Epidemiology of *Salmonella* infection in laying hens with special emphasis on the influence of the housing system

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Summary

From January 1st 2012 onwards housing of laying hens in battery cages will be forbidden in the EU and only alternative housing such as enriched cages and non-cage systems (barn, free-range and free-range organic) will be allowed. This ban aims to improve the welfare of laying hens, yet it has also initiated the question whether there aren't any adverse consequences of this decision on the spread and/or persistence of infectious (zoonotic) diseases in a flock.

In this paper an overview is given of all currently available information on the effect of the used housing system on the occurrence and epidemiology of *Salmonella* infections in laying hens.

Keywords: Laying hens, Salmonella, Epidemiology, Housing system

Introduction

Already since the late 1960's there was a growing public awareness concerning farm animal welfare which resulted in a consumer's aversion to eggs produced by laying hens housed in cages (Appleby, 2003). This finally resulted in a ban of the housing of laying hens in conventional battery cages in the EU, from January 1st 2012 onwards (Council Directive 1999/74/EC). From this date on, the housing of laying hens in the EU will be restricted to enriched cages and non-cage systems (barn, free-range and free-range organic). This ban on battery cages aims to improve the welfare of laying hens (Wall et al., 2004), yet it has also initiated the question whether there aren't any adverse consequences of this decision on the spread and/or persistence of infectious (zoonotic) diseases in a flock. This fear is based on the opinion that one of the big advantages of conventional battery cages is that, because hens are separated from their faeces, the risk for disease transmission through faeces can be minimized (Duncan, 2000). It has also been stated that the increased exposure of layers to environmental contamination in non-cage systems would increase the risk of contamination with *Salmonella* (Kinde et al., 1996; EFSA, 2005)

Salmonella remains an important cause of human disease worldwide. In Europe, *S*. Enteritidis and *S*. Typhimurium are the most commonly isolated serotypes in human cases of salmonellosis (EFSA, 2007) and contaminated eggs still remain the most important source of infection with *S*. Enteritidis for humans (Crespo et al., 2005; De Jong and Ekdahl, 2006).

The aim of this paper is to review all currently available information on the effect of the used housing system on the occurrence and epidemiology of *Salmonella* infections in laying hens.

Effect of the housing system on *Salmonella* prevalence

A number of observational studies, varying from small to very large sample sizes, have evaluated the effect of the housing system on the *Salmonella* prevalence. As often is the case, these studies have variable results going from a preventive effect of the cage system over no influence up to a risk factor.

One study showed a significant lower *Salmonella* prevalence in laying hens housed in cage systems in comparison deep litter systems (Mollenhorst et al., 2005). Yet it has to be mentioned that in this study the antibody response was used as the outcome variable. It is questionable whether a serological response is a good method to estimate the level of infection in laying hens. Garber et al., (2003) found that rearing pullets on floor resulted in a higher risk of being SE positive in comparison to cage rearing systems.

A number of studies were not able to demonstrate any significant effect of the housing system (cage versus non-cage) on the *Salmonella* occurrence (Schaar et al., 1997; Hald et al., 2002; Pieskus et al., 2008; Van Hoorebeke et al., 2009a).

In an earlier retrospective epidemiological study in Denmark, Mølbak and Neiman (2002) found that eggs from battery cages were associated with human S. enteritidis disease versus no association with alternative housing. Later on several studies have found that rearing flocks in cages is a significant risk factor compared to rearing in barns and free range systems (Methner et al., 2006; EFSA, 2007; Namata et al., 2008; Van Hoorebeke et al., 2009b; Huneau-Salaün et al., 2009).

A summary of all studies for which an estimation of the Odds Ratio (OR) was available or could be calculated based on the presented data is given in Table 1. Additionally the number of flocks included in the study is given to give an indication of the magnitude of the study. Results of table 1 are only indicative and one should be careful with comparing the results of the individual studies since often important methodological differences are present that may have influenced the results.

Table 1: An overview of all published observational studies that have evaluated the effect of the housing system on the prevalence of *Salmonella* Enteritidis infections.

comparison	Number of observations	OR	95% Confidence Interval	Comment	reference
Cage vs deep litter	1642	0.48	NA	serology	Mollenhorst et al., 2005
Cage vs free range	34	0.61	0.15-2.34		Schaar et al., 1997
Cage vs Aviary	8	1.28	0.51-3.21		Pieskus et al., 2008
Cage vs non-cage	30	2.11	0.28-15.77		Van Hoorebeke et al., 2009a
Cage vs non-cage	329	2.34	1.42-3.85		Methner et al., 2006
Cage vs non-cage	195	4.69	1.85-11.90		Van Hoorebeke et al., 2009b
Cage vs non-cage	3768	5.12	4.07-6.45		EFSA, 2007
Cage vs floor raised and free range	148	20.11 10.27	2.52-160.49 2.13-49.57	Dust samples Faeces samples	Namata et al., 2008
Cage vs on floor	519	35.1	12.2-101.1	All *Salmonella* spp.	Huneau-Salaün et al., 2009

Obviously it is difficult to summarize the above presented data into one single conclusion concerning the effect of the housing type (cage versus alternative housing system) given the large heterogeneity in the study objectives and designs. Nevertheless the majority of the studies clearly indicate that a cage housing system has an increased risk of being *Salmonella* positive in comparison to non-cage housing systems. Yet in most of the above cited studies it is also stated that the observed effect does not necessarily means that there is a causal relation chip between the housing type and the *Salmonella* infection or excretion level. On the contrary it is more likely that the housing system is a proxy of many other production characteristics such as magnitude of the flock or the herd, age of the building, probability of previous *Salmonella* infections on the farm, ….

Underneath a number of important production characteristics that may be both related to the housing system and the probability of a *Salmonella* infection are discussed.

Herd and flock size

As can be seen in table 2, cage flocks are normally larger flocks not only with more animals per flock but also with more flocks per herd. Moreover these different flocks in the same herd are often all in a different stage of production.

Table 2: Herd characteristics for different types of laying hen production based on the EFSA baseline study (EFSA 2007)

type	# flocks / herd mean (SD)	# birds / herd mean (SD)	# birds / flock mean (SD)
Cage	2.15 (1.87)	33600 (78000)	14700 (29000)
Floor raised	2.03 (1.50)	10600 (15500)	5900 (7500)
Free range standard	1.67 (1.31)	9500 (17500)	6000 (6100)
Free range organic	1.82 (1.66)	6300 (13000)	3300 (3000)

It has been shown in several studies that both the number of flocks, and thus the number of hens in a herd, and the number of hens in a flock are, independently from the production type, significant risk factor for *Salmonella* infections (Heuvelink et al., 1999; Mollenhorst et al., 2005; EFSA, 2007; Carrique-Mas et al., 2008b; Huneau-Salaün et al., 2009; Snow et al. 2008).

The presence of multiple flocks in one herd may enhance the risk of cross-contamination of infection from one flock to another (Carrique-Mas et al., 2008b). Especially since in multihouse systems there are often open communications between houses to give way to conveyor belts, feed pipes, passageways, etc. this close association of houses makes it easy for infections to be transferred from one flock to the other especially by rodents.

In the study of Carrique-Mas et al., (2008b) also the persistence of the infection was evaluated. It was found that the likelihood of persistence was, in contrast to the probability of infection, not significantly related to the number of birds present. Based on this the authors suggested that the effect of the flock size may have something to do with a greater risk of introduction of infection.

Stocking density

Often related to both production type and flock size is the stocking density. For many infectious diseases in production animals (Dewulf et al., 2006) it has been demonstrated that an increased stocking density also increases the prevalence of disease and the ease of spread. To our knowledge no results on this parameter are available in relation to *Salmonella* infections in laying hens. This maybe either due to the fact that this parameter was never studied or that it has been evaluated but was never found to be influential? For *Salmonella* infections in pigs the stocking density has been shown to be highly influential (Funk et al., 2001, Nollet et al., 2004).

Stress

Stressors such as re-housing (Hughes et al., 1989), thermal extremes (Thaxton et al, 1974), transport (Rigby and Petit, 1980), initiation of egg lay (Jones and Ambali, 1987), and molting (Holt, 2003) have all been shown to exacerbate infection problems in poultry. Also the housing conditions may influence the stress level in the animal. As in the effect of the housing system on Salmonella prevalence, also in the effect on the stress levels in animals some conflicting results are present. Some studies suggest that birds have less stress in cage systems (Koelkebeck et al., 1986; Craig et al., 1996) whereas other research found that hens housed in non-cage environments experience less stress (Hansen et al., 1993; Colson et al., 2008). It has been concluded that today, based on the few published, peer-reviewed studies which have conducted controlled comparisons among different housing systems there is not a clear distinction between housing systems based on the stress response of the hen.

In the framework of the EU Safehouse research project a number of studies on the effect of the housing system on the stress levels in birds are being carried out.

Age of the building and carry-over infections

The difficulty of coping with a *Salmonella* infection in a laying hen house is very well illustrated by the study of Carrique-Mas et al. (2008b) who revisited a number of *Salmonella* positive tested farms in a follow-on study. In 11out of the13 cage and 4 out of the 7 floor houses *Salmonella* was found again after restocking with a new batch of birds and cleaning and disinfection in between. These results clearly illustrates that under current practices, carry-over of infection from one production round to the next is very likely. It may even be one of the main sources of *Salmonella* infections in laying hen premises (Davies and Breslin, 2003). Although the sample size is too small to draw strong conclusions, the above results show that the rate of carry-over was lower in free-range houses in comparison to cage houses. This certainly illustrates that it is not necessarily more difficult to clean and disinfect the alternative housing systems.

In a recent study of Van Hoorebeke et al., (2009b) the age of the building was identified as a significant risk factor in the sense that the older the building is the higher the risk. This may be either an illustration of an accumulation of infection pressure or the result of the fact that older equipment is often more difficult to thoroughly clean and disinfect. Since it is believed that the challenge to new birds is dose dependant a more efficient cleaning and decontamination process may result in a lower infection level and thus less carry-over.

In general conventional battery cages are older than alternative production systems such as floor raised or free range systems (Van Hoorebeke et al., 2009b). It has also been shown that it is easier to decontaminate free range and barn systems than cage systems, possibly due to the more difficult access to the cages and associated machinery (Davies and Breslin, 2003).

Pests

The infection of a flock with *Salmonella* through vectors such as rodents, flies and beetles (Davies and Breslin, 2001; Carrique-Mass *et al*., 2009) and the reported capacity of some *Salmonella* strains isolated in poultry to develop a biofilm contributes to the survival of *Salmonella* in the environment of poultry houses (Marin *et al*., 2009).
It has been suggested that cage houses present a more attractive environment to such pests compared with free range systems since birds are restrained in cages and do not interfere with their movements (Carrique-Mas et al., 2008a,b).

Vaccination

The use of vaccination against *Salmonella* has a significant protective influence on the detection of *Salmonella* in laying hen flocks or at least on the shedding of *Salmonella*. The currently available vaccines claim to reduce both the shedding and colonization of the reproductive tract which leads to a decrease of the number of internally contaminated eggs (Gantois et al., 2006). However, *Salmonella* can still be found in the caeca and reproductive tract of a fairly large proportion of vaccinated hens (Davies and Breslin, 2004; Van Hoorebeke et al., 2009a) which implies the risk of a renewed shedding of the pathogen, especially at moments of stress such as the onset and final stages of lay or induced molting (Humphrey, 2006; Golden et al., 2008).
It is interesting to note that studies have reported disappearing of *Salmonella* after vaccination in free-range flocks but not in cage flocks (Hold et al., 2009). This may again be an indication of the lower challenge of birds in free range conditions. On the other hand it is stated that a move to alternative housing systems is not anticipated to have any impact on the effectiveness of one vaccine type over another (Davis and Breslin, 2003).
The difficulties in eliminating *Salmonella* from any part of empty hen houses plus the tendency of wildlife vectors to reintroduce the organisms, implies that the control measures such as vaccination will remain important in the foreseeable future, even with *Salmonella* free replacement stock (Wales et al., 2007).

Finding of exotic *Salmonella* species in outdoor production

Because *Salmonella* Typhymurium is much more common in wildlife, pigs and cattle, it has been postulated that free range laying flocks will be at greater risk of becoming infected with *Salmonella* Typhimurium than other production types (Carrique-Mas and Davies, 2008a). It has also been suggested that production systems where hens have access to outdoor facilities are more likely to become infected with less frequently (other than *S*. Enteritidis and Typhimurium) encountered *Salmonella* serovars. Yet this cannot be confirmed based on the EFSA baseline study where also cage systems had a significant higher risk of becoming infected with non- *Salmonella* Enteritidis or Typhimurium strains (EFSA 2007). Also in the recent large scale study of Van Hoorebeke et al., (2009b) this suggestion could not be confirmed.

Conclusions

Based on epidemiological data provided above it can be stated that it is highly unlikely that a move from conventional battery cage systems to alternative cage systems and non-cage housing systems for laying hens will result in an increases in *Salmonella* infection and shedding rather the opposite is expected. The true underlying causal mechanism that causes the prevalence of *Salmonella* to be generally lower in alternative housing systems in comparison to cage systems has not been identified yet. It is likely to be a combination of factors influencing the infection pressure in the herd.

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