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Investigation of the source of haemolytic *Escherichia coli* infecting weaned pigs

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SUMMARY

Attempts were made to discover the source of strains of haemolytic *Escherichia coli* infecting weaned pigs on a piggery. The organisms were not detected in the faeces of sows in the farrowing house, or in the in the faeces or intestinal tracts of slaughtered bacon pigs or sows. Sows held in a quarantine unit, and their offspring born in the unit, did not excrete haemolytic *E. coli* until after they were returned to the piggery.

The environment of the piggery was the most likely source of infection for weaned pigs, and routine cleaning and disinfection of the accommodation did not prevent infection. Unweaned pigs were however able to transfer haemolytic *E. coli* to a newly built, previously unused weaning house, and establish a cycle of infection.

INTRODUCTION

Post-weaning diarrhoea (PWD) is a common condition which occurs in pigs 3–10 days after weaning (Blood, Radostits & Henderson, 1983) and is thought to result from the action of haemolytic *Escherichia coli* which proliferate in the small intestines after weaning (Richards & Fraser, 1961; Kenworthy & Crabb, 1963). Only a limited number of serotypes of *E. coli*, which are either enterotoxigenic (Smith, 1976) or produce vero cell toxin (Hampson *et al.* 1986), have been associated with outbreaks of PWD (Dam & Knox, 1974). Such serotypes may also be found in the faeces of healthy weaned animals (Campbell, 1959; Svendsen, Larsen & Bille, 1974), and have been isolated from some cases of diarrhoea in newly born piglets. Strains of *E. coli* affecting neonatal pigs do, however, differ from PWD strains in that they usually possess the K88 adhesin (Svendsen, 1974).

The source of the haemolytic *E. coli* which infect weaned pigs has received little investigation. Infection may presumably be acquired from the contaminated environment of the weaning house, or else small numbers of the microorganisms may be carried undetected in the intestinal tract of unweaned pigs and these then proliferate selectively there after weaning. Evidence for the latter suggestion has been provided by Miller *et al.* (1984), who challenged 5-day-old piglets with a naladixic acid-resistant strain of haemolytic *E. coli* which was not detected in the faeces until it appeared suddenly after weaning. Unweaned piglets would them-

selves have to acquire infection with the appropriate serotypes either directly from the farrowing house environment, or from the sows. In studies of serotypes of haemolytic *E. coli* which were infecting piglets and causing diarrhoea in the first few days of life, both Arbuckle (1968) and Shreeve & Thomlinson (1971) noted that sows often excreted these organisms in faeces in the first few days after they were moved to the farrowing house. In both these studies it remained unclear whether this excretion was the result of sows becoming infected from the contaminated environment of the farrowing house, or of a sudden proliferation of a small population of these microorganisms which formed part of the sow's intestinal flora.

The present investigation was undertaken to examine these possibilities. Previous work demonstrated that haemolytic *E. coli* of two O-serotypes were involved in PWD in this piggery (Hampson *et al.* 1986), these being an O-138 type producing a heat-labile enterotoxin and an O-139 type producing a Shiga-like vero cell toxin. The O-138 isolates had urease activity but did not produce indole, whilst the O-139 isolates were urease-negative and were either negative or weakly positive for indole production. Since these two biochemical reactions gave 100% correlation with the two O-types on the piggery (Hampson *et al.*, submitted for publication), the two tests were used in the present study as a method for typing the PWD strains of haemolytic *E. coli*.

MATERIALS AND METHODS

Animals

The pigs used were Landrace × Large White sows and their progeny, obtained from the Pig Research Centre, Massey University.

Experiment 1

Eight piglets from each of two litters had rectal swabs taken daily from 2 days before weaning at 5 weeks of age until 10 days after weaning. Four animals from each litter were moved after weaning to conventional flat-deck weaner pens which had been in use for many years. This accommodation was routinely pressure-hosed, scrubbed and disinfected with a commercial potassium hydroxide solution between batches of pigs. The other four piglets from each litter were accommodated in similar pens situated in a newly built weaner house which had not previously housed pigs. Access to the latter pigs for feeding and sampling was restricted to one individual who had no other contact with pigs, and who wore clean overalls and boots which were kept in the new weaner house.

Experiment 2

At various intervals over a 2-year period, rectal swabs were collected from 17 sows and gilts on the piggery, daily from 2–3 days before until 2–3 days after farrowing.

Experiment 3

Swabs of rectal and ileal contents were taken from 32 randomly selected bacon-weight pigs (22–26 weeks old) and 5 sows, whilst they were being processed at a local abattoir.

Experiment 4

Two pregnant sows were thoroughly washed, bathed in a proprietary chlorhexidine/cetrimide disinfectant solution (Savlon, ICI) and transported to a poultry quarantine house approximately 3 km from the piggery. They were again washed, bathed in the disinfectant solution and then housed in separate pens. The unit had not previously housed pigs and had been cleaned and disinfected with formaldehyde gas before use. Staff who had no other contact with pigs cared for the sows. Rectal swabs were taken from the sows daily, and from their 20 offspring. The piglets were weaned at 5 weeks of age, and eight were transferred directly to a cleaned and rested room in the weaner house of the piggery, whilst the remainder were kept in a single pen in the quarantine unit. The latter pigs were transferred to a separate but similar room in the piggery when 7 weeks of age.

Bacteriology

Fresh swabs were plated on to split-layer agar plates containing 5% sheep red blood cells in the upper layer. After overnight incubation in air at 37 °C, the proportion of haemolytic coliform colonies was estimated. Selected isolates were identified as *E. coli* using the API 20E system, and were tested for urease production on Christensen's urea media, and indole production in peptone water after addition of Kovacs' reagent (Cowan, 1974).

RESULTS

Experiment 1

None of the 16 pigs had detectable haemolytic *E. coli* in their faeces before weaning, but 5 from those weaned to the old weaner house and 4 weaned to the new weaner house excreted haemolytic *E. coli* in the week after weaning. Representative isolates were urease-positive and indole-negative, i.e. probably O-138 type. One piglet from each group developed a mild diarrhoea.

Experiment 2

Occasional haemolytic colonies of *E. coli* were recovered from the faeces of sows and gilts examined in the farrowing house, but they were not of the two main types involved in PWD on this piggery.

Experiment 3

Small numbers of haemolytic *E. coli* were recovered from the faeces and intestinal contents of approximately 20% of the bacon-weight pigs. None of the isolates tested had urease and indole reactions typical of the two strains of haemolytic *E. coli* known to regularly proliferate in weaned pigs from this herd.

Experiment 4

Haemolytic *E. coli* were not found in the faeces of either of the sows or their piglets whilst they remained in the poultry quarantine unit. PDW was not observed in piglets in the quarantine unit. All the piglets which were weaned to the cleaned room on the piggery excreted haemolytic *E. coli* of the O-138 type at some

point in the week after weaning, and four developed post-weaning diarrhoea. After the remaining piglets were moved from the quarantine unit at 7 weeks of age, they all excreted haemolytic *E. coli* of the O-138 type, and three developed diarrhoea.

DISCUSSION

Piglets weaned either to a regular weaner facility, or to a newly constructed unused weaner house, first excreted haemolytic *E. coli* in their faeces after they arrived in the houses. Assuming that the environment of the new house was not accidentally contaminated with haemolytic *E. coli*, the results suggest that the pigs themselves carried the organisms into the weaner house. Further evidence for this was obtained in a previous study on this piggery, when four unweaned pigs were found to carry haemolytic *E. coli* of the O-138 type in their intestinal contents, with these microorganisms not being detected in their faeces (Hampson, Fu & Smith, submitted for publication).

Assuming that the unweaned pigs did carry infection to the weaning house, they themselves must either have been infected from the sow, or from the environment of the farrowing house. Studies by Arbuckle (1968) and Shreeve & Thomlinson (1971) indicated that sows may excrete haemolytic *E. coli*, of serotypes which affect young pigs, at the time that they are moved to the farrowing house. Shedding of haemolytic *E. coli* was not, however, demonstrated in another study where 12 gilts were monitored up to farrowing (Callear & Smith, 1966), and sows did not shed PWD strains in the present study. To confirm this observation, and because PWD strains had previously been detected in the intestinal contents but not the faeces of unweaned pigs, ileal contents and faeces were examined from slaughtered bacon pigs and a few sows. Again no PWD strain was found, suggesting that on this piggery it was not usual for adult animals to carry detectable numbers of these bacteria. This conclusion was strengthened by the observation that piglets born in the quarantine unit only excreted haemolytic *E. coli* after they were transferred to the weaner house in the piggery, although ideally this experiment should be repeated with more sows and their offspring to determine whether adult carriers do exist at a lower frequency.

It may be concluded that the environment of the weaner house appeared to be the most likely source of haemolytic *E. coli* infecting weaned pigs. Even where the weaning house was not contaminated, however, unweaned pigs could acquire infection in the farrowing house, presumably from previous environmental contamination, and carry it undetected into the weaning house. Routine cleaning and disinfection of the piggery under investigation was insufficient to break the cycle of infection.

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