



Some statistics on *E. coli* O157

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|--------------------------------------------------|----------------------|
| 8 O157 <i>eae</i> + <i>vtx1a</i> | NO HUS |
| 85 O157 <i>eae</i> + <i>vtx1a</i> + <i>vtx2c</i> | NO HUS |
| 7 O157 <i>eae</i> + <i>vtx1a</i> + <i>vtx2a</i> | 2/6 (33%) with HUS |
| 37 O157 <i>eae</i> + <i>vtx2a</i> | 11/37 (30%) with HUS |
| 45 O157 <i>eae</i> + <i>vtx2a</i> + <i>vtx2c</i> | 11/45 (24%) with HUS |
| 31 O157 <i>eae</i> + <i>vtx2c</i> | 1/31 (3%) with HUS |

HUSEC: *vtx2a* and *eae*

Current outbreak:

O157 *eae* + *vtx1a* + *vtx2a* 7/12 (58%) with HUS



4th external quality assurance (EQA-4) scheme & PT10 EU-RL for typing of VTEC in 2012-2013

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ECDC contracts for 4 years

ECDC framework contracts with the overall purpose of

- Performing surveillance of foodborne infections based on isolate characteristics (subtyping) → outbreak detection
- Ensure that comparable methods are used in all countries
- Pathogens covered:
 - Salmonella
 - Listeria
 - Vero cytotoxin-producing *E. coli*

CONTRACT 1: EQA PROGRAMMES

MICROBIOLOGICAL CHARACTERISATION SERVICES

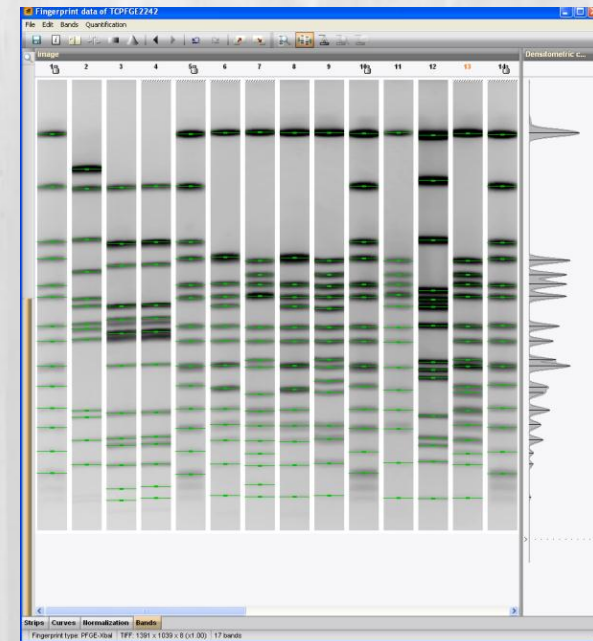


- **3 EQA programmes each year (4 years):**
 - >30 laboratories; 10-60 strains in each EQA
- **Methods:**
 - Pulsed-field gel electrophoresis (PFGE) for all 3 pathogens
 - MLVA for *Salmonella* Typhimurium
 - Phage typing of *Salmonella* (subcontractor: DTU-Food)
 - *E. coli* serotyping and virulence characterisation
 - *Listeria* serotyping – molecular and conventional (subcontractor: ANSES)
- **Trouble-shooting regarding PFGE and MLVA**
- **The MLVA method: maintain and extend the reference material**
- **Perform molecular typing for other countries, including data analysis and storage of strains**
- **Protocols, certificates, individual reports, report, scientific paper**
- **Since start Sept 1: protocols, invitation letter, selection of strains...**



CONTRACT 2: EXPERT MANAGEMENT (curator)

- Expert management of molecular typing data (curator)
- BioNumerics databases with molecular typing data maintained at ECDC, but as lab-experts we shall:
 - Data validation (quality, normalisation, band assignment)
 - Assign nomenclature (reference types)
 - Analyse for clusters
 - Give feedback to laboratories
 - Give expert advice to ECDC regarding interpretation of data, clusters/outbreaks, SOPs, etc
- Expected amount of typing data for curation
 - Salmonella: 5000 isolates per year
 - Listeria: 2000 isolates first year; 500 per following years
 - VTEC: 500 isolates per year
- **September-October: kick-off meeting pilot project, our input to SOPs...**





4th external quality assurance (EQA-4) scheme & PT10 EU-RL for typing of VTEC in 2012-2013

Strains and methods

Strains selected for the EQA-4 programme is based on five criteria:

- they represent commonly reported strains in Europe
- they remain stable during the preliminary testing period at the organising laboratory
- they are easy to type
- they represent the three different subtypes of *vtx1* and the seven different subtypes of *vtx2* in positive and stable strains
- the PFGE profile is stable and represent the diversity of the occurring VTEC strains in Europe



EQA-4 & PT10 includes the following methods

- molecular typing: Pulsed Field Gel Electrophoresis (PFGE)
- serotyping O:H
- virulence determination:
 - genotyping of VTEC virulence genes: *vtx1*, *vtx2*, *eae*, *aaiC* and *aggR*
 - subtyping of *vtx1* (*vtx1a*, *vtx1c* and *vtx1d*) and *vtx2* (*vtx2a* to *vtx2g*)
- phenotypic testing
 - production of Verocytotoxin/Shiga toxin
 - production of Extended Spectrum Beta Lactamases (ESBL)
 - production of β -glucuronidase
 - production of enterohaemolysin
 - fermentation of sorbitol



Test panel

- In total, 15 blinded *E. coli* test strains, including 11 strains that will be used for PFGE

If requested

- 9 reference control strains covering all the established variants of *vtx1* and *vtx2* genes
- the global standard strain S. Braenderup (if participation in EQA PFGE)



Deadlines

20th of Nov. 2012

Response to the invitation to ecoli.ega@ssi.dk

26th of Nov. 2012

-3rd of Dec. 2012

Shipment of test and control strains

25th of Feb. 2013

Deadline for submission of the results



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Head



Mia Torpdahl
Salmonella



Jonas Larsson
Listeria



Søren Persson
Curator