

## **Evaluation of two strategies for reducing the spread of *Salmonella* in commercial swine herds during the finishing phase and their incremental cost-effectiveness ratios**

### **Avaliação de duas estratégias para redução da infecção por *Salmonella* em rebanhos comerciais de suínos na fase de terminação e a razão incremental de custo efetividade**

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#### **Highlights:**

The use of a vaccine and a prebiotic for *Salmonella* control in swine was assessed.

Seroprevalence and carcass contamination were compared with the control.

Carcass contamination was 18.33 percentage points lower in the prebiotic group.

The use of prebiotic reduced seroprevalence by 48.5 percentage points.

The cost of reducing seroprevalence in prebiotic was 1.92 USD/ton.

#### **Abstract**

To achieve control of *Salmonella* contamination in pig carcasses, on-farm measures need to be better understood. Complementary strategies require research not only on their effectiveness but also on their financial impact. In this study, we evaluated the incremental cost-effectiveness ratio of two treatments for reducing *Salmonella* seroprevalence in commercial swine herds. Pigs treated with a prebiotic or a vaccine were studied and compared with pigs in an untreated control group. Each strategy was applied to three batches of pigs in a commercial integration system; the animals were followed from farrowing to the slaughterhouse, and their serologies upon arrival at finishing farms and before slaughter were evaluated. Additionally, carcass surface contamination was assessed for each strategy. The seroprevalence upon arrival at the finishing farm was lower than 3% in all groups. In the control and vaccine groups, the seroprevalence increased by more than 90 percentage points from the day of arrival at the finishing farm to four days before slaughter. Only the prebiotic treatment yielded a significant effect on preslaughter seroprevalence (a 49 percentage points reduction from that in the control). Carcass contamination was 0% in the prebiotic group, 18.33% in the control group and 29.16% in the vaccine group. Only prebiotics significantly reduced the seroprevalence of *Salmonella* in the

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studied herds, and the incremental cost-effectiveness ratio associated with prebiotic use was 1.92 USD to reduce seroprevalence by 10 percentage points per carcass ton.

**Key words:** Cost-effectiveness. Finishing herds. Prebiotic. *Salmonella*. Swine. Vaccine.

## Resumo

Para alcançar controle da contaminação por *Salmonella* em carcaças suínas, intervenções na produção primária precisam ser melhor compreendidas. Estratégias complementares requerem não só pesquisas acerca da sua efetividade, mas também dos custos implicados no uso de tais tecnologias. Para tanto, foi avaliada a razão de custo-efetividade incremental de dois tratamentos para reduzir a soroprevalência de *Salmonella* em rebanhos suínos comerciais. O uso de um prebiótico e de uma vacina foram comparados com um controle sem tratamento. Cada estratégia foi aplicada em três lotes de suínos em um sistema comercial de integração. Os animais foram acompanhados da maternidade até o abate e suas sorologias no dia do alojamento na terminação e quatro dias antes do abate foram avaliadas. Também, em cada estratégia, amostras de suabe de carcaça foram coletadas para avaliação da contaminação superficial. A soroprevalência no dia do alojamento na terminação foi menor do que 3% em todos os grupos, sendo que nos grupos controle e vacina a soroprevalência aumentou mais de 90 pontos percentuais quatro dias antes do abate. Apenas o uso do prebiótico levou a um efeito significativo na redução da soroprevalência pré-abate (49 pontos percentuais), quando comparado com o controle. A contaminação das carcaças no grupo prebiótico foi 0%, 18,33% no controle e 29,16 no grupo vacinado. Assim, apenas o prebiótico foi capaz de reduzir a soroprevalência nos rebanhos estudados com razão incremental de custo-efetividade de 1,92 USD para redução de 10 pontos percentuais na soroprevalência por tonelada de carcaça.

**Palavras chave:** Custo-efetividade. Prebiótico. Rebanhos de terminação. *Salmonella*. Suínos. Vacina.

## Introduction

Brazil is one of the largest swine producers in the world and was the fourth-largest exporter of pork in 2017 (Associação Brasileira de Proteína Animal [ABPA], 2018). Therefore, the quality and safety of Brazilian pork should be prioritized to allow increasing participation in the international market and to guarantee the health of consumers. In the European Union, *Salmonella enterica* management has been focused on monitoring pork, since pork consumption was associated with approximately 10% of human salmonellosis cases (Pires, Vigre, Makela, & Hald, 2010). Recently, a *Salmonella* monitoring program in pig carcasses was launched (Instrução Normativa nº 60, 2018) that will lead to the need for control measures throughout the production chain.

Although the hygiene of the slaughter process is considered the basis of any *Salmonella* control program, the relationship between preharvest

sanitary conditions and food safety highlights the need for farm-to-fork control programs, which necessarily integrate on-farm measures to enhance product quality (European Food Safety Authority [EFSA], 2010). According to Alban and Stärk (2005), efforts to control *Salmonella* should consider the epidemiological situation of each country, state or even company. The same authors highlight the importance of keeping a low proportion of swine finishing herds with high *Salmonella* levels to reduce carcass contamination. Baptista, Dahl and Nielsen (2010) have shown that if the number of seropositive animals could be kept below 50 per day, the prevalence of carcass contamination would remain, on average, below 3%. On the other hand, Argüello, Carvajal, Álvarez-Ordóñez, Jaramillo-Torres and Rubio (2014) reported no clear effect of classifying herds into levels of risk based on serology to avoid the contamination of holding pens and the slaughter line activities. In any case, seroconversion represents the exposure to *Salmonella*, and the

final seroprevalence is a measure of the spread of the infection. In this context, several countries have adopted seroprevalence as a risk indicator for carcass contamination (Alban et al., 2012; Mainar-Jaime, Cassanova-Jiges, Andrés-Barranco, & Vico, 2017; Sørensen, Alban, & Dahl, 2004).

In Brazilian herds, specifically those in the southern states, the estimated prevalence of *Salmonella* is high on farms and at slaughterhouses (Bessa, Costa, & Cardoso, 2004; Silva, Gotardi, Vizzotto, Kich, & Cardoso, 2006). *Salmonella* infection seems to spread more intensively in the finishing stage of production (Müller, Schwarz, & Cardoso 2009; Schwarz et al., 2009). Therefore, on-farm control measures and complementary interventions focused on this production phase should be better understood. Previous studies conducted in the same region of Brazil have reported encouraging results from alternative control strategies. Calveyra et al. (2012) have found promising results in pigs fed manno oligosaccharides, which act as prebiotics and competitively bind to type I *Salmonella* fimbriae, reducing intestinal colonization. An attenuated live vaccine based on *Salmonella* serovar Choleraesuis, tested by Schwarz, Kich, Kolb and Cardoso (2011), led to vaccinated animals having lower seroprevalence and excretion of *Salmonella enterica*.

There is no direct financial incentive in the Brazilian domestic market for controlling *Salmonella*. In this context, an evaluation of the incremental cost-effectiveness ratio plays an important role in the application of strategies to reduce pathogens. Thus, management decisions should consider the costs and the measurable impacts, such as the seroprevalence or carcass contamination, of each strategy. The objective of this study was to evaluate two strategies for reducing the spread of *Salmonella* during the finishing phase of

an intensive swine production system and to assess the incremental cost-effectiveness ratio of these measures against a control.

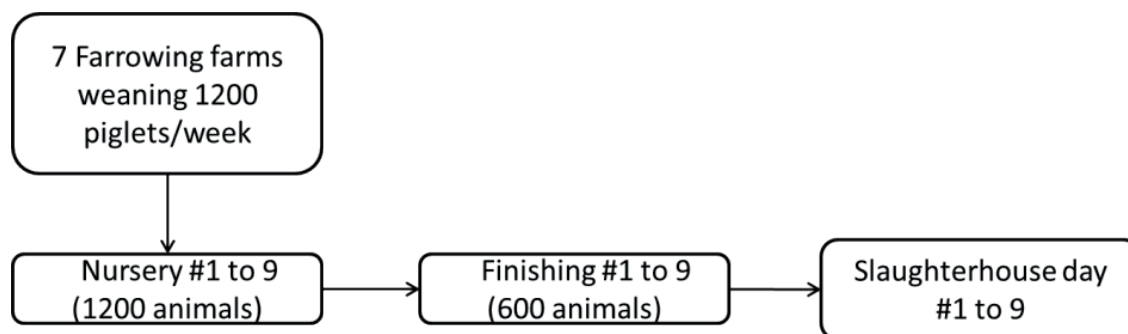
## Materials and Methods

### *Study population*

This study was conducted in the state of Santa Catarina, which is the source of almost 26% of the country's total swine production (Empresa Brasileira de Pesquisa Agropecuária [EMBRAPA], 2019). The study was conducted at a pork processing company with a vertically integrated production chain that rears piglets and pigs in segregated three-site systems. All the piglet production farms, nurseries, and finishing farms are operated as an all-in/all-out production system.

### *Experimental design*

Two strategies to reduce the spread of *Salmonella*, prebiotics and vaccine administration, were assessed and compared with a control. Each strategy was tested on three independent batches of pigs that were followed from farrowing, to the nursery, to the finishing farm. The same seven farrowing farms delivered the piglets in all batches to control for the effect of animal origin. At weaning, at an average weight of 7.5 kg (28 days), 1200 animals were transported to nurseries, where they stayed for 32 days and reached an average weight of 22 kg. Subsequently, 600 of these pigs were transported to finishing farms. After 110 days, at a weight of 120 kg, these animals were delivered to the same slaughterhouse (Figure 1). The control group was followed from August 2011 to February 2012; the vaccine group was followed from September 2011 to March 2012, and the prebiotic group was followed from June 2012 to December 2012.



**Figure 1.** Flowchart of the study design following the batches from the farrowing farm to the slaughterhouse.

For logistical reasons, the animals were not all transported in the same truck, but all the vehicles underwent the same cleaning and disinfection procedures. To be eligible, farms needed to adhere to good production practices (GPP), which were verified monthly using a standard and validated GPP checklist for organizational, biosecurity, hygiene and management procedures (supplementary material). This standard GPP checklist was applied in the control treatment and in both strategies. Among the eligible farms, nine agreed to participate, and the allocation of farms to treatments was made to accommodate the production flow.

### Prebiotic

A commercial prebiotic based on the active fraction of manooligosaccharides extracted from *Saccharomyces cerevisiae* yeast cells was added to the feed from weaning until slaughter. The concentration of the prebiotic in the feed (grams/ton) was 1600 from weaning to 63 days, 800 from 64 to 78 days and 400 from 100 to 173 days (i.e., six hours before slaughter).

### Vaccine

All animals in this group were provided with a commercial avirulent live vaccine based on *Salmonella* serovar *Choleraesuis* variety Kurzendorf. This strain had been attenuated by

several passages in pig neutrophils as previously described (Roof, Kramer, & Kunesh, 1992). This vaccine induces antibodies against somatic antigens O: 6, 7 (C<sup>1-4</sup>), which do not interfere with the serological test used in this study (i.e., the ELISA test, see the laboratory procedures section). Oral vaccinations were performed at the piglet production farms after 24 and before 72 hours after birth by the same veterinarian. In accordance with the manufacturer's guidelines, the use of antimicrobials three days before or after vaccine administration was avoided because it could interfere with the vaccine efficacy. When there were clinical reasons for antimicrobial treatment during the abovementioned interval, the left ears of the treated pigs were marked and they were excluded from the study.

### Blood sampling

To assess the within-batch prevalence, fifty-five blood samples were randomly taken per batch at two independent times: on the day of arrival at the finishing farms and four days before slaughter.

The sizes of these samples were calculated using an expected within-batch seroprevalence of 80% (Schwarz et al., 2009) for a population of 600 animals (i.e., mean herd size) with an absolute error of 10 % and a 95% confidence level with equation 1 below:

$$n = \frac{z_{\alpha/2}^2 [p(1-p)N]}{d^2(N-1) + z_{\alpha/2}^2 [p(1-p)]} \quad (1)$$

where  $n$  = number of samples,  $N$  = population size,  $z$  =  $z$ -value for  $\alpha = 0.05$ ,  $p$  = estimated prevalence, and  $d$  = absolute error.

#### *Bacteriology samples*

Upon arrival at the finishing farms, the same 55 animals that were sampled for serology had rectal swabs taken. For carcasses, the first 40 carcasses from each group entering the slaughterhouse were separated before cooling, and their surfaces were swabbed. Each batch was slaughtered in different weeks on the first day of the week at the beginning of the day (i.e., the first batch of the day) to avoid residual contamination on slaughterhouse surfaces. The sample size was calculated using equation 1, and the parameters were as follows: previous level of prevalence of 20%, population size of 4000 carcasses per day, absolute error of 12% and a 95% confidence level. The samples were taken from the ham, loin, belly and jowl areas by surface swabbing on four 100 cm<sup>2</sup> areas delimited by a sterile template that was changed for each area. Two sponges, hydrated in 10 mL of buffered peptone water (BPW) 1%, were used for each carcass and packed together as one sample.

Samples from the environment were taken prior to pig arrival by swabbing all floor pens with four overshoes, and the two samples for each facility were pooled. All drinkers and feeders were sampled by swab and sponge, respectively, and the two samples per facility were pooled. Each truck used was sampled prior to access by animals; piglet production farm facilities were sampled as a pool before the sows entered the pens, and lairage pens were sampled with overshoes before the arrival of the animals. All samples were soaked in BPW 1% and submitted to *Salmonella* isolation to evaluate the level of contamination to which animals were exposed in the facilities.

#### *Laboratory procedures*

Serum samples were identified and submitted to an in-house ELISA test based on somatic (O) antigens 1, 4, 5, and 12 of *Salmonella enterica* serovar Typhimurium with an optical density cut-off point of 0.169. At this cut-off point, the test has a sensitivity of 0.92 and a specificity of 1 (Kich et al., 2007).

Bacteriological samples were conditioned and pre-enriched in a sterile recipient with 225 mL of buffered peptone water (BPW) 1%, followed by selective enrichment in Rappaport-Vassiliadis medium with soya (Merck & Co., Inc.) and tetrathionate broths (Difco Laboratories, Detroit, USA) and isolation on a solid medium (xylose-lysine-tergitol 4 agar and brilliant green-phenol red-lactose-sucrose agar (Bencton-Dickson and Company) according to the protocol described in ISO 6579:2002/Amd 1:2007 (International Organization for Standardization [ISO], 2002). Each *Salmonella* isolate was serotyped at the National Reference Center, Instituto Oswaldo Cruz in Rio de Janeiro, Brazil.

#### *Statistical analysis*

A logistic regression model was fitted to evaluate the effect of the strategies ( $X_1$ ) on the serology before slaughter. Serology on the day of arrival at the finishing farm ( $X_2$ ) and the residual environmental contamination of trucks and facilities before the animals were placed at nurseries and finishing farms ( $X_3$ ) were tested as confounders in the logistic model. The status of the farms and trucks regarding *Salmonella* presence/absence after cleaning, disinfection and sanitary emptying were added together to compose the variable  $X_3$ . The models follow equation 2:

$$\text{logit}[y] = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 \quad (2)$$



where  $[y]$  is the odds of a *Salmonella*-positive result for each treatment.  $\beta_0$  is the regression intercept;  $\beta_1$  is the effect of each treatment on the output;  $\beta_2$  is the effect of serology on the day of arrival at the finishing farm on the output and  $\beta_3$  is the effect of residual contamination on the output. Variables were considered confounders when the estimated treatments changed by more than 20%. To compare the frequencies between groups, a Wald Chi-square test was applied with a Tukey post hoc correction for multiple comparisons. The analyses were performed with the PROC GENMOD procedure in SAS software, version 9.2 of the SAS System for Windows (SAS Institute Inc. Cary, NC, USA).

#### *Economic analysis*

The incremental cost-effectiveness ratio (ICER) was selected for comparison of the treatments and the control. In this method the costs, but not the benefits, are measurable in monetary terms (Organization for Economic Co-operation and Development [OECD], 2007; Food and Agriculture Organization of the United Nations [FAO],

2016). This technique consists of estimating the monetary costs of the different treatments (USD per ton of carcass weight) and comparing them in a nonmonetary manner (effect): in this case, the variation in preslaughter seroprevalence.

The study estimated the average total production costs for both the contracted pork processor and the finishers for the last three cycles (herds). Contractor costs included feed, piglets, veterinary and medicinal supplies, and logistics, whereas finishing costs include housing, equipment investment and maintenance, energy and heating, manure handling and labor. Quantitative data were collected from the pork processing company to estimate its costs and provide indicators of the production efficiency. The company's costs were assessed via qualitative interviews using a standardized and validated questionnaire and included the average technical efficiency, market prices and cash expenditures for the last three herds or 12 months (2012). The economic parameters are shown in table 1. Investments in biosecurity measures were not considered because all treatments met the same biosecurity requirements.

**Table 1**

**Production cost items and treatment costs in USD/ton of cold carcass used to estimate the incremental cost-effectiveness ratio**

Variable	Costs (USD/ton cold carcass)
Feed	949.94
Piglets	650.04
Veterinary and medicines	39.56
Energy and heating	8.36
Maintenance	2.97
Levies, insurance, inspection	4.67
Miscellaneous	8.48
Labor	62.70
Depreciation	30.66
Average interest costs	23.80
Interest on working capital	19.35
Vaccines	10.86
Prebiotics	9.31

To determine the ICER of decreasing seroprevalence by 10 percentage points, the total cost variation ( $\Delta TC$ ) in USD per ton of carcass weight was divided by the 10 percentage-point reduction in seroprevalence ( $\Delta P_{10}$ ) according to equation 3 below:

$$ICER = \left( \frac{\Delta TC}{\Delta P_{10}} \right) \quad (3)$$

## Results and Discussion

All rectal swabs taken from piglets on the day of arrival at finishing farms were negative, while positive samples from the environment and from trucks were detected in at least one sampling in all tested strategies (Table 2). Seroprevalence, which was lower than 3% on the day of arrival at the finishing farm, increased by more than 90 percentage points in the Control and Vaccine

groups and by approximately 48 percentage points in the Prebiotic group. These results corroborate the findings of Müller et al. (2009) and Silva et al. (2006) demonstrate that the finishing phase is a critical step for *Salmonella* amplification in Brazilian commercial herds. Although this experimental design there is no restriction in randomization of farms (i.e., blocking), there is no reason to believe that the independence of farms could bias the results since all farms are part of the same company and follow the same biosecurity practices that were evaluated monthly by a veterinarian. Additionally, for logistical reasons, the time of year in which each treatment was applied was not exactly the same, but the seasons were the same for the two treatments and the control (winter, spring and summer), and there is no reason to expect that there was a bias related to the effect of seasonality.

**Table 2**

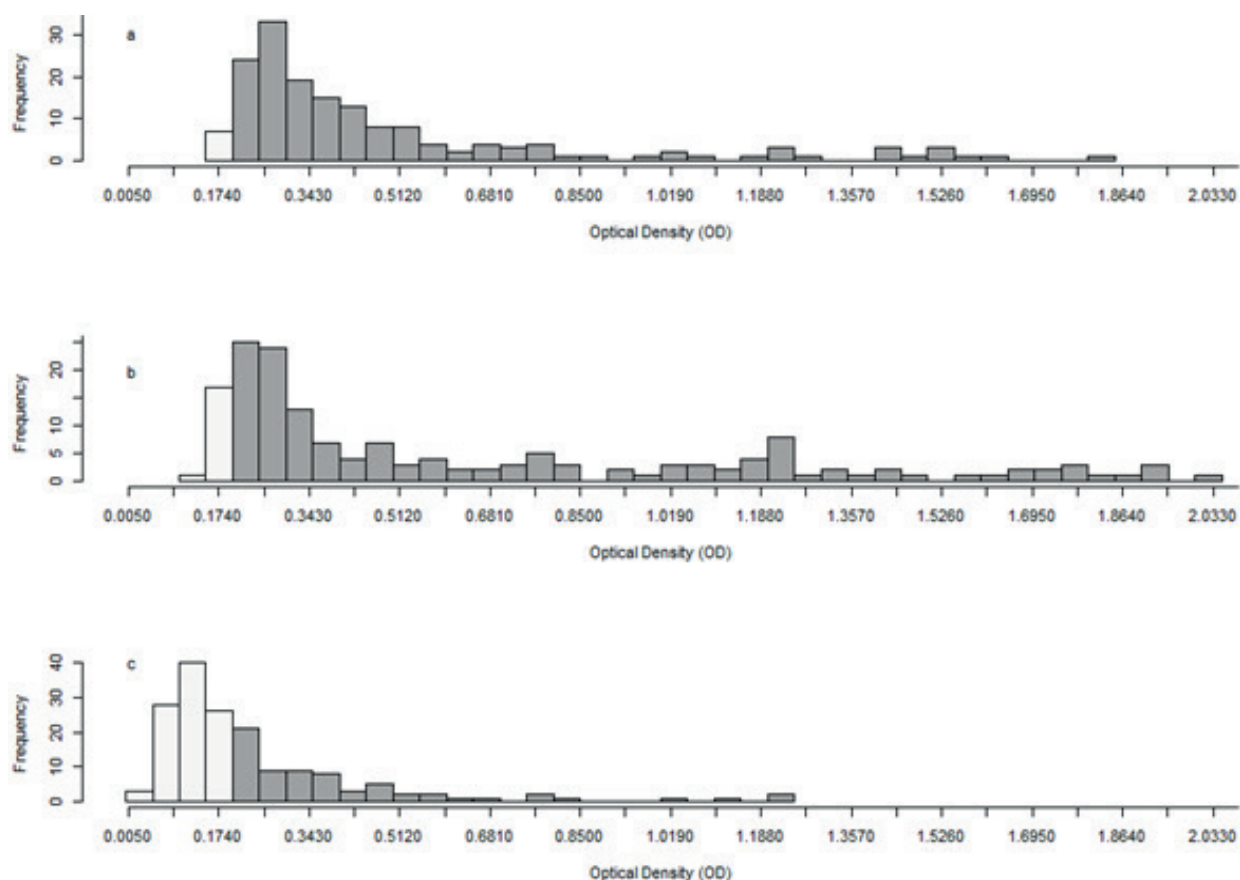
**Environmental samples (positive/total) taken before the arrival of animals during the experiment for all strategy groups**

Strategy	Piglet production farm	Nursery	Floor*	Drinker*	Feeder*	Trucks	Lairage
Control	(5/11)	(0/3)	(1/12)	(1/3)	(1/6)	(1/9)	(0/3)
Prebiotic	(6/17)	(0/3)	(0/12)	(0/3)	(0/6)	(1/9)	(1/3)
Vaccine	(3/16)	(2/3)	(1/12)	(1/3)	(1/6)	(1/9)	(0/3)

\* Samples from facilities of finishing farms.

All the optical densities were right skewed. In the Control and Vaccine groups, a large tail was observed, with optical density values reaching more than 1.8 in both groups (Figure 2a and b). On the other hand, the maximum value for optical density in the Prebiotic group was approximately 1.2 (Figure 2c). No confounding effect was attributed to the initial seroprevalence or to positive environmental samples; thus, only the effect of the treatment was included in the logistic regression model. The Prebiotic group exhibited a significantly lower seroprevalence when

compared to those in both Control and Vaccine groups ( $p$ -value<0.0001), while the seroprevalence did not significantly differ between the Control and Vaccine groups (Table 3). The lower seroconversion in the Prebiotic group indicates that these animals were less exposed to *Salmonella* infection during the finishing phase. In an experimental study, Calveyra et al. (2012) demonstrated that pigs inoculated with *Salmonella enterica* serovar Typhimurium and subjected to an in-feed treatment with probiotics presented a lower excretion of *Salmonella*.



**Figure 2.** Distributions of serological optical density (OD) four days before slaughter in the (a) Control group, (b) Vaccine group and (c) Prebiotic group. Light gray bars represent the distribution of negative samples, and dark gray bars represent the distribution of positive samples using 0.169 as the cut-off.

**Table 3**

**Relative frequency % and confidence interval (CI 95%) of seropositive animals and carcass contamination in each strategy**

Strategy	Serology upon arrival at finishing farm (n=165/group)	Sampling time	
		Preslaughter serology (n=165/group)	Carcass swab (n=120/group)
Control	3.0 (1 - 7.2) <sup>a</sup>	98.8 <sup>a</sup> (95.2-99.7)	18.33 <sup>a</sup> (12.1 - 24.6)
Prebiotic	2.4 (0.6 - 8.4) <sup>a</sup>	50.3 <sup>b</sup> (38 - 62)	0 <sup>b</sup>
Vaccine	1.2 (0.2-5.3) <sup>a</sup>	97 <sup>a</sup> (92-98.7)	29.16 <sup>a</sup> (19.51 - 38.8)

Different letters in the same column indicate significant differences calculated using a Wald chi-square p-value of 0.05 with Tukey correction.



According to Moran (2004), mannan oligosaccharides and mannoprotein act in two similar ways in Gram-negative bacteria: type I fimbria agglutination and cellular immune response modulation. This prebiotic is composed of the active fractions of mannan oligosaccharides and concentrates the mannose molecules (which are responsible for fimbria agglutination), thereby increasing the probability of a prebiotic-bacteria bond despite the bacteria-enterocyte interactions (Hooge, 2004). Furthermore, the effect of mannose on the promotion of specific bacteria, such as *Lactobacillus* sp. and *Bifidobacterium* sp., and the activation of dendritic cells and macrophages enhances cellular intestinal immunity (Moran, 2004). Therefore, our hypothesis is that the effect on seroprevalence detected in the Prebiotic group may be related to a reduced spread of *Salmonella* in the finishing stage.

However, even though they showed a reduced seroprevalence level, approximately 50% of the pigs belonging to the Prebiotic group still had contact with *Salmonella* and carried antibodies detected by the ELISA test. This suggests that other factors, particularly biosecurity failures, may have occurred during this period. These failures might be related to cross-contamination/infection between animals and the environment or the consumption of contaminated feed (Melo et al., 2011). Furthermore, experts from different countries have agreed on the importance of general hygiene, all-in/all-out management, feed contamination prevention, rodent control and good working practices for *Salmonella* control (Stärk et al., 2002). In this sense, biosecurity measures should be considered the basis of *Salmonella* control, which can be combined with treatments such as prebiotics to achieve effective *Salmonella* control in a shorter time span. Although Schwarz et al. (2011) reported a decrease in seroprevalence and *Salmonella* fecal shedding in swine herds immunized with the same vaccine, no difference in seroprevalence was observed between the Vaccine and Control groups in our study. The efficacy of a vaccine is influenced

by factors such as the different serotypes that cause infection and infection pressures and dynamics on the farms (EFSA, 2006). All these factors may have played a role in the failure of protection observed after the vaccination.

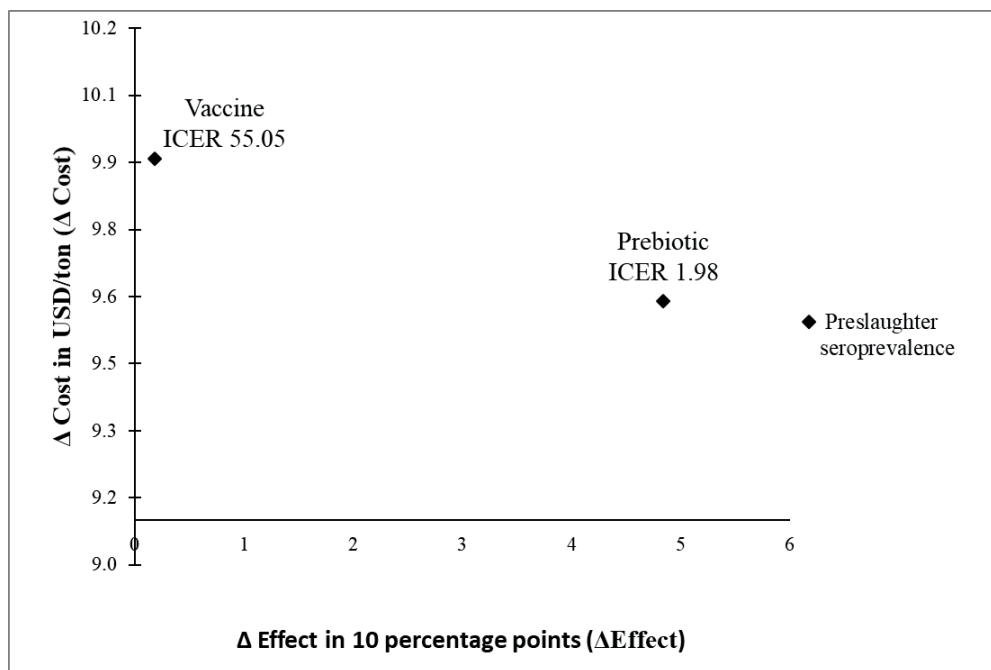
The frequency of carcass contamination was higher for the Control (19.1%) and Vaccine (27.1%) groups when compared with that in the Prebiotic group (0%) in this study. The positive carcass prevalence found in the Control group was similar to that in previous studies conducted in the same region (Kich et al., 2011). Although we are not able to confirm this correlation with our study design, the lower seroprevalence in the Prebiotic group, which represents a lower exposure rate on the farm, may have resulted in a lower number of harboring/shedding animals at slaughter, reducing the probability of contamination during dressing activities. This hypothesis is supported by previous studies (Baptista et al., 2010; De Busser et al., 2011; Silva, et al., 2012) demonstrating that batches with low seroprevalence had a lower probability of carcass contamination. Of the 56 *Salmonella* isolates from carcasses, only serovars Typhimurium and Anatum were identified. In carcasses belonging to the Control group, all strains belonged to serovar Typhimurium, while in the Vaccine group, 29 of 33 strains were *S. Anatum*. The remaining four strains were *S. Typhimurium*. Several *Salmonella* serovars have been reported in pig carcasses in Brazil, however *S. Typhimurium* is among the most prevalent in all studies (Bessa et al., 2004; Kich et al., 2011; Silva et al., 2012).

Carcass contamination/decontamination is a complex and iterative process influenced by many factors, such as batch prevalence levels and environmental contamination as well as slaughterhouse practices related to hygiene, cleaning and disinfection, and education and training (Argüello, Álvarez-Ordoñez, Carvajal, Rubio, & Prieto, 2013). To avoid bias related to slaughterhouse hygiene, pigs in this study were always the first batch slaughtered on Mondays,

after a complete cleaning and disinfection and two days of the slaughterhouse remaining empty. In this regard, the effect of hygiene practices was the same for all batches, and no further bias regarding residual contamination on slaughter equipment surfaces was identified in relation to differences in carcass contamination.

On average, the ICERs associated with the Prebiotic and Vaccine treatments were 1.92 and 60.36 USD/ton to reduce the seroprevalence by 10 percentage points, and the costs associated with attaining these effects are depicted in the cost-effectiveness graph (Figure 3). The costs

associated with the prebiotic did not impact the farmers' budgets because the costs were borne by the contractors (industry). Although the benefits of *Salmonella* control cannot be measured in monetary terms, these contractors also enjoy the benefits of control strategies through compliance with importer standards and by avoiding legal problems and brand threats due to public health impacts associated with *Salmonella*. Furthermore, the results observed here do not advocate the use of prebiotics without also ensuring biosecurity and good production practices. The vaccine did not reduce the seroprevalence compared with that in the control, so there was no need to calculate the ICER.



**Figure 3.** Incremental cost-effectiveness ratio (ICER) values for a 10 percentage points decrease in seroprevalence in USD per ton of carcass weight for the Vaccine and Prebiotic strategies.

Although further studies should be conducted to assess the effects and associated costs of other strategies to increase the body of information, this study indicated that prebiotic use may be considered a complementary control measure for *Salmonella* control. The use of prebiotics, in turn, should be considered in addition to biosecurity

programs, which are largely recognized as the basis of *Salmonella* control with a low impact on the production cost. Furthermore, good production measures are important in controlling not only *Salmonella* but all pathogens that cause economic losses in swine production.

## Conclusions

The vaccine was not effective in reducing the within-herd spread of *Salmonella* during the finishing phase. The prebiotic significantly reduced the spread of *Salmonella* in the studied herds, and the ICERs associated with 10% reductions in seroprevalence due to prebiotic use were estimated to be 1.92 USD/ton.

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## Conflict of interests

The authors declare no conflict of interests.

## References

- Associação Brasileira de Proteína Animal (2018). *Relatório anual 2018*. Retrieved December 3, 2019, from <http://abpa-br.com.br/setores/avicultura/publicacoes/relatorios-anuais/2018>
- Alban, L., Baptista, F. M., Møgelmoose, V., Sørensen, L. L., Christensen, H., Aabo, S., & Dahl, J. (2012). Salmonella surveillance and control for finisher pigs and pork in Denmark - A case study. *Food Research International*, 45(2), 656-665. doi: 10.1016/j.foodres.2011.02.050
- Alban, L., & Stärk, K. D. C. (2005). Where should the effort be put to reduce the Salmonella prevalence in the slaughtered swine carcass effectively? *Preventive Veterinary Medicine*, 68(1), 63-79. doi: 10.1016/j.prevetmed.2005.01.001
- Argüello, H., Álvarez-Ordoñez, A., Carvajal, A., Rubio, P., & Prieto, M. (2013). Role of slaughtering in Salmonella spreading and control in pork production. *Journal of Food Protection*, 76(5), 899-911. doi: 10.4315/0362-028X.JFP-12-404
- Argüello, H., Carvajal, A., Álvarez-Ordóñez, A., Jaramillo-Torres, H. A., & Rubio, P. (2014). Effect of logistic slaughter on Salmonella contamination on pig carcasses. *Food Research International*, 55, 77-82. doi: 10.1016/j.foodres.2013.10.041
- Baptista, F. M., Dahl, J., & Nielsen, L. R. (2010). Factors influencing Salmonella carcass prevalence in Danish pig abattoirs. *Preventive Veterinary Medicine*, 95(3-4), 231-238. doi: 10.1016/j.prevetmed.2010.04.007
- Bessa, M. C., Costa, M. da, & Cardoso, M. (2004). Prevalência de Salmonella sp em suínos abatidos em frigoríficos do Rio Grande do Sul. *Pesquisa Veterinária Brasileira*, 24(2), 80-84. doi: 10.1590/S0100-736X2004000200006
- Calveyra, J. C., Nogueira, M. G., Kich, J. D., Biesus, L. L., Vizzotto, R., Berno, L.,... Cardoso, M. (2012). Effect of organic acids and mannanoligosaccharide on excretion of Salmonella typhimurium in experimentally infected growing pigs. *Research in Veterinary Science*, 93(1), 46-47. doi: 10.1016/j.rvsc.2011.08.018
- De Busser, E. V., Maes, D., Houf, K., Dewulf, J., Imberechts, H., Bertrand, S., & De Zutter, L. (2011). Detection and characterization of Salmonella in lairage, on pig carcasses and intestines in five slaughterhouses. *International Journal of Food Microbiology*, 145(1), 279-286. doi: 10.1016/j.ijfoodmicro.2011.01.009
- Empresa Brasileira de Pesquisa Agropecuária (2019). *Estatísticas suínos no Brasil*. Retrieved December 3, 2019, from <https://www.embrapa.br/suinos-e-aves/cias/estatisticas/suinos/brasil>
- European Food Safety Authority (2006). Opinion of the Scientific Panel on biological hazards (BIOHAZ) related to "Risk assessment and mitigation options of Salmonella in pig production." *EFSA Journal*, 341, 1-131. doi: 10.2903/j.efsa.2006.341
- European Food Safety Authority (2010). Panel on biological hazards; scientific opinion on a quantitative microbiological risk assessment of Salmonella in slaughter and breeder pigs. *EFSA Journal*, 8(4), 1547-1590. doi: 10.2903/j.efsa.2010.1547
- Food and Agriculture Organization of the United Nations (2016). *Economic analysis of animal diseases. Animal production and health guidelines* (18nd ed.). Rome: Food and Agriculture Organization of the United Nations and World Health Organization.
- Hooge, D. M. (2004). Meta-analysis of broiler chicken pen trials evaluating dietary mannan oligosaccharide, 1993-2003. *International Journal of Poultry Science*, 3(3), 163-174. doi: 10.3923/ijps.2004.163.174
- Instrução Normativa nº 60 de 20 de dezembro de 2018. *Diário Oficial da União* (p. 3). Imprensa Nacional.

- International Organization for Standardization (2002). Detection of *Salmonella* spp. in animal faeces and in environmental samples from the primary production stage (ISO Standard No. 6579). Retrieved from <https://www.iso.org/standard/42109.html>
- Kich, J. D., Schwarz, P., Eduardo S, L., Coldebella, A., Piffer, I. A., Vizzoto, R., & Cardoso, M. (2007). Development and application of an enzyme-linked immunosorbent assay to detect antibodies against prevalent *Salmonella* serovars in swine in southern Brazil. *Journal of Veterinary Diagnostic Investigation*, 19(5), 510-517. doi: 10.1177/104063870701900508
- Kich, J. D., Coldebella, A., Morés, N., Nogueira, M. G., Cardoso, M., Fratomico, P. M.,... Luchansky, J. B. (2011). Prevalence, distribution, and molecular characterization of *Salmonella* recovered from swine finishing herds and a slaughter facility in Santa Catarina, Brazil. *International Journal of Food Microbiology*, 151(3), 307-313. doi: 10.1016/j.ijfoodmicro.2011.09.024
- Mainar-Jaime, R., Casanova-Higes, A., Andrés-Barranco, S., & Vico, J. (2017). Revisiting the role of pig serology in the context of *Salmonella* control programs in countries with high prevalence of infection preliminary study. *International Conference on the Epidemiology and Control of Biological, Chemical and Physical Hazards in Pigs and Pork*, Foz do Iguaçu, PR, Brazil.
- Melo, R. T., Guimarães, A. R., Mendonça, E. P., Coelho, L. R., Monteiro, G. P., Fonseca, B. B., & Rossi, D. A. (2011). Identificação sorológica e relação filogenética de *Salmonella* spp. de origem suína. *Pesquisa Veterinária Brasileira*, 31(12), 1039-1044. doi: 10.1590/S0100-736X2011001200001
- Moran, C. A. (2004). Functional components of the cell wall of *Saccharomyces cerevisiae*: applications for yeast glucan and mannan. In K. A. Lyons, T. P. Jacques (Ed.), *Nutritional biotechnology in the feed and food industries*. Lexington, Kentucky: CAB Direct.
- Müller, M., Schwarz, P., Kich, J., & Cardoso, M. (2009). Perfil sorológico e de isolamento de *Salmonella* sp. em suínos no início de terminação e ao abate. *Ciência Animal Brasileira*, 10(3), 931-937.
- Organization for Economic Co-operation and Development (2007). *Handbook for appraisal of environmental projects financed from public funds*. Paris: OECD Publishing.
- Pires, S. M., Vigre, H., Makela, P., & Hald, T. (2010). Using outbreak data for source attribution of human salmonellosis and campylobacteriosis in Europe. *Foodborne Pathogens and Disease*, 7(11), 1351-1361. doi: 10.1089/fpd.2010.0564
- Roof, M. B., Kramer, T. T., Kunesh, J. P., & Roth, J. A. (1992). In vivo isolation of *Salmonella choleraesuis* from porcine neutrophils. *American Journal of Veterinary Research*, 53(8), 1333-1336
- Schwarz, P., Calveira, J., Sella, A., Bessa, M., Barcellos, D. E. S. N., & Cardoso, M. (2009). *Salmonella* enterica: isolamento e soroprevalência em suínos abatidos no Rio Grande do Sul. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, 61(5), 1028-1034. doi: 10.1590/S0102-09352009000500003
- Schwarz, P., Kich, J. D., Kolb, J., & Cardoso, M. (2011). Use of an avirulent live *Salmonella Choleraesuis* vaccine to reduce the prevalence of *Salmonella* carrier pigs at slaughter. *The Veterinary Record*, 169(21), 553. doi: 10.1136/vr.d5510
- Silva, L. E., Gotardi, C. P., Vizzotto, R., Kich, J. D., & Cardoso, M. R. I. (2006). Infecção por *Salmonella* enterica em suínos criados em um sistema integrado de produção do sul do Brasil. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia*, 58(4), 455-461. doi: 10.1590/S0102-09352006000400001
- Silva, L. E., Dias, V., Ferronato, A., Guerra, P., Berno, L., Triches, N.,... Cardoso, M. (2012). Longitudinal dissemination of *Salmonella* enterica clonal groups through the slaughter process of *Salmonella*-positive pig batches. *Journal of Food Protection*, 75(9), 1580-1588. doi: 10.4315/0362-028X.JFP-11-515
- Sørensen, L. L., Alban, L., Nielsen, B., & Dahl, J. (2004). The correlation between *Salmonella* serology and isolation of *Salmonella* in Danish pigs at slaughter. *Veterinary Microbiology*, 101(2), 131-141. doi: 10.1016/j.vetmic.2004.02.016
- Stärk, K. D. C., Wingstrand, A., Dahl, J., Møgelmoose, V., & Lo Fo Wong, D. M. A. (2002). Differences and similarities among experts' opinions on *Salmonella* enterica dynamics in swine pre-harvest. *Preventive Veterinary Medicine*, 53, 7-20. doi: 10.1016/S0167-5877(01)00278-1