

Depts. of Animal Health & Zoonosis  
Fac. Vet. Med., Alexandria Univ.

## MICROBIOLOGICAL IMPURITIES OF AIR IN BROILER HOUSES

(With 4 Tables)

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(Received at 30/9/2001)

الملوثات الميكروبيولوجية للهواء في مساكن دجاج التسمين

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تم جمع ١٥٠ عينة هواء من داخل وخارج مساكن دجاج التسمين وذلك للفحص البكتريولوجي والميكولوجي وتقدير السمومة الداخلية للبكتريا بهذه المساكن. أظهرت النتائج أن العدد الكلي للبكتريا والفطريات والهواء خارج وداخل هذه المساكن كان  $1.3 \times 10^5$  ،  $6.4 \times 10^6$  ،  $7.2 \times 10^2$  ،  $9.7 \times 10^2$  و  $1.0 \times 10^7$  ،  $1.0 \times 10^9$  على الترتيب. كما تبين أن مستوى السموم الداخلية للبكتريا والهواء خارج وداخل المساكن كان  $1054$  و  $1098$  وحدة سموم داخلية لكل متر مكعب من الهواء على التوالي. كذلك أشارت النتائج التي تم الحصول عليها إلى مصادر مثل هذه الملوثات للهواء داخل مساكن الدجاج.

### SUMMARY

A total of 150 air samples were collected from outside and inside air of broiler houses and subjected to bacteriological, and mycological investigation as well as determination of endotoxins concentration. The obtained results revealed that the mean total bacterial and fungal count of outside and inside air, were  $1.3 \times 10^5$ ,  $6.4 \times 10^6$  and  $7.2 \times 10^2$ ,  $9.7 \times 10^2$ , respectively. In addition, isolation and identification of certain bacteria and fungi, was carried out. Moreover, the mean concentrations of endotoxins of outside and inside air samples of broiler houses were 1054 and 1098 Eu/m<sup>3</sup>. The results, also gave some indications on the sources of such microbiological impurities of air in poultry houses.

*Key words: Microbiological impurities in broiler houses.*

## INTRODUCTION

Inhalation of airborne microorganisms and their toxins has been recognized as an important factor in the prevalence of respiratory disease within the farm community.

The most common bacterial families suspended in air of animal and poultry houses were Enterobacteriaceae, Pseudomonaceae and the Neisseriaceae. Within the family Enterobacteriaceae, the species *Escherichia coli* and *Enterobacter agglomerans* were predominant (Zucker *et al.*, 2000). Air carry dust particles loaded with different species of microorganisms. Such biological impurities may be suspended and disseminated in surrounding environment. So, infection easily to be introduced into poultry farms.

Inhalation of organic dusts contaminated by endotoxin has been associated with the development of subjective symptoms and transient or chronic lung infection impairment in workers as well as with development of occupational chronic lung obstructive bronchial diseases (Rylander, 1987 and Bergmann & Masken, 1994).

Clark *et al.* (1983) found average endotoxin concentrations in poultry confinement units of  $0.31 \text{ ug/m}^3$ . Lacey and Dutkiewicz (1994) mentioned that a suggested threshold level of endotoxin for men is  $200 \text{ ng/m}^3$ .

Endotoxin persists in the air after death of airborne gram negative bacteria of short survival period (Muller *et al.*, 1980).

The main sources of airborne bacterial endotoxins are dust, manure and feed contaminated with gram negative bacteria (Draz, 1998).

The purpose of this study was to determine the total bacterial and fungal count, isolation and identification of bacteria and fungi as well as bacterial endotoxins in poultry houses.

## MATERIAL and METHODS

### 1- Sampling:

A total of 150 air samples, [75 from outside (about 10 meter) and 75 from inside (at the middle)] were collected from broiler houses using AGI-30 sampler (Brachmann *et al.*, 1964).

### 2- Determination of total bacterial count:

AGI-30 sampler were opened for 20 minutes at an air flow rate of 12.5 l/min. The air samples were cultivated on 5% sheep blood agar to obtain the total number of airborne aerobic bacteria.

**3- Determination of total fungal count of air:**

The air samples were cultivated on sabouroud's dextrose agar for 3-5 days at room temperature.

**4- Isolation and identification of bacteria and fungi:**

It was carried out according to the scheme described by Barnet and Hunter (1972); Edward and Ewing (1972); Cruickshank *et al.* (1975) and Onions *et al.* (1981).

**5- Determination of endotoxin concentration of air samples:**

Air samples were collected in 50 ml. Pyrogen free water. The amount of endotoxin in sampler fluids was determined by using LAL assay QLC-1000 (Bio Whittaker, Walkersville, MD) as recommended by the manufacture. The endotoxin values obtained from the sampler fluid were converted from EU/ml to EU/m<sup>3</sup> air using the air flow rate of the sampler and the sampling time.

## DISCUSSION

This study was planned to obtain information about the airborne bacteria and fungi in broiler houses as well as concentration of gm-negative bacterial endotoxins.

The data presented in Table (1) revealed that the mean counts of viable bacteria outside and inside broiler houses were  $1.3 \times 10^5$  and  $6.4 \times 10^6$  CFU/m<sup>3</sup> respectively. However, increasing the number of bacteria in air may be due to insufficient ventilation rate, overcrowding, poor sanitary condition and increased of the stirred dust particles suspended in air carrying microorganisms. Moreover, it was recorded that the degree of air pollution affected the mortality rate, feed consumption, weight gain, and carcass quality (Rudy, 1985).

The mean fungal count of air outside and inside broiler houses were  $7.2 \times 10^2$  and  $9.7 \times 10^2$  CFU/m<sup>3</sup>, respectively (Table 1). However, the sources of airborne microorganisms were incoming air, feed, and dust (Baikov and Petkov, 1986).

Table (2) shows bacteria isolated from air of outside and inside broiler houses. The isolated bacteria were identified as staphylococcus aureus (4.6%), Streptococcus faecalis (10%), Salmonella spp. (4%) [S.typhimurium (2%), S.enteritidis (1.3%), S.Newport (0.6%)], E.coli (11.3%), Klebsiella spp. (12%), Arizona spp (2-6%), Proteus rettgeri (1.3%), Proteus mirabilis (1.3%) and Pseudomonas spp. (4.6%).

Concerning the hygienic significance, Staphylococcus aureus is considered as an aetiologic agent in cases of wound infection,

Septicaemia, Spondylitis, Omphalitis, Emaciation and Bacterial endocarditis (Povar and Brownstein, 1947), *Streptococcus faecalis* is implicated in a variety of diseases of broilers, it cause acute streptococcal septicaemia of chickens with losses up to 50% (Norgaard and Mohler, 1902).

*Salmonella* species was incriminated in cases of pneumonia, egg peritonitis, enteritis, retained yolk sac, omphalitis and chronic respiratory diseases (Bhatia *et al.*, 1971; Verma & Adlaka, 1971). Infection by *Salmonella* enteritidis between groups of chicks was apparently transmitted principally by oral ingestion, perhaps from environmental surfaces contaminated by airborne movement of the pathogen (Gast *et al.*, 1998).

*E. coli* is responsible for various diseases causing major economic losses to poultry industry including colibacillosis, Hjarre's disease, coligranuloma, Peritonitis, salpingitis, Synovitis and Omphalitis (Verma and Adlaka, 1971). *Klebsiella* spp. are considered as opportunistic pathogens in the respiratory and urinary system (Cowan *et al.*, 1960). Arizona organisms can be localized in the brain of infected chicks leading to convulsions, blindness, ataxia, torticollis and mortality rate may reach to 10% (Silva *et al.*, 1980). Members of the genus proteus were implicated in omphalitis and unabsorbed yolk sac leading to mortality of chickens up to 10 days old (Bhatia *et al.*, 1971).

The data presented in Table (3) shows fungi isolated from air of outside and inside broiler houses. The recovered fungi were *Aspergillus niger* (21.3%), *Aspergillus flavus* (4.6%), *Aspergillus ochraceus* (2%), *Aspergillus nidulans* (1.3%), *Aspergillus candidus* (3.3%), *Alternaria* spp (2%), *Cladosporium* (2%), *Rhizopus* spp. (2.6%), *Mucor* spp. (10%), *Candida albicans* (7.3%); *Rhodotorula rubra* (1.3%) and *penicillium* sp. (14.6%).

Concerning the hygienic significance of isolated fungi. *Aspergillus* species are incriminated in poultry mycotoxicosis (Lovett, 1972), the acute form known as broader pneumonia, while the chronic form is characterized by respiratory and digestive manifestation (Refai *et al.*, 1992). *Penicillium* spp. are incriminated in some cases of gastrointestinal disturbances and poultry mycotoxicosis (Lovett, 1972).

*Fusarium* and *Alternaria* species are responsible for some cases of poultry mycotoxicosis (Forgaw *et al.*, 1966). *Cladosporium* species are implicated in some cases of dermatitis in poultry mycotoxicosis (Lovett, 1972). *Candida* species causes candidiasis in poultry which is

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characterized by infection of the upper alimentary canal particularly of the crop (Refai *et al.*, 1992).

It was found that the mean concentration of endotoxin outside and inside broiler houses were 1054 and 1098 EU/m<sup>3</sup> respectively (Table 4). Larsson *et al.* (1999) reported a concentration of endotoxin approximately 100 ng/m<sup>3</sup> in poultry houses. However, the flooring and feeding were predominant characteristics explaining variation in endotoxin exposure. In addition Wathes (1995) found a correlation between concentration of airborne endotoxin and obstructive pulmonary disease.

It can be concluded that the air inside broiler houses was found more polluted than outside air which act as a source of pollution for such houses. Also, the bad hygienic house was associated with increase in endotoxin concentration as well as number of bacteria and fungi. Moreover, activities performed inside poultry houses like feeding, cleaning, changing of the litter and others were associated with exposure to dust, bacteria, fungi and endotoxins. So, houses must be hygienically constructed, well ventilated with regular removal of waste materials as well as avoid overcrowding of birds and disruption of dust inside poultry houses.

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Table 1: Total bacterial and fungal count of air outside and inside broiler house:

Site of sampling	Total bacterial count (CFU/m <sup>3</sup> )			Total fungal count (CFU/m <sup>3</sup> )		
	Min.	Max.	Mean	Min.	Max.	Mean
Outside the house	2.9x10 <sup>5</sup>	5.7x10 <sup>6</sup>	1.3x10 <sup>5</sup>	2.2x10 <sup>2</sup>	9.8x10 <sup>3</sup>	7.2x10 <sup>2</sup>
Inside the house	3.7x10 <sup>3</sup>	8.6x10 <sup>6</sup>	6.4x10 <sup>6</sup>	9.4x10 <sup>2</sup>	6.5x10 <sup>3</sup>	9.7x10 <sup>2</sup>

Table 2: Bacteria isolated from air of outside and inside broiler house.

Isolate	Outside the house (N=75)		Inside the house (N=75)		Total (N=150)	
	+ve	%	+ve	%	+ve	%
<i>Staphylococcus aureus</i>	1	1.3	6	8.0	7	4.6
<i>Staphylococcus epidermidis</i>	2	2.6	3	4.0	5	3.3
<i>Streptococcus faecalis</i>	2	2.6	8	10.6	15	10.0
<i>Salmonella spp.</i>	-	0.0	6	8.0	6	4.0
<i>S. typhimurium</i>	-	0.0	3	4.0	3	2.0
<i>S. enteritidis</i>	-	0.0	2	2.6	2	1.3
<i>S. Newport</i>	-	0.0	1	1.3	1	0.6
<i>E. coli</i>	4	5.3	13	17.3	17	11.3
<i>Klebsiella</i>	3	4.0	15	20.0	18	12.0
<i>Arizona sp.</i>	1	1.3	3	4.0	4	2.6
<i>Proteus retergri</i>	-	0.0	2	2.6	2	1.3
<i>Proteus mirabilis</i>	-	0.0	2	2.6	2	1.3
<i>Pseudomonas spp.</i>	2	2.6	5	6.6	7	4.6

Table 3: Fungi isolated from air of outside and inside broiler house.

Isolate	Outside the house (N=75)		Inside the house (N=75)		Total (N=150)	
	+ve	%	+ve	%	+ve	%
<b>A) Mould:</b>						
<i>Aspergillus niger</i>	12	16.0	20	26.6	32	21.3
<i>Aspergillus flavus</i>	1	1.3	6	8.0	7	4.6
<i>Aspergillus ochraceus</i>	-	0.0	3	4.0	3	2.0
<i>Aspergillus nidulans</i>	-	0.0	2	2.6	2	1.3
<i>Aspergillus candidus</i>	1	1.3	4	5.3	5	3.3
<i>Penicillium spp.</i>	7	9.3	15	10.0	22	14.6
<i>Fusarium spp.</i>	1	1.3	4	5.3	5	3.3
<i>Alternaria spp.</i>	-	0.0	3	4.0	3	2.0
<i>Cladosporium spp.</i>	-	0.0	3	4.0	3	2.0
<i>Rhizopus spp.</i>	1	1.3	3	4.0	4	2.6
<i>Mucor spp.</i>	6	8.0	9	12.0	15	10.0
<b>B) Yeast and yeast like organisms:</b>						
<i>Candida albicans</i>	2	2.6	9	12.0	11	7.3
<i>Rhodotorula rubra</i>	-	0.0	2	2.6	2	1.3
<i>Torulopsis versatilis</i>	1	1.3	3	4.0	4	2.6
Unidentified fungi	3	4.0	5	6.6	8	5.3

Table 4: Concentration of airborne endotoxin outside and inside broiler house.

Site of sampling	Endotoxin unit (EU/m <sup>3</sup> )		
	Min.	Max.	Mean
Outside the house	744	600	1054
Inside the house	1380	4818	1098