Molecular epidemiology of *Salmonella* and *Campylobacter* contamination of poultry during transport and slaughter

Geertrui Rasschaert

Vakgroep Veterinaire Volksgezondheid & Voedselveiligheid

Promotor: Prof. Dr. De Zutter
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
Incidence of *Salmonella* and *Campylobacter* infections per 100 000 Belgian habitants
35 *Salmonella* infections/100,000
54 *Campylobacter* infections/100,000

true incidence is much higher! ca. x 10!

socio-economic impact:
- employment costs,
- GP care,
- medicines,
- laboratory costs,
- hospital care

complications mortality

reported cases in 2006

e.g. 27 million euro in Belgium for *Campylobacter*

persons becoming ill
persons seeking care
stool sample obtained
culture confirmed
cases reported
Sources of *Salmonella* infection

- eggs & egg products: 39%
- pork: 25%
- poultry meat: 21%
- beef: 10%

*(van Pelt et al., 1999)*

**Outbreak:** > 2000 persons eating pre-cooked chicken from a particular brand (Spain, 2005)
Sources of *Campylobacter* infection

- barbecue
- contact with pets or other animals
- overseas travel
- consumption of poultry meat
- cross-contamination in kitchen

- 40% reduction in *Campylobacter* infections during dioxin crisis

- outbreaks: 12 persons eating stir-fried chicken in a restaurant (UK, 1997)
  > 80 employees eating chicken salad in a canteen (Denmark, 2005)
Dr. Henk van der Zee, Food Inspectorate, the Netherlands
OVERVIEW

1. Situation of the problem

2. Characteristics of *Salmonella* and *Campylobacter*

3. Clinical aspects of *Salmonella* and *Campylobacter* infections

4. Poultry flocks

5. Aim of the study

6. Molecular discrimination of *Salmonella* isolates at serotype level

7. Materials and Methods

8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter

9. *Salmonella*: impact of the slaughter line contamination on carcass contamination

10. *Campylobacter*: contamination or colonization of poultry flocks after transport

11. Conclusions
2. Characteristics of Salmonella and Campylobacter

**Salmonella**

- **Family**: Enterobacteriaceae
- **Genus**: *Salmonella*
- **Species**:
  - *enterica*
  - *salamae*
  - *arizonae*
  - *diarrizonae*
  - *houtenae*
  - *indica*
- **Subspecies**:
  - *bongori*
  - *subterranea*

> 2500 serotypes – Kauffmann-White scheme

- e.g. *Salmonella enterica* subspecies *enterica* serotype Typhimurium
- or short *Salmonella* Typhimurium
• Gram-negative rods
• 0.7-1.5 µm – 2.0-5.0 µm
• usually motile – peritrichous flagella
• facultative anaerobic
• temp: 5°C – 46°C (opt. ± 37°C)
• pH: 3.8 – 9.5 (opt. ± 7)
Campylobacter

Family: Campylobacteraceae
Genus: Campylobacter
Species: 17 species and 6 subspecies

C. jejuni, C. coli, C. lari

= thermophilic campylobacters
• Gram-negative spirils
• 0.2-0.8 µm – 0.5-5.0 µm
• motile with corkscrew-like motion:
  monotrichous or amphitrichous
• micro-aerobic
• temp: 30°C – 45°C (opt. ± 37°C, ± 42°C)
• pH: 4.9 – 9.0 (opt. ± 7)

Source: www.microbes-edu.org/etudiant/campylo.html
ASM, cover, Infection and Immunity 74
Virginia-Maryland Regional College of Veterinary Medicine, Virginia
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. **Clinical aspects of *Salmonella* and *Campylobacter* infections**
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
## 3. Clinical aspects

<table>
<thead>
<tr>
<th></th>
<th><strong>Salmonella</strong></th>
<th><strong>Campylobacter</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Incubation period</strong></td>
<td>12 h to 5 days</td>
<td>18 h to 8 days</td>
</tr>
<tr>
<td><strong>Infective dose</strong></td>
<td>$10^5$-$10^{10}$ bacteria (&gt;100)</td>
<td>500 - 800 bacteria</td>
</tr>
<tr>
<td><strong>Symptoms</strong></td>
<td>non-bloody diarrhea, abdominal pain, mild fever &amp; headache, nausea &amp; vomiting</td>
<td>diarrhea (may be bloody), intense abdominal pain, fever &amp; headache, nausea &amp; vomiting</td>
</tr>
<tr>
<td><strong>Treatment</strong></td>
<td>self-limiting fluoroquinolones</td>
<td>self-limiting erythromycin</td>
</tr>
<tr>
<td><strong>Complications</strong></td>
<td>Reactive Arthritis</td>
<td>Reactive Arthritis, Guillain-Barré syndrome</td>
</tr>
</tbody>
</table>
Distribution of *Salmonella* serotypes from patients in Belgium in 2005

- *Enteritidis*: 45.30%
- *Typhimurium*: 33.70%
- *Andere*: 11.70%
- *Hadar*: 0.60%
- *Paratyphi B*: 0.70%
- *Manhattan*: 0.80%
- *Infantis*: 1.20%
- *Virchow*: 1.30%
- *Derby*: 1.40%
- *Brandenburg*: 1.60%
- *Ohio*: 1.80%

(SIPH, 2006)
Distribution of *Campylobacter* species from patients during 1995-2002 in Belgium

(Vandenberg *et al.*, 2004)
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
4. Poultry flocks

Vertical transmission:
**Horizontal transmission:**

- Feed and drinking water
- Insects
- Rodents
- Wild birds
- Human traffic & hygiene
- Other farm animals
Prevalence of colonized flocks

**Salmonella**
- top: 2 to 4 weeks
- shedders vs. carriers
- within-flock prevalence: variable
- prevalence: 50% - 33% (Belgian study)

**Campylobacter**
- top: 6 weeks (lag of 2 weeks)
- shedders
- within prevalence: 100%
- prevalence: 3 - > 90%
slaughter

- loading at 5 or 6 weeks
- transport
- processing (logistic slaughter !)
  - hanging
  - stunning: electrical shock, $\text{CO}_2$
  - killing
  - scalding
  - defeathering
  - evisceration: removal of crop, nek, internal organs
  - chilling
Putting on transport band
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
hanging
stunning
killing
scalding
defeathering
evisceration
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks

5. **Aim of the study**

6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
5. Aim of the study

study of contamination of poultry with *Salmonella* and *Campylobacter* during transport and slaughter by means of molecular tools

- molecular discrimination of *Salmonella* isolates at serotype level
- poultry meat contamination due to gastrointestinal colonization or cross-contamination

*Salmonella*: impact of slaughter line contamination on carcass contamination

*Campylobacter*: impact of container contamination on carcass contamination
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study

6. Molecular discrimination of *Salmonella* isolates at serotype level

7. Materials and Methods

8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter

9. *Salmonella*: impact of the slaughter line contamination on carcass contamination

10. *Campylobacter*: contamination or colonization of poultry flocks after transport

11. Conclusions
6. Molecular discrimination of *Salmonella* isolates at serotype level

Serotyping ↔ PCR-based techniques (e.g., Repetitive-sequence-based PCR)

- Experiment 1: 5 different primers tested
- Experiment 2: reproducibility
- Experiment 3: typeability & discriminatory power
- Experiment 4: stability
ERIC primer set (GTG)$_5$ primer

S. Ent. typhimurium O5+ lysate 11 PCR run 2
S. Ent. typhimurium O5+ lysate 12 PCR run 2
S. Ent. typhimurium O5+ lysate 13 PCR run 2
S. Ent. typhimurium O5+ lysate 11 PCR run 3
S. Ent. typhimurium O5+ lysate 12 PCR run 3
S. Ent. typhimurium O5+ lysate 13 PCR run 3
S. Typhimurium O5+ lysate 11 PCR run 1
S. Typhimurium O5+ lysate 12 PCR run 1
S. Typhimurium O5+ lysate 13 PCR run 1
S. Typhimurium O5+ lysate 11 PCR run 1
S. Typhimurium O5+ lysate 12 PCR run 1
S. Typhimurium O5+ lysate 13 PCR run 1
S. Hadar lysate 11 PCR run 3
S. Hadar lysate 12 PCR run 3
S. Hadar lysate 13 PCR run 3
S. Hadar lysate 11 PCR run 2
S. Hadar lysate 12 PCR run 2
S. Hadar lysate 13 PCR run 2
S. Hadar lysate 11 PCR run 1
S. Hadar lysate 12 PCR run 1
S. Infantis lysate 11 PCR run 3
S. Infantis lysate 12 PCR run 3
S. Infantis lysate 13 PCR run 3
S. Infantis lysate 11 PCR run 2
S. Infantis lysate 12 PCR run 2
S. Infantis lysate 13 PCR run 2
S. Infantis lysate 11 PCR run 1
S. Infantis lysate 12 PCR run 1
S. Infantis lysate 13 PCR run 1
S. Brandenburg lysate 11 PCR run 2
S. Brandenburg lysate 12 PCR run 2
S. Brandenburg lysate 13 PCR run 2
S. Brandenburg lysate 11 PCR run 3
S. Brandenburg lysate 12 PCR run 3
S. Brandenburg lysate 13 PCR run 3
S. Brandenburg lysate 11 PCR run 1
S. Brandenburg lysate 12 PCR run 1
S. Brandenburg lysate 13 PCR run 1

ca. 80%
92.5%
Conclusions

- clusters of isolates belonging to same serotype
- profiles of non-serotypeable isolates (molecular ‘serotyping’)
- profiles obtained by one PCR can be compared (or representative of each cluster)
- ERIC primer set and (GTG)$_5$ are equally useful for discrimination
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. **Materials and Methods**
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
7. Materials and Methods

**Bacteriological culture & Identification**

crop swabs, neck skins, gastrointestinal tracts (duodenum & ceca), ...
diluted in 1:10 BPW

**Salmonella**

- enrichment (16-20 h 37°C)
- Diassalm and RV (24 h 42°C)
- XLD (24 h 37°C)
- genus PCR
- ERIC-PCR
- serotyping
**Bacteriological culture & Identification**

crop swabs, neck skins, gastrointestinal tracts (duodenum & ceca), ...
diluted in 1:10 BPW

---

Campylobacter

1:10 dilution in preston

<table>
<thead>
<tr>
<th>↓</th>
<th>enrichment (24 – 48 h at microaerobic conditions at 42°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>↓</td>
<td>CCDA (24 h at 42°C)</td>
</tr>
<tr>
<td>↓</td>
<td>species-mPCR</td>
</tr>
</tbody>
</table>

---

C. jejuni

---

C. coli
Characterization of the isolates

**Salmonella:**

PFGE (SmaI, NotI, Spel)

**Campylobacter:**

FlaA PCR/RFLP

PFGE (SmaI, KpnI)
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. **Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter**
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
8. Contribution of gastrointestinal colonization & cross-contamination to carcass contamination during slaughter

**Salmonella**

**M&M:**
- 30 crop swabs
- 30 gastrointestinal tracts
- 20 neck skins

![Image of carcasses]

4 broiler slaughterhouses (x 3)

**Results:**
- 13% of flocks were colonized with *Salmonella* ↔ status (5%)
- 55% of flocks were contaminated with *Salmonella* after slaughter
  → Agona, Hadar, Infantis, Typhimurium, Virchow, Indiana, ...
### gastrointestinal contribution

#### external contamination

#### gastrointestinal tract

<table>
<thead>
<tr>
<th>Flock</th>
<th>NECK SKINS</th>
<th>CROP</th>
<th>DUODENUM</th>
<th>CECA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Flock</th>
<th>Serotype &amp; genotype</th>
<th>Serotype &amp; genotype</th>
<th>Serotype &amp; genotype</th>
<th>Serotype &amp; genotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Indiana I-1</td>
<td>Typhimurium T-1</td>
<td>Typhimurium T-1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Newport N-1</td>
<td>London</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>NT-1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>NT-1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Agona A-1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Hadar H-1</td>
<td>Hadar H-1</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## cross-contamination

<table>
<thead>
<tr>
<th></th>
<th>NECK SKINS</th>
<th></th>
<th>CROP</th>
<th></th>
<th>DUODENUM</th>
<th></th>
<th>CECA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Flock</td>
<td>Numb. /20</td>
<td>Serotype &amp; genotype</td>
<td>Numb. /3</td>
<td>Serotype genotype</td>
<td>Numb. /3</td>
<td>Serotype genotype</td>
</tr>
<tr>
<td>1</td>
<td>3</td>
<td>Hadar H-1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>2</td>
<td>Hadar H-1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2</td>
<td>Hadar H-1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>12</td>
<td>Agona A-2 Virchow V-1 Typhimurium T-2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>9</td>
<td>Agona A-2 Typhimurium T-2 Ealing Ea-1</td>
<td>1</td>
<td>Typhimurium T-2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>8</td>
<td>Agona A-2 Typhimurium T-2 NT-2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>6</td>
<td>Agona A-2, A-3 Typhimurium T-2 O4:d:-</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>Typhimurium T-2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>3</td>
<td>Typhimurium T-2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>3</td>
<td>Typhimurium T-2 O4:d:-</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Conclusions:

- gastrointestinal contribution is limited
- cross-contamination is high
- origin of majority of strains is unknown
- logistic slaughter is difficult
8. Contribution of gastrointestinal colonization & cross-contamination to carcass contamination during slaughter

**Campylobacter**

M&M:
- 30 crop swabs
- 30 gastrointestinal tracts
- 20 neck skins

\[ \text{3 broiler slaughterhouses (x 3)} \]

**Results:**
- 72% of flocks were colonized with *Campylobacter*
- 79% of flocks were contaminated with *Campylobacter* after slaughter
  \[ \rightarrow C. \text{jejuni (89%), C. coli (8.7%) and C. lari (2.3%)} \]
<table>
<thead>
<tr>
<th>Flock</th>
<th>Numb /20</th>
<th>Species &amp; genotype</th>
<th>Numb /3</th>
<th>Species genotype</th>
<th>Numb /3</th>
<th>Species genotype</th>
<th>Numb /3</th>
<th>Species genotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>18</td>
<td>C. jejuni G-1</td>
<td>1</td>
<td>C. jejuni G-1</td>
<td>3</td>
<td>C. jejuni G-1</td>
<td>3</td>
<td>C. jejuni G-1</td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>20</td>
<td>C. jejuni G-2</td>
<td>3</td>
<td>C. jejuni G-2</td>
<td>3</td>
<td>C. jejuni G-2 C. coli G-3</td>
<td>3</td>
<td>C. coli G-3</td>
</tr>
<tr>
<td>4</td>
<td>18</td>
<td>C. jejuni G-2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>7</td>
<td>C. jejuni G-2 C. coli G-3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>16</td>
<td>C. jejuni G-4</td>
<td>2</td>
<td>C. jejuni G-5</td>
<td>3</td>
<td>C. jejuni G-6</td>
<td>1</td>
<td>C. jejuni G-6</td>
</tr>
<tr>
<td>2</td>
<td>20</td>
<td>C. jejuni G-5 C. jejuni G-7</td>
<td>3</td>
<td>C. jejuni G-5</td>
<td>3</td>
<td>C. jejuni G-7</td>
<td>3</td>
<td>C. jejuni G-7</td>
</tr>
<tr>
<td>3</td>
<td>20</td>
<td>C. jejuni G-8 C. jejuni G-5</td>
<td>3</td>
<td>C. jejuni G-8</td>
<td>3</td>
<td>C. jejuni G-8</td>
<td>3</td>
<td>C. jejuni G-8</td>
</tr>
<tr>
<td>4</td>
<td>20</td>
<td>C. jejuni G-8</td>
<td>3</td>
<td>C. jejuni G-8</td>
<td>3</td>
<td>C. jejuni G-8</td>
<td>2</td>
<td>C. jejuni G-8</td>
</tr>
</tbody>
</table>
Conclusions for *Salmonella*:

- gastrointestinal contribution is limited
- cross-contamination is high
- origin of majority of strains is unknown
- logistic slaughter is difficult

Conclusions for *Campylobacter*:

- gastrointestinal contribution is high
- cross-contamination is limited
- origin of only a few strains is unknown
- flocks slaughtered on same day had same strains in intestines ???
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter

9. *Salmonella*: impact of the slaughter line contamination on carcass contamination

10. *Campylobacter*: contamination or colonization of poultry flocks after transport

11. Conclusions
Observation: origin of majority of strains is unknown
Hypothesis: slaughter line contamination

M&M:
- 6 flocks (3 slaughterhouses, x2)
- samples of slaughter line
- samples of first flock
  - gastrointestinal tracts
  - neck skins
- feathers
- containers
<table>
<thead>
<tr>
<th>Samples</th>
<th>Number</th>
<th>Serotype &amp; genotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scalding tank (+ water)</td>
<td>6/19</td>
<td>Typhimurium T-1 Blockley B-1 Paratyphi B P-1 Indiana I-1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plucking machine</td>
<td>8/15</td>
<td>Typhimurium T-1 Blockley B-1 Paratyphi B P-1 Indiana I-1 Montevideo M-1 M-2, M-3 Agona A-1, A-2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Evisceration</td>
<td>2/14</td>
<td>Typhimurium T-1 Paratyphi B P-1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crates</td>
<td>1/6</td>
<td>Typhimurium T-1</td>
</tr>
<tr>
<td>Feathers before scalding</td>
<td>3/3</td>
<td>Typhimurium T-1</td>
</tr>
<tr>
<td>Feathers after scalding</td>
<td>2/3</td>
<td>Agona A-2</td>
</tr>
<tr>
<td>Feathers during plucking</td>
<td>3/3</td>
<td>Montevideo M-1 Rissen</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neck skins after plucking</td>
<td>13/30</td>
<td>Typhimurium T-1, T-2 Blockley B-1 Paratyphi B P-1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neck skins after evisceration</td>
<td>15/30</td>
<td>Minnesota Mi-1</td>
</tr>
</tbody>
</table>
Results:

- 2/3 slaughterhouses contaminated on slaughter line before start slaughter activities:
  - shackles & wheels, plucking machine, scalding
  - containers

strains from *Salmonella* colonized flocks in the week before

\[ \text{slaughter line} \]

\[ \text{contamination of flocks} \]
OVERVIEW

1. Situation of the problem
2. Characteristics of *Salmonella* and *Campylobacter*
3. Clinical aspects of *Salmonella* and *Campylobacter* infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of *Salmonella* isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. *Salmonella*: impact of the slaughter line contamination on carcass contamination
10. *Campylobacter*: contamination or colonization of poultry flocks after transport
11. Conclusions
10. *Campylobacter*: contamination or colonization of poultry flocks after transport

**Observation:** flocks slaughtered on same day had same strains in intestines

**Hypothesis:** flocks become colonized during transport

**M&M:**
- 7 *Campylobacter*-free flocks
- at the farm: just before depopulation
  - samples from cecal droppings & breast
  - samples from 5 containers
- at the slaughter house:
  - samples from breast, feet, head
  - samples from crop & gastrointestinal tract
**Results:**
-25/35 container *Campylobacter* positive (54% *C. jejuni*, 46% *C. coli*)
-15 by direct plating
-10 by enrichment

<table>
<thead>
<tr>
<th>Flock 1</th>
<th>Cont. 1</th>
<th>Cont. 2</th>
<th>Cont. 3</th>
<th>Cont. 4</th>
<th>Cont. 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>90</td>
<td>5</td>
<td>3</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Flock 2</td>
<td>+</td>
<td>+</td>
<td>1</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Flock 3</td>
<td>82</td>
<td>12</td>
<td>2</td>
<td>14</td>
<td>27</td>
</tr>
<tr>
<td>Flock 4</td>
<td>5</td>
<td>NC</td>
<td>+</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Flock 5</td>
<td>+</td>
<td>NC</td>
<td>1</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flock 6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flock 7</td>
<td></td>
<td>+</td>
<td></td>
<td></td>
<td>+</td>
</tr>
</tbody>
</table>

*cfu/cm²*
Results:
- 25/35 container *Campylobacter* contaminated (54% *C. jejuni*, 46% *C. coli*)
- 3/7 flocks became colonized in last days before slaughter
  - no colonization, no co-colonization
  - limited external contamination

<table>
<thead>
<tr>
<th>Birds at the farm</th>
<th>5 Containers</th>
<th>Birds at slaughterhouse (external contamination)</th>
<th>Birds at slaughterhouse (intestines)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>C. jejuni</em> G-1</td>
<td><em>C. jejuni</em> G-2</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. jejuni</em> G-2</td>
<td><em>C. jejuni</em> G-3</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. coli</em> G-3</td>
<td><em>C. jejuni</em> G-4</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. jejuni</em> G-4</td>
<td><em>C. jejuni</em> G-5</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. jejuni</em> G-5</td>
<td><em>C. jejuni</em> G-6</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. jejuni</em> G-6</td>
<td><em>C. jejuni</em> G-7</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>C. jejuni</em> G-7</td>
<td><em>C. jejuni</em> G-8</td>
<td></td>
</tr>
</tbody>
</table>
OVERVIEW

1. Situation of the problem
2. Characteristics of Salmonella and Campylobacter
3. Clinical aspects of Salmonella and Campylobacter infections
4. Poultry flocks
5. Aim of the study
6. Molecular discrimination of Salmonella isolates at serotype level
7. Materials and Methods
8. Contribution of gastrointestinal colonization and cross-contamination to carcass contamination during slaughter
9. Salmonella: impact of the slaughter line contamination on carcass contamination
10. Campylobacter: contamination or colonization of poultry flocks after transport
11. Conclusions
11. Conclusions

**Salmonella**
- 13% of flocks colonized; 55% of flocks contaminated after slaughter
- cross-contamination was important
- caused by contamination of the slaughter equipment

**Campylobacter**
- 72% of flocks colonized; 79% of flocks contaminated after slaughter
- carcass contamination caused by gastrointestinal colonization
- transport containers often contaminated → limited external contamination