# A Novel Technique for *E. coli* Testing of Beef and Pork Carcasses

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## **Summary and Implications**

In January 1997, the USDA-FSIS mandated Escherichia coli (EC) testing in all processing plants to monitor surface contamination of beef, pork, and poultry carcasses. A novel technique that uses membrane filtration (MF) and m-ColiBlue24 (mCB) to determine the amount of EC contamination on a carcass was developed. Sponges from pork carcasses and excision samples from beef carcasses were analyzed using mCB and compared with standard methods. Samples from 77 pork carcasses were plated both on mCB and pour plating on Violet Red Bile (VRB) agar to obtain Total Coliform (TC) counts. There was no significant difference between the counts. The mean values for mCB and VRB agar were 7.4 colony forming units (CFU)/15 cm<sup>2</sup> and 6.1 CFU/15 cm<sup>2</sup>, respectively. The paired t-test revealed a t=0.52 and P=0.61. Excision samples from 57 spiked beef carcasses were used to compare mCB with both TC and EC Petrifilm<sup>TM</sup>. The mean TC count on mCB was  $1.6 \times 10^4$  CFU/cm<sup>2</sup> and  $9.3 \times 10^3$  CFU/cm<sup>2</sup> on TC Petrifilm. The paired *t*-test gave a t=2.4 and t=2.4count on mCB was 9.3 x 10<sup>3</sup> CFU/cm<sup>2</sup> and 3.2 x 10<sup>3</sup> CFU/cm<sup>2</sup> on EC Petrifilm. The t=3.5 and P<0.01. The new technique that uses mCB detected more TC and EC than both types of Petrifilm. Furthermore, the t values suggest that there is a significant treatment effect.

### Introduction

Over the past 30 years there have been dramatic changes and improvements in methods for the determination of bacterial pathogens (1). Many of these changes were brought about to ensure the microbiological safety of foods (3). However, there continues to be an increase in the number of cases of foodborne disease associated with newly emerging and reemerging pathogens (1). In response to this increase, the USDA-FSIS mandated EC testing in all processing plants to monitor surface contamination of beef, pork, and poultry carcasses. This mandate, in conjunction with HACCP, was implemented to decrease pathogens of animal origin.

A novel technique has been developed to monitor EC contamination on carcasses through MF and mCB. mCB is a MF medium that simultaneously detects TC and EC

in a 24-hour period (2). Studies were conducted on carcasses to compare mCB to standard methods.

#### **Materials and Methods**

The samples were taken off pork carcasses at a local processing facility. The sampling was carried out according to the procedure described by the USDA-FSIS (5). The samples were stomached with 20 ml of buffer peptone water and 1 ml was pour plated on VRB (Difco) with overlay. The pour plating procedure was performed according to the Compendium of Methods for the Microbiological Examination of Foods (4). In addition, 1 ml of the stomached liquid was added to 10 ml of buffered peptone water and filtered through a 0.45-micron filter. The filter was placed on absorbent pad saturated in mCB (Hach, Loveland,CO) and incubated at 35°C for 24±4 hours.

A 30-cm<sup>2</sup> excision sample was taken from 57 different beef carcasses. Each of the samples was stomached with 100 ml of buffer peptone water and the appropriate dilutions were made. Then, 1 ml was placed on TC Petrifilm (3M), EC Petrifilm, and filtered as described above. The procedure was performed according to manufacturer's instructions. These samples were part of an in-plant study on interventions. Some of the samples were intentionally contaminated, resulting in artificially high populations.

### **Results and Discussion**

The mean value obtained from the 77 pork sponges was 7.4 and 6.1 CFU/15 cm<sup>2</sup> on mCB and VRB, respectively. The results of the paired *t*-test that compared TC from the pork samples on mCB and VRB are presented in Table 1. The value obtained from the paired *t*-test was 0.52 and the probability of a larger t (P >) was 0.61. These results suggest no treatment effect.

There were various levels of presumptive EC recovered from the pork samples by using mCB. Of 77 samples, 60 had undetectable levels of EC, seven had 1 CFU, five had 2 CFU, one had 6 CFU, three had 21 CFU, and one had 53 CFU. All of the counts were based on 15 cm<sup>2</sup>.

The mean value from the TC was 4.2 log 10 CFU/cm<sup>2</sup> on mCB and 4.0 log 10 CFU/cm<sup>2</sup> on TC Petrifilm. The paired *t*-test results from the TC and EC counts off the beef excision samples also are expressed in Table 1. When the TC from mCB was compared with TC Petrifilm, the paired *t*-test indicated a value of 2.4 and a P>0.02. The paired *t*-test and P>T gave values of 3.5 and <0.01 when the EC from mCB were compared with EC Petrifilm. The mean value of EC was 4.0 log 10 CFU/cm<sup>2</sup> on mCB and 3.5 log 10 CFU/cm<sup>2</sup> on EC Petrifilm. The paired *t*-test results from both of the comparisons suggest a treatment effect.

#### References

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Table 1. Statistical analysis (P>0.05) of the results from the pork sponges and the beef excision samples.

<u>Comparison</u>	<u>N</u>	Paired t-test*	<u>P&gt;t**</u>
mCB VS VRB	77	0.52	0.61
mCB VS TC Petrifilm	57	2.4	0.02
mCB VS EC Petrifilm	57	3.5	<0.01

<sup>\*</sup>A paired *t* test value under two suggests no significant difference.

<sup>\*\*</sup>Probability of a larger *t*. High probability suggests there is no treatment effect. Low probability suggests there is a treatment effect.