

Improvements in processing hygiene indicator and microbial hazard levels in Australian finisher pigs from 1996 to 2010

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Abstract

In 2010 a national pig carcass baseline survey was undertaken to help determine the impact of regulation on processing hygiene and hazard levels and to inform national standard setting. The results are compared to those from a 1996 baseline survey of Australian finisher pigs. For *E. coli* a reduction in prevalence of around 10% (29.3% in 1996 to 20.7% in 2010) and an average reduction in concentration of 1.5 log₁₀ cfu/cm² were observed. Similarly, average concentration of TVC reduced by 1.9 log₁₀ cfu/cm² over the same period. Despite more stringent sampling techniques utilised in the 2010 survey, the carcass hygiene indicator data indicates a substantial improvement in dressing hygiene compared to the 1996 survey. *Salmonella* levels remain low (0.4%; 95% CI: 0.0 – 2.1%) by international standards. This substantial improvement in dressing hygiene over the past 14 years is most likely a result of the mandating of HACCP in Australian abattoirs in the late 1990s that has in part driven investment in processing effectiveness.

Introduction

In 1996/97 a national microbiological baseline survey of 680 Australian pig carcasses was conducted. This involved swab sampling pig carcasses on three x 20 cm² areas (Coates et al 1997). The study found low levels of *Salmonella* and *Y. enterocolitica* (1% and 0.15% respectively), no detections of *C. jejuni*, *L. monocytogenes* and *E. coli* O157 and relatively high levels of *S. aureus* (14.9% with a carcass toxigenic strain prevalence of 7%).

Since that time, international guidelines have been developed for determining carcass microbiological levels, the so-called ESAM (*E. coli* and *Salmonella* Monitoring) system (Anon 2000). Given that the original survey was conducted 14 years prior, and utilised a different methodology, the results could no longer be relied upon to demonstrate the quality of Australian pig meat in the event of a food borne disease outbreak or satisfy new and existing export market requirements.

Materials and Methods

A total of 294 finisher pig carcasses were sampled by swabbing at 6 major pig abattoirs in Australia. Carcasses were sampled on 2 occasions approximately 6 months apart (March/April and August/September) using the standard ESAM sponge method and a single sampler. The number of samples allocated to each abattoir was in proportion to that abattoir's throughput. Overall, the 6 abattoirs represented approximately 60% of the national finisher pig kill.

For each abattoir a similar number of their available ESAM *Salmonella* test results, which straddled both sampling periods, were assessed. If there was sufficient ESAM data around each survey sampling date, half the ESAM sample results were taken prior to each survey date and half after. Otherwise, for the first survey sampling period more ESAM data was taken prior to the date and for the second survey sampling period more was taken after the sampling date (to avoid crossover of ESAM data).

Carcasses were aseptically sampled after a minimum of 12 hours chilling in accordance with AQIS Meat Notice 2003/6 (Anon 2003), using the sponge method and stainless steel 100 cm² template. Swabs were placed immediately into an esky with ice packs and shipped overnight to the laboratory for testing within 24 hours.

Total Viable Count (TVC) and *E. coli*

The sponge swabs were massaged in a stomacher for 60 seconds. Ten-fold serial dilutions of the swab diluent were

prepared in Peptone Saline Solution (Oxoid, Thebarton, South Australia). Aliquots (1 mL) from each serial dilution were inoculated onto 3M™ Petrifilm™ Aerobic Plate Count Plates and 3M™ Petrifilm™ E. coli/Coliform Count Plates (3M Corporation, St Paul, Minnesota) and incubated at 35°C for 48 hours for the aerobic count and 35°C for 24 hours for E. coli. Colonies were identified and counted as per manufacturer's instructions with the limit of detection being 10 cfu/mL.

E. coli O157:H7

After removing aliquots of the diluent for Total Aerobic and E coli counts, carcass swabs were tested for the presence of E coli O157:H7 using the BIOCONTROL VIP® Gold for EHEC single step immunoassay (BIOCONTROL). 50 mL of pre-warmed modified Tryptone Soya Broth was prepared as per the manufacturer's instructions and added to the sponge swab. This was incubated for 18 to 28 h at 35 - 37°C. The culture (1 mL) was transferred to the sample addition well of a VIP Gold unit. This was incubated at room temperature for 10 min and examined as per the manufacturer's instructions. Provisionally positive isolates were sent to the Melbourne Diagnostic Unit (MDU) for confirmation.

Results

The microbiological results for this survey are compared with the 1996 survey in Table 1. For log₁₀ TVC cfu/cm² there was a significant difference between abattoirs (p<0.001), but no consistent seasonal effect. For E. coli Abattoir 2 had only 1 positive result and the prevalence estimate was significantly lower than all other establishments (p=0.02). Excluding Abattoir 2, there were significant differences in the mean log₁₀ E. coli cfu/cm² between all abattoirs (p=0.02). For Salmonella, routine ESAM sampling for the 6 abattoirs for a similar number of samples over the survey period detected Salmonella in 1/266 (0.4%) samples (Table 1)

Table 1. Comparison of the finisher carcass microbiological survey results 1996 (Coates et al 1997) versus 2010

Year	# abattoirs ^a	# samples	Mean log ₁₀ TVC/cm ² (SD)	# E. coli pos (%)	Mean log ₁₀ E. coli/ cm ² (SD)	# Salmonella pos (%)
1996	18	680	3.8	(29.3)	0.71	(1)
2010	6	294	1.93 (0.7)	61 (20.8)	-0.79 (0.4)	1 (0.4) ^b

^a there has been significant rationalisation in the pig processing industry since 1996

^b from ESAM records for corresponding time period. Serovar = *S. infantis*. SD = standard deviation

None of the 294 samples in 2010 were positive for E. coli O157:H7 (prevalence < 1.3%). Five samples were identified as suspect following screening with BIOCONTROL VIP®, however on confirmation were determined to be negative.

Discussion

Comparisons with other studies, both in Australia and overseas are difficult as they often use variable methodologies (eg sponge vs. swabs; pre- vs. post-chill) and swabbed areas (eg ESAM 300cm² vs. EU 400 cm²) and target different organisms (eg. ESAM E. coli vs. EU Enterobacteriaceae). Increasing the swabbed area can be expected to increase the detection (prevalence) of an organism, and counter intuitively with high counts it can decrease the estimate of cfu/cm² (Miraglia et al, 2005).

In 1996 the Australian pork industry funded a microbiological benchmarking study of 680 finisher carcasses at 18 Australian abattoirs as part of a Pig Meat Hygiene Program. Care must be exercised, however, in comparing the results of that first National Pig Carcass Microbiology Survey (Coates et al 1997) with this present study. At the time (pre-USA Megaregs) there was no internationally accepted carcass swabbing protocol so there were a number of differences methodologies between the 2 surveys, vis à vis

- The previous study utilised a wet-dry swab method (Kitchel et al 1973) on 3 x 20cm² areas. This contrasts with the present study using the internationally accepted ESAM sponge method on 3 x 100cm² areas (Anon 2000).
- Carcasses were sampled 30 minutes after the final wash (rather than after 12 hours chilling).
- The previous study relied on company employees (compared with a single survey sampler).
- The previous study sampled randomly over a 12 month period (compared with just 2 intensive sampling periods 6 months apart).

Despite these caveats, overall, the present survey can be considered to have been carried out under more stringent conditions, so any improvements can be considered real. The differences in the results between the 2 studies are summarised in Table 1.

Total Viable Count (TVC)

A high TVC is an indication of poor general hygiene and/or poor temperature control. In this study the overall mean log₁₀ TVC was 1.93 cfu/cm² for finishers. There was no consistent sampling period effect between abattoirs, suggesting that temporal differences are simply a reflection of day-to-day processing variability.

The results compare favourably with those found in previous studies in Australia and overseas. Since 1996 there has been substantial improvement in dressing hygiene as indicated by a 1.9 reduction in the mean log₁₀ TVC (Table 1, Fig 1). In 1996 the worst 10% of finisher carcasses fell in the TVC range of 3.8 to 5.4 log₁₀ cfu/cm² (Coates et al 1997). In the present study only the worst 2/294 (0.7%) of carcasses were found in a similar range.

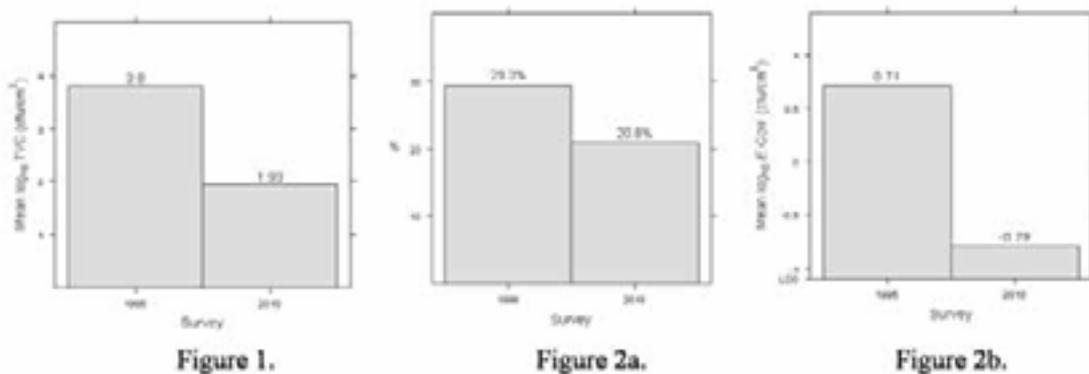


Figure 1. Comparison of mean log₁₀ TVC cfu.cm² between the 1996 (Coates et al 1997) and 2010 carcass benchmarking surveys. Figure 2a & 2b. Comparison of mean *E. coli* percent positive and mean log₁₀ *E. coli* cfu.cm² between the 1996 and 2010 carcass benchmarking surveys

E. coli

In the US, Australia and Asia, *E. coli* is tested for as an indication of faecal contamination and poor general hygiene. In this study the overall prevalence for finishers was 20.8%, and the mean log₁₀ cfu/cm² was low at -0.79 compared with 29.3% and 0.71 in 1996 (Table 1, Fig 2a, Fig 2b). A 13 month Swedish baseline survey of 541 pig carcasses at the 10 largest abattoirs in Sweden estimated an

E. coli prevalence of 57% with a mean log₁₀ of 0.05 (Lindblad et al 2007) and a Taiwanese 2003 national baseline survey of 1650 pigs from 39 abattoirs reported a prevalence of 87.5% (Yeh et al, 2005).

E. coli O157:H7

E. coli O157:H7 was not detected in this study. Recently, a New Zealand study isolated *E. coli* O157:H7 both from 1% of 100 NZ domestic pig carcasses and from 3.1% of 65 imported Australian pig meat samples (Wong et al 2009). The sample numbers in this study, however, would have been able to detect this organism at lower prevalence than the NZ study and hence previous NZ results are not supported by the results of this national survey.

Salmonella

A total of 266 Salmonella ESAM test results from the survey abattoirs were examined covering the survey period, of which only 1 was positive giving a prevalence of 0.4%. This reflects the 1996 survey in which Salmonella was isolated from 1% of 680 carcasses (Coates et al 1997) and a 7 year summation of the pig carcass National ESAM database from 2000 to 2006 which reported an overall Salmonella carcass prevalence of 1.88% (Hamilton et al 2007) that continues to decline significantly to 2010 (A Kiermeier pers comm.).

In the late 1990s HACCP was mandated in pig abattoirs in Australia (Anon 1996/38). Subsequent investment to improve processing effectiveness and efficiency are inextricably linked with these dramatically enhanced regulatory requirements.

Conclusion

- There have been substantial improvements in the indicators of carcass hygiene between 1996 and 2010 as measured by TVC and E. coli prevalence.
- The relatively good Salmonella status of Australian finisher carcasses was confirmed.
- E. coli O157:H7 contamination does not appear to be an issue with Australian pig carcasses and the findings do not support the 3.1% prevalence reported by NZ.

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