



UNDER THE MICROSCOPE

The Sources of E. coli Contamination of Ground Beef in a Commercial Beef Processing Plant

Mueen Aslam, Gordon G. Greer, Frances Nattress and Colin Gill, Agriculture and Agri-Food Canada, Lacombe Research Centre, Lacombe, Alberta T4L 1W1
Phone: 403-782-8106 email: aslamm@agr.gc.ca

Introduction

Quantitative data obtained by cultural methods have limited value in determining the sources of contamination in beef processing plants. However, molecular typing of strains can determine the distribution of *E. coli* among various processing locations by tracking genetically related strains. A study using a molecular typing approach had suggested that *E. coli* O157:H7 isolated from faecal and hide samples were genetically similar to those found on carcasses after slaughter in a beef processing plant (Barkocy-Gallagher et al., 2002). However, the presence of low numbers of pathogenic strains

makes it difficult to determine the sources of these contaminants. There is also a lack of data to support a direct link between *E. coli* strains associated with animals and those found on carcasses at various processing steps and in commercial ground beef.

Objective of study

To determine the distribution and sources of generic *E. coli* during commercial beef processing, using random amplification of polymorphic DNA (RAPD).

Materials and Methods

During summer (2001) and winter (2002), swab samples were obtained from hides (H), washed carcasses (WC), conveyers used for moving portions of carcasses (CE), beef trimmings (BT), and 25 g of ground beef (GB) samples were obtained at a commercial beef processing facility. Generic *E. coli* were isolated using hydrophobic grid membrane filtration with direct plating on SD-39 agar (Entis and Lerner, 1997).

The RAPD technique was used to analyze *E. coli* isolates from each of the 5 sample locations. Pulsed-field gel electrophoresis (PFGE) was used to confirm the results obtained with RAPD.

Results and Discussion

Table 1 shows the percent of *E. coli* isolates that were shared among various sample locations. These findings agree with the results of previous studies conducted in pig processing plants (Bouvet, et al., 2002; Warriner et al., 2002) which used molecular typing methods to identify genetically related strains of *E. coli* throughout the pork processing line. It was suggested that *E. coli* contamination can be transferred among carcasses, meat and equipment. In the present study the majority of *E. coli* isolated from the hides (81% isolates), washed carcasses (83% isolates), and conveyers (60% isolates) were genetically related to those isolated from ground beef indicating that ground beef contamination in a processing plant may come from several sources. However, it is not clear whether identical strains are carried through the process on product, or whether they represent environmental contaminants.

There were some *E. coli* strains (11% and 21% for summer and winter samples, respectively) that were found only during various beef processing stages and in ground beef samples and were distinct from those found on the hides. This suggests that hides of incoming animals might not directly contribute *E. coli* to ground beef as no genetically related *E. coli* was recovered from only ground beef and hides. However, an indirect role of the hide in spreading *E. coli* throughout the plant seems likely, as most genetically samples from the conveyers were obtained from drive mechanisms, rollers and idlers after routine cleaning and before related *E. coli* recovered from the hides were also found in all other sample locations. Therefore the processing environment, in part, may be responsible for ground beef contamination.

In this study a few *E. coli* were recovered from various sample locations which did not share a close genetic relatedness and therefore were termed as unique. These unique *E. coli* were recovered from the hide samples as well as from the other sample locations in the beef processing environment (Fig.1) indicating that the *E. coli* populations from these sources were diverse in nature. In a previous study (Aslam et al., 2003) it was reported that hides and carcasses of animals could harbour *E. coli* genotypes which are not found in their fae-

ces. The sources of unique *E. coli* types in the beef processing plant deserve further investigation.

PFGE is used as a gold standard in epidemiological investigations to determine if disease-causing strains match those isolated from suspected sources. The rationale for using PFGE in the present study was to establish the validity of RAPD typing. Fig.2 shows RAPD and PFGE patterns of *E. coli* which were found to be genetically different by both methods. A good correlation between the results obtained with both methods confirmed that the RAPD method was a valid approach to differentiating the isolates recovered in this study. The majority of isolates were differentiated into corresponding genetic types by both RAPD and PFGE, which is consistent with the findings of a previous study (Khan et al. 2002). Although comparable results can be obtained with both methods, PFGE was more discriminatory.

Conclusions

This study has demonstrated that *E. coli* found on the hides of incoming cattle as well as in the beef packing environment are responsible for ground beef contamination. It was found that the conveyers were also an important source of meat contamination and there is considerable opportunity for cross-contamination among various locations in the beef packing plant. Molecular typing methods are valuable tools for identifying the sources of contamination during beef processing and packing and a combination of RAPD, used as a screening tool, followed by more detailed analysis using PFGE would be a good approach. The findings of this study stress the importance of improvements to processing hygiene post-carcass chilling to minimize the contamination of ground beef.

References

- Aslam, M., Nattress, F., Greer, G., Yost, C., Gill, C. and McMullen, L. 2003. Origin of contamination and genetic diversity of *Escherichia coli* in beef cattle. *Applied and Environmental Microbiology* 69, 2794-2799.
- Barkocy-Gallagher, G.A., Arthur, T.M., Siragusa, G.R., Keen, J.E., Elder, R.O., Laegreid, W.W. and

CANADIAN MEAT SCIENCE ASSOCIATION

Koohmaraie, M. 2001. Genotypic analysis of *Escherichia coli* O157:H7 and O157 nonmotile isolates recovered from beef cattle and carcasses at processing plants in Midwestern states of the United States. *Applied and Environmental Microbiology* 67, 3810-3818.

Bouvet, J., Montet, M.P., Rossel, R., Le Roux, A., Bavai, C., Ray-Gueniot, S., Mazuy, C., Atrache, V. and Vernozy-Rozand, C. 2002. Effects of slaughter processes on pig carcass contamination by verotoxin-producing *Escherichia coli* and *E. coli* O157:H7. *International Journal of Food Microbiology* 77, 99-108.

Entis, P. and Lerner, I. 1997. 24-hour presumptive enumeration of *Escherichia coli* O157:H7 in foods

by using the ISO-GRID^a method with SD-39 agar. *Journal of Food Protection* 60, 883-890.

Khan, A., Das, S.C., Ramamurthy, T., Sikdar, A., Khanam, J., Yamasaki, S., Takeda, Y., and Nair, G. B. (2002) Antibiotic resistance, virulence gene, and molecular profiles of Shiga toxin-producing *Escherichia coli* isolates from diverse sources in Calcutta, India. *Journal of Clinical Microbiology* 40, 2009-2015.

Warriner, K., Aldsworth, T.G. Kaur, S. and Dodd, C. E.R. 2002. Cross-contamination of carcasses and equipment during pork processing. *Journal of Applied Microbiology* 93, 169-177.

Table 1. Percentage of *E. coli* isolates that were shared between two sample locations.

Comparison		% Shared isolates	
Source A	Source B	Summer	Winter
Ground beef	Hide	81	43
	Washed carcass	83	66
	Beef trimmings	86	58
	Conveyers*	60	38
Hide	Washed carcass	81	46
	Beef trimmings	78	46
	Conveyers*	52	40
Washed carcass	Beef trimmings	82	46
	Conveyers*	83	23
Beef trimmings	Conveyers*	62	42

*Conveyers = Samples from the conveyers were obtained from drive mechanisms, rollers and idlers after routine cleaning and before the start of processing.

CANADIAN MEAT SCIENCE ASSOCIATION

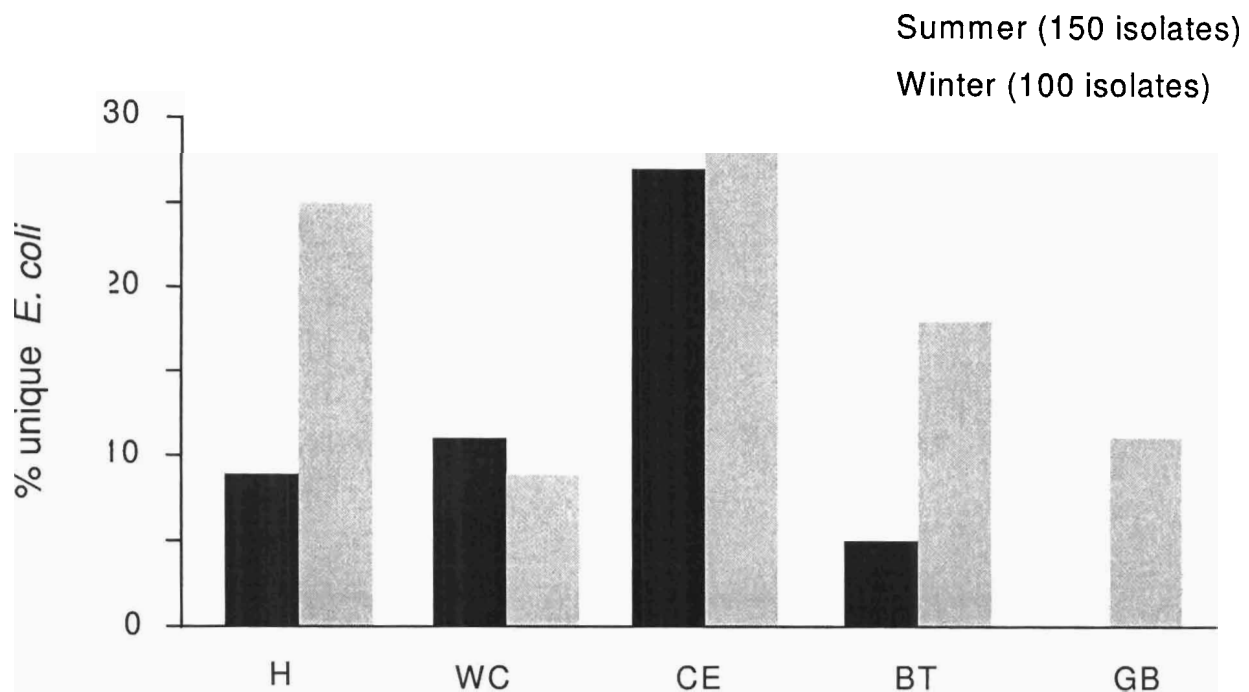


Figure 1. Percentage of unique *E. coli* isolates that were recovered from a commercial beef processing plant during summer and winter. H= Hide, WC= Washed carcass, BT= Beef trimmings, GB= Ground beef, CE= Conveyers

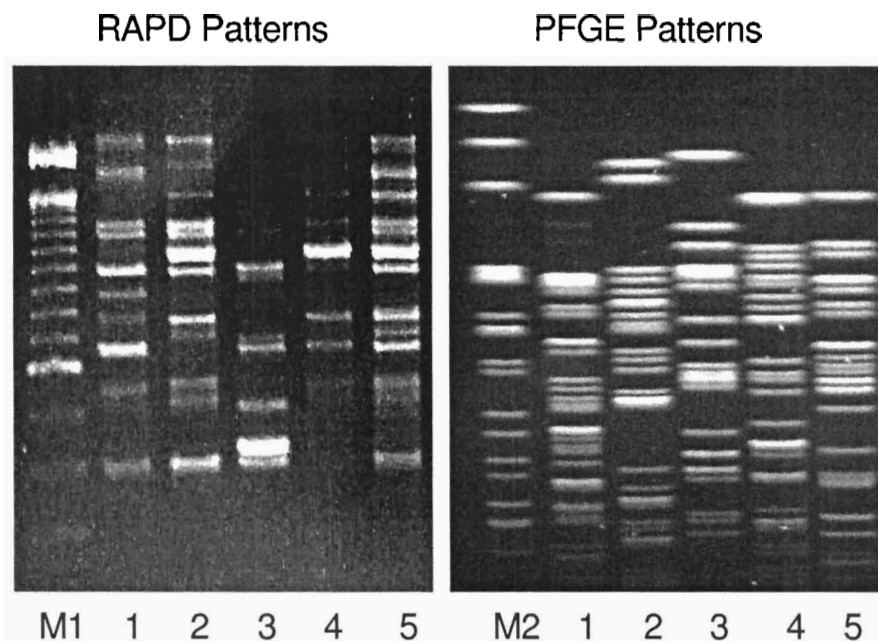


Figure 2. RAPD and PFGE patterns of *E. coli* isolates (lanes 1-5) recovered from a commercial beef processing plant. M1 = 100-base pair DNA ladder. M2 = *Salmonella Newport* as marker.