. (0%)
- *Rhizopus* spp. (1.7%).

The fungal species isolated from nasal swabs of dairy farm workers were as follows:

- *A. fumigatus* (6.7%)
- *A. niger* (5%)
- *A. flavus* (6.7%)
- *P. citrinum* (3.3%)
- *Cladosporium* spp. (5%)
- *Mucor* spp. (0%)
- *Alternaria* spp. (0%)
- *Curvularia* spp. (1.7%).

The comparable percentages of these species in nasal swabs of beef farm workers were as follows:

- *A. fumigatus* (10%)
- *A. niger* (6.7%)
- *A. flavus* (8.3%)
- *P. citrinum* (5%)
- *Cladosporium* spp. (10%)
- *Mucor* spp. (3.3%)
- *Alternaria* spp. (1.7)
- *Curvularia* spp. (0%).

Similar fungal species, but at low frequencies, have been isolated from hand swabs and sputum of poultry farm workers in Egypt (34). In contrast, higher occurrences of certain isolated fungal species (*Cladosporium*, *Penicillium* and *Alternaria* species) have been detected in nasal and skin specimens of patients with upper respiratory tract and allergic manifestations (8). *A. fumigatus* is an important cause of allergic alveolitis, asthma, pulmonary aspergillosis and possibly mycotoxicosis (20). *Cladosporium* species is associated with cerebral chromomycosis, allergy and mycotic keratitis (13). Some other fungi (*A. niger*, *A. flavus*, *P. citrinum* and *Curvularia* spp.) may cause allergic bronchopulmonary mycoses (14). *Alternaria* spp. is incriminated in sinusitis, allergic rhinitis, keratitis and maxillary osteomyelitis whereas *Mucor*, *Rhizopus* and *Absidia* spp. are

allergenic and can cause mucromycosis in lungs, nasal sinuses, brain, eyes and the skin of immuno-depressed individuals (13).

Conclusions

From the above results, it can be concluded that there is a high frequency of bacterial and fungal species in hand and nasal swabs of beef cattle workers compared to dairy workers. This may be attributed to the low level of hygiene and high degree of air pollution as indicated by higher total bacterial and fungal counts. This, in turn, results in the contamination of the hands of the workers and in the colonisation of nasal mucosa. In addition, poor hygiene and the presence of muddy floors, high stocking densities and

inadequate ventilation on beef cattle farms can be considered to be additional contributing factors (5).

In conclusion, airborne micro-organisms on cattle farms in Egypt could lead to acquired infections. Cattle sheds, particularly those for beef cattle, represent important sources of different zoonotic micro-organisms. To minimise the potential airborne risks, regular cleaning and disinfection, hygienic disposal of collected manure, adequate ventilation and correct stocking densities are required. Masks and gloves should be used by workers who should be trained by biosafety personnel to decrease the risk of occupational infections in the future.

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