

## The use of a *Salmonella* Typhimurium live vaccine to control *Salmonella* Typhimurium in fattening pigs in field and effects on serological surveillance

Lindner, T\*, Springer, S., Selbitz, H.-J.

Impfstoffwerk Dessau Tornau (IDT) GmbH, Streetzer Weg 15 a, D-06862 Rodleben, Germany, Phone: +49 34901 885 435 Fax: +49 34901 885 327

\*corresponding author: [thomas.lindner@idt-direct.de](mailto:thomas.lindner@idt-direct.de)

### Abstract

This field study was designed to evaluate the use of a live-attenuated *Salmonella* Typhimurium vaccine in pigs in respect of efficacy against *S. Typhimurium* at time of slaughter and the effect on serological herd monitoring using a commercial mixed LPS-ELISA.

About 1289 slaughtered pigs (805 of non vaccinated groups and 484 of vaccinated groups) were investigated by bacteriological and serological examination (1149 pigs).

The study showed the efficacy of an oral vaccination with a live-attenuated *Salmonella* Typhimurium vaccine in reducing the number of *Salmonella* carrying pigs at slaughter without a detectable interference with the serological monitoring of *Salmonella* (using a cut off at 40% OD level).

### Introduction

The work carried out in this study was designed to evaluate efficacy of a live *Salmonella* Typhimurium vaccine in a pig population with low prevalence of *Salmonella* Typhimurium and no clinical signs of salmonellosis. Furthermore the impact of the vaccination on the monitoring of the herd with a commercial ELISA kit test was investigated.

### Material and Methods

The study was conducted as sequential trial in a commercial pig herd with no signs of clinical salmonellosis but subclinical infection with *Salmonella* Typhimurium.

The swine population was a fattening one with piglet production and rearing under "all-in-all-out"-management. During the first period of this study 805 nonvaccinated pigs were sampled at slaughter. At the same time sows and their offspring were vaccinated. Thereafter 484 vaccinated pigs were examined in the same way.

The vaccine used was a commercial available live *Salmonella* Typhimurium vaccine (SALMOPORC®) and was based on a double-attenuated strain of *S. Typhimurium*, phage type DT 9, containing the serotype-specific plasmid of 60 MDa. This strain can be distinguished from field strains of the same serotype on the basis of its auxotrophy (ade- and his-), using a rapid test (*Salmonella* Diagnostic Kit from IDT GmbH) and molecular biology methods (Schwarz et al., 1995).

As far as possible all sows were given one dose of the vaccine ( $\geq 5 \times 10^8$  cfu) at 6 and again at 3 weeks ante partum by the subcutaneous route. All viable piglets of the vaccinated sows were given a single dose by the oral route on day 21 and 42 post partum.

Culturing of ileocaecal lymph nodes was done to demonstrate the presence of *Salmonella* (qualitative) using standard microbiological culturing. Bacteriological culture carried out by 18 hours pre-enrichment at 37°C Buffered Peptone Water (BPW; Merck), 48 hours selective enrichment at 37°C in RVS medium (Merck), with subculturing on to Rambach and XLT4 agar (Merck) plates after 24 and 48 hours culture. The plates were incubated for 24 hours at 37°C and suspect colonies confirmed by standard biochemical and serological tests (SIFIN, Berlin). The wild-type strain distinguished from the vaccine strain using the IDT *Salmonella* Diagnostic Kit.

Blood samples were examined by a Mixed-LPS-ELISA (SALMOTYPE®, Labor Diagnostik, Leipzig) using a cut off at 40% OD level.

### Results

In the bacteriological examination 64 of 805 non-vaccinated pigs showed *Salmonella* Typhimurium at slaughter whereas 10 of 484 of vaccinated pigs did so (Table 1). All isolates of the vaccinated

pigs have been wild strains. The Odd Ratio for being tested positive in bacteriological examination for non vaccinated pigs was 4.09 (ci95%: 2.05 – 9.03,  $p=3.38E-6$ ). The risk factor was not being vaccinated. The result suggested that vaccination reduced the number of Salmonella carrying pigs at slaughter.

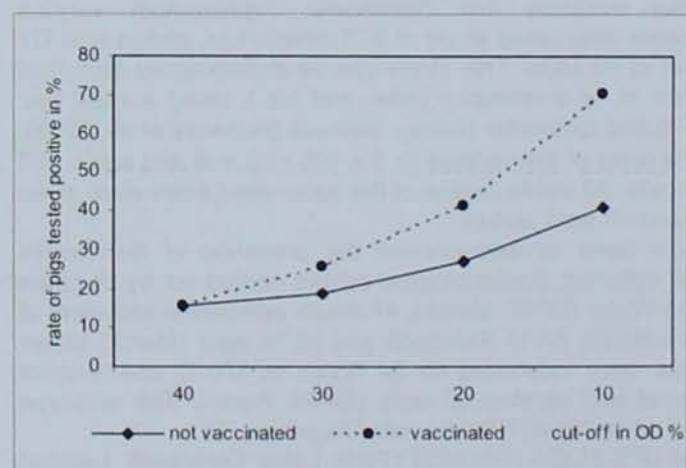
The serological examination of blood samples taken at slaughter revealed 127 of 802 pigs tested positive among the non-vaccinated groups and 55 of 347 pigs tested positive of the vaccinated groups. The OR for being tested positive in ELISA at 40% OD level was 1.0 (ci95%: 0.70 – 1.44,  $p=1.0$ ). The results of the serological examination showed that the oral vaccination didn't increase the number of pigs tested positive by ELISA.

Table 1: Bacteriological and Serological results among vaccinated and not vaccinated pigs

Date	Group	No Lymph nodes tested	Lymph tested positiv	Rate (%) of pos. lymph node samples	No Serum samples	No positiv samples (40%OD)	Rate (%) of pos. serum samples
2407	non-vaccinated	165	10	6.06	160	45	28.13
2108	non-vaccinated	100	3	3.00	92	26	28.26
2011	non-vaccinated	69	3	4.35	100	7	7.00
0412	non-vaccinated	118	1	0.85	100	16	16.00
1112	non-vaccinated	88	5	5.68	70	19	27.14
0502	non-vaccinated	135	16	11.85	-	-	-
1202	non-vaccinated	109	15	13.76	93	6	6.45
0904	non-vaccinated	83	0	0.00	90	5	5.56
3004	non-vaccinated	103	21	20.39	97	3	3.09
0907	vaccinated	95	0	0.00	98	12	12.24
2307	vaccinated	150	4	2.67	50	7	14.00
3007	vaccinated	124	2	1.61	100	15	15.00
0608	vaccinated	115	4	3.48	99	21	21.21
<b>non vaccinated (Total)</b>		<b>805</b>	<b>64</b>	<b>7.95</b>	<b>802</b>	<b>127</b>	<b>15.84</b>
<b>vaccinated (Total)</b>		<b>484</b>	<b>10</b>	<b>2.07</b>	<b>347</b>	<b>55</b>	<b>15.85</b>

Considering lower cut off levels (%OD) than 40% resulted in a rising rate of pigs tested positive among the vaccinated group than in the control group (Figure 1). That trend increases with lowering of the cut off level.

Figure 1: The influence of the cut off level (%OD) that has chosen in the Salmonella ELISA on the rate of pigs tested positive at slaughter in vaccinated and non vaccinated pigs.





## Discussion

This study demonstrates that the vaccination of pig with a live attenuated *Salmonella* Typhimurium vaccine (SALMOPORC®) was efficacious in reducing the number of *Salmonella* Typhimurium positive pigs at slaughter when used in a herd with low level of prevalence of *Salmonella*. This result corresponds to other trials using that *Salmonella* live vaccine in herds with high prevalence of *Salmonella* Typhimurium (LINDNER, 2001). The serological control of both groups (ELISA with cut off level 40% OD) showed nearly a similar overall rate of pigs tested positive whereas the bacteriological examination revealed lower rate of *Salmonella* Typhimurium carrying pigs among the vaccinated animals. But both methods show a low prevalence of the herd. Using lower cut off levels to be more stringent in the control of *Salmonella* may cause a change the category of vaccinated herds earlier than in non-vaccinated herds. That may be seen as an effect of the vaccination itself or of a different immune response of vaccinated pigs compared to non-vaccinated pigs to *Salmonella* present in the environment during fattening period.

## Conclusions

The vaccination in a herd with low level of prevalence of *Salmonella* following that scheme proposed for the vaccine will be efficacious in reducing *Salmonella* Typhimurium in fattening pigs and will not have an adverse impact on the classification into risk categories as long as *Salmonella* ELISA are used at the 40% OD level. When using cut off levels lower 40% OD there may be an influence on to the classification into risk categories. In situations where a bacteriological monitoring is used there will be no influence. This has to be considered when preparing control programs based on serological surveillance of pig herds.

## References

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