Available online at www.sciencedirect.com
ScienceDirect
Journal of Theoretical Biology 242 (2006) 755–763
Journal of Theoretical Biology
www.elsevier.com/locate/yjtbi

A model of *Salmonella* infection within industrial house hens

K. Prévost[a,b,1], P. Magal[a,*], C. Beaumont[b]

[a]*Faculté des Sciences et Techniques, Université du Havre 76085 Le Havre, France*
[b]*INRA Département de Génétique Animale, Station de Recherches Avicoles, 37380 Nouzilly, France*

Received 24 January 2006; received in revised form 27 March 2006; accepted 18 April 2006
Available online 9 May 2006

Abstract

Salmonella is one of the major sources of toxi-infection in humans. Incidences of human salmonellosis have greatly increased over the past 20 years and this can largely be attributed to epidemics of *Salmonella enteritidis* phage type 4 within poultry. The main concern with this bacterium is the existence of silent carriers, i.e. animals harbouring *S. enteritidis* without expressing any visible symptoms. In this article, we formulate a model for *S. enteritidis* transmission in hen houses, considering both the hens and the environmental bacterium contamination. By considering the hen's individual development of the disease, we build a model for the production of eggs contaminated by *S. enteritidis*. The objectives are to analyse the dynamic of the disease, and to provide understanding of measures to avoid the endemicity of *S. enteritidis* in industrial hen houses.

Keywords: Salmonella infection; Models; Reproductive number; Asymptotic behaviour

1. Introduction

Salmonella is one of the major sources of toxi-infection in humans and, in France, the most common one (Bouvet et al., 2002). The incidence of human salmonellosis has increased greatly over the past 20 years and this can mostly be attributed to epidemics of *Salmonella enteritidis* phage type 4 in poultry in numerous countries (Barrow et al., 2003; Guard-Petter, 2001). The association between egg consumption and *S. enteritidis* outbreaks is a serious international economic and public health problem (Centers for Disease Control and Prevention, 2000, 2003; Guard-Petter, 2001; Patrick et al., 2004). Transmission to hens may originate from contaminated food or water or by contact with wild animals. But the main concern with this bacterium is the existence of silent carriers, i.e. animals harbouring *S. enteritidis* without expressing any visible symptoms. These animals can, in turn, transmit the bacterium to their flock-mates through horizontal transmission or to their offspring by vertical transmission. However, they are difficult to distinguish from healthy animals, thus are responsible for transmission to human beings. To control this zoonosis, a number of prophylactic means have been developed. Vaccinations have a general effect and may reduce animal contamination and rate of excretion of the bacterium through the faeces (Zhang-Barber et al., 1999). Other methods aim to reduce the introduction of the bacterium into the gut. This is the case for competitive exclusion, which is based on the early implementation of an adult-type intestinal flora which competes with *S. enteritidis* (Rabsch et al., 2000; Rantala and Nurmi, 1973) or acidification of feed which deters bacterial growth. Genetic methods may also be successful in increasing resistance to systemic disease (Bumstead and Barrow, 1988) or carrier-state (Beaumont et al., 1999), thus reducing the need for antibiotic treatments and the risk of antibioresistance. However, the efficiency of these methods was most often measured after experimental inoculation, thus comparing *S. enteritidis* contamination rates at a given interval after inoculation and neglecting the dynamics of bacterial

*Corresponding author.
E-mail address: magal.pierre@wanadoo.fr (P. Magal).
[1]The Ph.D. grant of this author was partially supported by the Région Haute Normandie.

0022-5193/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.jtbi.2006.04.019

dissemination within the flock. The objective of this study is to derive models of Salmonella transmission and use them to choose the best strategies to limit the rate of contamination within egg production.

We develop a model to compare the impact of those prophylactic methods, on the dynamics of *S. enteritidis* transmission both within the laying flock of hens and in the environment. This model is derived from a model incorporating spatial structure which is necessary to understand the contamination process (see Prévost et al., 2006). Here, hens are implicitly assumed to remain within their cages, thus are motionless, while the bacteria diffuse in the environment. The contamination of susceptible hens is determined by contact between hens and bacteria in the environment. When the diffusion process for bacteria is sufficiently fast the model can be simplified to another model presented in Section 2. The model also incorporates the main features of the hens contamination status, distinguishing between three steps: digestive contamination, systemic infection (when systemic organs such as liver or spleen are contaminated after translocation of the bacterium through the digestive barrier), and bacterial clearance leading to recovery. Furthermore, we use this epidemic model for hens to describe the percentage of contamination within egg production. To our best knowledge, such a problem has not been explored yet.

In this article, we first present the model of *S. enteritidis* transmission, and compare it to existing data. We build a model for production of contaminated eggs which will also be compared to data. We investigate the dependency of the model to parameters.

2. Models

2.1. Epidemic model formulation

Let $S(t)$ be the number of susceptible hens at time t, $I^D(t)$ the number of hens suffering from digestive contamination at time t (i.e. D-infectious), $I^S(t)$ the number of hens suffering from systemic contamination (i.e. S-infectious) at time t, and $R(t)$ the number of recovered hens (i.e. having been able to eliminate all bacteria). The status D-infectious is assumed to be a transient status, so that infected hens will first be D-infectious, and after some time, become S-infectious hens. In the model, the total number of hens is constant and equal to $\overline{N}$, so we have $S(t)+I^D(t)+I^S(t)+R(t)=\overline{N}, \forall t \geqslant 0$. Let $C(t)$ be the bacterial environmental contamination (i.e. bacterial load within hen house) at time t. We assume that the transmission rate (the rate at which susceptible becomes D-infectious) is proportional to the total number of bacteria. This transmission is represented by the term $-\kappa C(t)S(t)$. We also suppose that D-infectious and S-infectious animals shed bacterium in the environment (by an excretion process). This flux of excreted bacterium is hypothesized to be equal to $\beta_E I^D(t)+\beta_I I^S(t)$.

The transfer between stages is summarized in Fig. 1.

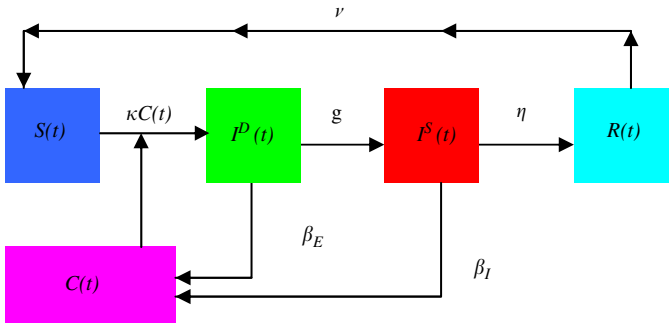

Fig. 1. Flow diagram of epidemic populations of hens.

The different steps and the bacterial load are coupled into the following system:

$$\frac{dS(t)}{dt} = -\kappa C(t)S(t) + \nu R(t),$$
$$\frac{dI^D(t)}{dt} = \kappa C(t)S(t) - gI^D(t),$$
$$\frac{dI^S(t)}{dt} = gI^D(t) - \eta I^S(t),$$
$$\frac{dR(t)}{dt} = \eta I^S(t) - \nu R(t),$$
$$\frac{dC(t)}{dt} = \beta_E I^D(t) + \beta_I I^S(t) - \lambda C(t),$$

where initial conditions are $(S^0, I^{D^0}, I^{S^0}, R^0, C^0) \in [0, +\infty)^5$.

2.2. Interpretation of parameters and validation of the model

We give some interpretation of different parameters of model and fix values to simulate infection. As an average size concerning the flock of fowl, $\overline{N} = 20\,000$ was chosen. The initial environmental contamination level, C^0 was chosen within the range of experimental inoculation doses 10^6–10^9 CFU (colony for unit) as reported by Gast (1993), Gast et al. (1997), and Protais et al. (1996). In the following the various parameters which have been included into the model will be described:

- κ: The exposition rate κ modulates the transmission of the infection. Fitting different values of this parameter allows to take into consideration different types of infections (from food, fresh water, or experimental inoculation).
- g: A distinction was made between D-infectious and S-infectious animals to take into account the efficiency of the digestive barrier. To include this efficiency, we defined the parameter g which determines the rate of translocation through the digestive barrier. Its value was chosen to fit the interval between inoculation and the value of the maximum percentage of infection reported in Gast et al. (1997).

- η: The recovery rate η depends on the development of innate or acquired immunity, and mechanisms of bacterial clearance, which result in the decrease and finally to the elimination of bacterial contamination. Value of this rate was determined according to the asymptotic proportion of *S*-infectious animals showed in Gast et al. (1997), Protais et al. (1996), and values given by an expert (G. Salvat, personal communication).
- ν: The ν parameter is a characteristic of the immune protection. Indeed, the efficiency of vaccines (especially with live vaccine) show that immunization reduces the risk of infection but does not preclude it. This underlines the interest in testing the effect of this parameter on disease evolution. Value of this rate was selected according to the asymptotic values of *S*-infectious animals in Protais et al. (1996).
- $-\lambda$:$-\lambda$ is the growth rate of bacteria in the environment. Hollinger (2000) observed a diminution of the bacterial load so that we employed a negative value for $-\lambda$ to simulate this observation. *S. enteritidis* are often transmitted by a wild animal or by transient contamination of feed or water. So we assume that the bacterial load decreases with time.
- β_E: β_I Values of excretion rate of *D*-infectious (β_E) and *S*-infectious (β_I) hens are very difficult to estimate. Literature gives no explicit data but many hypotheses. Since experiments have shown that excretion remains at low levels (see for example Tilquin et al., 2005), we assume that the number of excreted bacteria remains very low in comparison to the bacterial load. The hypothesis was made that *D*-infectious animals excrete less than *S*-infectious animals.

To validate the model, we distinguish two types of initial (i.e. at time $t = 0$) inoculation. We first consider the case where all the animals are *D*-infectious at time $t = 0$. In this case, the exposition rate has no influence on the behaviour of the model. We use the data of Gast et al. (1997) to fit parameters and to compare the coherence of the result of the model to those of experimental infection.

We also consider the case where only a portion of hens are inoculated at time $t = 0$, and we use the data of Nakamura et al. (1993). The values of all parameters and the initial conditions for both cases are summarized in Table 1.

Since Gast et al. (1997) inoculated 36 hens through oral administration of *S. enteritidis* the initial conditions were fitted at (0,36,0,0,0) to simulate this process. In their experiment, the frequency of isolation of *S. enteritidis* from fecal samples declined steadily from a peak value of 87.7% of inoculated hens 6 days post-inoculation (*p.i.*) to 31.9% 24 days *p.i.*. In our simulated data, a similar dynamic for infection was obtained with a peak of 75% at 6 days and a frequency of 36.6% 24 days *p.i.* (Fig. 2 and Table 2). The result of the simulation can also be compared to other articles. For example, the model is coherent with observations of Bichler et al. (1996). They inoculated hens with 10^{10} bacteria and observed a peak of swab contamination 3 days after inoculation. The model also fits with Shivaprasad et al. (1990) who detected *S. enteritidis* 4 and 7 days

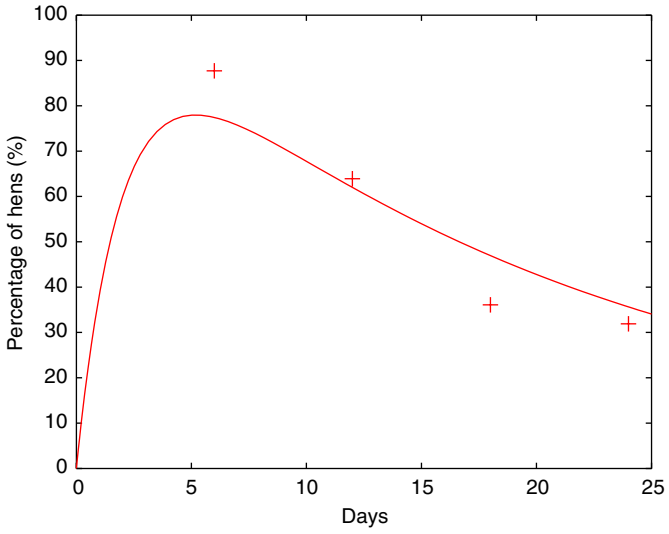

Fig. 2. *S*-Infectious computed percentage (red curve) and data of Gast et al. (1997) (+ symbol).

Table 1
Description and values of model's parameters

Parameters	Description of the parameters	Total infection	Partial infection
S^0	Number of susceptible hens at time 0	0	8
I^{D0}	Number of *D*-infectious hens at time 0	36	8
I^{S0}	Number of *S*-infectious at time 0	0	0
R^0	Number of recovered hens at time 0	0	0
C^0	Bacterial load at time *0*	0	10^5
κ	Exposition rate	0.01	
ν	Recontamination rate	0.002	
g	Rate at which digestive contaminated becomes systemically contaminated	0.5	
η	Recovery rate	0.0048	
β_E	Excretion rate of *D*-infectious hens	0.03	
β_I	Excretion rate of *S*-infectious hens	0.1	
$-\lambda$	Growth rate of bacteria in the environment	−0.1	

post-inoculation in systemic (liver, spleen), reproductive (ovule and oviduct), or digestive (caecum, jejunum and colon) organs and observed contamination in all but one (in ovule). Thus showing that, after 4 days *p.i.*, most *S*-infectious infected animals have already undergone systemic infection. Within partial initial inoculation, the exposition rate plays an important role. In Nakamura et al. (1993), 16 laying hens were assigned to cages that shared a water supply. Of those, 8 hens were inoculated at 7 months of age with 10^5 CFU of *S. enteritidis* and served as the source of infection for the uninoculated hens that shared the drinking water. To simulate this process, initial conditions were equal to $(8,8,0,0,10^5)$. Fig. 3 and Table 3 compares the data of Nakamura et al. (1993) and our simulated data.

Table 2
Table of values for Gast et al. (1997) and model's results

Days	Observations made by Gast et al. (1997)	Simulated data
6	87.7%	75%
12	63.9%	63%
18	36.1%	48%
24	31.9%	36%

Table 3
Table of values for Nakamura et al. (1993) and model's results

Days	Observations made by Nakamura et al. (1993)	Simulated data
3	75%	71%
6	87.5%	77.5%
10	68.75%	68%
13	68%	62%

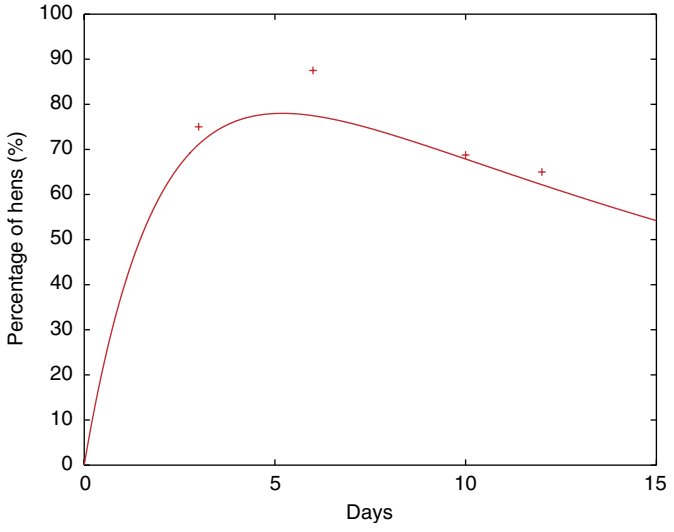

Fig. 3. *S*-Infectious computed percentage (red curve) and data of Nakamura et al. (1993) (+ symbol).

2.3. Model for egg contamination

For the model describing the contamination of eggs with egg production, we assume that the number of contaminated eggs produced by *D*-infectious hens is negligible, since the *Salmonella* level of contamination of *D*-infectious hens is very small. Therefore, in the sequel we only focus on the output of contaminated eggs by *S*-infectious hens which take into consideration the individual development of the disease for each hen. In fact, as we need to follow the history of *S*-infectious hen, we introduce the age of *S*-infection. The age of *S*-infection is defined as the time since when the hens have become *S*-infectious. Let $i^S(t,a)$ be the density of *S*-infectious hens with respect to the age of *S*-infection a, at time t. Assuming that there is no *S*-infectious hens at time $t=0$, we rewrite the third equation of the first model as follows:

$$\frac{\partial i^S(t,a)}{\partial t}+\frac{\partial i^S(t,a)}{\partial a}=-\eta i^S(t,a), \quad \text{for } a\in(0,+\infty),$$
$$i^S(t,0)=gI^D(t),$$
$$i^S(0,.)=0.$$

Let $I^S_{ec}(t)$ be the number of *S*-infectious hens, which lay contaminated eggs at time t. We assume that the probability to produce contaminated eggs (either by direct yolk contamination because of ovarian infection or by contamination of shell at oviposition through faeces) only depends on the age of *S*-infection. Let $p_{ec}(a)$ be the probability, for an *S*-infectious hen with age of *S*-infection a, to produce contaminated eggs. With the above assumption, we obtain the following formula:

$$I^S_{ec}(t)=\int_0^{+\infty} p_{ec}(a)i^S(t,a)\,\mathrm{d}a.$$

At the individual level, the probability $p_{ec}(a)$ describes the ability to produce contaminated eggs. Here, we assume that this probability $p_{ec}(a)$ has the following form

$$p_{ec}(a)=\exp(-\theta a)(1-\exp(-\gamma a)),$$

where the parameter γ is the slope of $p_{ec}(a)$ at 0, and the parameter θ is an exponential growth rate of $p_{ec}(a)$ when a tends to infinity. Fig. 4 depicts the graph of $p_{ec}(a)$ with the fitted values of γ and θ.

The density $i^S_{ec}(t,a)$ of *S*-infectious hens laying contaminated eggs at time t and age of *S*-infection a is thus given by

$$i^S_{ec}(t,a)=p_{ec}(a)i^S(t,a),$$

and satisfies the following age-structured model

$$\frac{\partial i^S_{ec}(t,a)}{\partial t}+\frac{\partial i^S_{ec}(t,a)}{\partial a}=-(\eta+\theta)i^S_{ec}(t,a)+\gamma\exp(-(\gamma+\theta)a)i^S(t,a),$$
$$i^S_{ec}(t,0)=0,$$
$$i^S_{ec}(0,.)=0.$$

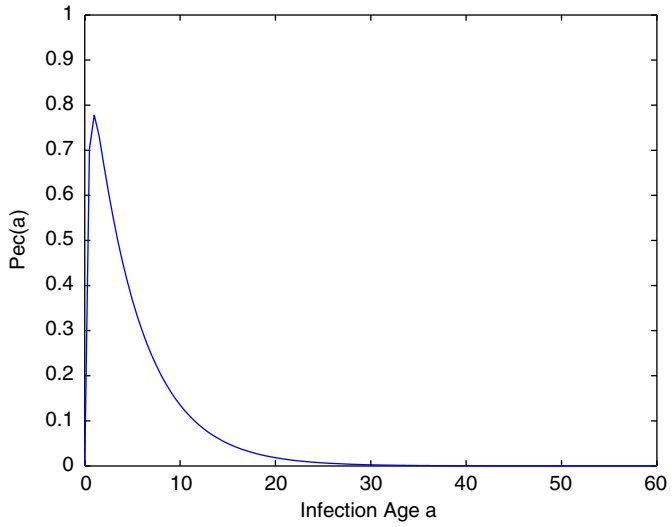

Fig. 4. Evolution of the probability for an S-infectious hen to lay contaminated eggs with respect to the age of S-infection ($\gamma = 3$, $\theta = 0.2$).

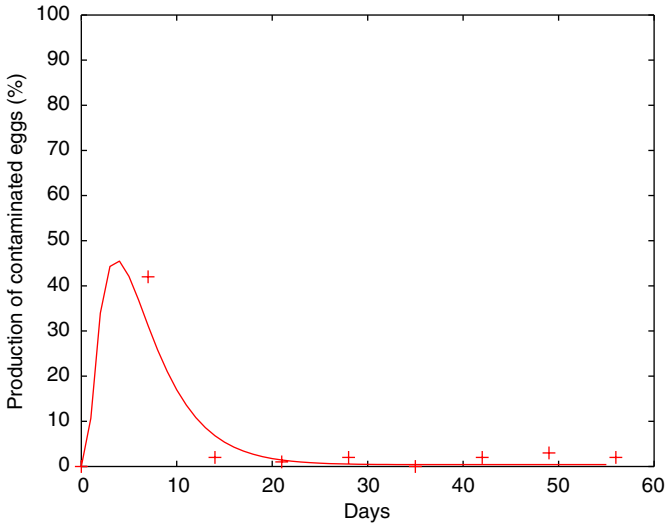

Fig. 5. Production of contaminated eggs: model (red curve), data (+ symbol).

Using this model, and the above definition of $p_{ec}(a)$, we obtain

$$\frac{dI^S_{ec}(t)}{dt} = -(\eta+\theta)I^S_{ec}(t) + \int_0^t \exp(-(\gamma+\theta+\eta)a)gI^D(t-a)\,da.$$

and, in turn, the following system to describe the number of S-infectious hens producing contaminated eggs:

$$\frac{dI^S_{ec}(t)}{dt} = -(\eta+\theta)I^S_{ec}(t) + Y(t),$$
$$\frac{dY(t)}{dt} = -(\gamma+\eta+\theta)Y(t) + \gamma g I^D(t).$$

where

$$y(t) = \int_0^t \exp(-(\gamma+\theta+\eta)a)gI^D(t-a)\,da$$

is an auxiliary variable.

Let b be the rate of eggs laid per hen. Since the total number of hens is assumed to be constant (equal to $\overline{N}$), the total number of eggs produced (whether contaminated or not) during a time step Δt, is given by

$$N_{egg} = b\Delta t\overline{N}.$$

We suppose that each hen produces one egg per day, and $\Delta t = 1$ day. Thus b is fixed to 1. During a time interval going from t to $t+\Delta t$, the number of contaminated eggs is now given by

$$N_{contam,egg} = \int_t^{t+\Delta t} bI^S_{ec}(s)\,ds.$$

The above formula is then used to compute the number of contaminated eggs laid per day (i.e. with $\Delta t = 1$ day).

To validate the model the data of Bichler et al. (1996) were used. In Bichler et al. (1996), 30 White Leghorn laying hens were orally inoculated with 10^{10} CFU of *S. enteritidis*. In 41% of eggs, yolk was culture positive during the first week *p.i.*; this percentage significantly decreased during the second week *p.i* to 2.3%, and remained between 1.2% and 3.8% until week 7. Our model simulations lead to similar results (Fig. 5 and Table 4).

In Fig. 5, after 55 days *p.i.*, it remains a very low but not null percentage of contaminated eggs. This is also coherent with Protais et al. (1996), in which a low percentage of contaminated ovaries and oviducts was observed 6 weeks *p.i.*.

3. Effect of various parameters on the asymptotic behaviour of *salmonella* infection

As further detailed in Prévost et al. (2006), when $v = 0$ the disease and the contaminant always go to extinction. Thus, when t goes to infinity, the population of hens converges to $E = (S^\infty, 0, 0, \overline{N} - S^\infty)$, where $S_\infty > 0$ is uniquely and explicitly determined by the initial conditions. When $v > 0$ the extinction and endemicity depend on the reproductive number R_0 (which is computed for the original ordinary differential equation system presented in Section 2.1) given by

$$R_0 = \overline{N}\,\frac{\kappa}{\lambda}\left(\frac{\beta_E}{g} + \frac{\beta_I}{\eta}\right).$$

R_0 measures the number of secondary *S. enteritidis* infections generated by a (D- or S-) infectious hen. So when $R_0 > 1$, epizooty will persist, and when $R_0 > 1$, there will be extinction of the disease. In the formula for R_0, the quantity $(\overline{N}(\kappa/\lambda))$ corresponds to the contribution of the bacteria to the disease in the poultry population and $((\beta_E/g) + (\beta_I/\eta))$ corresponds to the contribution of D-infectious and S-infectious hens to the bacterial contamination in the environment. More precisely, we have the

Table 4
Table of values for Bichler et al. (1996) and model's results

Days	Observations made by Bichler et al. (1996)	Simulated data
7	41%	45%
14	2%	7%
21	1%	1.4%
28	2%	0.5%
35	0%	0.4%
41	2%	0.4%
48	3%	0.4%
55	2%	0.4%

following interpretation:

$$\overline{N}\,\frac{\kappa}{\lambda} = \overline{N}\kappa \int_0^{+\infty} \mathrm{e}^{-\lambda t}\,\mathrm{d}t$$

is the average number of S-infectious produced by one bacterium and,

$$\frac{\beta_E}{g} = \beta_E \int_0^{+\infty} \mathrm{e}^{-gt}\,\mathrm{d}t \left(\text{respectively}: \frac{\beta_I}{\eta} = \beta_I \int_0^{+\infty} \mathrm{e}^{-\eta t}\,\mathrm{d}t\right)$$

is the average number of bacteria produced by one D-infectious (respectively S-infectious) hen.

When $R_0 \leqslant 1$, the only non-trivial steady state E_1 is given by $S = \overline{N}$, $I^D = 0$, $I^S = 0$, $R = 0$, and $C = 0$. In this case, D-infectious, S-infectious, recovered as well as the bacterial load go extinct. An illustration of this case is given in Fig. 6(A)). When $R_0 > 1$, we have in addition an endemic equilibrium given by

$$S = \frac{\overline{N}}{R_0}, \quad I^D = \frac{\overline{N}\kappa}{gR_0}\,C, \quad I^S = \frac{\overline{N}\kappa}{\eta R_0}\,C, \quad R = \frac{\overline{N}\kappa}{\nu R_0}\,C,$$

and, for the bacterial load,

$$C = \frac{(R_0 - 1)}{\kappa\left((1/g) + (1/\eta) + (1/\nu)\right)}.$$

In that case, the epizooty is endemic. An illustration of this case is given in Fig. 6(B).

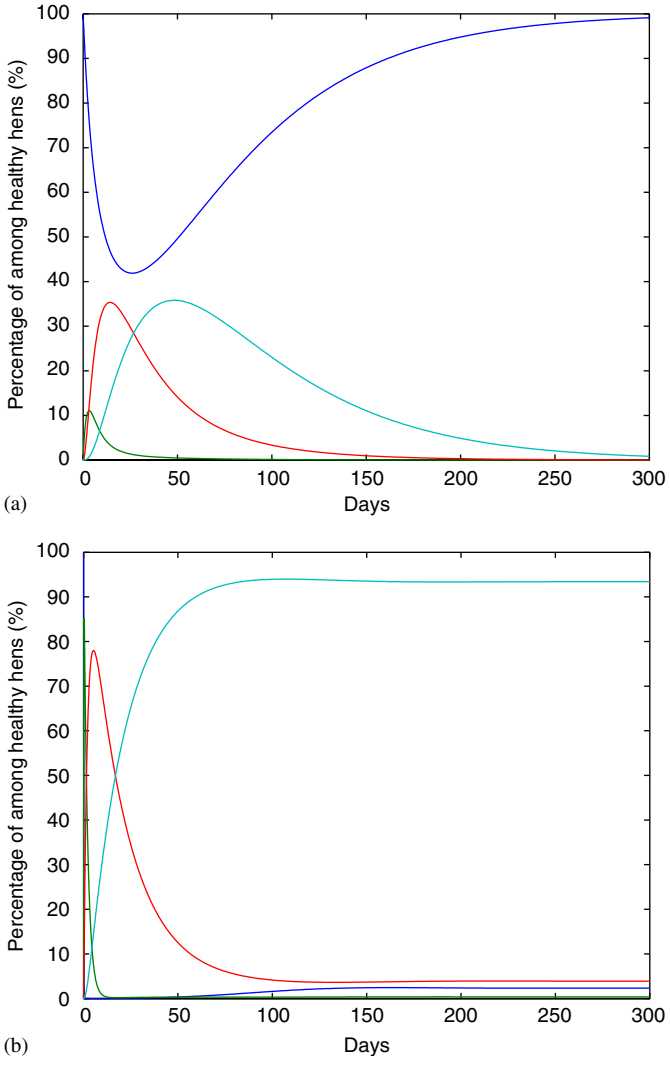

Fig. 6. (a) Evolution of epizooty when $R_0 \leqslant 1$ ($\kappa = 10^{-6}$ and $\nu = 0.02$), (b) evolution of epizooty when $R_0 > 1$ ($\kappa = 10^{-4}$), with $N = 20\,000$ and $C^0 = 10^5$, for susceptible (blue curve), D-infectious (green curve), S-infectious (red curve), and recovered hens (cyan curve).

4. Impact of parameters on endemicity of *S. enteritidis* carrier-state

4.1. Effect of the recovery rate

When η increases the maximal contamination level decreases. From Fig. 7, we can see that when $R_0 < 1$ the maximal infection rate decreases from 83.59% when $\eta = 3 \times 10^{-2}$ to 7.8% when $\eta = 3$. When $R_0 > 1$, we observe a similar behaviour. Moreover, when $R_0 < 1$, the recovery rate is also linked to the duration of epizooty. Higher values of the recovery rate will provide shorter periods of epizooty. With the aforementioned values of the parameters, the percentage of contaminated eggs decreased very quickly. Even when there is endemicity of the *S. enteritidis* infection, almost no more contaminated eggs can be laid at an interval *p.i.* longer than 30 days. This observation is coherent with the very low percentage of contaminated eggs observed by Humphrey (1994).

4.2. Effect of the recontamination rate

In the endemic case, the recontamination rate ν does not modify the initial dynamic of the infection but has a strong impact on the later level of epizooty. When the recontamination rate decreases, the final level of infection decreases. For example in Fig. 8, the level decreases from 39% when $\nu = 2 \times 10^{-2}$ to 0.7% when $\nu = 2 \times 10^{-4}$. This recontamination rate is dependent on the immune defense. A null value of ν corresponds to a perfect immunization, while positive value means that the hens may get ill again after bacterial clearance. When comparing the evolution of

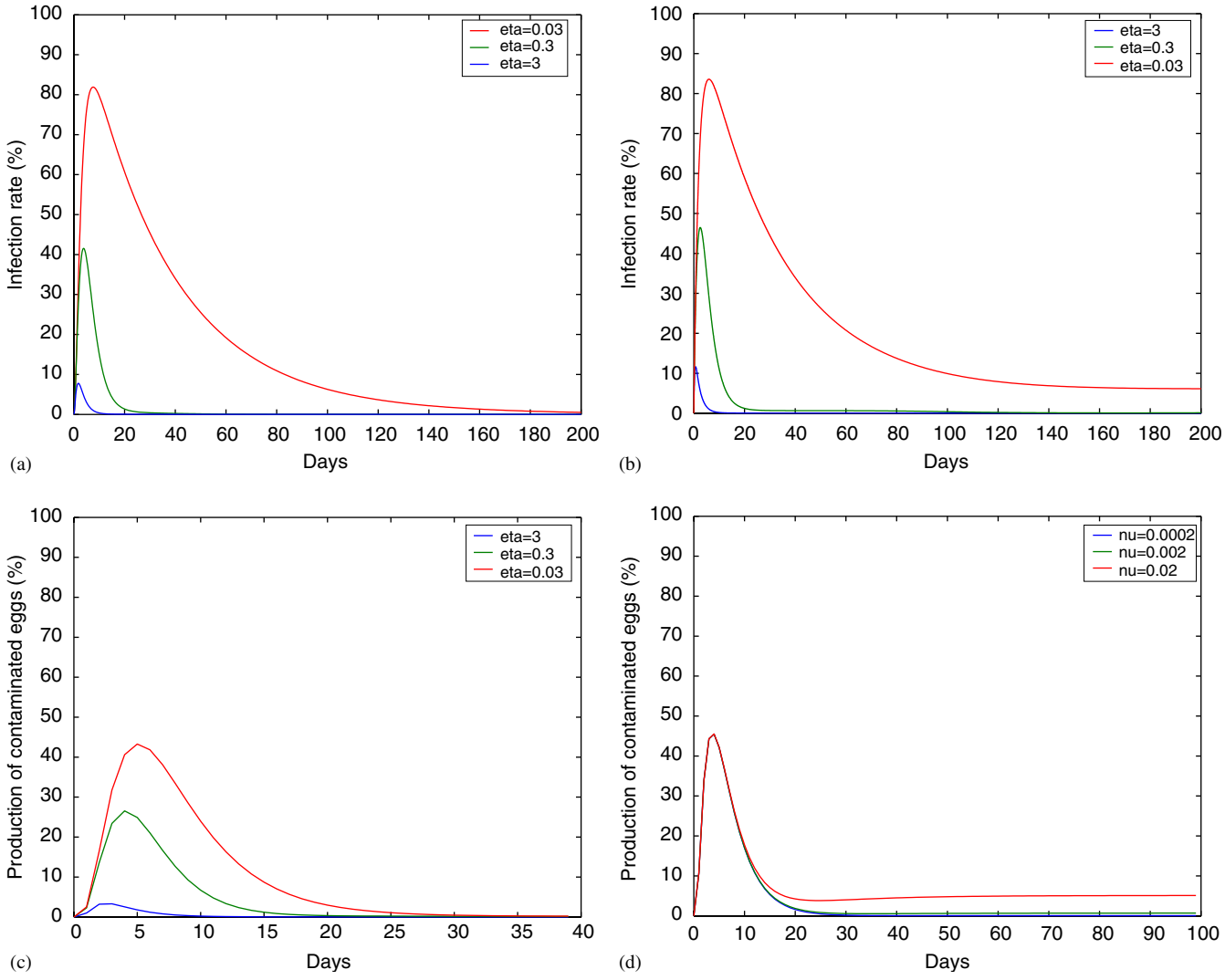

Fig. 7. Evolution over time of the percentage of *S*-infectious hens and of the production of contaminated eggs when (a) (respect. (c)) $R_0 < 1$ with $\kappa = 10^{-6}$ and when (b) (respect. (d)) $R_0 > 1$ with $\kappa = 10^{-3}$.

four poultry lines after inoculation, Protais et al. (1996) observed large differences in both infection peaks and in the clearance rate. The most resistant meat-type line was much less contaminated four weeks after inoculation (with only 12.5% and 25% of contaminated spleens and ceaca) than the L2 egg-type line where more than half the liver and more than three quarters of liver and spleen were still contaminated.

Since the peaks of infection were similar, the differences could originate variations in clearance ability but also in immune defense.

This hypothesis is reinforced by observations of lower immune responses in the L2 egg-type line (Proux et al., 2002). Reducing recontamination rate by vaccines or genetic selection seems to be a good approach for limiting the persistence of *Salmonella*. When the recontamination rate is large (Fig. 8), about 5% of eggs are contaminated in later steps of infection, in relation with the high final level of *S*-infectious hens (39%).

5. General discussion

In this article, we have presented a model describing the dynamics of both Salmonella transmission within the laying flock of hens, and *Salmonella* in the environment. This model is derived from a previous model which incorporated spatial structure, this being a necessary condition to understand the contamination process (see Prévost et al., 2006). At the level of hens, there are four possible steps of contamination: (1) susceptible, (2) *D*-infectious (suffering from digestive contamination), (3) *S*-infectious (suffering from systemic contamination), and (4) recovered. At the bacterial level, we take into account the bacterial contamination of the environment, assuming that transmission is due to the contact between susceptible hens and bacteria. *S. enteritidis* excretion by *D*-infectious and *S*-infectious hens contribute to the environmental load of bacteria and thus to the risk of transmission. Production of contaminated eggs was also modelized starting from the

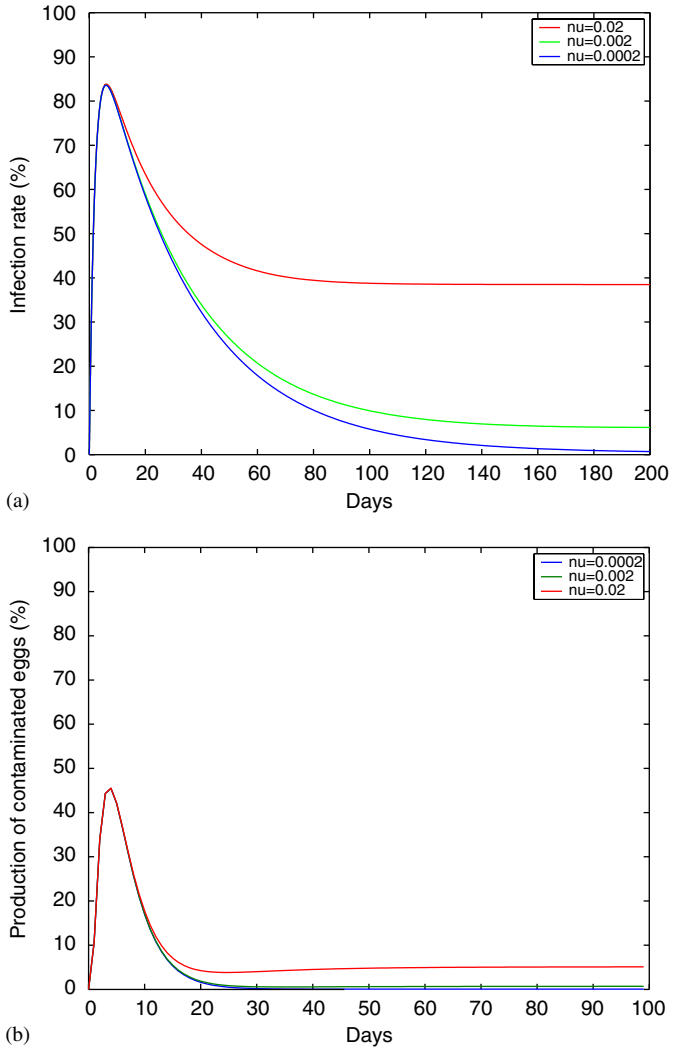

Fig. 8. Evolution over time post-inoculation of *S*-infectious hens and evolution of production of contaminated eggs with ($R_0 > 1$, $\kappa = 10^{-3}$ and $N = 20\,000$).

individual level development of the disease for *S*-infectious hens. Results obtained with the model were validated to experimentally observed data. In particular the results from Gast et al. (1997) were used to fit the rate of translocation of digestive barrier g, the recovery rate η,and the recontamination rate v. We also used the data of Nakamura et al. (1993) to derive the exposition rate κ. The probability of laying contaminated eggs $p_{ec}(a)$ was fitted with data from Bichler et al. (1996). All these fitted values allowed to obtain a reasonably good approximation of the data.

In addition, the results on the asymptotic behaviour of *Salmonella* infection were provided. Endemicity or extinction of the disease depends first on the parameters whether the recontamination rate v is equal to 0 or strictly positive. When $v = 0$, immunization after recovery is perfect and leads to the extinction of the disease and of the contaminant. Asymptotically the population of hens is only composed of susceptible and recovered animals. When $v > 0$, the extinction or the persistence of the disease depends on the reproductive number

$$R_0 = \overline{N}\,\frac{\kappa}{\lambda}\left(\frac{\beta_E}{g} + \frac{\beta_I}{\eta}\right).$$

The prevention of endemicity requires $R_0 \leqslant 1$, and in this case the disease and the contaminant always go to extinction and the population of hens is asymptotically composed with susceptible ones (Fig. 6(A)). Conversely, when $R_0 > 1$, there is endemicity of disease and persistence of the contaminant in the environment (Fig. 6(B)). This key parameter R_0 is dependent on the size of the flock since the more animals there are in a given flock the higher is the probability for an infected animal to transmit the infection, as long as the bacteria are able to reach any animal within the flock. Since R_0 depends on all parameters controlling the contamination except the initial number of bacteria C^0, it may be reduced by an action on some or all of them. The κ parameter should largely influence R_0; its value is dependant on the ability of the intestinal flora to resist to the introduction of a new bacteria. The main effects of a competitive exclusion are observed in young chicks, before they acquired an intestinal flora (Schneitz and Mead, 2000) but the incorporation in food of organic acid (Humphrey and Lanning, 1988) or other supplements as yolk powder (Kassaify and Mine, 2004) may also be efficient in the adults. The λ parameter is mainly dependant on classical rules of hygiene and disinfection that should not vary much in well-managed farms. Since the value of β_E is smaller than that of β_I and according to R_0 definition, it should be more efficient to increase the η rather than the g parameter. This result enhances the interest of increasing resistance to the *Salmonella* carrier-state (Beaumont et al., 1999), i.e. the η parameter rather than to *Salmonella* systemic contamination (as in Mariani et al., 2001), that is the g parameter.

To further investigate the impact of the different parameters on the evolution of infection, numerical simulations were used. The amplitude of epizooty mainly depends on η (the recovery rate). Increasing η (i.e. increasing the ability to clear bacteria) has a favourable effect on both the maximal prevalence and the duration of epizooty (Fig. 7(A)). Moreover, in case of endemicity, increasing the recovery rate also reduces the final percentage of *S*-infectious hens (Fig. 7(B)) and the proportion of contaminated eggs (Fig. 7(C)). The same holds when epizooty persists (Fig. 7(D)). Indeed, it should be feasible to increase η by genetic selection since the percentage of hens having cleared *S. eneritidis* 4 weeks after an experimental contamination by the oral route is heritable (Beaumont et al., 1999). The genes controlling it are currently investigated (Tilquin et al., 2005).

Numerical simulations also highlighted the influence of the recontamination rate on the persistence of the epizooty. Decreasing this parameter (i.e. decreasing the risk for a recovered animal to become susceptible again) results in a reduced final percentage of *S*-infectious hens

(Fig. 8(A)) and to very low percentage of contaminated eggs (Fig. 8(B)). Indeed, immune responses varied for a given model of experimentation with a particular fowl line (Proux et al., 2002). Selection should be feasible, since it has been proved to be efficient on antibody production with a large variety of antigens (Pinard-Van der Laan et al., 1998). Thus our study already shows the basic importance for prevalence and egg contamination levels, of clearance ability and immune response. Both should be considered to improve animal health and food safety.

Our model allows the understanding of the relationship between genetic or vaccine strategies and the production of eggs contaminated by *S. enteritidis*. It also permits to quantify the number of contaminated eggs produced in one industrial hen house. In the risk assessment of *S. enteritidis* in eggs, reducing the prevalence of *S. enteritidis* in poultry flocks was directly proportional to the reduction in risk to human health (FAO, 2002). Nevertheless, as described by those authors, many other aspects need to be considered in order to understand the incidence of *Salmonella* in Human.

Acknowledgements

The authors thank Jocelyne Protais and Gilles Salvat for helpful comments and discussions on Salmonella propagation.

References

Barrow, P.A., Mead, G.C., Wray, C., Duchet-Suchaux, M., 2003. Control of food-poisoning in poultry—Biological options. Worlds Poult. Sci. J. 59, 373–383.

Beaumont, C., Protais, J., Guillot, J.F., Colin, P., Proux, K., Millet, N., Pardon, P., 1999. Genetic resistance to mortality of day-old chicks and carrier-state of hens after inoculation with *Salmonella enteritidis*. Avian Pathol. 28, 131–135.

Bichler, L.A., Nagaraja, K.V., Halvorson, D.A., 1996. *Salmonella enteritidis* in eggs, cloacal swab specimens, and internal organs of experimentally infected white leghorn chickens. Am. J. Vet. Res. 57 (4), 489–495.

Bouvet, P.J.M., Fougerat, I., Guesnier, F., Guibert, V., K'Ouas, G., Lenormand, P., Metz, L., Ruckly, C., Grimont, P.A.D., 2002. Human Salmonellosis Surveillance in France: recent data from the National Reference Center. In: Colin, P., Clément, G. (Eds.), International Symposium Salmonella and Salmonellosis. Ploufragan; 2002/05/29-31, 175–180. Ispaia, Ploufragan (FRA), pp. 411–416.

Bumstead, N., Barrow, P.A., 1988. Genetics of resistance to *Salmonella typhimurium* in newly hatched chicks. Br. Poult. Sci. 29, 521–529.

Centers for Disease Control, 2000. Outbreaks of *Salmonella* serotype *enteritidis* infection associated with eating raw or undercooked shell eggs-United States, 1996–1998. (2000). Morb. Mortal. Wkly. Rep. 49, 73–79.

Centers for Disease Control, 2003. Outbreaks of *Salmonella* serotype *enteritidis* infection associated with eating shell eggs-United States, 1999–2001. (2003). Morb. Mortal. Wkly. Rep. 51, 1149–1152.

FAO [Food and Agriculture Organization of the United Nations], 2002. Risk assessments of Salmonella in eggs broiler chickens. See http://www.fao.org/documents/

Gast, R.K., 1993. Detection of *Salmonella enteritidis* in experimentally infected laying hens by culturing pools of egg contents. Poult. Sci. 72, 267–274.

Gast, R.K., Porter, R.E., Holt, P.S., 1997. Applying tests for specific yolk antibodies to predict contamination by *Salmonella enteritidis* in eggs from experimentally infected laying hens. Avian Dis. 41, 195–1997.

Guard-Petter, J., 2001. The chicken, the egg and *Salmonella enteritidis*. Environ. Microbiol. 3 (7), 421–430.

Hollinger, K., 2000. Epidemiology and Salmonellosis. In: Wray, C., Wray, A. (Eds.), Salmonella in Domestic Animals. CABI Publishing, New-York, USA, pp. 341–353.

Humphrey, T.J., 1994. Contamination of egg shell and contents with *Salmonella enteritidis*: a review. Int. J. Food Microbiol. 21, 31–40.

Humphrey, T.J., Lanning, D.G., 1988. The vertical transmission of salmonellas and formic acid treatment of chicken feed: a possible strategy for control. Epidemiol. Infect. 100, 43–49.

Kassaify, Z.G., Mine, Y., 2004. Nonimmunized egg yolk powder can suppress the colonization of *Salmonella typhimurium*, *Escherichia coli* O157 : H7, and *Campylobacter* jejuni in laying hens. Poult. Sci. 83 (9), 1497–1506.

Mariani, P., Barrow, P.A., Cheng, H.H., Groenen, M.A.M., Negrini, R., Bumstead, N., 2001. Localization to chicken Chromosome 5 of a novel locus determining salmonellosis resistance. Immunogenetics 53, 786–791.

Nakamura, M., Nagamine, N., Takahashi, T., Suzuki, S., Kijima, M., Tamura, Y., Sato, S., 1993. Horizontal transmission of *Salmonella enteritidis* and effect of stress on shedding in laying hens. Avian Dis. 38, 282–288.

Patrick, M.E., Adcock, P.M., Gomez, T.M., Altekruse, S.F., Holand, B.H., Tauxe, R.V., Swerdlow, D.L., 2004. *Salmonella enteritidis* infections, United States, 1985–1999. Emerg. Infect. Dis. 10 (1), 1–7.

Pinard-van der Laan, M.H., Siegel, P.B., Lamont, S.J., 1998. Lessons from selection experiments on immune response in the chicken. Poult. Avian Biol. Rev. 9, 125–141.

Prévost, K., Beaumont, C., Magal, P., 2006. Asymptotic behaviour in a *Salmonella* infection model (submitted).

Protais, J., Colin, P., Beaumont, C., Guillot, J.F., Lantier, F., Pardon, P., Bennejean, G., 1996. Line differences in resistance to *Salmonella enteriditis* PT4 infection. Br. Poult. Sci. 37, 329–339.

Proux, K., Jouy, E., Houdayer, C., Protais, J., Dibb-Fuller, M., Boscher, E., Gillard, A., Gracieux, P., Gilbert, F., Beaumont, C., Duchet-Suchaux, M., 2002. Reliable ELISAS showing differences between resistant and susceptible lines in hens orally inoculated with Salmonella enteritidis. Vet. Res. 33, 23–33.

Rabsch, W., Hargis, B.M., Tsolis, R.M., Kingsley, R.A., Hinz, K-H., Tschäpe, H., Bäumler, A., 2000. Competive exclusion of Salmonella enteritidis by Salmonella gallinarum in Poultry. Emerg. Infect. Dis. 6 (5), 443–448.

Rantala, M., Nurmi, E., 1973. Prevention of the growth of *Salmonella infantis* in chicks by the flora of the alimentary tract of chicken. Br. Poult. Sci. 14, 627–630.

Schneitz, C., Mead, G., 2000. Competitive exclusion. In: Wray, C., Wray, A. (Eds.), *Salmonella* in Domestic Animals. CABI Publishing, New-York, USA, pp. 301–322.

Shivaprasad, H.L., Timoney, J.F., Morales, S., Lucio, B., Baker, R.C., 1990. Pathogenesis of *Salmonella enteritidis* infection in laying chickens. I. Studies on egg transmission, clinical signs, fecal shedding, and serologic responses. Avian Dis. 34, 548–557.

Tilquin, P., Barrow, P.A., Marly, J., Pitel, F., Plisson-Petit, F., Velge, P., Vignal, A., Baret, P.V., Bumstead, N., Beaumont, C., 2005. Genome scan for quantitative trait loci affecting the Salmonella carrier-state in the chicken. Genet. Select. Evolut. 37, 539–561.

Zhang-Barber, L., Turner, A.K., Barrow, P.A., 1999. Vaccination for control of *Salmonella* in Poultry. Vaccine 17 (20–21), 2538–2545.