Ongoing Research and Outreach Efforts Targeted at Non-O157 STEC

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Introduction

Since the first *E. coli* O157:H7 outbreak in 1982, research efforts in the U.S. and in other countries have been devoted to increase our understanding of:

- its prevalence in cattle, beef, and other foods (e.g., fresh leafy vegetables)
- its infection to humans
- its pathogenic factors
- possible identifications of pre- and post-harvest control measures to reduce its prevalence in cattle and infection to humans

Introduction

Non-O157 STEC outbreaks started to emerge:

- Argentina (1982 – 1991)
  - 433 cases (ground beef)
  - O1, O2, O15, O25, O75, and O111
- Italy (1992)
  - 9 cases (ground beef)
  - O111:H^{-}
- Canada (1992)
  - 6 cases (raw milk)
  - O80:H^{-}, O91:H14, O103:H2, O119:H25, O132:H^{-}, and O146:H21

Introduction

Non-O157 STEC outbreaks started to emerge:

- U.S. (MT; 1994)
  - 4 cases (raw milk)
  - O104:H21
- Australia (1994 – 1995)
  - 161 cases (beef sausage)
  - O111:H7, O111:H–, O157:H–, and O160:HUT
- Germany (2000)
  - 6 cases (beef sausage)
  - O26:H11

Introduction

Pathogenic STEC produce one or more virulence factors:

- Shiga toxin 1 (Stx1)
- Shiga toxin 2 (Stx2)
- α-hemolysin (HlyA)
- EHEC-hemolysin (EHEC-HlyA)
- Intimin

These virulence factors are encoded by various genes:

- stx_1
- stx_2,
- *hly*A
- EHEC-*hly*A
- *eae*

Introduction

Cattle as a Reservoir of STEC

STEC strains are not host specific

STEC have been shown to be more prevalent in cattle than in other animals

STEC infection in humans has been traced, in most cases, to cattle, their products (especially beef), and vegetables or water contaminated with cattle feces

Non-O157 STEC prevalence in beef cattle: up to 70.1%

- STEC strains belonged to 341 serotypes
- ~ 36% of these serotypes are pathogenic

Non-O157 STEC prevalence in dairy cattle: up to 74.0%

- STEC strains belonged to 152 serotypes
- ~ 49% of these serotypes are pathogenic

Our Research

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Objectives

Main Objective

To identify on-farm factors that influence prevalence of O157 and non-O157 STEC in cattle

Specific Objectives

1) To assess prevalence, human health risks, and pre-harvest control measures of STEC in beef and dairy cattle in various production systems in *Nevada* and *California* over 1 year

2) To integrate the knowledge gained from achieving the first objective and from published reports on pre-harvest control measures into an education program on food safety with emphasis on developing pre-harvest control strategies to assure beef safety

Methods & Results

Nevada

- Small size operations (~ 100 cattle tested per ranch)
- Dairy heifers, beef heifers (pasture), beef heifers (range), and culled beef cows
- STEC prevalence rates ranged from 4.0 to 22.7%
- *Serotypes*

  O6:H49, O6:H–, O8:H–, O26:H–, O39:H–, O105:H–, O113:H–, O116:H–, O118:H–, O138:H–, O141:H–, O157:H7, and OUT:HUT

HUS

Other illnesses

Methods & Results

California

- Larger-scale operations ranging in size from 13,000 to 46,000 cattle for feedlots, from 38 to 1,300 cows on pasture, from 65 to 225 cows on the range, and an average herd size of 713 cows and heifers for dairy farms

- Prevalence rates ranged from 1.9 to 4.3% in feedlot cattle (*n = 642*), from 1.9 to 5.0% in cattle grazing irrigated pastures (*n = 638*), from 0.7 to 18.6% in those grazing rangeland forages (*n = 774*), and from 0.8 to 3.2% in dairy cattle (*n = 1,268*)

- *Serotypes*

  The STEC isolates from beef cattle in the feedlot, beef cattle on pasture, beef cattle on the range, and dairy cattle belonged to 14, 13, 35, and 16 serotypes, respectively

Results

California

Serotypes - Beef cattle in the feedlot

O86:H19, O114:H2, O125:H19, O127:H19, O136:H12, O136:H–, O153:H–, O157:H7, O165:H7, OUT:H5, OUT:H12, OUT:H20, OUT:H–, and OUT:HUT

Serotype - Beef cattle on pasture

O1:H2, O5:H16, O5:H–, O26:H8, O26:H11, O84:H–, O103:HUT, O111:H8, O125:H2, O125:H19, O137:H16, O157:H7, and O169:H19

Serotype - Beef cattle on the range

O1:H2, O5:H–, O26:H11, O39:H–, O84:H2, O84:H–, O86:H2, O96:H19, O111:H16, O111:H–, O116:H2, O116:H36, O125:H2, O125:H16, O125:H19, O125:H27, O125:H28, O125:H–, O127:H2, O127:H19, O127:H28, O128:H2, O128:H16, O128:H20, O146:H21, O157:H7, O158:H16, O158:H19, O158:H28, O166:H2, O166:H6, O166:H20, OUT:H2, OUT:H19, and OUT:H–

Serotypes - Dairy cattle

O15:H–, O116:H–, O125:H20, O127:H19, O128:H20, O136:H2, O136:H10, O136:H12, O136:H19, O136:HUT, O157:H7, O166:H6, OX13:H19, OX13:H20, OUT:H7, and OUT:H–

Results

California

Of the 161 STEC isolates:

27 O157

134 non-O157

83.2% non-O157 STEC

Pathogenicity of the non-O157 isolates

- All lethal to Vero cells
- 78 had and expressed only stx_1
- 16 had and expressed only stx_2
- 40 had stx_1 and stx_2
  - 3 expressed only stx_1
  - 2 expressed only stx_2
  - 35 express both stx_1 and stx_2
- 10 had and expressed *hly*A
- 84 had EHEC-*hly*A but only 56 expressed it
- 53 had *eae*

Results

Because STEC strains lacking the attaching and effacing gene or the hemolysin genes have been shown to cause human illnesses (Neill, 1997), it was suggested that these genes are not absolutely required for pathogenicity and each STEC strain should be considered a potential EHEC (Bürk et al., 2002).

Results

California

29 Serotypes – Not reported previously in cattle or their products

O86:H2, O86:H19, O114:H2, O116:H2, O116:H36, O125:H2, O125:H16, O125:H19, O125:H20, O125:H27, O125:H28, O125:H–, O127:H2, O127:H19, O127:H28, O128:H16, O128:H20, O136:H10, O136:H19, O137:H16, O158:H19, O158:H28, O166:H2, O166:H6, O166:H20, O169:H19, OX13:H19, OX13:H20, and OUT:H20

Results

Examples of the on-farm factors tested:

Season, water (e.g., source, location, and cleanliness), animal factors (e.g., sex, age, source, parity, stage of lactation, and health), pen size, body weight, shelter type, manure handling, dietary factors (e.g., diet composition, feed ingredients, bunk type, location, and cleanliness)

Factors with high potential to decrease STEC prevalence:

Dairy

Feeding soybean meal as the protein supplement

Feedlot

Maintaining heavier cattle, clean feed bunks, and increasing dietary forage from 10 to 15%

Results

Factors with high potential to decrease STEC prevalence:

Irrigated pasture

Offering running drinking water (streams or springs versus ponds or ditches) and shortening the calving season (≤ 2 months)

Range

Animal factors

Decreasing stock density (≤ 1 cow/acre), early separation of calves (≤ 6 mo), increasing the size of calving pasture (> 120 acres), and absence of diarrheic calves (2 to 4 mo) prior to fecal sampling

Dietary factor

Molasses supplementation to pregnant cows

Outreach

Our past and current efforts:

The prevalence and pre-harvest control data from our studies have been incorporated into:

1) outreach publications such as:

- The annual extension proceedings (Cattlemen's Update) published by the University of Nevada-Reno
- Other miscellaneous publications

2) Presentation to farmers, ranchers, farm advisors, and extension specialists by Dr. Atwill (Extension Veterinarian)

Our future efforts:

With new funding, we plan to establish a food safety website focusing on STEC to provide a continuously updated database on STEC prevalence in U.S. cattle, pathogenicity of the isolates, and pre- and post-harvest control measures with high potential to decrease cattle carriage and contamination of their edible products with these foodborne pathogens

Questions