

Diarrhoea in Neonatal piglets caused by enterotoxigenic K99-positive *E. coli*; immunization of sows using a K99 vaccine.

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In the Netherlands diarrhoea in neonatal piglets due to enterotoxigenic *E. coli* was a major problem in pig husbandry. After the introduction of LT-88 vaccines losses were reduced rapidly.*

In spite of well performed vaccination schedules with these vaccines a gradual increase of the frequency of neonatal diarrhoea (N.D.) in piglets has been noticed over the last few years.

The last two years we have examined 6 farms with N.D. Examination of samples (fecal and/or post mortal) for the presence of enterotoxigenic *E. coli* was done by routine bacteriological methods.* For the detection of the K99 antigen in faecal samples the ELISA also was applied.* The problems on all 6 farms were caused by K99⁺ enterotoxigenic *E. coli*. The OK type 09 K30 K99 (ST⁺) was the predominant type.

In spite of the treatment with antibiotics and/or chemotherapeutics the problem persisted on all farms but one. Therefore it was decided to vaccinate sows with a home-made whole bacterial cell vaccine containing the K99 antigen. The vaccine strain (a K12 transconjugate, that harbors the K99 plasmid of reference strain B41) was cultured in 10 ml trypticase soy broth (T.S.B.) for 24 hours at 37°C; a subculture was made in 2 l. of TSB (20 hrs., 37°C), which subsequently was inoculated into 30 l. of T.S.B., cultivated for 24 hours at 37°C in a fermentor. The formalinized culture was harvested, washed twice with PBS (pH 7.2) and resuspended in 0,15 M NaCl.

The suspension was emulsified with an oil phase (W/O 1 : 1), the final vaccine contained 10⁸ cells/ml and was administered twice to sows in a single dose of 2 ml (i.m.).

The response of vaccination in sows serum and milk was followed by ELISA.

Results of one farm are shown in table 1.

Table 1

number of	before vaccination			after first vac.			after second vac.		
	tot.	pos.	neg.	tot.	pos.	neg.	tot.	pos.	neg.
sows									
serum	20	2	18	10	10	0	16	16	0
milk	10	3	7	6	3	3	9	7	2

The presence of K99 positive *E. coli* in faeces of scouring piglets is shown in Table 2.

Table 2

K99 positive *E. coli* in faeces of scouring piglets (detected by ELISA)¹.

number	before vaccination			after vaccination		
	total ² of piglets	pos.	neg.	total of piglets	pos.	neg.
	52	47	5	49	0	49

1. same farm as table 1.

2. minimal 10 litters

Morbidity in neonatal piglets declined from 90% to 20% of the litters and the mortality rate in all the piglets until weaning from 35% to 15%.

It is concluded that in the Netherlands there is a need for the incorporation of the K99 antigen in the *E. coli* vaccines for pigs.

Selected references:

- Rozemond, H., *E. coli* enterotoxigenicosis in Unweaned Piglets. IV. Evaluation of Results Obtained on Using an Adjuvant Vaccine in the Field. Tijdschr. Diergeneesk., deel 101, afl. 9, 1976, (P.481).
- Guinée, P.A.M., Jansen, W.H., Wadström, T. and Sellwood, R., 1981. In: Proceeding E.C. Workshop: Diagnostic technics for enteropathogenic agents associated with neonatal diarrhoea in calves and pigs (P.163). Eds. P.W. de Leeuw and P.A.M. Guinée.
- Ellens, D.J., de Leeuw P.W., Rozemond, H. (1979) Detection of the K99 antigen of *E. coli* in calf faeces by enzyme-linked immunosorbent assay (ELISA). Vet. Quarterly 1. 169-175.