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An individual-based model for *Salmonella* transmission along the pig production chain

Stefanie Hotes, Imke Traulsen and Joachim Krieter

Institute of Animal Breeding and Husbandry, Christian-Albrechts-Universität zu Kiel, Kiel, Germany

Abstract

The aim was to develop an individual-based model for the transmission of *Salmonella* from farrowing farm to slaughterhouse. The present study concentrated on farrowing sows as the initial source of *Salmonella* transmission. The model was evaluated with a sensitivity analysis using a Plackett-Burman design. Three levels (minimum, default, maximum values) for all input factors were considered. The deviations from the default prevalences caused by the extreme values did not balance each other for several significant input factors. For these factors, the relation between input factor and regarded health states was not linear.

Results showed that the probability of effective contact, to restart shedding, the shedding duration and the sow herd prevalences as well as their distribution across farrowing farms determined the *Salmonella* prevalence at slaughter. The study emphasised the importance of vertical and horizontal transmission. Low *Salmonella* prevalences obtained after nursing caused prevalences at slaughterhouse up to 12 %.

Keywords: stochastic modelling, *Salmonella*, pig production, Plackett-Burman design

Introduction

Salmonellosis is a major problem in most countries in the world (WHO 2007). The World Health Organization (WHO) has reported that diseases of zoonotic origin are of particular concern in the European Union (EU). Recent estimates predict that temperature-sensitive infectious disease, such as food-borne infections, will become more important in the coming decades (WHO 2010). Frequent cases of human salmonellosis in Germany have induced the Federal Institute for Risk Assessment (BfR) to emphasise the avoidance of raw meat, in particular raw pork in any form (BfR 2005). *Salmonella* prevalence in slaughter pigs in the EU is estimated at about 10 %. Country prevalences range from no positive findings in Finland up to a prevalence of 29 % in Spanish slaughter pigs (EFSA 2008). The 2003 European regulation for the control of *Salmonella* and other specified food-borne zoonotic agents (EC No 2160/2003) regulates that national control programmes have to be established to provide the detection of zoonosis and specific control measures. The German implementation focuses on the finishing stage. Only if prevalence at slaughter age exceeds 40 %, the pigs purchased from farrowing stage should be tested as well. However, pig deliveries are certainly critical points for dynamics of *Salmonella* transmission. Bacteria can spread across farms due to the enhanced contact structure. Previously published models have concentrated on the spread within a single farm (Ivanek *et al.* 2004, Hill *et al.* 2008, Lurette *et al.* 2008) but have not considered the production and contact structures in detail (van der Gaag *et al.* 2004, Wehebrink *et al.* 2007).

The objective of the present study was to develop an individual-based model for the transmission of non-clinical Salmonellosis within the pork supply chain. Modelling was to reveal the most important factors for *Salmonella* spread with regard to the trade relationships between the production stages. The current paper represents a further development of the simulation study developed by Krieter (2004).

Material and methods

The developed simulation model describes the spread of non-clinical *Salmonella* infection (henceforth referred to as »infection«) within the pork supply chain from farrowing stage to slaughter. The model is stochastic with a discrete time step of one week. Comparable to all-in all-out production, the one-week time step is defined as the basic unit for regrouping tasks and moving pigs between branches of production. Only transports and processes at slaughterhouses are detached from the one-week rhythm of the model. The present study focused on the transmission of *Salmonella* from sow to pig and subsequently from pig to pig. The epidemiological process in the model describes the spread of *Salmonella* over a 52-week period with a prior burn-in also of 52 weeks. The burn-in preceded the simulation in order to initiate the trade relationships and to fill the model with pigs. At starting point, the model contains only farms and sows because output data has to be based on slaughter pigs whose whole lives were to be monitored. Without burn-in, the first pigs would reach slaughter age at week 28. The quantity and quality of the output data would be low. The burn-in ensures a continuous flow of slaughter pigs and thereby improves the results. The program is written in the object-oriented language C++. Routines from the NAG C library (NAG 2001) are used to generate random numbers. Random numbers are drawn to determine farm characteristics and trade relationships as well as individual infection and course of disease as described below.

Trade relationships

The model contains a network of farrowing and finishing farms as well as a slaughterhouse which are linked by pig deliveries. The model considers two types of farrowing farms:

1) conventional farrowing farms with supply relationships (PP_S) to one (PP_S_one), two (PP_S_two) or three finishing farms (PP_S_three);
2) farrow-to-finishing farms without supply relationships to a specialised finishing farm (PP_F).

At the finishing stage, fatteners buying pigs from one (F_one), two (F_two) or three (F_three) conventional farrowing farms are distinguished. Figure 1 illustrates several possible trade relationships. The only information obligatory for the network concerns the farrowing farms and contains the sow herd size (varying from 100 to 350 sows with a mean of 181 sows in the current study), the suckling period (four weeks by default for the current study) and the production cycle (mix of one and three weeks in the current study). Prior to every replication a regeneration of the trade relationships is performed considering the stated distributions (Table 1, no. 1-3). The regeneration implicates the classification of the farrowing farms into

Table 1
Definition of the input proportions, probabilities and parameters used in the model

No.	Definition	Notation	Default values			Minumum values	Maximum values	Source
			proportions	probabilities	parameter			
1	Proportion of farrow-to-finishing farms to conventional farrowing farms	PP_F	0.25			0.1875	0.3125	a
		PP_S	0.75			0.8125	0.6875	a
2	Proportion of conventional farrowing farms selling their pigs to one, two, or three fatteners	PP_S_one	0.7			0.775	0.625	b
		PP_S_two	0.2			0.15	0.25	b
		PP_S_three	0.1			0.075	0.125	b
3	Proportion of finishing farms buying the pigs from one, two, or three farrowing farms	F_one	0.85			0.8875	0.8125	a
		F_two	0.14			0.105	0.175	a
		F_three	0.01			0.0075	0.0125	a
4	Proportion of farrowing farms with low, middle, and high sow herd prevalence	$PP_{lowPrev}$	0.8			0.85	0.75	c
		$PP_{middlePrev}$	0.15			0.1125	0.1875	c
		$PP_{highPrev}$	0.05			0.0375	0.0625	c
5	Proportion of non-shedder to shedder sows if…							
	sow herd prevalence is low	S_l_nonShedder	0.99			0.9925	0.9875	b
		S_l_shedder	0.01			0.0075	0.0125	b
	sow herd prevalence is middle	S_m_nonShedder	0.95			0.9625	0.9375	b
		S_m_shedder	0.05			0.0375	0.0625	b
	sow herd prevalence is high	S_h_nonShedder	0.9			0.925	0.875	b
		S_h_shedder	0.1			0.075	0.125	b
6	Probability of effective contact within compartment between pen i and j if pigs are in…							
	the same pen	$p_{ij}=p_w$		0.0014		0.00105	0.00175	d
	pens adjacent to each other	$p_{ij}=p_b$		0.0009		0.000675	0.001125	d
	pens in same row but not adjacent	$p_{ij}=p_b/3$		0.0003		0.000225	0.000375	d
	pens in opposite row	$p_{ij}=p_b/100$		0.000009		0.00000675	0.00001125	d
	Probability of effective contact at transport	p_m		0.0004		0.0003	0.0005	b
	Probability of effective contact at lairage	p_l		0.0004		0.0003	0.0005	b
7	Probability to restart shedding…							
	basically	P_{rs}_basic		0.2		0.15	0.25	e
	due to stress	P_{rs}_stress		0.4		0.3	0.5	e
8	Proportion of *Salmonella* units remaining at lairage	r_l	0.3			0.225	0.375	b
9	Probability to lose *Salmonella* at evisceration	P_e		0.75		0.5625	0.9375	f
10	Probability of contaminating contact at slaughterhouse	p_c		0.1		0.075	0.125	f
11	Proportion of *Salmonella* units remaining at tools	r_c	0.3			0.225	0.375	f
12	Weibull-distributed shedding duration with							
	shape parameter	α			2.36	2.36	2.36	d
	scale parameter	β			27.8	15.05	40.6	d

Source a: based on ZDS (2009), b: assumed, c: based on QS GmbH classification (Struck 2009), d: Hill *et al.* (2008), e: Ivanek *et al.* (2004), f: based on van der Gaag *et al.* (2004)

farms with a low ($PP_{lowPrev}$), middle ($PP_{middlePrev}$), or high sow herd prevalence level ($PP_{highPrev}$). Based on this classification each sow on the farm has a certain probability to become a shedder (S_shedder) or non-shedder (S_nonShedder) (Table 1; no. 4 and 5). The health states of the sows are not regarded in more detail and remain unchanged for the respective replication.

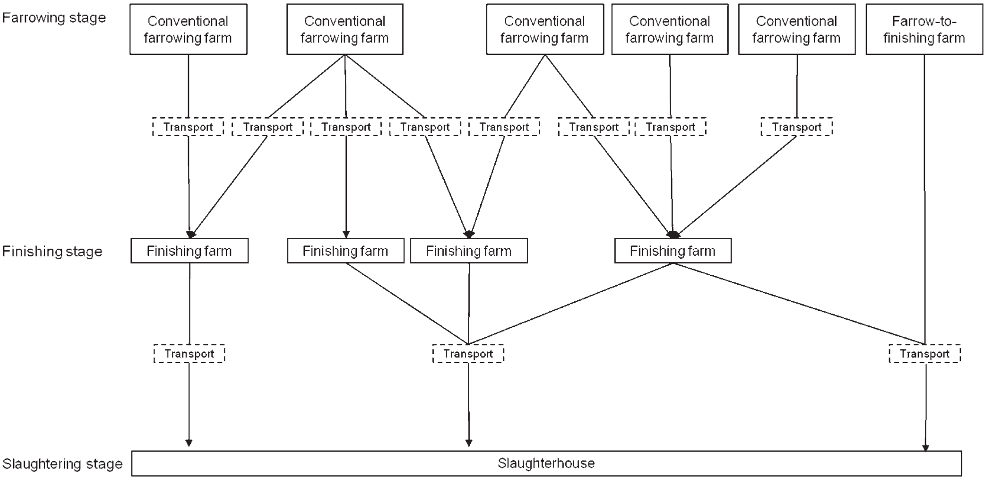

Figure 1
Illustration of possible trade relationships within the simulated pork supply chain

Health and contamination states

The program is individual-based i.e. each pig is followed from farrowing to chilling at slaughter. During its lifetime the pig's health status is updated every week. As described by Lurette *et al.* (2008), four mutually exclusive health states are distinguished: susceptible pigs free of *Salmonella* and thus non-shedding and seronegative (S-); seronegative, shedding pigs (I-); seropositive, shedding pigs (I+), and seropositive non-shedding but *Salmonella*-bearing pigs (C+) (Figure 2). The latent period between *Salmonella* ingestion and shedding falls below the model basis of one week (Lurette *et al.* 2008) and is therefore neglected. The seroconversion is assumed to last about two weeks (van der Gaag *et al.* 2004). Subsequent to seroconversion, the pig stays seropositive. The possibility of recovery is not considered. Even if a return to seronegativity should exist in reality, it is assumed to last longer than the lifetime of the pigs (Lurette *et al.* 2008). The only change in health state based on the carrier stage is to restart shedding and become I+ again. The risk that the carrier pigs restart shedding is always present (P_{rs}_basic) but increases if the pigs are exposed to stress (P_{rs}_stress) (Table 1; no. 7). Whether it is the first shedding period (I- until C+) or a subsequent one (starting from C+: I+ to C+), the shedding duration is assumed to be Weibull-distributed (Hill *et al.* 2008) with a mean shedding duration of about four weeks (Table 1, no. 12). All piglets are born susceptible to infection and infection is immediately possible within the farrowing crate. The aspect of passive immunity of newborn piglets from a sow's antibodies is however neglected, due to the lack of precise information.

The health state of the pig does not change post-slaughter (Figure 2). But slaughter pigs lose bacteria due to careful evisceration or become contaminated on their skin surfaces due to the slaughter sequence (direct contact of carcasses) or contaminated tools (cross-contamination) (Table 1; nos 8-11).

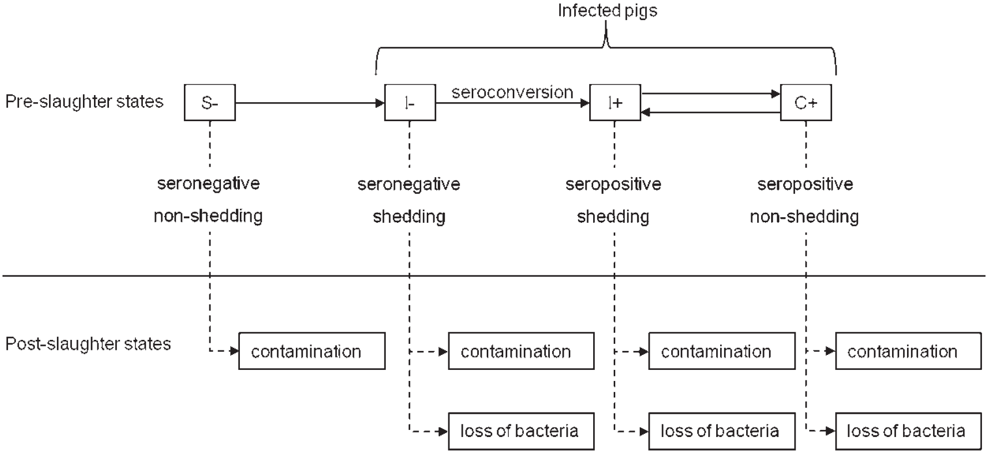

Figure 2
Health and contamination states of pigs pre- and post-slaughter

Probability of infection and contamination

The modelling of infection is based on the Reed-Frost epidemic model, which describes the probability $P(t)$ of a susceptible pig having effective contact with any of $E(t)$ excreting pigs during the period $[t-1, t]$ (Rubel 2005, Hill *et al.* 2008).

$$P(t) = 1-(1-p)^{E(t)} \tag{1}$$

Effective contact is defined as contact between an infectious individual and a susceptible one, which produces a newly infected individual (Bailey 1975). The probability for an effective contact is given by p.

The current program considers the sows of the farrowing farms as the initial sources of *Salmonella* infection. Consequently, *Salmonella* is transmitted only from sow to pig and subsequently through transmission from pig to pig without *Salmonella* entry via rodents, birds, feed et cetera. In the following, the possibilities for effective contact between two individuals are described in detail.

Infection at farm

Assuming that all pigs housed in pens of the same compartment have contact with each other, the probability of an effective contact increases with an increasing number of shedding pigs. Contact can happen via faecal or airborne transmission. Formula 2 describes the probability P_{ij} of a susceptible pig in pen i becoming infected because of $E_j(t)$ excreting pigs in pen j during the period [t-1, t] (Hill *et al.* 2008):

$$P_{ij}(t) = 1-(1-p_{ij})^{E_j(t)} \tag{2}$$

The probability of effective contact between pens i and j (p_{ij}) depends on the distance between these pens (Table 1; no. 6). The number of pens within a compartment varies between farms but all compartments consist of two opposite rows. During nursing, transmission is assumed to be reduced to pen level. This assumption is based on the limited contacting of new born piglets to piglets of other pens.

Infection at transport

In an agglomerated pig producing area such as Northern Germany, transport from farrowing farm to finishing farm does not exceed a few hours. Precise data about new infections during transport are missing. Pigs are stressed due to transport and prior fasting whereby carrier pigs could restart shedding. This is implemented in the program by an increased probability for shedding reactivation (P_{rs}_stress; Table 1; no. 7). Hence, in the model, the increased number of shedding pigs increases the probability of a susceptible pig becoming infected at the fattening compartment (P_{ij}). This mainly compensates for the missing infection opportunity during transport to the finishing stage.

In contrast, transport to slaughterhouse is not followed by an extensive housing period. But transport to slaughterhouse lasts much longer compared to transport to finishing farm, which increases the probability of effective contact at the lorry (p_m; Table 1; no. 6). The probability P_m of a susceptible pig becoming infected due to effective contact with any of E_m(t) excreting pigs at transport to slaughterhouse is represented by:

$$P_m(t) = 1-(1-p_m)^{E_m(t)} \tag{3}$$

An increased probability of shedding restart is also considered at transport to slaughterhouse (Table 1; no. 7).

Infection at lairage

After transport to the slaughterhouse, the pigs are housed at lairage until the slaughter process starts. Comparable to infection at the farm and during transport the probability of infection at lairage (p_l; Table 1; no. 6) increases with an increasing number of *Salmonella*-shedding pigs in the proximity. The lairage is neither cleaned intensively nor disinfected before a new group of pigs is housed. Comprehensive cleaning is only possible at the end of a working day. Insufficient cleaning during the day increases the probability of infection at lairage. In addition to the number of shedding pigs in the current group (E_l(t)), the *Salmonella* output of all previous groups has to be considered (R_l).

$$P_l(t) = 1-(1-p_l)^{E_l(t)+R_l(t)} \tag{4}$$

$$\text{with} \quad R_l(t) = (E_l(t-1) + R_l(t-1)) \times r_l$$

r_l represents the proportion of *Salmonella* units remaining at lairage after a superficial cleaning (Table 1; no. 8).

Loss of bacteria and contamination at slaughter line

After death, the pigs are no longer able to change their health state, however surface contamination and loss of bacteria is possible. The probability of losing *Salmonella* (P_e) depends on how carefully evisceration is performed (Table 1; no. 9). Carcass contamination could happen via direct contact of carcasses or cross-contamination due to soiled equipment and tools. Whether a pig becomes contaminated at the slaughter process depends preliminarily on the probability of a contaminating contact (p_c; Table 1; no. 10) and the *Salmonella* status of the previously slaughtered pigs ($E_c(t)$). Furthermore, pigs already slaughtered could have contaminated the equipment and tools whereby bacteria could skip carcasses or contaminate a whole array of subsequent carcasses.

$$P_c(t) = 1-(1-p_c)^{E_c(t)+R_c(t)} \tag{5}$$

$$\text{with} \quad E_c(t) = \begin{cases} 0 \text{ if Salmonella status of previous slaughtered pig} = S- \\ 1 \text{ if Salmonella status of previous slaughtered pig} = I-, I+ \text{ or } C+ \end{cases}$$

$$R_c(t) = (E_c(t-1) + R_c(t-1)) \times r_c$$

R_c describes the impact of all previously slaughtered pigs with r_c representing the proportion of *Salmonella* units which move from carcass to carcass (Table 1; no. 11).

Input data availability

For the present study most of the input data were obtained from literature or advisory services (Table 1). No information was available about how many farrowing farms selling their pigs to one, two or three fatteners, the respective sow herd prevalence, the probability of an effective contact at transport and lairage and the proportion of *Salmonella* units remaining at lairage (Table 1; no. 2, 5, 6 and 8). Hence, information was acquired by discussion with experts.

Number of replications

For the executed appraisal, the decision on how many replications were needed to obtain sufficient model output, was based on the variance which would appear in the output data due to a certain number of replications. First, the model was run with default, minimum, and maximum values[1] 200 times each (Table 1). Based on these 200 replications, 30 packages of 5, 10, 20, 30, ... 100 replications were randomly sampled with replacement. This bootstrapping was executed for minimum, maximum and default values. The variance of every package was calculated. Afterwards, the standard deviation of every package size, consisting of 30 variances each, was also calculated. Figure 3 shows the falling trend of standard deviation with increasing package size. Finally, the package size with 30 replications was considered as sufficient. There was a relatively sharp decline for the maximum graph up to 20 replications and for the default graph up to 30 replications. The further trend slowed down in decline. This indicates that the variance was no longer influenced by the number of replications.

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[1] Default, minimum, and maximum values represent the three factor levels of the Placket-Burman design explained later.

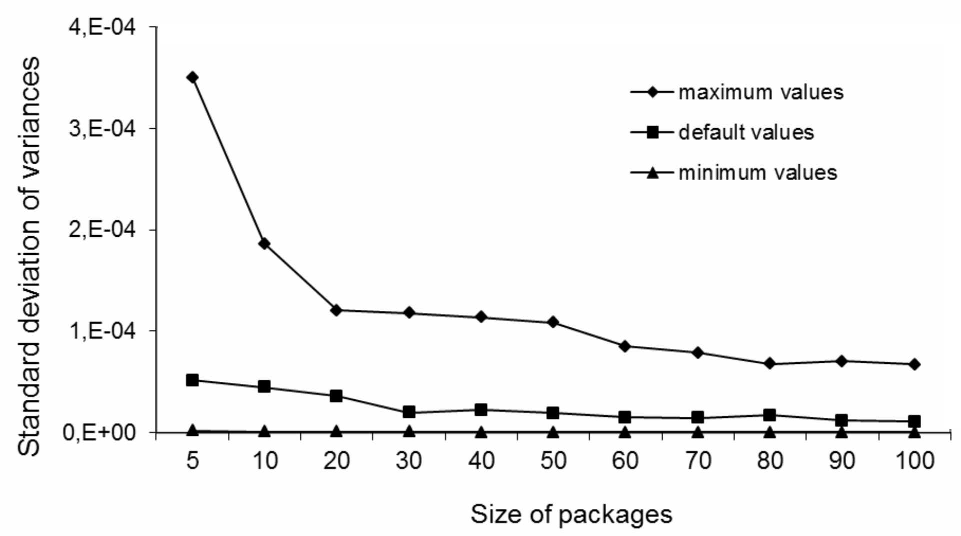

Figure 3
Standard deviation of variances related to the size of packages – obtained via bootstrapping method

Sensitivity analysis

A sensitivity analysis was performed to evaluate the model. A Plackett-Burman design (P-B design) was used to identify the most important input factors and to estimate their impact on the allocation of health states. Originally, P-B designs are two-level fractional factorial designs of resolution III for studying up to k=N-1 components in N runs (Montgomery 2005). To compare three factor levels in the analysis, the basic P-B design, considering maximum (+) and default (0) values, had to be reflected. Therefore, the maximum values were replaced with the minimum (-) values (Figure 4a and 4b) (Vanaja & Shobha Rani 2007).

In general, resolution III designs confound the main effects with two-factor interactions. To ensure that the main effects are not confounded with two-factor interactions, these designs have to be reversed (Barrentine 1996; Montgomery 2005). Hence, the basic and reflected designs were carried out again but extreme and default values were switched (Figure 4c and 4d). This full fold-over technique broke the alias links between all the main effects and their two-factor interactions and resulted in a design of resolution IV (Montgomery 2005). All together, 64 assemblies (four designs with 16 assemblies each) were performed considering the default, minimum and maximum values of the twelve factors described in Table 1. Default values of the probabilities were increased and decreased by 25 % to obtain minimum and maximum values, respectively. Concerning proportions, minor parts were increased by 25 % whereas the major part was decreased accordingly. For the Weibull distribution, the standard deviation was subtracted and added to the default value. Output data of the P-B design was analysed using the MIXED procedure of the SAS program package Version 9.1 (SAS Institute Inc., Cary, NC, USA). Linear models were fitted for the four health states, the transmission paths of infection and for the carcass contamination as well as for the loss of *Salmonella* due to careful evisceration (Figure 5). For every health state six models were formulated to analyse the variances over the course of time (Block A of Figure 5). To analyse *Salmonella* transmission paths, seven models were formulated (Block B of Figure 5) and for the loss of bacteria and the carcass contamination one model each was added (Block C of Figure 5). All 33 linear models considered the same twelve explanatory variables which represented the input factors of the simulation model. Analyses were done via F-Test, least square means and confidence limits. Corresponding to the P-B designs, the analysis had to consider three levels for every input factor. To adjust for these multiple comparisons, the *P*-values and confidence limits were accomplished with the Bonferroni adjustment.

a) Basic design

Assembly	Input factors				
	1	2	3	…	12
1	+	0	0		0
2	+	+	0		+
3	+	+	+		0
⋮					
16	0	0	0		0

b) Reflected design

Assembly	Input factors				
	1	2	3	…	12
17	-	0	0		0
18	-	-	0		-
19	-	-	-		0
⋮					
32	0	0	0		0

c) Reversion of the basic design

Assembly	Input factors				
	1	2	3	…	12
33	0	+	+		+
34	0	0	+		0
35	0	0	0		+
⋮					
48	+	+	+		+

d) Reversion of the reflected design

Assembly	Input factors				
	1	2	3	…	12
49	0	-	-		-
50	0	0	-		0
51	0	0	0		-
⋮					
64	-	-	-		-

Figure 4
Basic, reflected and reversed Plackett-Burman designs

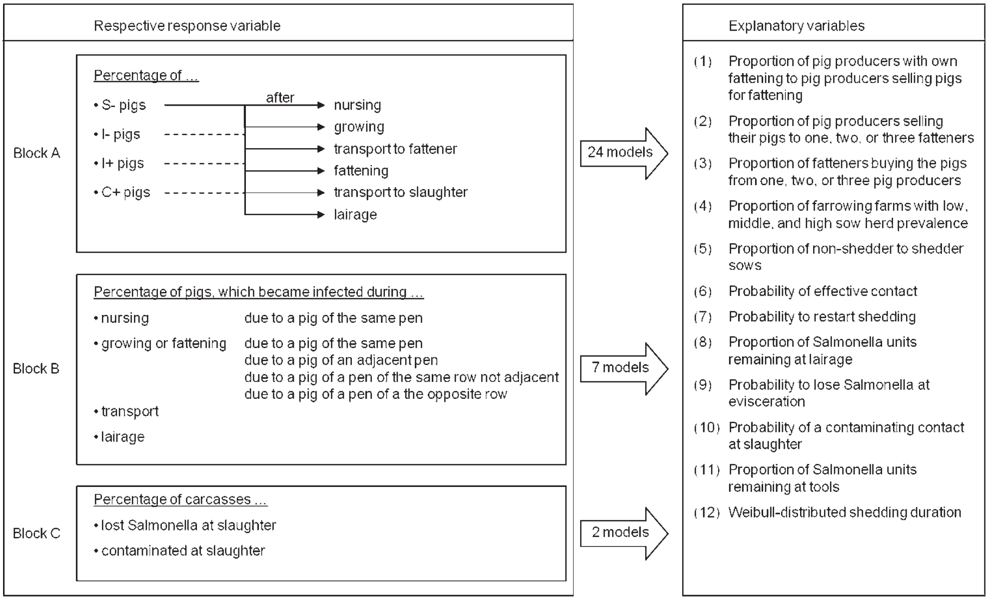

Figure 5
Outline of the formulated models for sensitivity analysis

Results

First analyses considered only the assemblies 16, 48 and 64 of the P-B design, to receive an impression of the prevalence generated by the model. These three assemblies described the model output if all input factors were set to default, maximum and minimum values, respectively. After nursing, the percentages of infected piglets were very similar, but differences in prevalence increased in the course of time.

- Minimum values: (assembly 64) prevalence after nursing: 0.04 % after lairage: 0.50 %
- Default values: (assembly 16) prevalence after nursing: 0.08 % after lairage: 4.84 %
- Maximum values: (assembly 48) prevalence after nursing: 0.14 % after lairage: 11.95 %

The proportion of infected pigs which lost *Salmonella* due to evisceration represented the given probabilities of about 56 % (minimum values), 75 % (default values) and 94 % (maximum values). The carcass contamination never exceeded 2.32 %. (Results not presented.)

Further analyses were based on all 64 assemblies. The significance of the input factors for the response variables are shown in Table 2. The presentation of the results for all 33 analyses would be too expansive. Hence, the presentation of the health state models is limited to the susceptible pigs (S-) (Table 2; Block A). This group represents exactly the opposite to the infected pigs ((I-) + (I+) + (C+). Hence, factors with significant impact on the percentage of susceptible pigs influenced the amount of infected pigs, respectively. Factors 4-6 in Table 2 show that from nursing to slaughter the »Proportion of farrowing farms with low, middle, and high sow herd prevalence«, the »Proportion of non-shedder to shedder sows«, as well as the »Probability of effective contact« have a significant impact on the percentage of susceptible pigs (Table 2; Block A). While the first two factors determined the amount of *Salmonella* units brought to model, the probability of effective contact effected the transmission dynamic of the present bacteria. Due to an average shedding duration of four weeks, Factor No. 12 did not become significant until growing. Even if pigs became infected at the week of birth, shedding duration probably exceeded the nursing time. Subsequently, the probability to restart shedding (no. 7) cannot be significant prior to that. Pigs have to finish shedding before they can restart. The factor »Proportion of *Salmonella* units remaining at lairage« (no.8) became significant at lairage.

The similarities between the significant factors for the percentage of susceptible pigs (Table 2; Block A) and the transmission paths (Table 2; Block B) were reasoned. The transmission paths were determined by effective contacts, which decreased the number of susceptible pigs.

Block C of Table 2 shows the significant input factors for the percentages of pigs which lost *Salmonella* at evisceration or became contaminated during slaughter process, respectively. Note that all pigs were considered to estimate the percentage of pigs which lost *Salmonella*; even susceptible pigs. Hence, additionally to the »Probability to lose *Salmonella* at evisceration« (no. 9) also factors influencing the percentage of infected pigs were significant. The same applies to the percentage of contaminated carcasses. Next to the »Probability of contaminating contact at slaughterhouse« (no. 10) and the »Proportion of *Salmonella* units remaining at tools« (no.11), all factor groups influencing the amount of infected pigs were significant.

Table 2
Results of the F-Test examining the significance of the input factors for the percentage of susceptible pigs (Block A), for the transmission path (Block B), as well as for the percentage of *Salmonella* loss and carcass contamination (Block C)

No.	Definition	Block A (limited to S-) % of susceptible pigs after...						Block B % of pigs, which became infected during...							Block C % of carcasses...	
								nursing	growing or fattening							
		nursing	growing	transport to fattener*	fattening	transport to slaughter	lairage	same pen	same pen	adjacent pen	same row	opposite row	transport	lairage	lost *Salmonella* at slaughter	contaminated at slaughter
1	Proportion of farrow-to-finishing farms to conventional farrowing farms	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
2	Proportion of conventional farrowing farms selling their pigs to one, two, or three fatteners	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
3	Proportion of finishing farms buying the pigs from one, two, or three farrowing farms	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
4	Proportion of farrowing farms with low, middle, and high sow herd prevalence	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
5	Proportion of non-shedder to shedder sows	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
6	Probability of effective contact	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
7	Probability to restart shedding	ns	ns	ns	<0.0001	<0.0001	<0.0001	ns	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
8	Proportion of *Salmonella* units remaining at lairage	ns	ns	ns	ns	ns	0.01	ns	ns	ns	ns	ns	ns	<0.0001	ns	0.006
9	Probability to lose *Salmonella* at evisceration	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	<0.0001	ns
10	Probability of contaminating contact at slaughterhouse	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	<0.0001
11	Proportion of *Salmonella* units remaining at tools	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	<0.0001
12	Shedding duration	ns	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	ns	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

significance level = 0.01, *New infections are not possible at transport to fattener. Hence, estimates for the percentage of susceptible pigs are identical to estimates after growing.

To illustrate the progress of infection, Figure 6 shows the least square means and their confidence limits for the percentage of susceptible pigs depending on factor groups 4-6 and 12. The most effective contacts happened during fattening where pigs spent most of their lives (Figure 6a). Figure 6b shows the importance of the sow herd prevalences at pig producing stage. Comparing the deviations from the default values, it became clear that the decrease in susceptible pigs caused by the maximum values exceeded the increasing effect of the minimum values. The same applied to the proportion of non-shedder to shedder sows (Figure 6c). In contrast, the relation between shedding duration and percentage of susceptible pigs was linear. Increasing and decreasing effects balanced each other out (Figure 6d).The influence of transport-stress on the shedding reactivation is shown in Figure 7. The number of seropositive, shedding pigs (I+) increased especially at transport to slaughter, which increased the probability of a susceptible pig becoming infected.

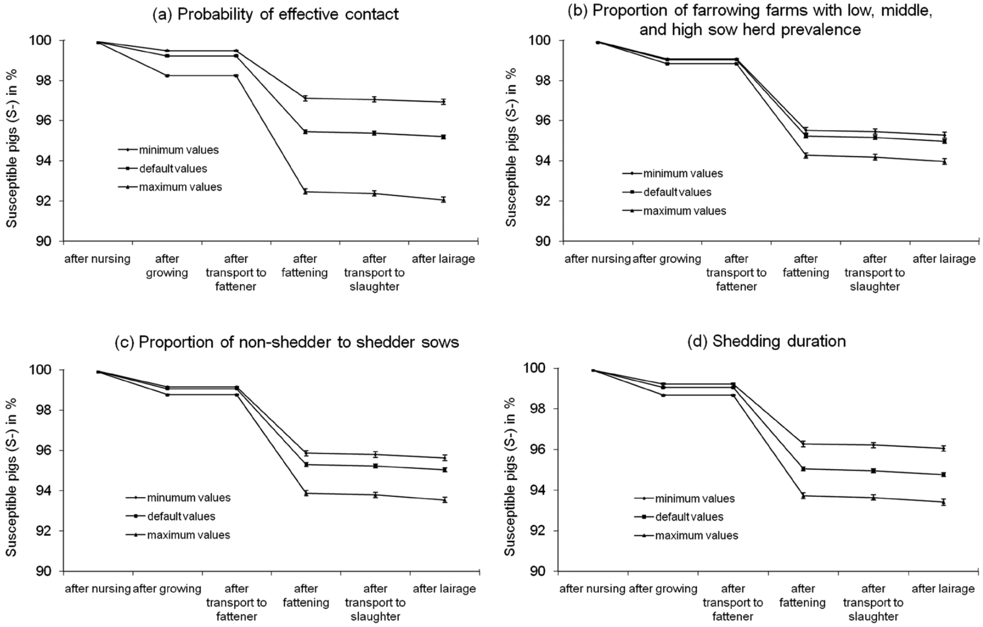

Figure 6
Least square means and confidence limits of the percentage of susceptible pigs depending on (a) the probability of effective contact, (b) proportion of farrowing farms with low, middle, and high sow herd prevalence, (c) the proportion of non-shedder to shedder sows, and (d) the shedding duration

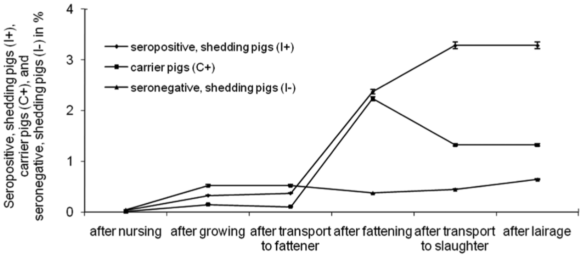

Figure 7
Least square means and confidence limits of the percentage of infectious, seronegative pigs, infectious, seropositive pigs, and carrier pigs depending on the probability to restart shedding (default value)

Discussion

Transition model

The transition model considered both farm level and slaughter stage. The consideration of the four pig's health states was adequate for farm level. Until transport to slaughter, neglecting of the latent period between *Salmonella* ingestion and bacteria shedding was considered to be unproblematic since duration fell below the considered time step of one week. But *Salmonella* ingestion at transport to slaughter resulted immediately in the shedding of bacteria since an intermediate health state was missing. Hence, the chance of a susceptible pig having become infected at lairage might have been overestimated. But infection at lairage represented about 5 % in the maximum case. Overestimation was neither of little consequence for carcass contamination. The passive immunity of newborn piglets was not considered either. Lurette *et al.* (2008) described the maternal protective factor as one of the most influential parameters on *Salmonella* prevalence in delivered pigs but precise information about this effect is missing. In support, Nollet *et al.* (2005) could not prove the direct transmission of *Salmonella* from the sows to their piglets at farrowing barn, which confirms passive immunity. But they demonstrated similarities between the isolates found in the sows and those found during growing, finishing and at slaughter. Hence, even if passive immunity of newborn piglets exists, the sow may play a significant role in the indirect transmission of *Salmonella* to growing and finishing pigs (Nollet *et al.* 2005). In the present study, not even 0.15 % of the piglets had effective contact during nursing. The differences between minimum, default, and maximum values were very small after nursing but increased in the course of time. In the maximum scenario, slaughter pig prevalences of 12 % were reached. This prevalence seemed to be very high, considering that the sows were the only initial infection source in the model presented. But it has to be considered that within the model no infection remained undetected, not even prior to seroconversion. In contrast, empirical prevalences based on antibody detection may never represent all infected pigs at a particular time (Battenberg 2007). Nevertheless, the obtained slaughter pig prevalences emphasize the impact of the sow for *Salmonella* entrance and the subsequent transmission. The transmission dynamic depends extremely on how many piglets come into contact with *Salmonella* during nursing.

Number of replications

The determination of the number of replications is important and difficult at once. Vynnycky & White (2010) emphasise that the number of replications is usually limited by the realisation time. If several models are to be compared, the determinations rely on the number of replications which are required to analyse statistical differences between the models (Chung 2004). But the present study did not compare models. Instead, the number of replications was of concern, which ensured that enough random numbers were used to represent the underlying distribution.

Usually, if this number of replications is obtained, the variance of the output does not decrease due to additional replications. Variations are casual and balance each other. This point was determined to be reached with 30 replications. Some might prefer 80 replications due to further flattening within the trend of the maximum graph, but because of the already small level of standard deviation the additional realisation time seemed to be unjustified.

Sensitivity analysis

Due to the relative high number of input factors a screening design was appropriate to identify the most important factors and evaluate the model based on their estimated effects. P-B designs are screening designs, which estimate unbiased main effects in the smallest design possible (Vanaja & Shobha Rani 2007). The limited number of runs minimised the realisation time and offered the comparison of three levels instead of two. Thus, it could be shown that the relationship between several factors and response variable is non-linear. An increase in the probability of effective contact, a higher proportion of pig producers with middle and high sow herd prevalence, or more shedder sows decreased the percentage of susceptible pigs much more as could have been increased by the minimum values (Figure 6).

The problem that the main effects are confounded with two-factor interactions in a P-B design was solved by reversing the designs. The resulting fold-over pairs were of resolution IV, which did not confound the main effects and two-factor interactions (Box *et al.* 1978, Montgomery 2005). But in contrast to traditional designs of resolution IV, a P-B design does not allow the estimation of interactions between factors (Vanaja & Shobha Rani 2007). Hence, it cannot be ruled out that there are significant relationships between factors which remained undetected.

The estimated main effects of the model met expectations. To avoid that biologically nonrelevant differences become significant, the significance level for the F-Test was reduced to 1 %. In simulation designs with large sample sizes smallest differences become significant. According to Ivanek *et al.* (2004) the probability of effective contact (no. 6), the probability of restarting shedding (no. 7), as well as the shedding duration (no. 12) were proven to be significant. Hill *et al.* (2008) point out that the most effective control strategies are those that reduce the probability of effective contact between pens. The relevance of the proportion of farrowing farms with low, middle, and high sow herd prevalence (no. 4) as well as the importance of the proportion of non-shedder to shedder sows (no. 5) cannot be compared exactly with other studies, since previous models known by the authors did not consider the sow as initial source of *Salmonella* infection in detail. But van der Gaag *et al.* (2004) proved that a higher starting prevalence at farrowing stage results in more infected animals. Furthermore, Ivanek *et al.* (2004) state that *Salmonella* transmission is influenced by the prevalence among weaners.

For the contamination and loss of bacteria at slaughter, detailed information and simulation studies are rare. Van der Gaag *et al.* (2004) describe the slaughterhouse as one of the most important stages in the supply chain to reduce the prevalence of *Salmonella*-contaminated carcasses. Accordingly, the present study showed that a lot of infected pigs lose bacteria due to careful evisceration. But these pigs remain seropositive and increase the herd prevalence even if there is no risk of humans becoming infected. The herd prevalence determines the classification of farms within quality assurance systems and is therefore of paramount importance, especially for the fatteners. The present study showed that efforts to decrease the herd prevalence should be focused on *Salmonella* entry and the transmission via effective contacts between pigs. A conceivable measure to reduce *Salmonella* entry via farrowing sows might be the vaccination of sows and piglets as well the removal of faeces from the pen. Furthermore, the susceptibility of pigs to present bacteria should be reduced. Common recommendations are the acidification of feed or water, rodent control, intensive cleaning and disinfection, et cetera. Subsequent studies will expand the presented model to horizontal entry of *Salmonella* and will analyse the effectiveness of several prevalence-reducing strategies.

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Corresponding author:

Stefanie Hotes
email: shotes@tierzucht.uni-kiel.de

Institute of Animal Breeding and Husbandry, Christian-Albrechts-Universität zu Kiel, Olshausenstraße 40, 24098 Kiel, Germany