The influence of housing systems on the air quality and bacterial eggshell contamination of table eggs

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ABSTRACT: This paper compares two different housing systems for laying hens producing table eggs, namely a conventional cage system and an aviary, during three summer months, starting from the 20th week of the production cycle. Research was focused on airborne bacteria, fungi and dust levels and on the bacterial eggshell contamination. Levels of airborne bacteria determined in the aviary system were many times higher and ranged from 6.2×10^4 CFU/m^3 to 8.9×10^4 CFU/m^3, and the levels of airborne fungi ranged from 1.6×10^4 to 1.9×10^4 CFU/m^3, while the levels of airborne bacteria and fungi determined in the conventional cage system ranged from 1.6×10^4 to 2.5×10^4 CFU/m^3 and from 0.8×10^4 to 1.3×10^4 CFU/m^3, respectively. Microbial air contamination was associated with eggshell contamination, with the levels in the aviary ranging from 5.4×10^3 to 9.6×10^3 CFU/eggshell and those in the conventional cage system ranging from 2.3×10^3 to 3.6×10^3 CFU/eggshell. Airborne dust levels in the aviary and conventional cage system ranged from 3.2 to 4.6 mg/m^3 and from 0.7 to 1.2 mg/m^3, respectively. From the aspect of animal welfare and behavioural requirements, alternative systems, i.e. aviaries, appear more acceptable; however, they are not satisfactory from hygienic aspects because of a higher content of airborne pollutants which can represent a greater risk of horizontal contamination of the egg content.

Keywords: conventional cage system; aviary; air hygiene; eggshell; microorganisms

Housing systems for laying hens producing table eggs have changed significantly in recent years. As of 1 January 2012 conventional cage systems will be prohibited and replaced by alternative housing systems (enriched cage systems, aviaries, free-range systems) (EU Directive, 1999).

Cages are the most common housing systems for hens intended for table egg production. Welfare

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problems associated with this particular housing system result from hindered physical activities. Conventional cages restrict freedom of movement and prevent hens from meeting their behavioural requirements such as wing-flapping, nest building, scratching for food, dust bathing, etc. Such restrictions lead to various frustrations, feather pecking and other abnormal behavioural patterns. It has been demonstrated that the poultry kept in cage systems had lower weight and higher bone fragility than the poultry kept in free-range systems. Alternative systems, such as enriched cages, aviaries, free-range systems, should provide the most natural environment possible for the keeping and breeding of laying hens and for egg production. At the same time, such systems should reduce to a minimum any stressful conditions, in particular pain, suffering, discomfort and fear (Vučemilo, 2008).

Many studies have shown that laying hen mortality rates are higher in alternative systems than in conventional cage systems, due to their higher exposure to various diseases, feather pecking and cannibalism, combined with problems in maintaining optimum microclimate conditions, higher feed consumption and increased feed waste. Percentage of dirty and cracked eggs is higher in alternative housing systems, because a part of these eggs are laid on the floor. Also, the eggs are lighter and feed conversion is less efficient. Obviously, the enriched systems, aviaries and other alternative housing systems for laying hens are much better from the animal welfare aspect. However, they pose threat to their immediate environment, in particular the risk of air pollution with bacteria, fungi, dust, endotoxins and ammonia, which is significantly higher in alternative than in conventional systems (Rodenburg et al., 2005; Vučemilo et al., 2007a, 2008).

Hygiene is an important link, not only in terms of health and production performance but also in terms of food safety. In alternative systems where the birds move freely in their environment, a significant amount of dust originating from litter is created, having as a consequence air contamination by microorganisms and endotoxins (Hartung, 1994; Wathes, 1994). It has been demonstrated that the facilities with litter have ten times more airborne bacteria and twenty to thirty times more bacteria on the eggshells as compared to the cage housing systems (De Reu et al., 2005). Eggshell contamination by aerobic bacteria is generally higher in eggs coming from alternative systems as compared to those coming from enriched or conventional cage systems (De Reu et al., 2008). Fiks-van Niekerk (2005) pointed out high eggshell contamination in an alternative system as well as a positive correlation between the total airborne bacteria count in the housing system and the initial eggshell contamination, as reported by Protais et al. (2003a,b). De Reu et al. (2006a) reported the significantly higher average eggshell contamination ($P < 0.001$) by aerobic bacteria in eggs coming from alternative housing systems as compared to those coming from conventional ones, in particular 5.46 against 5.08 log CFU/eggshell. They also determined a positive but statistically insignificant correlation between the initial bacterial eggshell contamination and the level of airborne bacteria in the housing system.

De Reu et al. (2006b) and Messens et al. (2007) proved that higher eggshell contamination led to a greater possibility of microorganism penetration and egg content contamination. The number of airborne microorganisms in laying hen dwellings could represent a higher risk of horizontal eggshell contamination as well as contamination of the egg content. This was the reason why this research focused on determining the air quality and eggshell bacterial contamination in conventional cage systems and aviaries.

MATERIAL AND METHODS

Research was performed on two laying hen farms, in different dwellings in which hens were at the 20^{th}, 45^{th}, and 65^{th} production week, in the area of the Zagreb County. Conventional cage systems housed about 17 000 laying hens of the Shaver hybrid and aviaries housed about 6 000 laying hens of the Lohman hybrid. Feeding, *ad libitum*; watering, ventilation, lighting and manure removal were controlled automatically. Light was provided for 16 hours per day from 05:00 h to 21:00 h.

In the conventional housing system eggs were collected manually while in the aviary the nests were located on the upper tier and the eggs were transported by a conveyor belt to the sorting line.

Over a three-month period, sampling was performed at regular intervals by means of Merck MAS 100 microbial air sampling system (Merck KgaA, Darmstadt, Germany). At the same time, air temperature (t, °C), relative humidity (RH, %) and air flow rate

(w, m/s) were measured with Testo 400 (Testo Inc. Lenzkirch, Germany). Dust samples were collected from filters (Whatman International Ltd., Maidstone, UK) by means of SKC pump (SKC Ltd., Blandford Forum, UK). The airflow rate was 4.0 l/min. Filters were weighed before and after sampling in a controlled laboratory at air temperature of 22°C and relative humidity of 45% (± 5%). Air was sampled on a commercially available nutrient agar. Columbia agar was used for bacteria and Sabouraud agar (Biolife, Milan, Italy) for fungi. Plates with the usual bacterial nutrient Columbia agar were then incubated for 24 hours in an incubator at a work temperature of 37°C. The material sampled on Sabouraud agar was incubated for 5 days at 22°C. The grown colonies (CFU/m^3) were calculated by a mechanical optic colony counter, and results were corrected by use of the respective table and mathematical equation (Anonymous, 1998). In the conventional housing system, air samples were collected at five points along the central corridor, at the level of the second tier. In the aviary, air samples were also taken at five points, along the passage between the tiers. All measurements were performed from 9 a.m. to 1 p.m.

Microclimate indicators and air samples were taken eight times over three summer months, starting from the 20th week of the production cycle.

Every time the eggshell surface of 15 eggs was swabbed two hours after laying with a sterile wet cotton swab, previously immersed in sterile saline. Then the swabs were stored in a refrigerator at 4°C and delivered to the laboratory for further processing according to the procedure described by Quinn et al. (1994).

Determined values of measured parameters were processed by computer programmes Microsoft Excel and Statistica 7. Before analysis we log-transformed data and afterward they were returned into normal distribution. The procedure also included descriptive statistical analysis and determination of statistical significance level of 1 and 5%, ($P < 0.01$, $P \leq 0.01$ and $P < 0.05$) by Student's t-test (Anonymous, 1994; Petz, 2002).

RESULTS AND DISCUSSION

Microorganisms are always present in animal housing facilities, most of them being saprophytes that are most frequently present in dust particles or aerosols. They originate from animals, feed, litter, floor and other surfaces, excreta, secretions, etc. Such airborne microorganisms remain in the air for a shorter or longer period of time, depending on the size and ventilation of the facility, on the air flow rate and the carrier. Viability and infectivity of different microorganisms depend on a number of factors, including relevant physical and biochemi-

Table 1. Descriptive statistical analysis of airborne bacteria and fungi levels (CFU/m^3) in housing systems A and B

Sampling time	Microorganisms (CFU/m^3)	n	X	Min.	Max.	SD	s
Production week 20	bacteria A	8	2.1×10^4	0.8×10^4	3.1×10^4	0.99	0.35
	fungi A	8	0.8×10^4	0.2×10^4	2.2×10^4	0.63	0.22
	bacteria B	8	6.2×10^4	1.1×10^4	9.2×10^4	2.88	1.02
	fungi B	8	1.6×10^4	0.4×10^4	2.7×10^4	0.65	0.23
Production week 45	bacteria A	8	2.5×10^4	0.9×10^4	3.3×10^4	1.01	0.36
	fungi A	8	1.2×10^4	0.7×10^4	2.2×10^4	0.53	0.19
	bacteria B	8	8.6×10^4	5.9×10^4	11.0×10^4	1.67	0.59
	fungi B	8	1.8×10^4	0.9×10^4	3.7×10^4	0.89	0.31
Production week 65	bacteria A	8	1.6×10^4	0.6×10^4	3.8×10^4	1.19	0.42
	fungi A	8	1.3×10^4	0.5×10^4	2.2×10^4	0.61	0.22
	bacteria B	8	8.9×10^4	4.5×10^4	13.4×10^4	2.96	1.05
	fungi B	8	1.9×10^4	0.8×10^4	3.7×10^4	1.03	0.36

A = conventional cages; B = aviary; X = mean values; SD = standard deviation; s = standard error of the standard deviation

Table 2. Descriptive statistical analysis of microorganism levels on eggshells coming from housing systems A and B

Sampling time	Microorganisms (CFU/eggshell)	*n*	X	Min.	Max.	SD	s
Production week 20	A	15	2.3×10^3	3.9×10^2	7.2×10^3	4 741 395	2 177.47
	B	15	9.6×10^3	5.4×10^3	1.7×10^4	9 791 141	3 129.08
Production week 45	A	15	3.6×10^3	3.6×10^2	1.0×10^4	12 263 507	3 501.93
	B	15	8.2×10^3	2.4×10^2	2.1×10^4	29 068 367	5 391.51
Production week 65	A	15	2.3×10^3	1.2×10^2	9.8×10^3	8 106 738	2 847.23
	B	15	5.4×10^3	1.5×10^2	1.7×10^4	23 726 650	4 871.00

A = conventional cages; B = aviary; X = mean values; SD = standard deviation; s = standard error of the standard deviation

cal properties of the aerosol, and on the microclimate complex inside the facility.

Comparative examination of sanitary conditions in the conventional cage system and in the aviary has shown a significant difference in the air quality in terms of airborne bacteria, fungi and dust levels as well as in terms of bacterial/fungal count on the eggshells. The airborne bacteria count in the conventional cage system ranged from 1.6×10^4 to 2.5×10^4 CFU per m^3 while in the aviary it was significantly higher and ranged from 6.2×10^4 CFU per m^3 to 8.9×10^4 CFU per m^3, which was confirmed by Student's *t*-test at a 1% statistical significance level ($P < 0.01$) (Table 1, Table 4). Similarly, the airborne fungi count in the conventional cage system ranged from 0.8×10^4 to 1.3×10^4 CFU/m^3, and in the aviary from 1.6×10^4 to 1.9×10^4 CFU/m^3 (Table 1). It was stated that this count was statistically more significant ($P < 0.01$) in the aviary at the initial stage of the production cycle (20^{th} week) (Table 4). In the conventional cage system with a 72-week production cycle, Vučemilo et al. (2007b) reported that the airborne bacteria count ranged from 7.9×10^3 CFU/m^3 at an early stage of the cycle to 1.2×10^4 CFU/m^3 at a later stage of the cycle, and the fungi count ranged from 6.8×10^3 CFU per m^3 to 1.0×10^4 CFU/m^3. Similar differences in the load of airborne microorganisms were reported by a number of authors, e.g. by Saleh et al. (2003), who examined airborne bacteria counts in three different housing systems for laying hens producing table eggs. These authors reported bacterial counts of 2.2×10^6 CFU/m^3 in aviaries, 0.3×10^6 CFU/m^3 in conventional cage systems and 0.1×10^6 CFU per m^3 in enriched cage systems. Protais et al. (2003a) and De Reu et al. (2006a) determined on average 4 log CFU/m^3 of airborne bacteria in dwellings with conventional cages, which is 100 times less than in aviaries (> 6 log CFU/m^3).

Airborne dust levels in the conventional cage system ranged from 0.7 to 1.2 mg/m^3. In aviaries, airborne dust levels ranged from 3.2 to 4.6 mg per m^3 (Table 3). Airborne dust levels in aviaries increased from one week to another and throughout the monitoring period they were statistically significantly higher ($P \leq 0.01$) than the levels recorded in con-

Table 3. Descriptive statistical analysis of airborne dust levels in housing systems A and B

Dust	Dust level (mg/m^3)	*n*	X	Min.	Max.	SD	s
Production week 20	A	4	0.9	0.8	0.9	0.06	0.03
	B	4	3.2	2.9	3.4	0.29	0.14
Production week 45	A	4	1.2	1.1	1.2	0.05	0.03
	B	4	3.7	3.5	3.8	0.17	0.08
Production week 65	A	4	0.7	0.6	0.7	0.06	0.03
	B	4	4.6	4.5	4.6	0.06	0.03

A = conventional cages; B = aviary; X = mean values; SD = standard deviation; s = standard error of the standard deviation

Table 4. Student's *t*-test indicating statistically significant difference between the levels of microorganisms in the air and on the eggshells and the airborne dust levels determined in two different housing systems

Sampling time	Pollutants	*n*	SD	*t*	*P*
Production week 20	airborne bacteria A/B	8	2.231	–5.173	0.001
Production week 45		8	1.967	–8.708	0.000
Production week 65		8	3.246	–6.356	0.000
Production week 20	airborne fungi in A/B	8	0.641	–3.568	0.009
Production week 45		8	0.757	–2.245	0.060
Production week 65		8	1.430	–1.188	0.274
Production week 20	microorganisms on eggshells A/B	15	3 129.08	–7.464	0.000
Production week 45		15	5 391.51	–2.794	0.009
Production week 65		15	4 871.00	–2.133	0.042
Production week 20	airborne dust in A/B	4	0.071	–9.021	0.012
Production week 45		4	0.212	–15.814	0.004
Production week 65		4	0.354	–55.154	0.000

*$P < 0.05$

ventional cage systems (Table 4). Martensson (1995) found out total airborne dust levels of 2.0 mg/m^3 and 3.4 mg/m^3 in a conventional housing system and an alternative housing system, respectively. Similar observations were reported by Mirtensson and Pehrson (1997), pointing out that such results indicate many times higher air contamination by dust in alternative housing systems as opposed to cage housing systems. Similar results were reported by Ellen et al. (2000), who determined four to five higher air contamination in aviaries compared to cage systems, while Michel and Huonnic (2003) measured 15 times higher air contamination in aviaries than in conventional cage systems (31.6 compared to 2.3 mg/m^3). Zoons et al. (2005) reported five times higher air dust contamination in aviaries in comparison with cage systems (10.1 versus 2.1 mg/m^3).

Microclimate parameters in both systems were within the limits recommended for this particular category of poultry, just the air flow velocity in the aviaries was above the upper limit values (up to 0.7 m/s).

Many studies confirmed that the total count of airborne bacteria in a hen housing system was in positive correlation with the initial eggshell bacterial contamination (Protais et al., 2003a; De Reu et al., 2006a). In our study 2.3×10^3 to 3.6×10^3 CFU per eggshell of microorganisms were determined in the conventional cage system, while in the aviary this count ranged from 5.4×10^3 to 9.6×10^3 CFU per eggshell (Table 2). Statistical data processing confirmed that throughout the monitoring period the amount of microorganisms on eggshells coming from the conventional housing system was significantly lower ($P < 0.01$) (Table 4). De Reu et al. (2006a) reported significantly higher ($P < 0.001$) initial eggshell contamination and total aerobic bacterial count in alternative systems compared to conventional ones, namely 5.5 compared to 5.1 log CFU/eggshell. Similar values were measured in researches of Protais et al. (2003a,b) and De Reu et al. (2005, 2006b).

In our investigation much lower values of air pollutants in general, as well as of microorganisms on the eggshell were determined in comparison with other researches. This could be related with the higher ventilation level in summer months. Similarly like the others, we determined a significantly higher number of airborne microorganisms in aviaries in comparison with conventional systems and consequently a higher microorganism number on the eggshell from aviaries.

CONCLUSION

As a general conclusion, air and eggshell contamination by bacteria, fungi and dust is higher in aviaries than in conventional cage systems. Bacterial

count in the aviary air ranged from 6.2×10^4 CFU per m^3 to 8.9×10^4 CFU/m^3, the level of airborne fungi ranged from 1.6×10^4 to 1.9×10^4 CFU/m^3, while in the conventional cage system airborne bacteria ranged from 1.6×10^4 to 2.5×10^4 CFU per m^3, and airborne fungi from 0.8×10^4 to 1.3×10^4 CFU per m^3. The amount of microorganisms on the eggshell in aviaries ranged from 5.4×10^3 to 9.6×10^3 CFU/eggshell and in conventional cage systems this amount ranged from 2.3×10^3 to 3.6×10^3 FU/eggshell.

The airborne dust level ranged from 3.2 to 4.6 mg per m^3 and from 0.7 to 1.2 mg/m^3 in aviaries and conventional housing systems, respectively.

From the aspect of animal welfare and behavioural requirements, alternative systems, i.e. aviaries, appear more acceptable; however, they are not satisfactory from hygienic aspects because of a higher content of airborne pollutants which can represent a greater risk of horizontal contamination of the egg content.

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