

Clusterdetection and interpretation using cgMLST and SNP analysis

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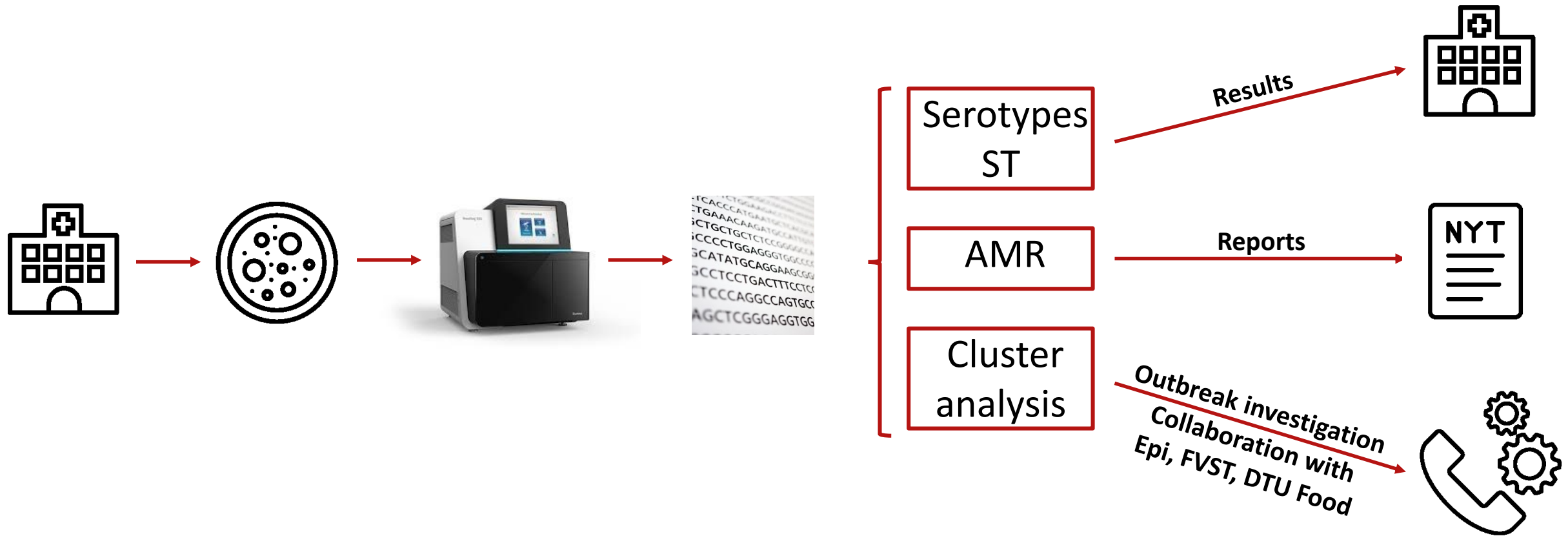


Content

- **Surveillance of *Salmonella* in Denmark**
 - Outbreak definition and communication
 - Cluster analysis and detection
 - Examples on cluster detection
- **Surveillance of *Campylobacter* in Denmark**

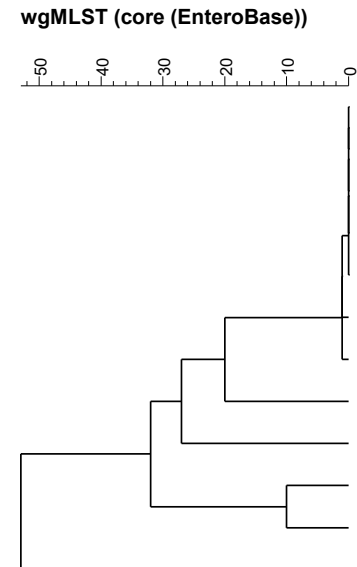
WGS-based surveillance of *Salmonella* in Denmark

- Real time surveillance since 2018
- Flow from Hospital to SSI



Outbreak definitions and communication

- **Outbreak definition**
 - Genetic cluster of ≥ 4 isolates within 3 months
- **Communication**
 - Genetic cluster ID
 - Outbreak ID
- **Retrospective analysis**
 - Cases to existing cluster – new event
 - Same cluster ID but different outbreak ID



ST19#65
FUD2102

EPI

- Outbreak investigation
- Patient interview
- Contact with food authorities

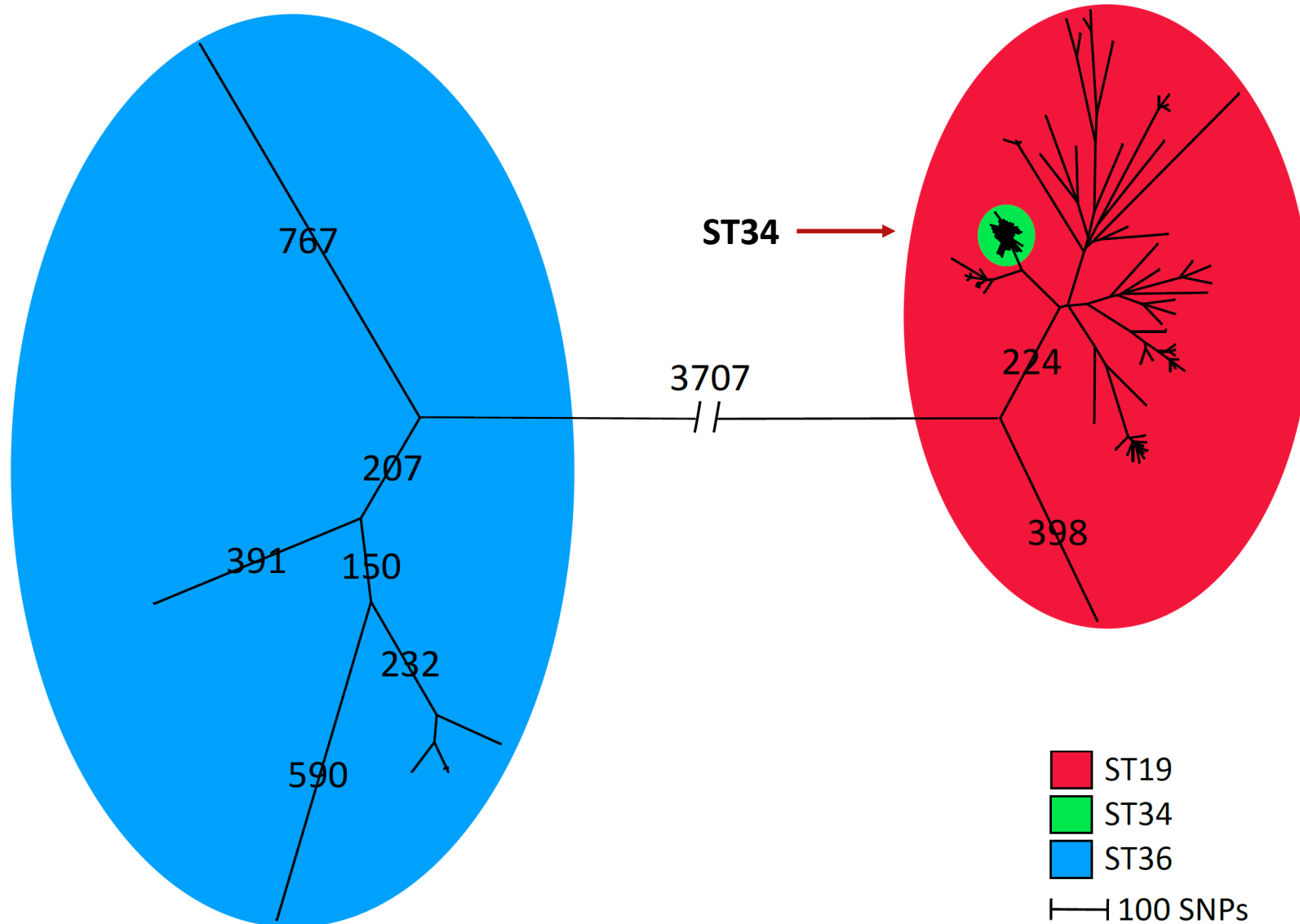
Cluster analysis and detection

- **Cluster detection**
 - cgMLST and single linkage clustering
 - Applied Maths/Enterobase scheme - Bionumerics
 - Cluster cut-off
 - ≤ 3 AD
 - ≤ 1 AD for clonal types (Enteritidis ST11 and monophasic Typhimurium ST34)
- **Inclusion criteria - not one fits all**
 - Type, time, place
 - Typing method (SNP, cg/wgMLST)
 - Clustering methods

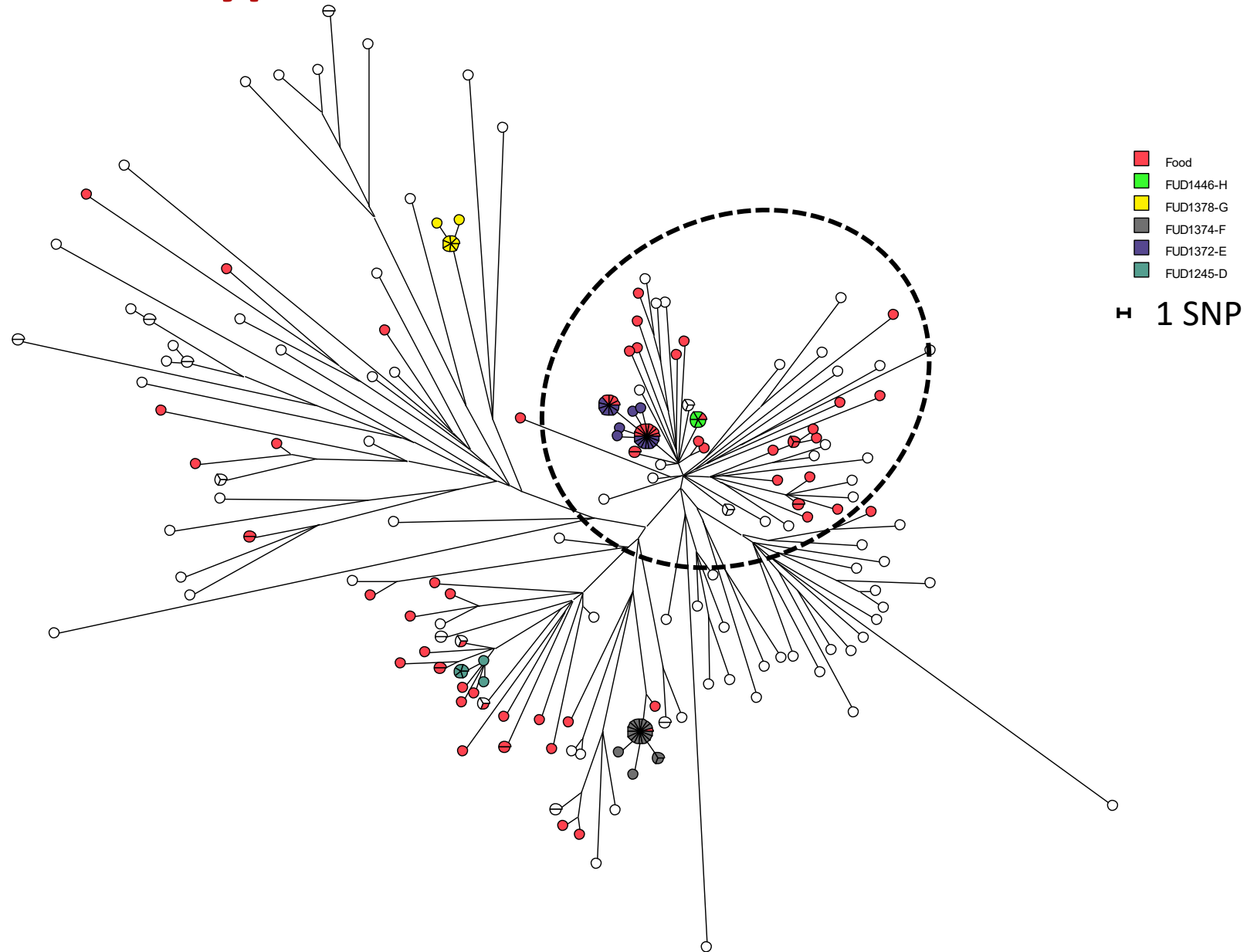
Clonal types

- **Enteritidis ST11 and monophasic Typhimurium ST34**
 - New clones
 - Little genetic diversity
 - Challenging cluster analysis
 - Supporting analysis – SNP or wgMLST
 - Additional analysis – AMR, plasmids, prophages
 - Other clustering methods
 - More strict cluster cut-off

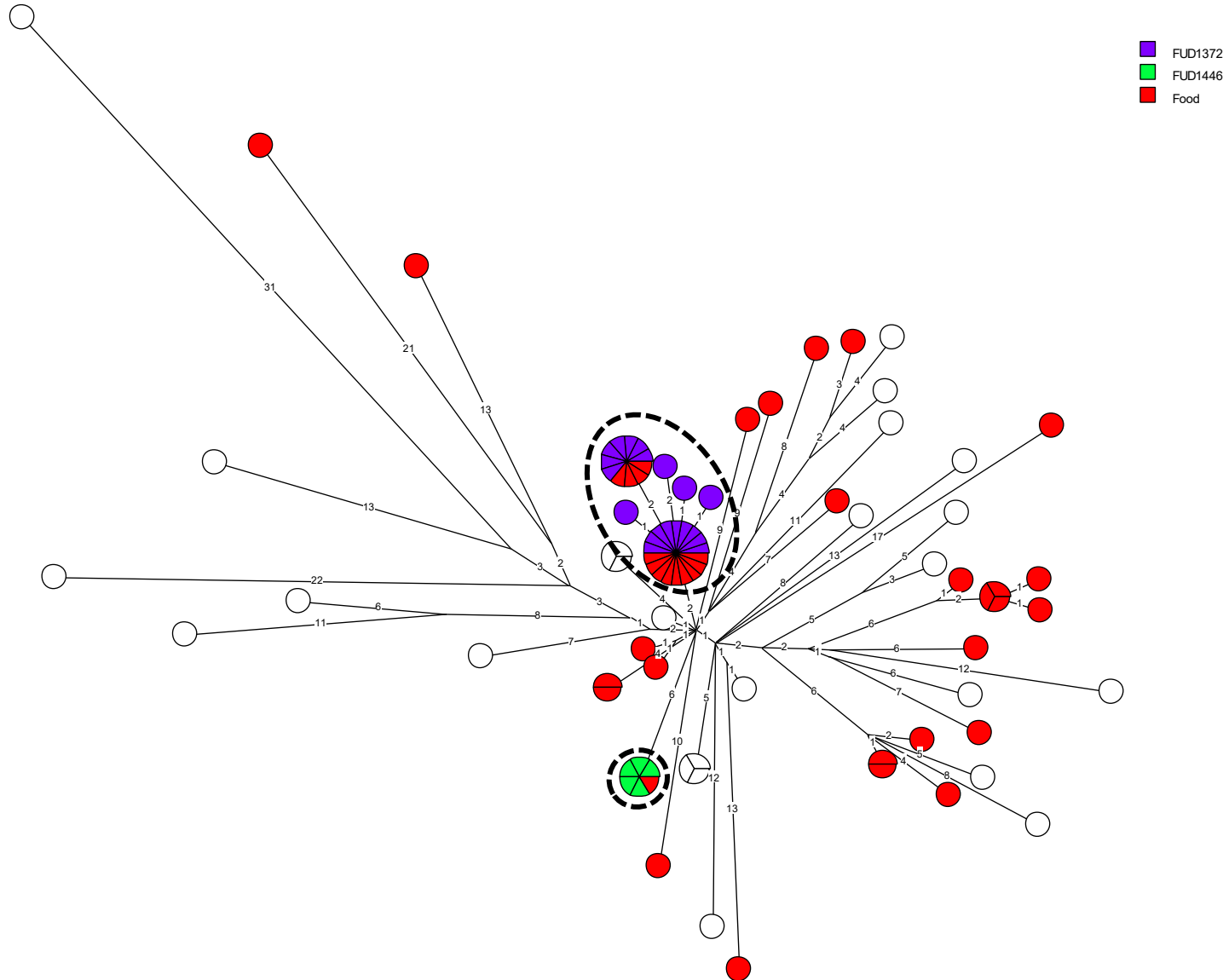
Clonal types – *S. Typhimurium* ST34



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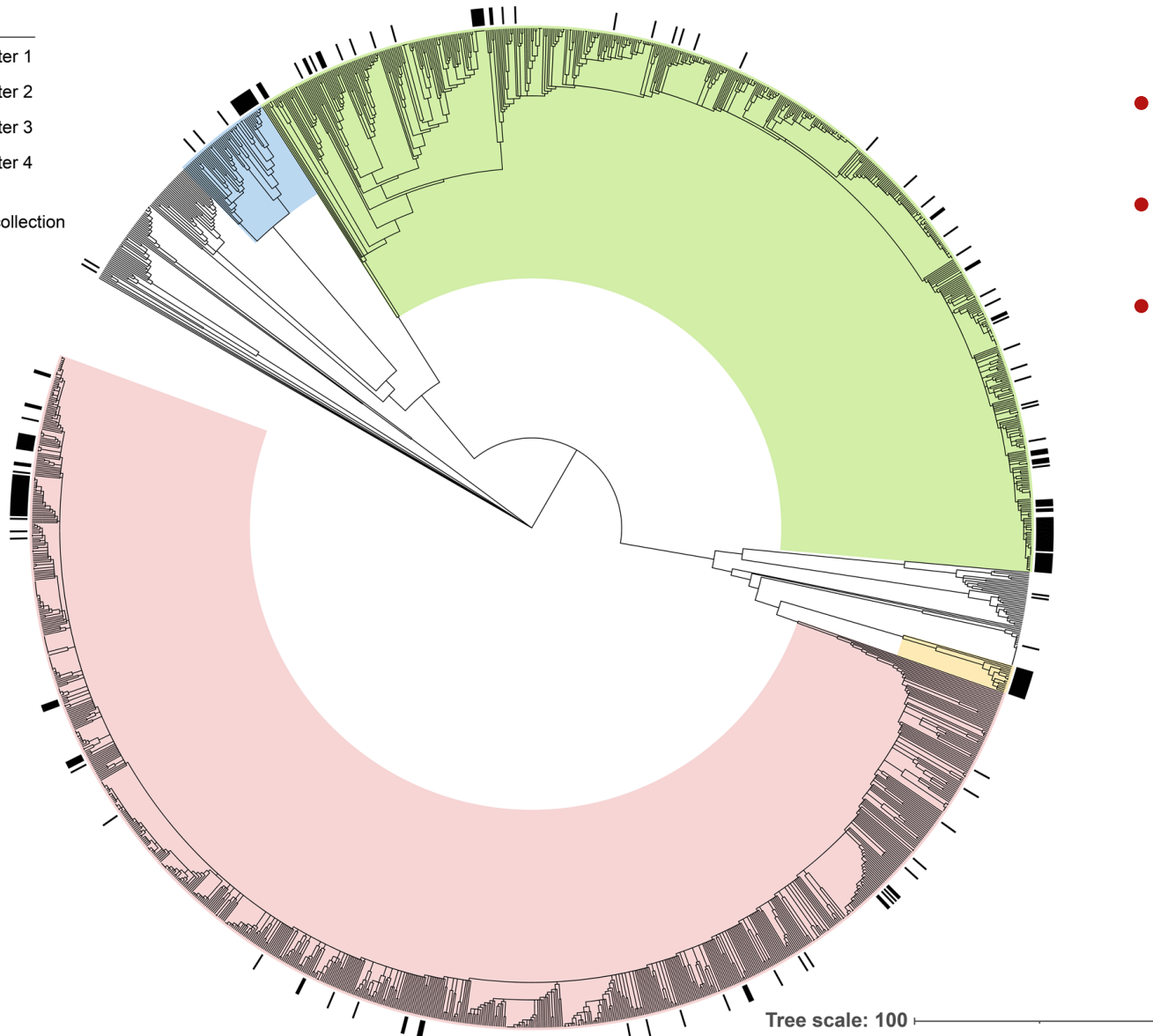


- **Outbreak FUD1372**
 - Defined by small plasmid
- **Outbreak FUD1446**
 - Defined by specific epi-data

Clonal types – *S. Enteritidis* ST11

Clusters

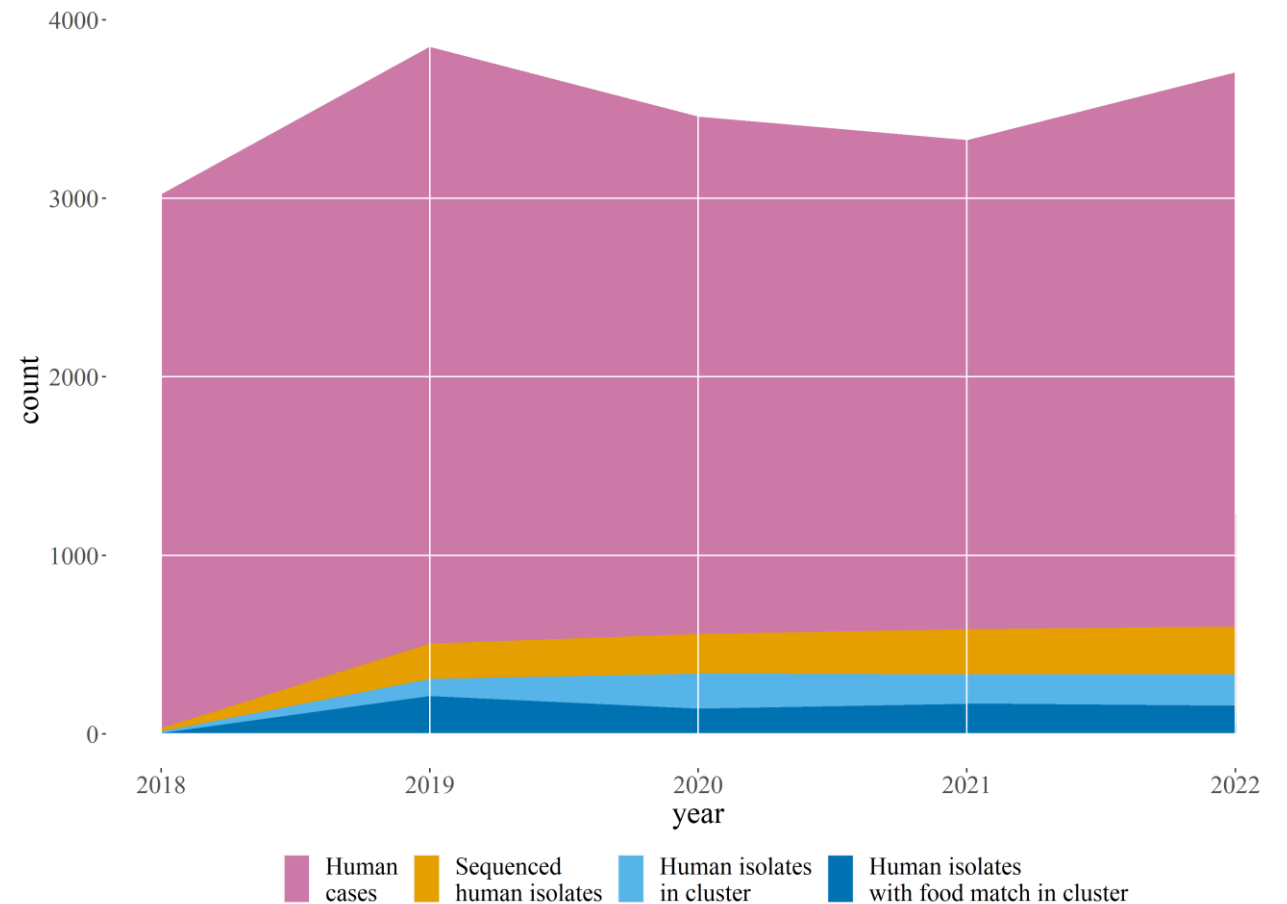
- Cluster 1
- Cluster 2
- Cluster 3
- Cluster 4
- DK collection

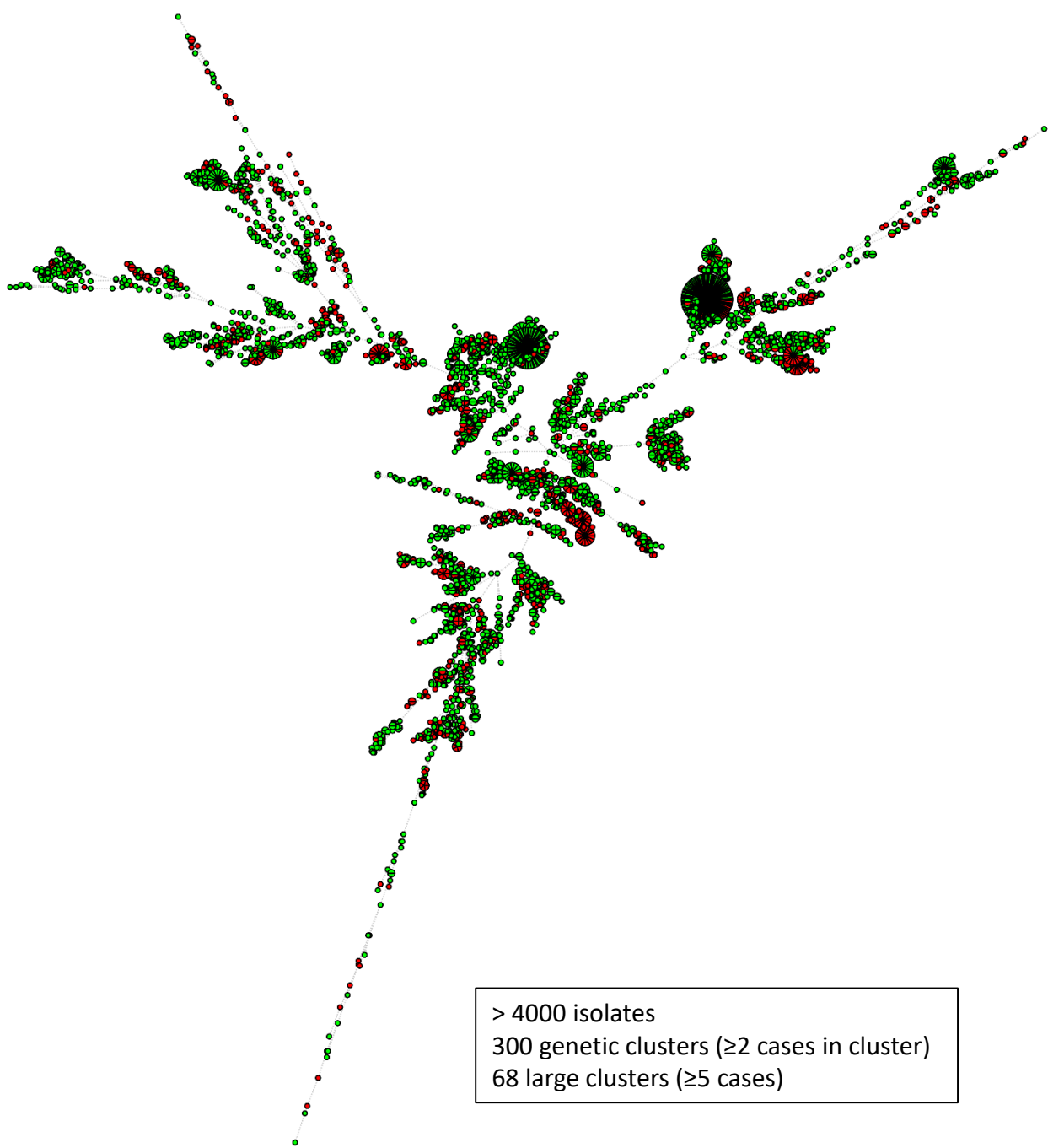


- 2 major clones in Europe
- Extremely little diversity
- **Cluster definition**
 - Strict cut-off
 - UPGMA instead of single linkage
 - Additional wgMLST analysis

WGS-based surveillance of *Campylobacter jejuni/coli*

- Performed routinely since 2019
- Isolates from 10-15% of human cases
- 4/5 regions of Denmark represented
- Food data analyzed routinely at SSI
 - To find food matches





> 4000 isolates
300 genetic clusters (≥ 2 cases in cluster)
68 large clusters (≥ 5 cases)

- *Campylobacter* overall very diverse
- cgMLST and single linkage
 - ≤ 4 AD cut-off
- $\sim 50\%$ of human isolates form clusters
 - mostly small, some big
- 25-30% of human isolates match to food isolates (mostly chicken)

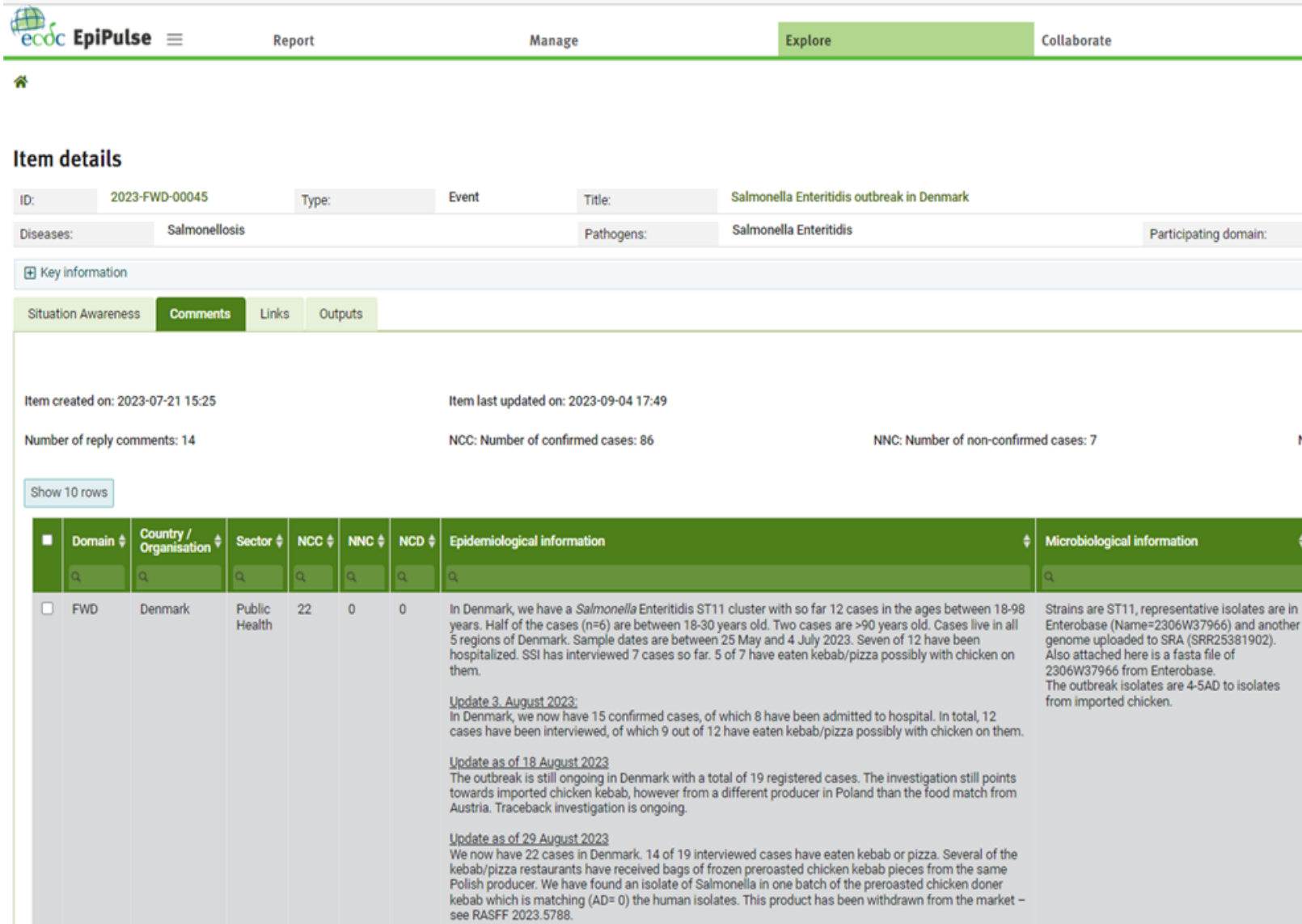


Concluding remarks

- **Not one fits all**
 - Interpretation of results from cluster to cluster
 - Find the method that fits the data and types seen in your country
 - Do validation using well defined outbreaks
- **Clear definitions and communication is important**
 - Clear information on tools and methods
 - Clear information on cluster and outbreak definition (type, time, place)
 - Better understanding between lab and epi
 - Better understanding between countries
- **Epi data is extremely important**
- **It's a working progress – ongoing evaluation of data is needed**

- **Questions?**
 - Is it better to include too many or too few?

Epipulse event – Outbreak of Enteritidis ST11



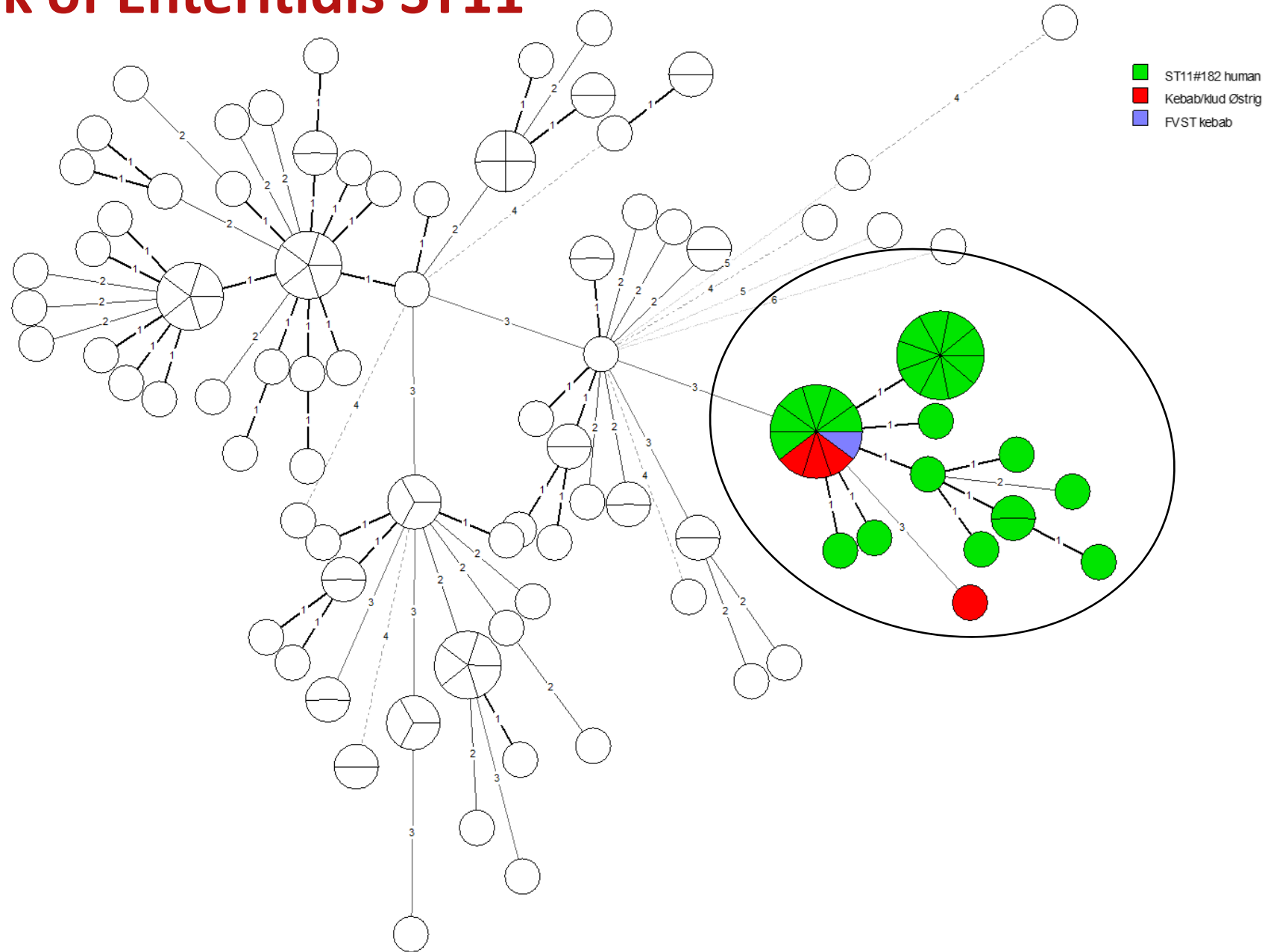
The screenshot shows the EpiPulse interface with the following details:

- Item details:** ID: 2023-FWD-00045, Type: Event, Title: Salmonella Enteritidis outbreak in Denmark
- Diseases:** Salmonellosis, **Pathogens:** Salmonella Enteritidis, **Participating domain:**
- Key Information:** Situation Awareness, Comments, Links, Outputs
- Item created on:** 2023-07-21 15:25, **Item last updated on:** 2023-09-04 17:49
- Number of reply comments:** 14, **NCC:** Number of confirmed cases: 86, **NNC:** Number of non-confirmed cases: 7
- Table:** A table with columns for Domain, Country/Organisation, Sector, NCC, NNC, NCD, Epidemiological information, and Microbiological information.

Domain	Country / Organisation	Sector	NCC	NNC	NCD	Epidemiological information	Microbiological information
FWD	Denmark	Public Health	22	0	0	<p>In Denmark, we have a <i>Salmonella</i> Enteritidis ST11 cluster with so far 12 cases in the ages between 18-98 years. Half of the cases (n=6) are between 18-30 years old. Two cases are >90 years old. Cases live in all 5 regions of Denmark. Sample dates are between 25 May and 4 July 2023. Seven of 12 have been hospitalized. SSI has interviewed 7 cases so far. 5 of 7 have eaten kebab/pizza possibly with chicken on them.</p> <p><u>Update 3. August 2023:</u> In Denmark, we now have 15 confirmed cases, of which 8 have been admitted to hospital. In total, 12 cases have been interviewed, of which 9 out of 12 have eaten kebab/pizza possibly with chicken on them.</p> <p><u>Update as of 18 August 2023</u> The outbreak is still ongoing in Denmark with a total of 19 registered cases. The investigation still points towards imported chicken kebab, however from a different producer in Poland than the food match from Austria. Traceback investigation is ongoing.</p> <p><u>Update as of 29 August 2023</u> We now have 22 cases in Denmark. 14 of 19 interviewed cases have eaten kebab or pizza. Several of the kebab/pizza restaurants have received bags of frozen pre-roasted chicken kebab pieces from the same Polish producer. We have found an isolate of <i>Salmonella</i> in one batch of the pre-roasted chicken doner kebab which is matching (AD= 0) the human isolates. This product has been withdrawn from the market – see RASFF 2023.5788.</p>	Strains are ST11, representative isolates are in Enterobase (Name=2306W37966) and another genome uploaded to SRA (SRR25381902). Also attached here is a fasta file of 2306W37966 from Enterobase. The outbreak isolates are 4-5AD to isolates from imported chicken.

- National outbreak of ST11 related to chicken kebab
- Epipulse - event
 - Epidata
 - Mikrobiological data
 - Attached sequence
- Turned into a international outbreak

Outbreak of Enteritidis ST11





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Polsk kyllingekebab er kædet sammen med dansk salmonellaudbrud

Siden maj er 22 personer registreret i Danmark med den samme salmonellatype, som også har gjort folk syge i andre europæiske lande. Nu har efterforskningen vist, at importeret kyllingekebab fra Polen også er smitekilden til de danske tilfælde.



Internationalt udbrud af *Salmonella* Enteritidis ST11 relateret til Polsk kyllingekebab

Pernille Gymoese, FBI – Fødevarebårne Infektioner