

Isolation of fimbriated F18 and Shiga-toxin producing *Escherichia coli* associated with oedema disease in post-weaned pigs in Malaysia

Kalaiwaney Muniandy¹, Lee Pheng Ong¹, See Mun Tuam¹, Seetha Jaganathan¹, Pow Yoon Choo², Bee Lee Ong², Peck Toung Ooi², Chiou Yan Tee³, Shiao Pau How³, Wei Hoong Loh³, Jin Wee Lee³, Kuan Lim Goh³
¹Asia-Pacific Special Nutrients Sdn. Bhd., Petaling Jaya, Selangor, Malaysia.
²Universiti Putra Malaysia, Serdang, Selangor, Malaysia
³Rhone Ma Malaysia Sdn. Bhd., Petaling Jaya, Selangor, Malaysia
chiouyan.tee@rhonema.com

Introduction

Oedema disease (ED) is an enterotoxemic disorder caused by specific pathotypes of *Escherichia coli* (*E. coli*) which affects primarily rapidly-growing nursery and weaned pigs. ED in pigs is associated with the colonization and adherence of the intestine cells by host-specific highly-adapted pathogenic *E. coli* strains with F18 fimbriae. The fimbriae are encoded by *fedab*⁺ and *fedac*⁺ genes predominantly found in ED strains of *E. coli* and diarrhoea-causing enterotoxigenic *E. coli* (ETEC) respectively (3). ED strains in addition, possess *stx2e* genes located in plasmids that produces Shiga toxins (1, 2). On absorption of the Shiga toxins from the intestinal lumen into the blood vessels, vascular cell damage occurs leading to oedema in particular the sub-mucosa of the stomach and mesocolon, ataxia and sudden death.

Materials and Methods

A survey was conducted to identify Shiga-toxin producing *E. coli* (STEC) isolates in post-weaners in Malaysia from February to April 2013. Faecal and organ samples were collected from 19 farms with about 500 standing sow population in 6 states in Malaysia namely Penang, Perak, Selangor, Melaka, Johor and Sarawak. The targeted animals were post-weaners (3-14 days post weaning), where there is sudden death in healthy pigs with swollen eyelids and/or with ataxia. Samples such as mesenteric lymph nodes, tonsils, jejunum with content, spleen, kidney and brain stem were tested for bacterial isolation and nucleic acid extraction then followed by multiplex PCR. Samples were cultured on MacConkey and on sheep blood agar. A total of 377 representative *E. coli* strains on blood agar were purified, determined for haemolysis and identified. PCR was conducted on the haemolytic *E. coli* for the presence of STEC (*stx2e* (2)). Positive Shiga-toxin *E. coli* strains were then analyzed for the presence of the *fedab* and *fedac* genes (1).

Results

A total of 59 haemolytic *E. coli* isolated from 8 farms were subjected to a singleplex PCR-based method for *stx2e*⁺ and a multiplex PCR-based method for the detection of *fedab*⁺ and *fedac*⁺. 7 of the 8 farms with haemolytic *E. coli* were found to be positive for *stx2e* genes. A total of 5 farms were positive for *stx2e*, *fedab*⁺ and *fedac*⁺ genes (Table 1).

Table 1. Microbiological and PCR findings

Farm	Haemolytic <i>E. coli</i>	<i>stx2e</i>		<i>fedab</i>		<i>fedac</i>	
		+ve	-ve	+ve	-ve	+ve	-ve
A	0	0	0	0	0	0	0
B	8	1	0	1	0	1	1
C	0	0	0	0	0	0	0
D	0	0	0	0	0	0	0
E	5	4	2	0	2	0	0
F	2	2	2	0	2	0	0
G	0	0	0	0	0	0	0
H	0	0	0	0	0	0	0
I	0	0	0	0	0	0	0
J	0	0	0	0	0	0	0
K	9	3	1	2	1	2	2
L	0	0	0	0	0	0	0
M	26	9	7	2	7	2	2
N	4	3	0	0	0	2	2
O	0	0	0	0	0	0	0
P	2	0	0	0	0	0	0
Q	3	3	1	0	1	0	0
R	0	0	0	0	0	0	0
S	0	0	0	0	0	0	0
Total	59	25	13	5	13	7	7

Discussion

This study demonstrated that Shiga-toxin producing *E. coli* (STEC) carrying *fedab*^{+/+} and *fedac*^{+/+} genes were detected from haemolytic *E. coli* associated with clinical findings of oedema disease (ED) in post-weaned pigs. This finding concludes that virulence genes were present in certain pig farms in Malaysia which leads to ED.

References

- [1] Barth S et al. 2011. J Vet Diagn Invest 23, 454-464.
- [2] Osek J. 2003. J of App Microb 95, 1217-1225.
- [3] Imberechts et al. 1992. Vet Microbiol 31, 221-233.