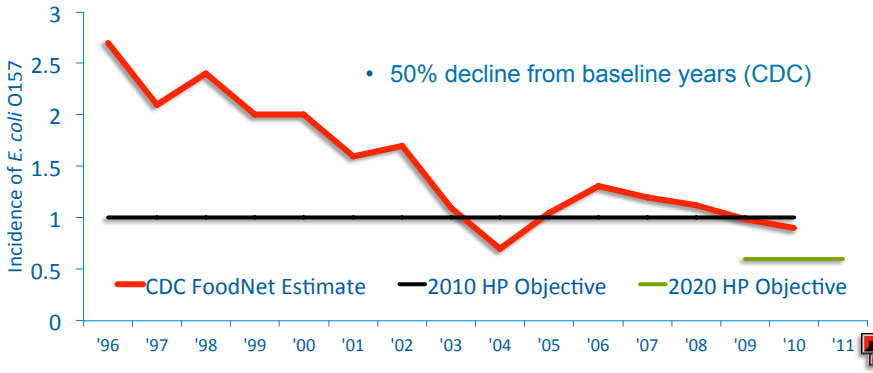
TEXAS TECH UNIVERSITY
Guy H. Loneragan, BVSc, PhD
International Center for Food Industry Excellence

Control of Food-borne Pathogens in Cattle: Challenges and Opportunities

Southwest Beef Symposium
Lubbock, Texas 16JAN2013

E. coli O157 and Beef

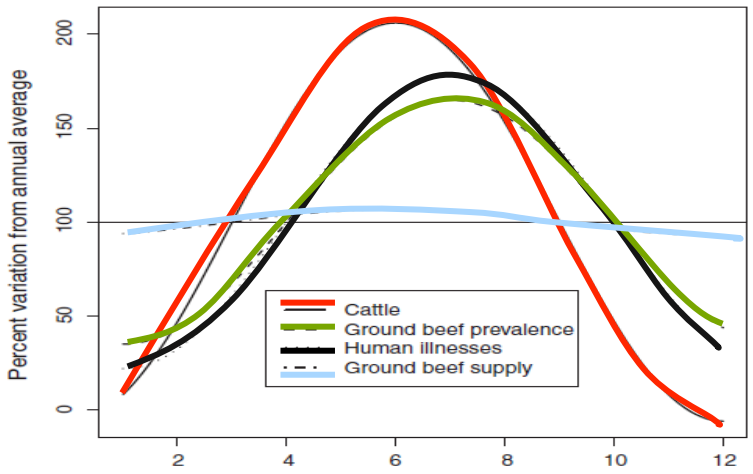
- Informed regulatory oversight and industry implementation of PR/HACCP plans have resulted in greatly improved microbial process control
- Observed across various metrics



E. coli O157 and Beef

- Many slaughter plants now excel at microbial process control
  - Diminishing opportunities for further improvement during slaughter/fabrication
  - Most cases now from non-beef sources
    - Cattle likely ultimate source of *E. coli* O157
- To move the needle further, need to start focusing on controls in live-animals
  - To what extent does pre-harvest control of pathogens further improve public health?

E. coli O157, Beef & Public Health



- Williams et al. *FPD* 2010

E. coli O157, Beef & Public Health

Pre-Harvest → Harvest/Processing → Consumers

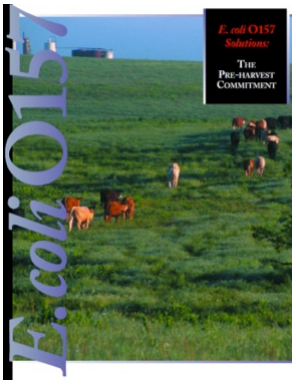
- Qualitative and logical relationship
  - Supported by empirical evidence
  - But hard to quantify impact
- More quantitative becoming available
  - Withee et al. *FPD* 2009 – streamlined model
  - Dodd et al. *JFP* 2011 –pre-harvest to harvest *E. coli* O157 (interventions efficacy)
  - Ebel et al. *JFP* 2004 – FSIS risk assessment

Best Practices

PROGRESSIVE BEEF

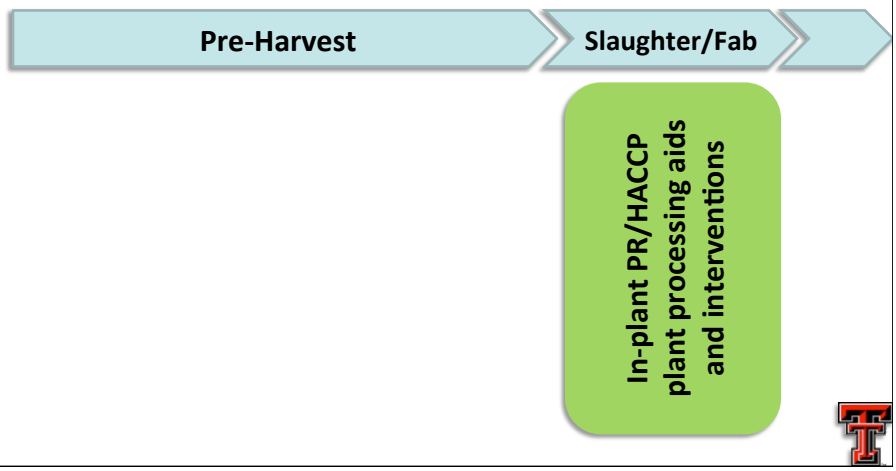
BMG
BEEFMARKETINGGROUP

- BIFSCo pre-harvest BP
  - *E. coli* Summit in 2003
    1. Clean feed
    2. Clean water
    3. Appropriate environment
    4. Relative freedom from pest
- Viewed as necessary foundation for specific interventions to be successful



Pre-harvest Interventions

- Majority of efficacy data about STEC O157
  - Increasing data on *Salmonella* and non-O157 STEC



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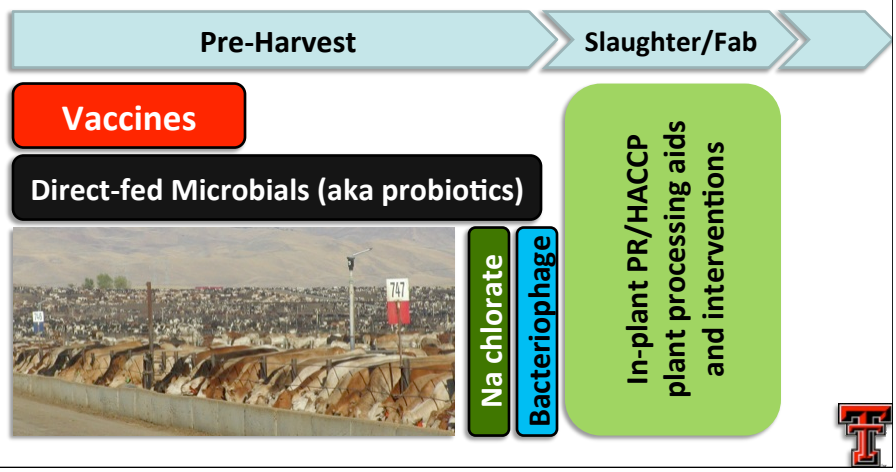


Vaccine Technologies

- Several vaccines trialed or proposed
  - Translation of 2 products relatively advanced

1. Epitopix/Pfizer Animal Health vaccine
   - Subunit vaccine of siderophore receptors and porin proteins (SPR)
     - Mechanism by which bacteria acquire iron
2. Bioniche Food Safety vaccine
   - Subunit vaccine based on Type III Secretion System (T3SS)
     - Includes several proteins required for enterocyte attachment and effacement

Vaccine Technologies

SRP vaccine (*Epitopix/Pfizer Animal Health*)

- Thomson et al. *FPD* 2009;6:871-7
  - 85% reduction in prevalence
  - 98% reduction in concentration
- 2010 commercial studies
  - 40% reduction in feces (2 doses)
    - Reduced number of combos of beef trimmings assoc. with positive test
  - 65% reduction on hides (1 dose)
- 2011 studies
  - 50 to 60% reduction in feces in completed study (Renter et al. and Loneragan et al.)
  - 75% reduction in 'high shedders'

Vaccine Technologies

T3SS vaccine (*Bioniche Food Safety*)

- Canadian regulatory agency has reviewed data and granted a full license
  - Label indication: 'For vaccination of healthy cattle as an aid in the reduction of shedding of *Escherichia coli* O157'
- Not yet licensed in the US
  - Might never be licensed…
- Peer-reviewed publications published in respected journals support efficacy

Vaccine Technologies

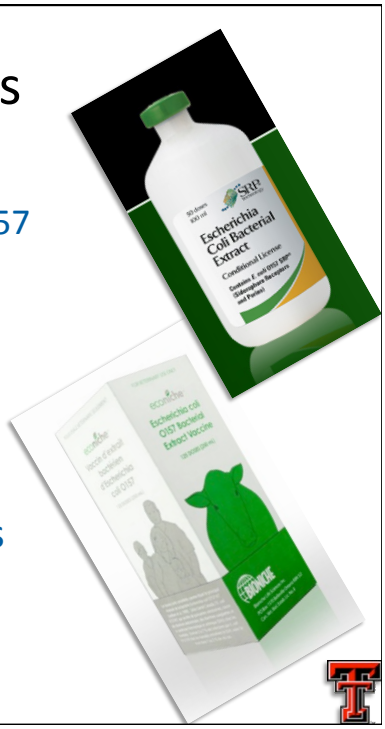
T3SS vaccine (*Bioniche Food Safety)*

Source of slide: David Smith, UNL

Reference	Regimen	Study Design		Outcome	OR	*P*-value
		Vaccination	Sampling			
Potter et al 2004	3 dose	Day 0, 21 and 42	Daily for 14 days post challenge	Feces	**0.35**	0.04
			6 samples: wks 0, 3, 6, 9, 12, 15	Feces	**0.36**	0.04
Peterson et al 2007	0 dose	n/a	7 samples: wks 0, 3, 6, 9, 12, 15 and 18	Feces	**0.36**	<0.01
	1 dose	Day 42			**0.25**	<0.01
	2 dose	Day 0 and 42			**0.27**	<0.01
	3 dose	Day 0, 21 and 42			**0.21**	<0.01
Peterson et al 2007	3 dose	Day 0, 21 and 42	5 samples: wks 0, 8, 10, 13, 14	Feces	**0.81**	0.57
				TRM	**0.01**	<0.01
Smith et al 2008	2 dose	14 – 104 days apart	4 samples: 3 wks apart	ROPES	**0.59**	<0.01
Smith et al 2009	2 dose	Day 0 and 32	3 samples: wks 11, 13, 16	Feces	**0.35**	<0.01
			4 samples: wks 11, 13, pre and post shipping	Hides	**0.43**	0.01
			Regional vaccination	TRM	**0.69**	0.63
	2 dose	Day 0 and 32	Comingling	Feces	**0.48**	0.01
				Hides	**0.67**	0.33
Smith et al 2009	3 dose	Day 0, 21 and 42	5 samples: wks 0, 9, 11, 13, 15	Feces	**0.5**	<0.01
Smith et al 2009	2 dose	Day 0 and 42	5 samples: wks 0, 9, 11, 13, 15	TRM	**0.07**	<0.01
Moxley et al 2009	2 dose	Day 0 and 42	5 samples: wks 0, 9, 11, 13, 15	Feces	**0.66**	0.20
	3 dose	Day 0, 21 and 42	5 samples: wks 0, 9, 11, 13, 15	Feces	**0.34**	<0.01
Allen et al 2011	3 dose	Day 0, 21 and 42	Daily for 14 days post challenge	Feces	**0.18**	<0.05

Vaccine Technologies

- Compelling body of evidence
  - Aid in the control of *E. coli* O157
- Efficacy is imperfect but nevertheless robust across a variety of study designs
  - Dose response observed
  - Snedeker et al *ZPH* 2011
- Can these imperfect vaccines have an impact?



Direct-Fed Microbials

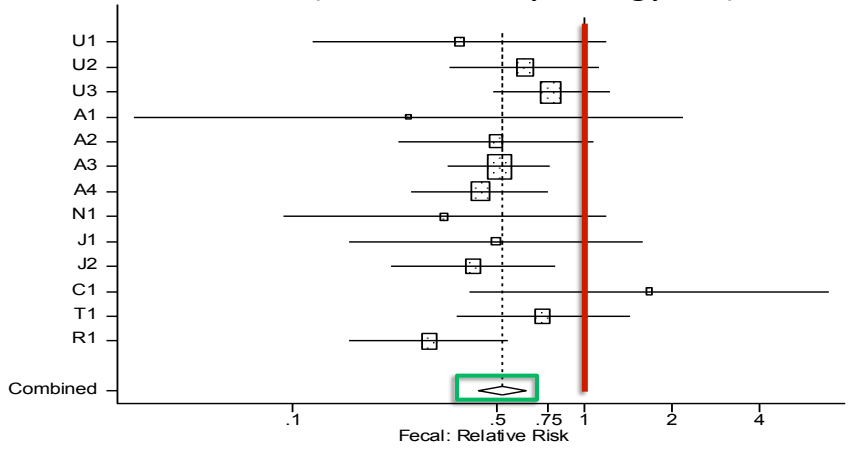
- Frequently referred to as probiotics
- GRAS (approval) for use in cattle
  - No label claim against food-borne pathogens
- Thoroughly evaluated against *E. coli* O157
  - More data for *Salmonella* and non-O157 STEC
  - Strain specific
  - Dose response
- Broadly adopted product
  - Nutrition Physiology Company

BOVAMINE®

NPC NUTRITION PHYSIOLOGY CO.,LLC

Direct-Fed Microbials

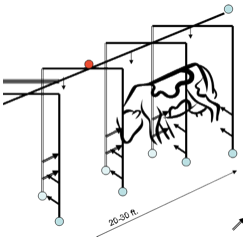
Strain NP51 (*Nutrition Physiology Co*)



Relative Risk	Efficacy	Lower 95% CL	Upper 95% CL	*P* value
Feces 0.53	47%	0.44	0.63	<0.01
Hides 0.60	40%	0.49	0.75	<0.01

Bacteriophage

- Biological control using targeted selection of lytic phages
  - Product available from Elanco Food Solutions (*Finalyse*)
- Preliminary field data on STEC O157 encouraging
  - Week-on/week-off study
  - Trim positives reduced 56% (*P*=0.06)
- Expanding cocktail of phages to cover non-O157 STEC



Finalyse
Elanco

Source of Data: Patrick Mies

Efficacy of Pre-harvest Interventions

- A variety of interventions have shown consistent efficacy
- Imperfect (<100%) efficacy – none will be a silver bullet
  - Efficacy nevertheless robust across study settings/designs
- Can adoption of these imperfect intervention(s) have a favorable impact?

Quantitative Risk Assessment

Scott Hurd Farm-to-Fork Model

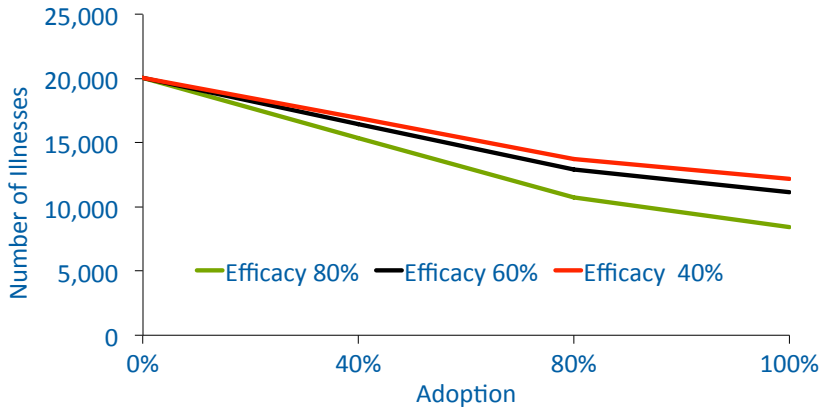
- Quantitative risk assessment
  - Farm to fork with various measures of ***impact***
    - Pre-publication

Scenario	Reduction (prevalence)	Reduction (log_{10} cfu/g)
A	40	0.3
B	60	0.3
C	80	1.0

Quantitative Risk Assessment

Scott Hurd Farm-to-Fork Model

Number of STEC O157:H7 Illnesses Due to Consumption of Ground Beef

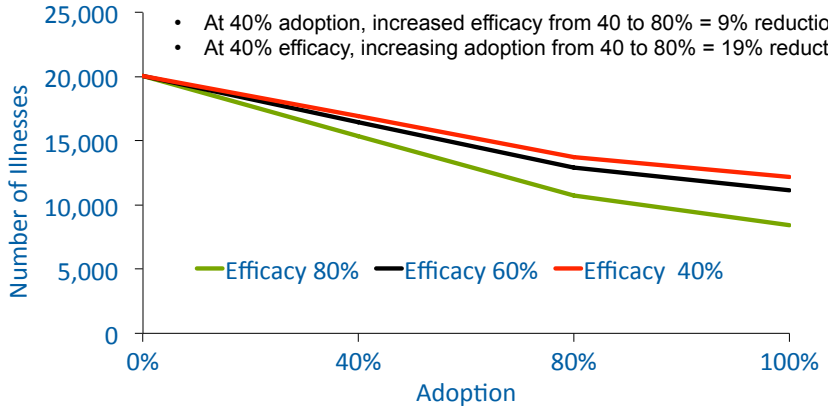


Quantitative Risk Assessment

Scott Hurd Farm-to-Fork Model

Number of STEC O157:H7 Illnesses Due to Consumption of Ground Beef

- At 40% adoption, increased efficacy from 40 to 80% = 9% reduction
- At 40% efficacy, increasing adoption from 40 to 80% = 19% reduction



An Opportunity to Impact

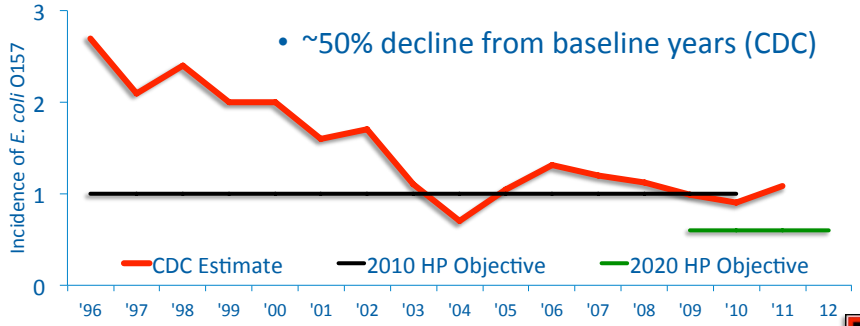
- Model built on best available data
  - All models contain some degree of uncertainty
- The model allows us to estimate the likely ***impact*** of pre-harvest control of *E. coli* O157 based on both ***efficacy*** & extent of ***adoption***
  - ***Impact = Efficacy * Adoption***
- A poorly efficacious intervention is expected to have an impact if broadly adopted
  - Adoption may be more important than efficacy
  - Yet we generally focus on efficacy

Summary

- We have robust (albeit imperfect) tools that can reduce prevalence of *E. coli* O157
  - Our best available models inform us they will likely improve public health
- Yet we tend to focus on efficacy
  - 'Wait for a better product'
  - Maybe if we believe control is important, we need to start focusing on those factors that will facilitate adoption
    - Broad adoption of even a poorly efficacious product appears to have a meaningful benefit

Human Incidence of *E. coli* O157

- Many abattoirs are now excelling at microbial process control
  - Achieving achievable control
- ***Impact*** observed across various metrics



Reported Human cases / 100,000

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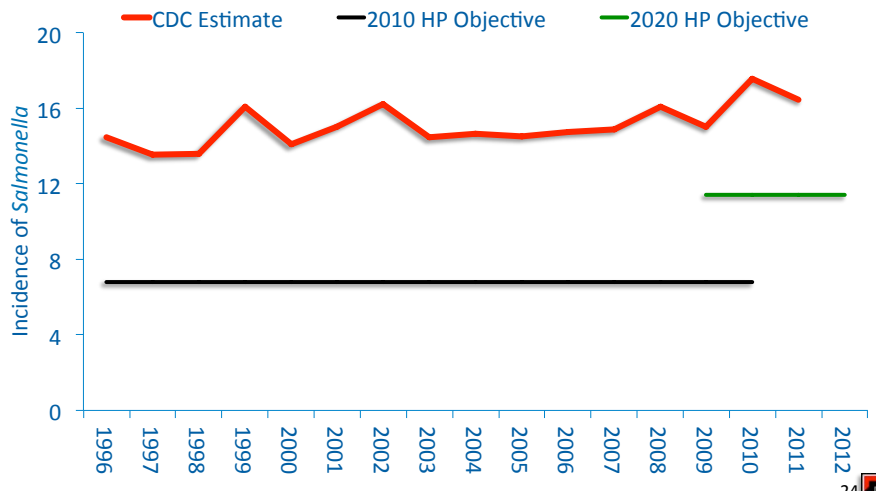
Human Incidence of *Salmonella*



Reported Human cases / 100,000

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E. coli O157: USDA/FISIS

- >90% decline from 2001
- 10,000 to 14,000 tests per year
- Many abattoirs are now excelling at microbial process control
  - Achieving achievable control

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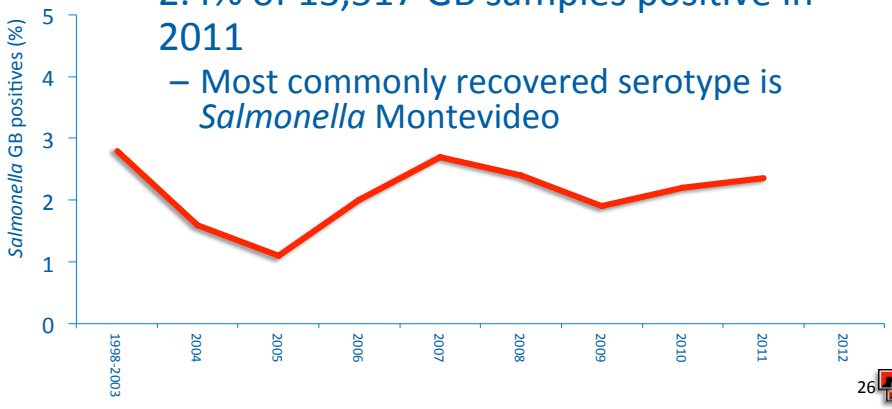
Salmonella: USDA/FSIS

- 2.4% of 13,517 GB samples positive in 2011
  - Most commonly recovered serotype is *Salmonella* Montevideo



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A Hypothesis was Presented

- 2010: A packer asked NCBA to work out if the issue is with *Salmonella* in lymph nodes?
- 2008 paper: *Salmonella* in 1.6% of lymph nodes
  - Cull-bulls 3.9 versus 0.35% in fed cattle
- PLN are beef when present at 'usual proportions'
  - Bypasses assumed route of contamination
  - Feces/Hides >>>> peripheral LN >>>> ground beef
- NCBA invested Beef Checkoff to explore this

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Surveillance Studies

Funded by the Beef Checkoff

- Packing-plant surveillance populations
  1. Cattle from feedlots
  2. Cows culled from dairy and beef herds
  - Cattle that passed USDA inspections
- Sep 2010 to Oct 2011
  - Samples collected 6 times throughout year
  - ~75 nodes per abattoir per time
- Feb 2012 to Dec 2012
  - More plants, more sample periods

United States Department of Agriculture

National Institute of Food and Agriculture

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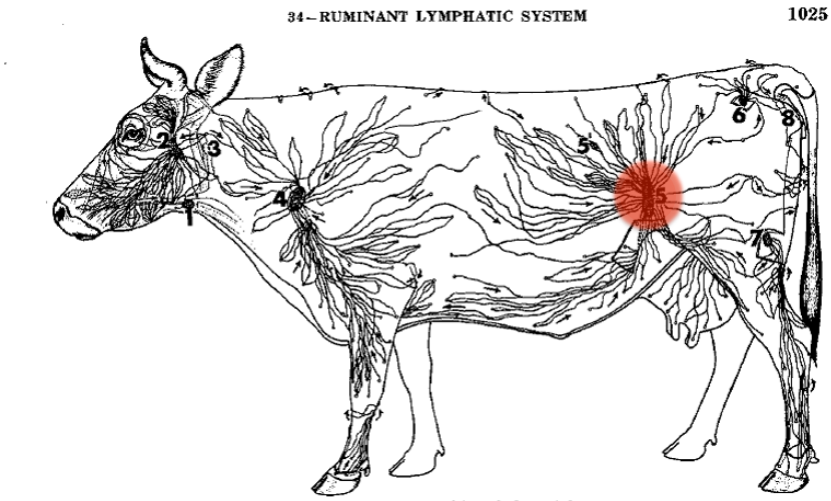

FIGURE 34–1. Superficial lymph flow of the cow.

1, Mandibular ln.; 2, parotid ln.; 3, lateral retropharyngeal ln.; 4, superficial cervical ln.; 5, subiliac ln.; 5′, lnn. of paralumbar fossa; 6, gluteal ln.; 7, popliteal ln.; 8, tuberal ln. (After Baum, 1912.)

From: Ruminant Lymphatic System (Saar and Getty) In Anatomy of the Domestic Animals. Eds Sisson and Grossman

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2010-2011 Surveillance

- Plants from which samples were received:
  - 5 in Texas (categorized as southern)
  - 2 in Nebraska (categorized as northern)
  - 1 in California (included in northern category)
    - Most plants have 6 collection windows; 2 each in:
    - Sep-Nov, Feb-Mar, Jul-Sep
- 3,327 lymph nodes assayed
  - 8.0% positive
  - Accepted for publication

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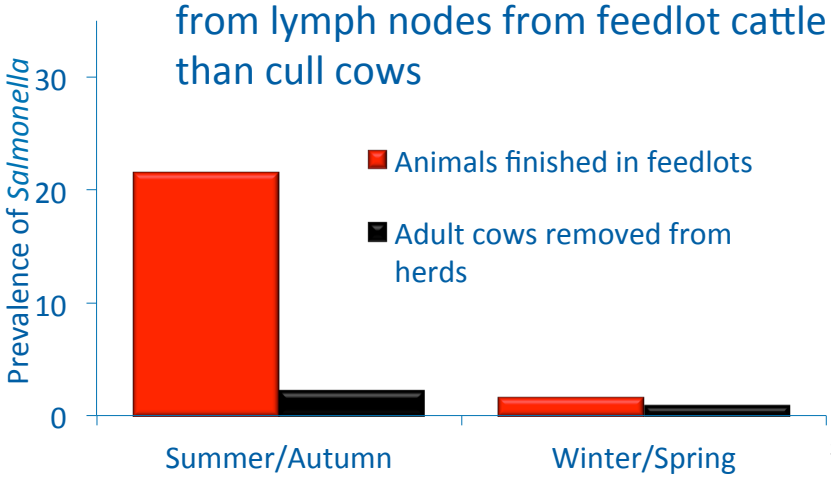
2010-2011 Surveillance

Funded by the Beef Checkoff

- 9 times more likely to be recovered from lymph nodes from feedlot cattle than cull cows



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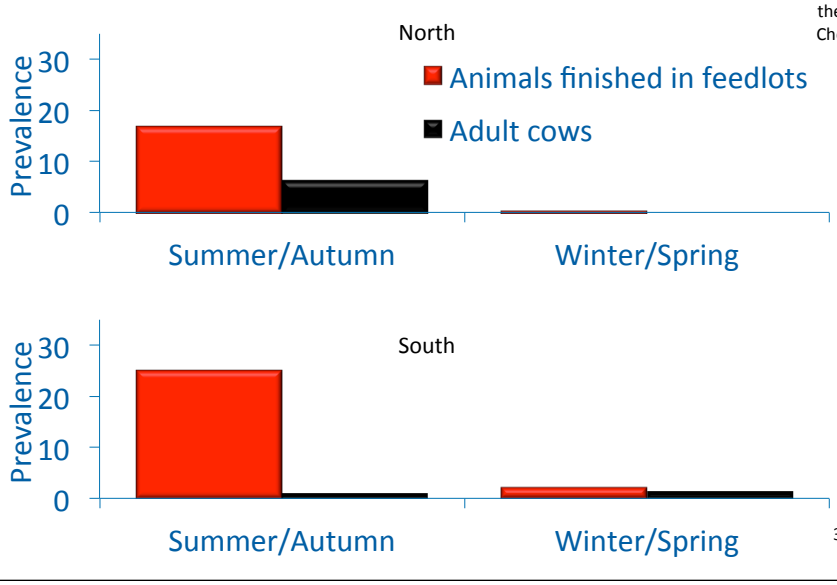
2010-2011 Surveillance

Funded by the Beef Checkoff



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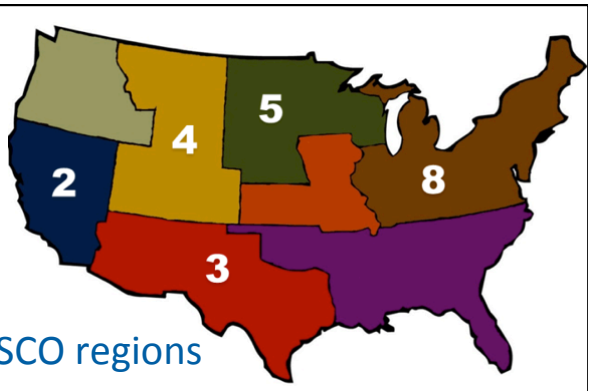
2012 Surveillance



- 12 Plants by BIFSCO regions
  - 5 in region 3
  - 3 in region 2
  - 2 in region 4
  - 1 each in regions 8 & 5
- 4,764 lymph nodes assayed
  - 6.0% positive

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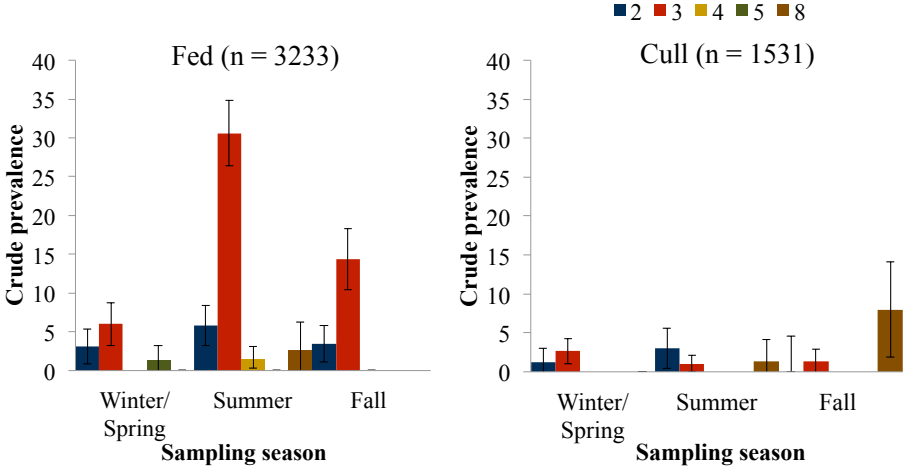
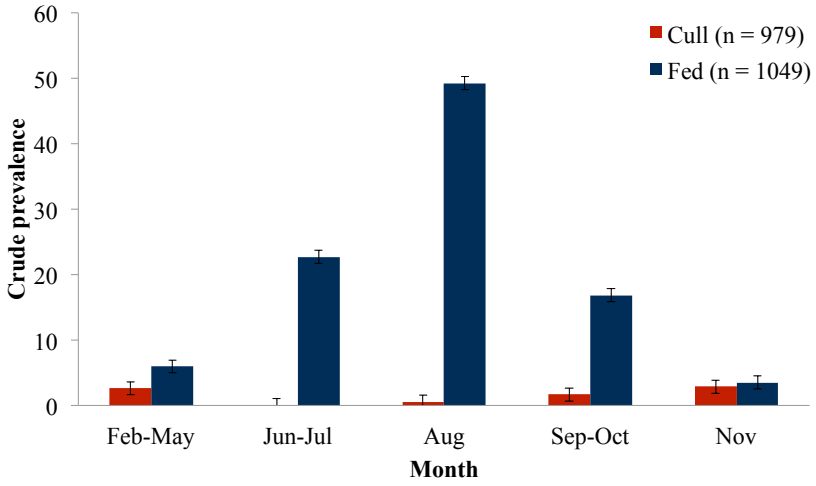
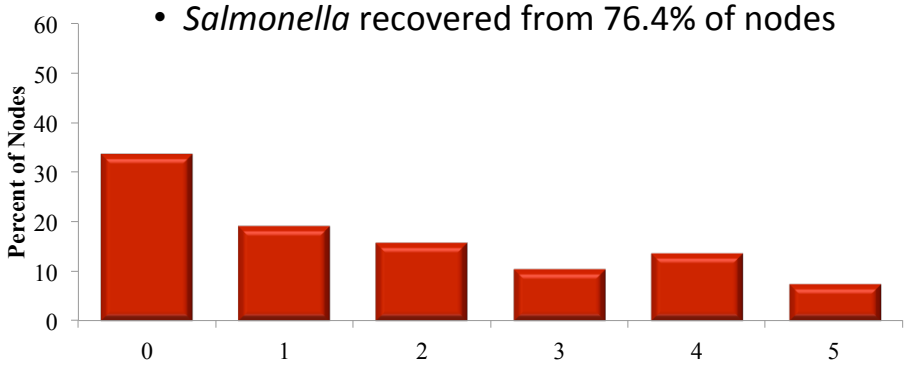
Prevalence by region and season



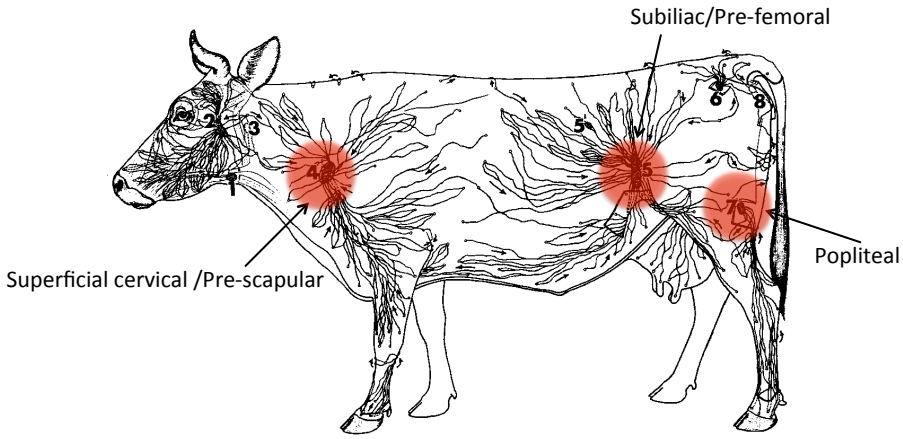
Prevalence in region 3 by month



- A single subiliac lymph node collected from 300 carcasses across 12 pens of cattle
  - *Salmonella* recovered from 76.4% of nodes



0: positive & le 1 log10 cfu/g
1: > 1 log10 cfu/g & le 2 log10 cfu/g
2: > 2 log10 cfu/g & le 3 log10 cfu/g
3: > 3 log10 cfu/g & le 4 log10 cfu/g
4: > 4 log10 cfu/g & le 5 log10 cfu/g
5: > 5 log10 cfu/g



Multiple lymph nodes per carcass

Visit	Carcasses Sampled	Fecal prevalence	Node prevalence	Positive in 1 or more	Positive in all 6
03OCT	15	80%	58.9%	100%	20%
17OCT	30	100% (n=6)	56.1%	96.7%	23.3%
24OCT	20	95%	15.8%	50%	5%
31OCT	35	47.8% (23)	9.1%	37.1%	0%

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Summary

- *Salmonella* recovered from lymph nodes
  - Varies by region, season, and animal type
  - Feedlot cattle more than cull cows (summer/fall)
    - Routinely recovered from >30% of PLN
- While less common in cull cows, more likely to be *Salmonella* Newport or Typhimurium
- Feedlot cattle serotypes more closely match what FSIS finds in its regulatory samples

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Salmonella and PLN

- Traditional paradigm is that *Salmonella* escapes the gut and disseminates systemically via lymphatic>vasculature system
  - Not all the data support this concept
- We hypothesize that a transdermal route of infection for *Salmonella* is the primary route by which the PLNs become infected
  - Biting flies, skin lesions, footrot, etc.

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How Might We Approach Control?

- In-plant peripheral lymph node removal

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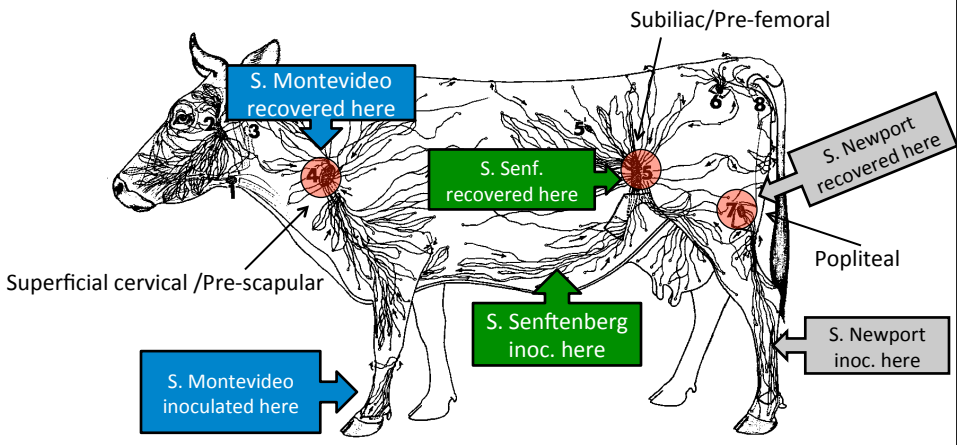
Trans/Intradermal Route of Infection

Tom Edrington USDA/ARS/SPARC/ FFSRU

- Development of a trans/intradermal challenge model
- Mimic real-world observations

Source of photos Tom Edrington



- Slide prepared by Tom Edrington

Challenge Model Studies to Date

Tom Edrington

- ***Oral*** challenge
  - *S.* Newport recovered from 4.2% of vaccinate peripheral nodes and 54.2% of control nodes
  - Oral challenge studies not very rewarding
- ***Transdermal*** challenge
  - *S.* Newport recovered from 33% of vaccinate peripheral nodes and 67% of control nodes

Summary

- For *E. coli* O157, assumed carcass was sterile when left feedlot and contaminated in the plant
  - Packers took 'ownership'
- *Salmonella* in PLNs means that carcass is not sterile
  - Very different perspective
- Consensus that in plant controls are insufficient
  - Pre-harvest efforts needed
- Early in the research process
  - Evidence we can control it

Disclaimer

- I come with conflicts of interest!
- My opinions are influenced by my research
  - Observational studies
  - Experimental studies
    - Sponsored by USDA, beef industry (e.g., Checkoff), companies [e.g., biopharmaceutical or assay])
- Sponsorship to provide continuing education at state, national, and private meetings
  - Expenses and sometimes honoraria
- Consulting and service on advisory boards for companies and associations
  - Expenses and sometimes a fee for service

TEXAS TECH UNIVERSITY
College *of* Agricultural Sciences & Natural Resources

- Thanks for the invitation to present
- Colleagues and funding
  - Dayna Harhay, Tom Edrington
  - Sara Gragg, Hattie Webb, Mindy Brashears, Marie Bugarel, and Kendra Nightingale
  - Beef Checkoff Program
  - USDA/NIFA/NIFSI
    - Contract # 2011-51110-31081
    - Texas Tech & USDA/ARS
  - Pfizer Animal Health
  - Intramural (Texas & USDA/ARS)

- Contact Information:

Guy.Loneragan@TTU.edu
Texas Tech University
+1 (806) 742-2805 x 268

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