Microorganisms Causing Foodborne Disease

*Enterobacteriaceae* Family
Genus *Salmonella*

Family *Enterobacteriaceae*

- Gram-negative
- Non-sporeforming rods
- Facultatively anaerobic
- Oxidase negative
- Glucose fermentative
- Generally motile

*Peritrichous flagella*
**Salmonella**

- Named after Dr. D. E. Salmon
- First described *Salmonella cholerae-suis*
- Currently only 2 species are recognized
  - *S. bongori*
  - *S. enterica*
    - 6 subspecies
      » enterica
      » salamae
      » arizonae
      » diarizonae
      » houtenae
      » Indica

**Principal biochemical characteristics**

- Lactose negative
- Lysine decarboxylase positive
- H₂S positive
- Urease negative

- 0.8% of strains can ferment lactose
- A few strains do not produce H₂S

**Salmonella**

- Salmonellae are separated in somatic groups (O antigens) and flagellar types (H antigens)
- About 2,400 serovars (or serotypes) have been described
- Example of correct nomenclature:
  - *Salmonella enterica* subsp. *enterica* serovar Typhimurium
  - Abbreviated: *Salmonella* Typhimurium
- After having defined by full name
- Also there is a phage-based classification
  - Phage types
Growth characteristics

- Temperature
  - Minimum 7°C
  - Maximum 47°C
  - Optimum 37°C
- pH
  - Minimum
    - Acetic acid 4.7
    - HCl 4.05
  - Optimum 7.0
- aw
  - Minimum 0.93

Heat resistance

- Non-thermoduric
- S. Senftenberg 775W is heat resistant
  - $D_{71}$ in milk = 0.09 min
  - $D_{71}$ for S. Typhimurium in milk = 0.003 min
- Heating mestrum affects heat resistance
  - $D_{70}$ for S. Typhimurium in chocolate sauce = 11.3 – 17.5 h.

Ecology

- Most salmonellae infect a wide range of animal species
- Some serovars are host adapted
  - S. Enteritidis PT4, S. Pullorum, S. Gallinarum in poultry
  - S. Cholerae-suis in pigs
  - S. Typhy in humans
Clinical features

- Enteritis
  - Diarrhea, nausea and fever
- Incubation period
  - 6 - 48 h
- Infectious dose
  - 1 to 10^6 cells

Pathogenesis

- Pathogenesis
  - Adhesion to epithelial cells of the ileum via mannose-resistant fimbriae
  - Bacterial uptake by host cells (endocytes)
  - Passage through epithelial cells within a vacuole, where the bacterium multiplies
  - Influx of inflammatory cells leading to the release of prostaglandins
  - Prostaglandins activate adenylate cyclase which produces fluid secretion to the intestinal lumen

Activity in foods

- Low \( a_w \) enhances the survival of *Salmonella*
  - The lower the \( a_w \) the greater the survival rate

![Survival of Salmonella in marigue powder as affected by \( a_w \)](image)
Survival of *Salmonella* in different foods

<table>
<thead>
<tr>
<th>Commodity</th>
<th>Temperature</th>
<th>Survival time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Butter</td>
<td>-23 to 25°C</td>
<td>&gt;10 weeks</td>
</tr>
<tr>
<td>Milk</td>
<td>Room temp.</td>
<td>6 months</td>
</tr>
<tr>
<td>Ice box</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Various vegetables</td>
<td>2 to 4°C</td>
<td>&gt;28 days</td>
</tr>
<tr>
<td>Chocolate</td>
<td>Room temp.</td>
<td>Months</td>
</tr>
</tbody>
</table>

Foods associated with *Salmonella* infection

- Eggs
  - *Salmonella* serovar Enteritidis
- Meat
  - *S. Typhimurium*
- Fruits and vegetables
  - *S. Montevideo* (tomatoes)
  - *S. Poona* (cantaloupes)
  - *S. Newport* (mangoes)
- Cheese
  - *S. Zanzibar*
- Chocolate
  - *S. Eastbourne*
- Milk
  - *S. Typhimurium*
  - *S. Newbrunswick*

Transmission routes

*Figure 7.8 The Salmonella cycle of infection (Reproduced with permission from WHO, 1983)*
### S. Enteritidis
- Associated with eggs
- Present in the yolk at oviposition
- Food associated:
  - Homemade mayonnaise w/raw eggs
  - Tartar sauce
  - Egg nogg
  - Milk shakes
  - Mousse
  - Ice cream
- Control
  - Decreasing S. Enteritidis populations at the farm
  - Refrigerating eggs
  - Egg pasteurization or irradiation

### S. Typhimurium DT 104
- First reported in the UK in 1984
- Increasing prevalence
- Isolated from food animals
  - As well as other domestic and wild animals
- R-factor as a natural DNA component
  - Typically resistant to ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline
  - R-type ACSSuT

### DT 104
- Resistant to multiple antibiotics
- UK reported case-fatality rate = 3%
  - 0.1% for other non-typhoid salmonellae
- R-type ACSSuT present in 30% of human isolates in the U. S.
  - Resistance to trimethoprim, fluoroquinones and ciprofloxacin is being reported
Outbreak of salmonellosis caused by DT 104 in the U.S.

- Nebraska, 1996
- 19 cases
  - Elementary school children
- 3 required hospitalization
- Vehicle implicated: cold chocolate milk
  - Relative Risk=8.2
- Isolates were R-type ACSSuT
- Phage typing confirmed DT 104

Isolation methods

- Methods include enrichment prior to plating on selective media
- Most methods for food analysis also include a pre enrichment
- Enrichment in a non-selective broth
  - Lactose broth
- After enrichment, a rapid method can be used
  - Immunoassays (ELISA, 1-2-3)
  - DNA probes

Isolation methods (cont.)

- If rapid method indicates + test, *Salmonella* must be confirmed by plating method
- Examples of enrichment broths
  - Tetrathionate broth
  - Selenite cystine broth
  - Rappaport-Vassiliadis
- Examples of plating media
  - Brilliant green agar
  - XLD agar (xylose lysine deoxycholate)
  - Bismuth sulfite agar
Isolation methods (cont.)
Typical colonies on plating media are subjected to identification tests
• Biochemical tests
  - TSI agar
  - LIA agar
  - Urea broth
• Serological tests
  - Polyvalent O antiserum
  - Serogrouping and serotyping