

# **On-going Cornell research on *Listeria monocytogenes* transmission at retail**

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# **Collaborative effort between Cornell and New York State Department of Agriculture and Markets (Dan Rice, Joe Corby)**

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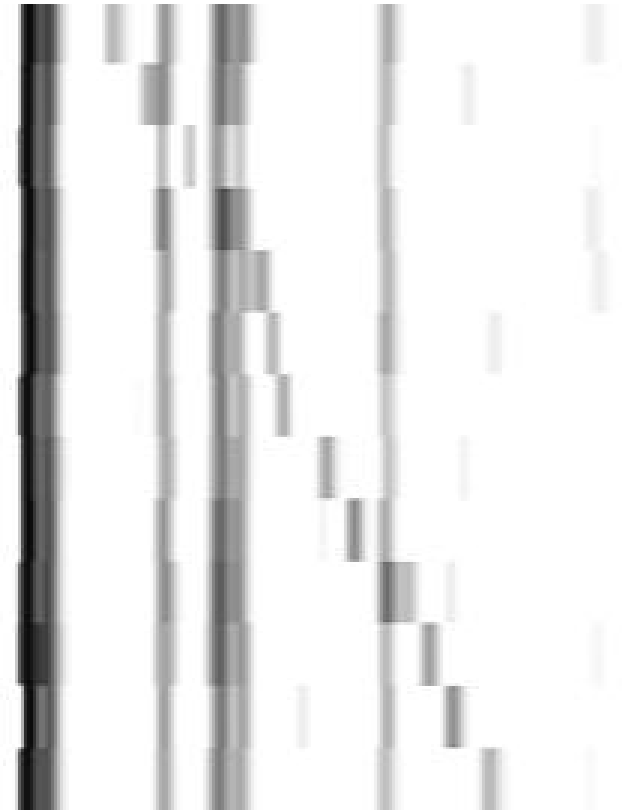
# *L. monocytogenes* strain differentiation (subtyping/fingerprinting)

- Tools which allow sensitive differentiation of bacterial subtypes
  - Detection of contamination sources
- Strain differentiation methods commonly applied to *L. monocytogenes* include serotyping, ribotyping, Pulsed Field Gel Electrophoresis (PFGE), DNA sequencing
  - While serotyping differentiates only 13 subtypes (serotypes), molecular methods generally differentiate more than >100 *L. monocytogenes* subtypes

# Ribotyping

## Examples of different *L. monocytogenes* ribotypes

Listeria monocytogenes  
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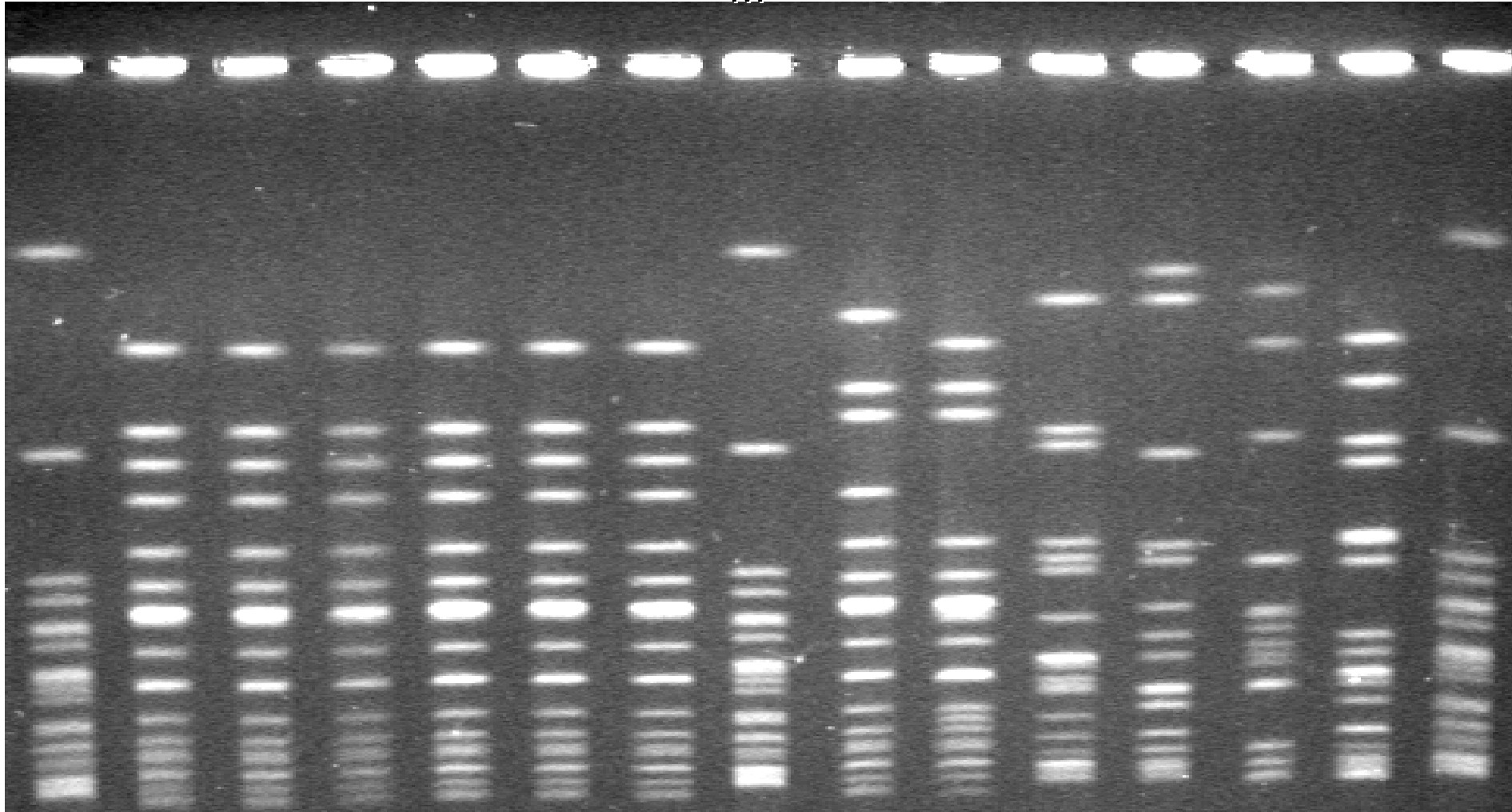


# Pulsed Field Gel Electrophoresis

M

M

M



# *L. monocytogenes* prevalence

Pristine environments: 1.3% (n=900)

Urban environments: 7.3% (n=900)

## Ruminant farms

- Bovine farms with listeriosis cases: 24.35% (n=616)
- Bovine farms without listeriosis cases: 20.06% (n=643)
- Small ruminant farms with listeriosis: 32.92% (n=322)
- Small ruminant farms without listeriosis: 5.89% (n=475)

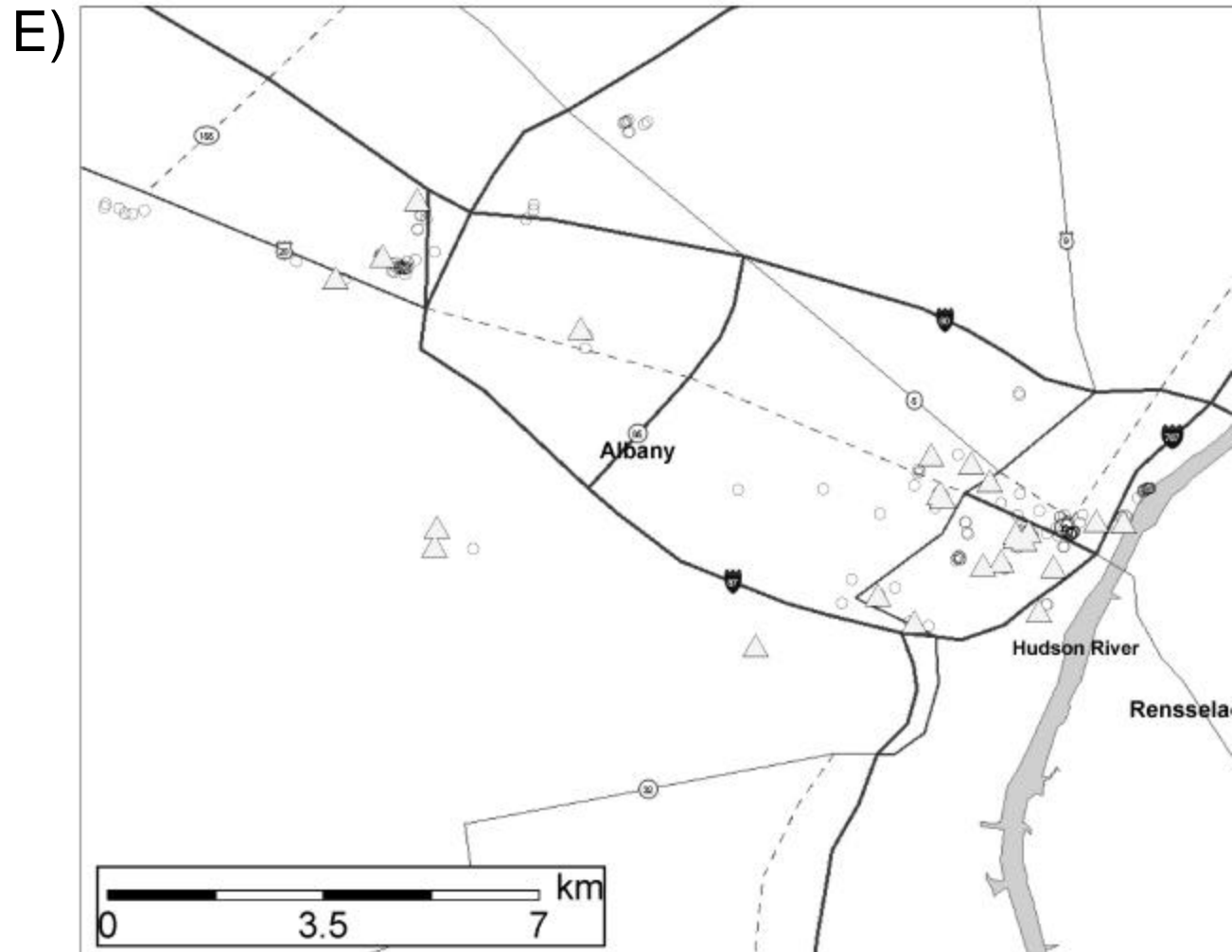
## Raw foods

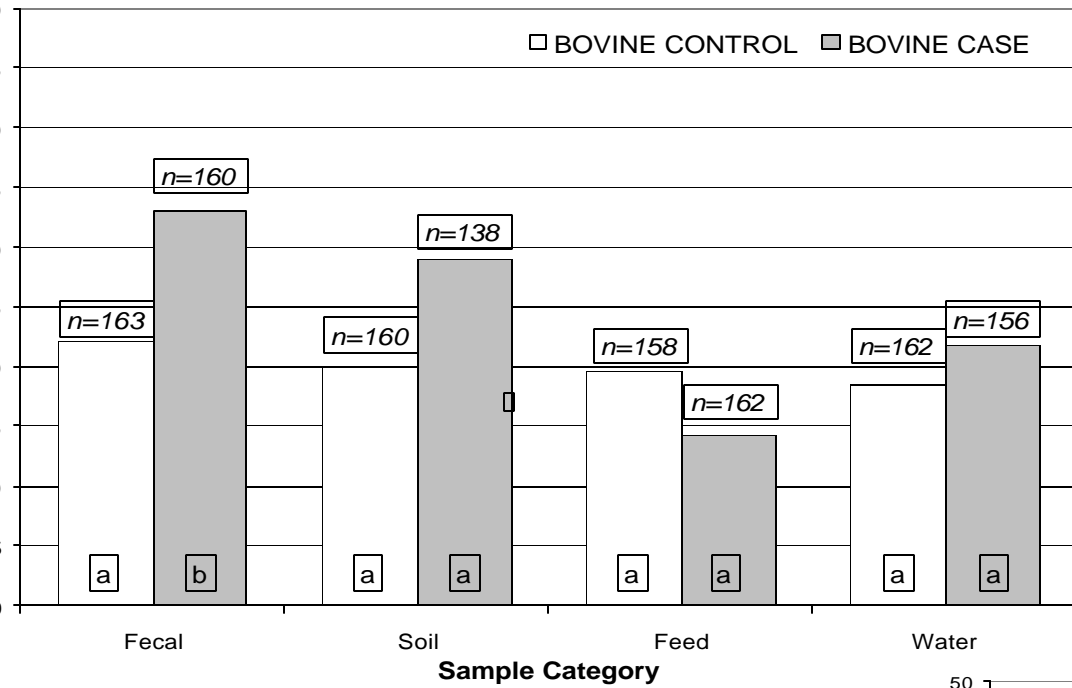
Food processing environments: from <0.1% to 30% or more

Ready-To-Eat foods: 0.17 – 4.7 % (Gombas et al., 2004)

# *L. monocytogenes* in urban environments

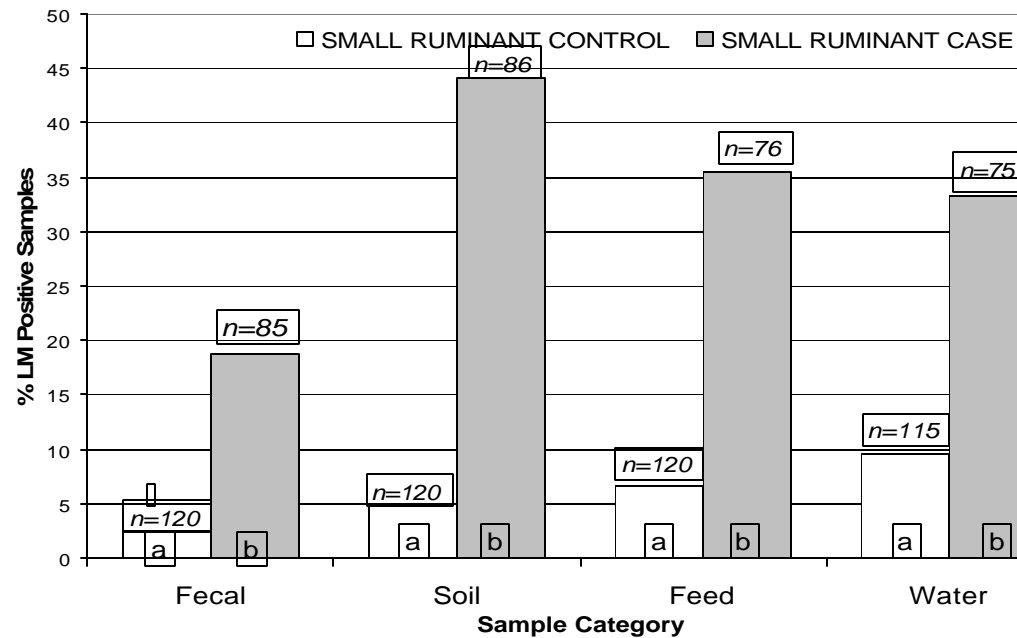
- Albany, NY
  - 214 samples tested
  - 27 positive for LM
  - 10 isolates were ribotype DUP-1038B (over three samplings and >1 year)





← Cattle

Small ruminants →



| ant AI            | 3/1/01 | 3/21/01 | 4/18/01 | 5/15/01 | 6/13/01 | 7/9/01 | 8/7/01 | 10/2/01 | 11/1/01 | 12/4/01 | 2/14/02 | 3/11/02 | 4/9/02 | 5/7/02 | 6/5/02 | 7/1/02 | 8/1/02 | 8/28/02 | 9/24/02 | #####  | 12/2/02 | # |
|-------------------|--------|---------|---------|---------|---------|--------|--------|---------|---------|---------|---------|---------|--------|--------|--------|--------|--------|---------|---------|--------|---------|---|
| Product           |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
|                   | L.spp  | L.spp   | 1052A   | 1042B   | -       | 1039A  | 1039C  | 1039C   | -       | -       | 1038B   | L.spp   | L.spp  | L.spp  | -      | L.spp  | L.spp  | L.spp   | 1042C   | 1027A  | 1052A   | 1 |
|                   | 1 of 6 | 3 of 6  | 2 of 6  | 1 of 6  | 6 of 6  | 1 of 6 | 1 of 6 | 1 of 6  | 6 of 6  | 6 of 6  | 1 of 6  | 3 of 6  | 2 of 6 | 2 of 6 | 6 of 6 | 2 of 6 | 1 of 6 | 2 of 6  | 1 of 6  | 1 of 6 | 1 of 6  | 1 |
| Environment       |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| Drain             | -      | 1043A   | -       | 1052A   | 1045B   | 1045B  | 1039C  | 1039C   | 1043A   | 1043A   | L.spp   | 1052A   | 1039C  | 1048A  | 1039C  | 1043A  | 1043A  | -       | 1043A   | 1062A  | 1027A   | 1 |
| Apron             | 1062A  | 1062A   | -       | -       | -       | -      | 1052A  | -       | 1043A   | -       | -       | -       | -      | -      | -      | -      | -      | -       | L.spp   | 1044A  | -       |   |
| knife             |        |         |         |         |         | 1043A  |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| Washed Enviroment |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| Drain             | 1039C  | 1043A   | 1042B   | 1039C   | L.spp   | L.spp  | L.spp  | L.spp   | 1043A   | L.spp   | 1043A   | -       | 1043A  |        |        | -      | 1043A  | -       | L.spp   | -      | 1039C   | 1 |
| Drain             | 1043A  | -       | 1043A   | -       | 1043A   | 1039C  | -      | L.spp   | -       | 1043A   | 1042C   | 1042C   | 1042C  | L.spp  | 1043A  | 1042C  |        | 1043A   | 1052A   | 1038B  | 1052A   | 1 |
| ooler Floor       | 1062A  | L.spp   | -       | -       | 1043A   | -      | L.spp  | L.spp   | 1052A   | 1043A   | L.spp   | -       | -      | L.spp  | 1052A  | L.spp  | -      | -       | -       | 1058B  | -       | 1 |
|                   |        |         |         |         |         |        |        | L.spp   |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| mat               |        |         |         |         |         |        |        |         |         |         | L.spp   | 1052A   |        |        |        |        |        |         |         |        |         |   |
| art wheels        | L.spp  | 1043A   | -       | 1052A   | 1027A   | 1043A  | L.spp  | 1043A   | 1043A   | 1052A   | 1052A   | -       | -      | -      | -      | -      | -      | -       | -       | -      | -       |   |
| nder Slicer       | -      | -       | -       | -       | -       | -      | L.spp  | L.spp   | -       | -       | -       | -       | -      | -      | -      | -      | -      | -       | -       | -      | -       |   |
| iding Door        | -      | -       | -       | -       | -       | -      | -      | -       | -       | -       | -       | -       | -      | -      | -      | -      | -      | -       | -       | -      | -       |   |
| Contact Surfaces  |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| oves              | -      | -       | -       | -       | -       | -      | -      | -       | L.spp   | 1043A   | -       | -       | -      | -      | -      | -      | -      | -       | -       | -      | -       |   |
| Slicer            | -      | -       | -       | -       | -       | -      | -      | -       | -       | -       | 1027A   | -       | -      | -      | -      | -      | -      | -       | -       | -      | -       | 1 |
| Skinner           | -      | -       | -       | -       | L.spp   | -      | -      | -       | -       | -       | -       | -       | -      | -      | -      | -      | -      | -       | -       | 1039C  | -       |   |
| DeBoner           | -      | -       | -       | 1042B   | -       | -      | -      | -       | -       | -       | -       | -       | -      | -      | -      | L.spp  | -      | 1043A   | 1044A   | 1044A  | 1044A   | 1 |
| Gal. Table        | -      |         |         |         |         |        |        |         |         |         | -       | L.spp   | -      | -      | -      | L.spp  | L.spp  | L.spp   | -       | -      | 1044A   |   |
| dirty             |        |         |         |         |         |        |        |         | 1062A   |         |         |         |        |        |        |        |        |         |         |        |         |   |
| clean             |        |         |         |         |         |        |        |         | 1043A   | 1044A   |         |         |        |        |        |        |        |         |         |        |         |   |
| Mixer             |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        | L.spp  | L.spp   | L.spp   | -      | -       | 1 |
| Table             |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
| Washed Product    |        |         |         |         |         |        |        |         |         |         |         |         |        |        |        |        |        |         |         |        |         |   |
|                   | 1 of 6 | 1 of 6  | 6 of 6  | 6 of 6  | 6 of 6  | 1 of 6 | 1 of 6 | 1 of 6  | 6 of 6  | 1 of 6  | 1 of 6  | 6 of 6  | 1 of 6 | 1 of 6 | 1 of 6 | 1 of 6 | 6 of 6 | 1 of 6  | 6 of 6  | 1 of 6 | 6 of 6  | 1 |
|                   | L.spp  | L.spp   | -       | -       | -       | 1062A  | L.spp  | L.spp   | -       | 1043A   | 1042C   | -       | L.spp  | 1042C  | L.spp  | 1052A  | -      | L.spp   | -       | L.spp  | -       | 1 |

# *L. monocytogenes* in retail environments

- Subtyped 98 food and 40 environmental *L. monocytogenes* isolates collected from retail establishments in New York State between 1997 and 2002
  - Isolates collected from 50 different retail establishments were characterized.
- 16 retail establishments showed evidence for persistence of one or more specific *L. monocytogenes* strains as indicated by isolation of the same *EcoRI* ribotype from food and/or environmental samples collected in a given establishment on different days
- 17 ribotypes were found among human clinical isolates as well as among food and environmental isolates

# **Distribution of *Listeria monocytogenes* Molecular Subtypes among Human and Food Isolates from New York State Shows Persistence of Human Disease–Associated *Listeria monocytogenes* Strains in Retail Environments**

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**ABSTRACT**

# *L. monocytogenes* in retail environments - example

| st. code | Source      | Sample Description   | Date Collected (No. isolates) | Ribotype  | Persistent Ribotype (Lineage) |
|----------|-------------|----------------------|-------------------------------|-----------|-------------------------------|
|          | Food        | Oven roasted turkey  | 08/17/00                      | DUP-1062A | DUP-1062A (II)                |
|          | Food        | Smoked turkey breast | 08/25/00                      | DUP-1062A |                               |
|          | Environment | Swab; display cooler | 08/29/00                      | DUP-1062A |                               |
|          | Environment | Swab                 | 08/29/00                      | DUP-1062A |                               |
|          | Environment | Swab                 | 08/29/00                      | DUP-1062A |                               |
|          | Environment | Swab; deli case      | 09/21/00                      | DUP-1062A |                               |
| /        | Environment | Swab                 | 10/27/00                      | DUP-1053A | Not applicable                |
|          | Environment | Swab                 | 10/27/00                      | DUP-1053A |                               |
|          | Food        | Ham                  | 11/07/00                      | DUP-1042C | DUP-1042C(I)                  |
|          | Environment | Swab                 | 11/20/00                      | DUP-1042C |                               |
|          | Environment | Swab                 | 11/20/00                      | DUP-1042C |                               |
|          | Environment | Swab                 | 12/04/00                      | DUP-1042C |                               |
|          | Environment | Swab                 | 12/04/00                      | DUP-1042C |                               |

# On-going prospective study on *L. monocytogenes* in retail

- The goal of this project is to collect and test for LM environmental sponge samples from 120 retail establishments over 12 months (10 establishments/month)
  - in each month sponge samples are collected from 5 establishments where NYS Ag & Mkts collected sliced deli meats and from 5 establishments where NYS Ag & Mkts collected store made salads
- Sampling visits are part of NYS Ag & Mkts regular routine surveillance, only change is that environmental sponges will be tested in addition to scheduled food sample

# Environmental samples collected

- Slicer *or* bowl, spoon, cutting boards, tables (e.g., utensils used to prepare salads)
- Sponge from deli case
- Sponge from sink interior in deli/salad area
- Drain in deli area
- Drain in raw meat area
- Drain in raw seafood area
- Floor in a dry aisle
- Sponge from dairy case
- One sponge sample from wheels of 4 grocery carts (if no carts are available sponge sample from floor near store entrance will be taken)
- Two optional sponge samples, including, if possible, one sponge sample from a drain in the produce preparation area and one from a drain in a restaurant/café located in the retail operation

# Data so far

- Total of 550 environmental sponges; 71 positives (12.9%)
- 32 stores had at least one environmental sample positive
  - 2 stores also had positive deli salad samples
  - 1 environ pos: 9 stores
  - 2 environ pos: 13 stores
  - 3 environ pos: 5 stores
  - 4 or more environ pos: 5 stores
- 24 stores had no environmental sample positive
  - None of these stores had product positive samples

# Positive sites (56 stores)

- Salad prep food contact surface: 2 positive
- Deli slicer: 1 positive
- Deli case: 1 positive
- Dairy case: 6 positive
- Floor drain deli area: 6 positive
- Floor drain raw meat area: 15 positive
- Produce area drain: 14 positive
- Seafood area drain: 5 positive
- Cart wheels: 4 positives
- Dry isle: 3 positives

# Subtyping results (completed for 16 stores with >1 LM positive sample)

5 stores: all different subtypes

- One store with positive RTE salad had different subtypes in product and environment (deli sink)

One store: drains in produce, seafood, and bakery area all same subtype

One store: drains in raw meat and produce area, dry isle floor, and sink in deli area all same subtype

One store: drain in raw meat and produce area same subtype

One store: drain in raw meat area and sink in deli area same subtype

# More subtyping results.....

One store: floor in deli area and dairy case same subtype

One store: shopping cart wheels and sink in meat area same subtype

One store: shopping cart wheels and dry isle floor same subtype

One store: shopping cart wheels produce prep area same subtype

One store: floor drain in deli area and sink in deli area same subtype

One store: deli salad utensils and sink in deli area same subtype

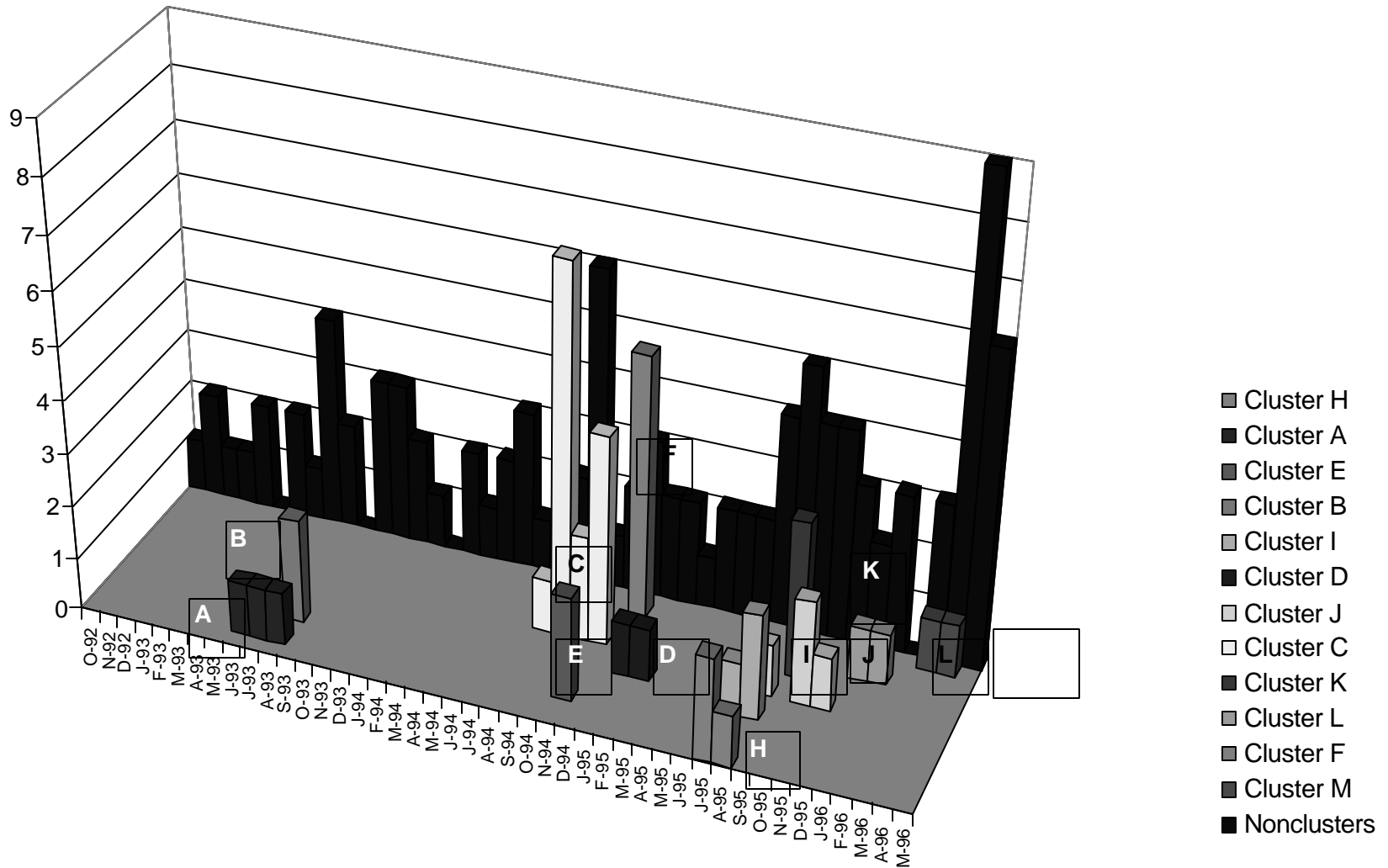
One store: shopping cart wheels and sink in meat area same subtype

11 stores with same subtype in multiple sites

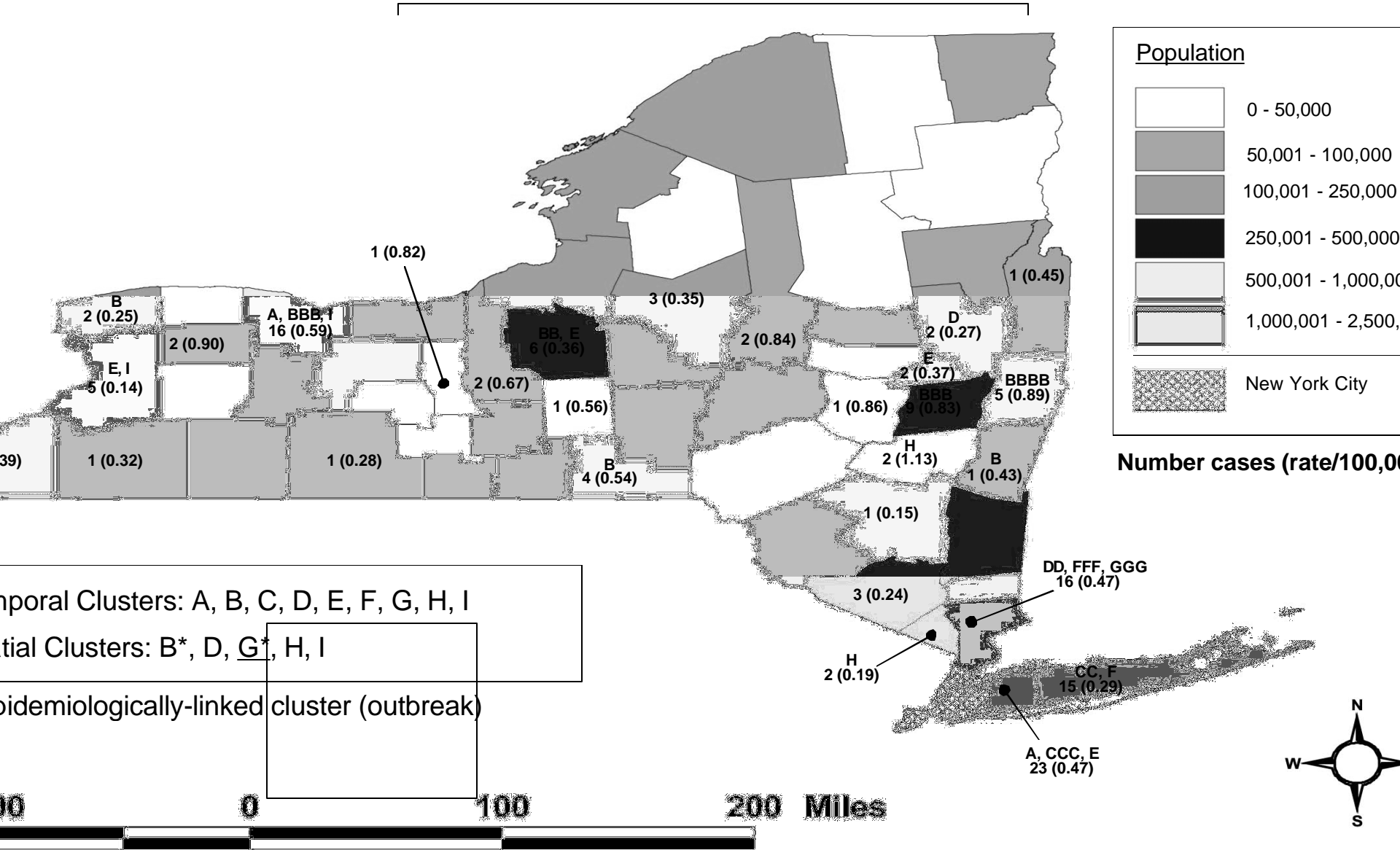
# Human listeriosis?

On-going subtyping of human listeriosis cases conducted in collaboration with NYS Department of Health and New York City Department of Health

# Patterns of human listeriosis



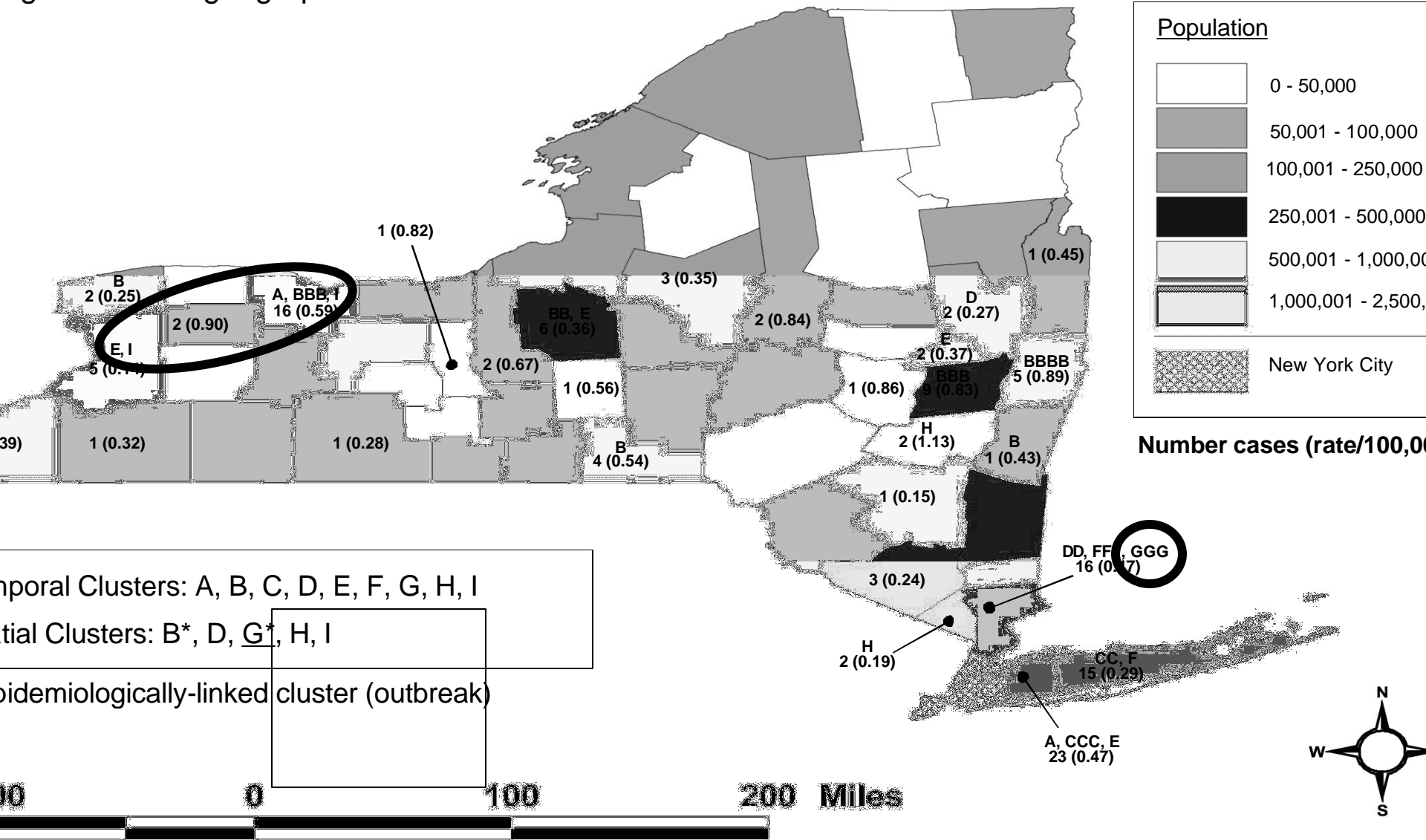
# Geographic Distribution of Human Listeriosis Cases



# Geographic Distribution of Human Listeriosis Cases

Example: Cluster G

Single localized geographic cluster



# Summary and conclusions

*L. monocytogenes*, including subtypes that have been linked to human disease, can persist in natural and urban environments, on farms, in food processing plant and in retail environments

- Persistence is likely to also occur in other environments (e.g., restaurants, consumer homes)

More human listeriosis cases than previously assumed may represent small outbreaks

- Persistence in food chain is likely to be critical for occurrence of listeriosis outbreaks

Lm contamination in retail operations appears to be dominated by positives in drain samples and multiple sample sites often have appear to have the same subtype

- Improved sanitation procedures and procedures to reduce cross contamination can likely reduce risk of RTE product contamination

# Future plans – LM in retail

- Complete current study including subtyping data and statistical analyses
- Longitudinal study in selected volunteer retail operations
  - Include temperature audits, questionnaires on sanitation procedures, employee training
- Develop training programs (employees and managers)
- Develop mathematical models incorporating processor, retail, and at home contamination to evaluate relative impact on human listeriosis
- Study “at home” LM persistence and contamination?