

# COMPARISON OF *SALMONELLA* RECOVERY RATES BY USING PLATING AND POLYMERASE CHAIN REACTION METHODS ON SAMPLES OBTAINED FROM A ONE YEAR SURVEILLANCE PROGRAM IN AN INTEGRATED PIG PRODUCTION SYSTEM



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## INTRODUCTION

Many methods exist to recover *Salmonella* spp in food or non-food materials. Cultural methods comprise different phases consisting in pre-enrichment, selective enrichment, plating on selective solid media and finally, identification. One of the major drawbacks is the non-detection of viable, non-culturable bacteria, that still might be responsible for disease in the field (Candrian, 1995). The same author also notes how time-consuming are classical methods, in comparison with DNA hybridisation techniques. In last decades various direct and indirect methods have been marketed. Among these, PCR methods have several advantages, including: a sensitivity and a specificity estimated at nearly 100 %, high rapidity and automatism. The aim of the present work was to compare two different *Salmonella* recovery methods in term of efficiency, rapidity and reliability.

## MATERIAL

- 14 pig herds located in the Southern part of Belgium, were followed
- from 1<sup>st</sup> November 1999 to 31<sup>st</sup> October 2000
- 6,800 slaughter pigs produced each year
- one herd, belonging to the production system and composed of only 600 sows, provides all weaned piglets

## METHODS

**Table 1 : Sampling plan**

Sample matrix	Method of sample	Volume of sample
Feeds	Random sampling when loading the lorry.	25 g of meal
Breeding	1x / month (every farrow should be inspected)	5 faeces samples pooled in 25 g
Weaned pigs (→ 20 kg)	Once on every batch 8 days before going out	5 faeces samples pooled in 25 g
Fattening	Twice on every batch (after 2 months and 4 months of fattening)	5 faeces samples pooled in 25 g
Slaughterhouse	Once on every batch	5 samples of contents of large intestine pooled in 25 g
Carcasses	Once on every batch	Pool of 5 surface swabs (5 X 600 cm <sup>2</sup> ) <sup>(1)</sup>
Cutting room	Random sampling	25 g of retail cut
Mincing room	Random sampling	25 g of ground minced meat
Butcheries	Random sampling	25 g of ground minced meat

### Classical bacteriological analyses (« Diassalm »)

A 18 hours Buffered Peptone Water (BPW) pre-enrichment step is operated at 37 °C (25 g + 225 ml BPW). After a streak on Diassalm and incubation at 42 °C during 24 hours, isolation is achieved on XLT4 medium. Suspected colonies appear red with a dark centre disc following a 22 hour incubation at 37 °C. Confirmation is achieved with classical biochemical and serological methods.

### PCR protocol (Probelia™)

Marketed PCR protocol was modified only for fecal matter by adding a second overnight enrichment in Rappaport-Vassiliadis broth after the BPW pre-enrichment

### Test values

Since both methods were used on the same samples, it was possible to calculate a relative sensitivity and a relative specificity for different types of matrixes using Diassalm method as reference. *Kappa* coefficients were also calculated.

<sup>(1)</sup> same pigs as those sampled for contents of large intestine. Areas of carcass swabbing were adapted from areas chosen by Korsak et al. (1998).

## RESULTS

**Table 2 : Relative sensitivity (SER) and specificity (SPR) for *Salmonella* recovery using PCR method at different sampling places**

	n	SER	SPR	<i>Kappa</i>		n	SER	SPR	<i>Kappa</i>
Feeds	239	61.9 %	98.2 %	0.657	Fattening pigs 2 m	121	66.7 %	89.6 %	0.48
Pregnant sows	60	37.5 %	92.3 %	0.315	Fattening pigs 4m	114	43.8 %	93.9 %	0.408
Lactating sows	149	20 %	93.8 %	0.093	Contents of large intestine	110	36 %	96.7 %	0.344
Weaned pigs	69	0 %	94.0 %	-0.04	Pork meat	259	91.7 %	95.5 %	0.625

## DISCUSSION & CONCLUSION

PCR relative sensitivity of faeces average 46 %, it was very low for contents of large intestine at slaughterhouse and was excellent for pork meat. This constitutes a major drawback, as it does not guarantee certitude for a negative result. Thus, to know *Salmonella* spp status of pigs before slaughtering, this method is not suitable to detect positive herds. In contrast, PCR relative specificity averages 90 % and seemed not to be affected by faeces. For *Kappa* coefficients, values superior to 0.6 were only obtained for animal feed and pork meat

**Probelia™ method appeared only reliable for pork meat and animal feeds.**

## REFERENCES

- Candrian, U., 1995. Polymerase chain reaction in food microbiology. J. Microbiol. Meth. 23,89-103.  
 Korsak, N., Daube, G., Ghafir, Y., Chahed, A., Jolly, S., and Vindevogel, H., 1998. An efficient sampling technique used to detect four foodborne pathogens on pork and beef carcasses in nine Belgian abattoirs. J. Food Prot. 61,535-541.

## ACKNOWLEDGEMENT

This project is financially supported by DG6 from the Belgian federal ministry of agriculture and DGA from Walloon regional ministry.