



# Whole genome sequencing initiatives and activities at the European Centre for Disease Prevention and Control

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# Outline of the presentation – ECDC activities comprising WGS



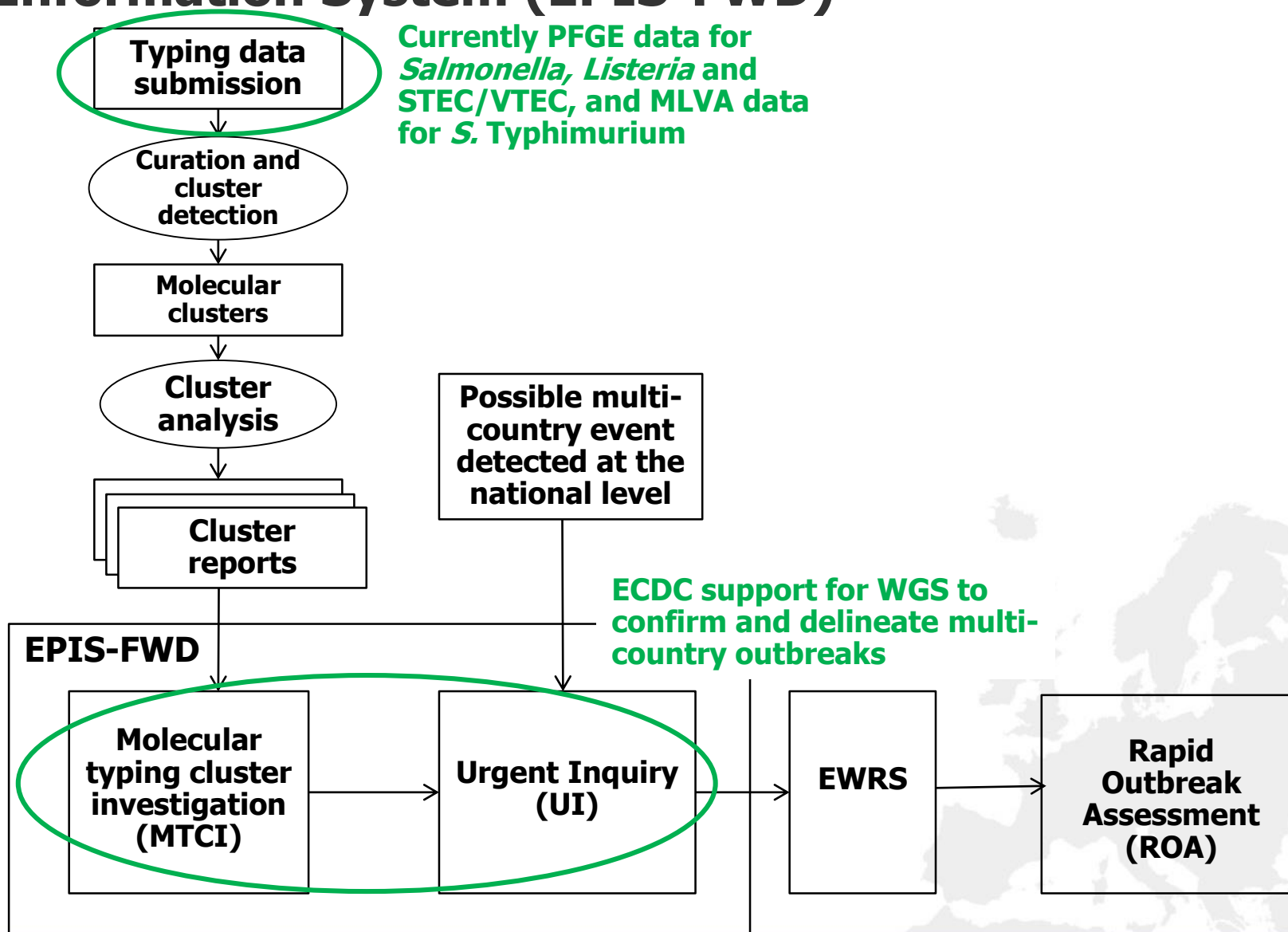
1. Background on ECDC FWD surveillance involving molecular typing data
2. ECDC-supported WGS services to support multi-country outbreak investigations
  - Case example: *Salmonella* Enteritidis MLVA type 2-9-7-3-2
3. FWD-NEXT expert group
4. Future plans for WGS in EU/EEA-level FWD surveillance

# Working objectives for molecular typing of the FWD bacteria



1. Early detection of multi-country FWD outbreaks and/or dispersed clusters
2. Detection and investigation of transmission chains and relatedness of microbial clones
3. Identification of persistent FWD strains causing human infections in EU and likely originating from continuous sources
4. Facilitation of trace back and trace forward investigations
5. Identification of new risk factors as strains can be linked more accurately to epidemiological data

# FWD surveillance in Epidemic Intelligence Information System (EPIS-FWD)



# WGS implementation in EU/EEA Member States – survey in May 2015

## Status of WGS implementation

	WGS has already replaced existing methods	WGS complement existing methods	WGS will complement existing methods in the next year or two	No plans to apply WGS-based methods	Not Known	Total
<i>Salmonella</i>	0	3	11	13	1	28
<i>Listeria</i>	1	5	9	11	2	28
STEC/VTEC	1	9	8	12	3	27

# Process for WGS support to FWD cross-border outbreak investigations



- Trigger: Signal where WGS may be helpful to verify and/or delineate a multi-country outbreak
- Involved Member States are asked if they are willing to share their WGS data or ship isolates for WGS to ECDC's contractor (SBS)
- WGS data is collected and analysed
- Results are shared with the involved countries
- Results may lead to further public health action and hopefully further investigations with food/veterinary sector

# Outbreak investigations supported with WGS



Pathogen	Signal	Total WGS (No of MS)	SBS	Own WGS	Cluster
<i>S. Oranienburg</i>	Urgent inquiry	237 (11)	72 (7)	165 (4)	46 (5)
<i>S. Typhimurium</i> 3-17-10-NA-211	Molecular typing	41 (8)	36 (6)	5 (2)	14 (6)
<i>S. Enteritidis</i> 2-9-7-3-2	Urgent inquiry	119 (6)	25 (3)	94 (4)	88 (3)
<i>S. Braenderup</i> (on-going)	Urgent inquiry	148 (9)	52 (4)	96 (6)	45 (3)

## Status:

- *S. Oranienburg*: No new cases detected since Dec 2015; scientific article in progress
- *S. Typhimurium* 3-17-10-NA-211: No new cases detected since Dec 2015
- *S. Enteritidis* 2-9-7-3-2: No new cases detected since Feb 2015; abstracts sent to I3S, FAO-GMI and ESCAIDE conferences. Bioinformatics pipeline comparison in progress
- *S. Braenderup*: WGS data available from 9 countries, data analysis on-going

# Urgent Inquiry on *S. Enteritidis*: Summary of events

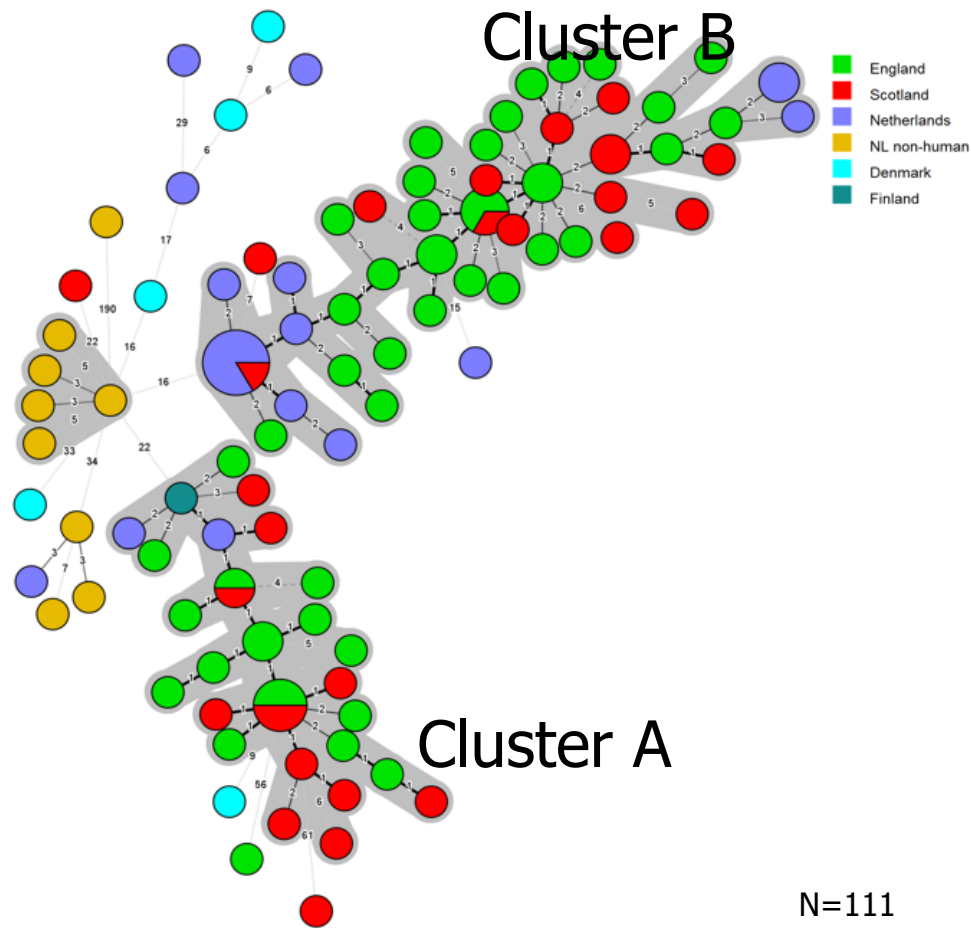
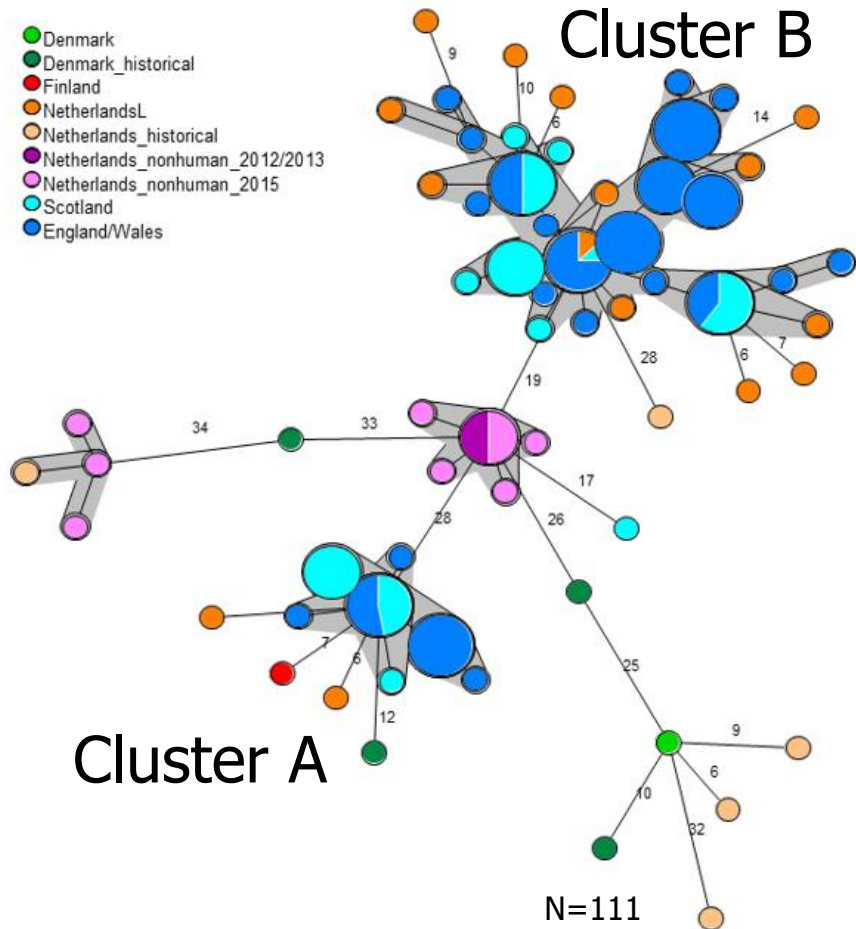


- January 2016: Scotland reported an unexpected increase in *S. Enteritidis* PT8 MLVA type 2-9-7-3-2
  - Cases also reported in England and Wales by Public Health England, whose WGS analysis reveals two distinct clusters (clusters A and B)
  
- Ten other countries replied to the urgent inquiry
  - Denmark, Finland and the Netherlands reported recent cases with the same MLVA type
  
- ECDC offered WGS support to affected countries
  
- WGS data analysis performed in parallel at Public Health England, Health Protection Scotland and the Dutch RIVM



# Methodology

- In-house SNP-based analytical approach
  - Used for the case definition, confirmed cases within 5 SNPs of the two clusters
- Ad hoc core genome MultiLocus Sequence Typing (cgMLST) approach using SeqSphere software
- Open access enterobase cgMLST scheme (<http://enterobase.warwick.ac.uk>)



Minimum spanning tree by ad-hoc cgMLST approach using SeqSphere with *S. Enteritidis* P125109 as reference, 3353 loci (RIVM)

Minimum spanning tree by cgMLST using the Enterobase platform (HPS)

# Results and conclusions from *S. Enteritidis* MLVA type 2-9-7-3-2 investigation



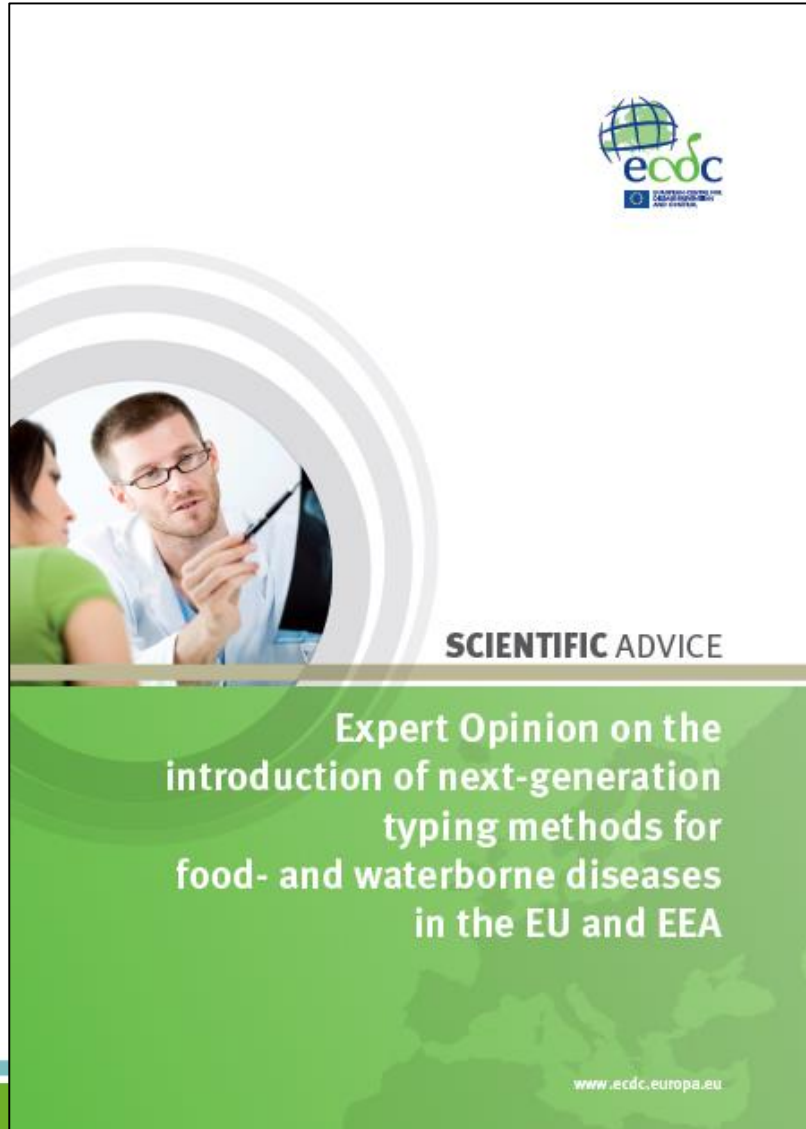
- A multi-country outbreak of *S. Enteritidis* PT8 with MLVA profile 2-9-7-3-2 had been ongoing in several EU countries since at least July 2015
- The presence of two distinct WGS clusters with a similar distribution in time and place suggests that there are potentially two outbreaks or alternatively a single outbreak which could be associated with two or more vehicles of infection
- All three analytical pipelines used confirmed the outbreak and existence of two distinct WGS clusters, although with some differences
- A more systematic comparison between the analytical pipelines will be performed

# Lessons learned from WGS support during multi-country investigations



- WGS is very effective for detecting and delineating outbreak clusters
- Epidemiological follow-up needs to be done in parallel to WGS
- Timeliness of WGS (shipping, sequencing & analysis) still need to be improved
- Data sharing and transfer to be streamlined
- Results analysis and interpretation
  - bioinformatic pipelines, cluster cut-offs, different sequencers, co-analysis of WGS and epi data, phylogenetic trees, visualisation software...
- Clear sampling strategy is very important

# Expert Opinion on the introduction of next-generation typing methods for food- and waterborne diseases



## Content:

1. Sample provision and sequencing
2. WGS data analysis for public health
3. Collaboration between organisations
4. Conclusions and future steps

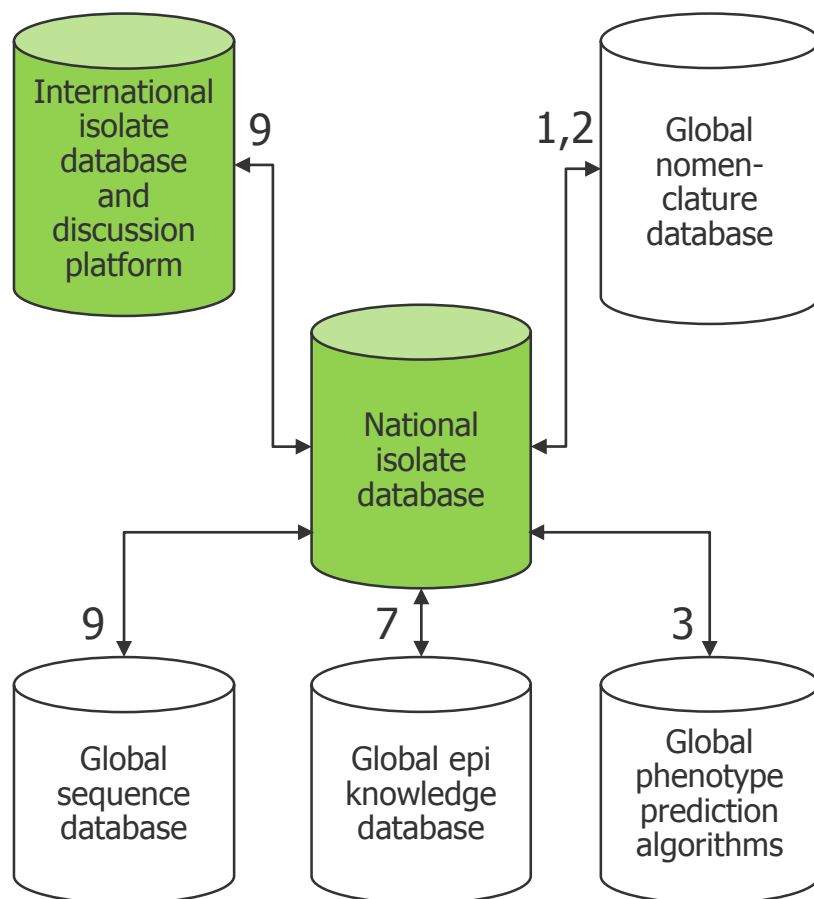
Link: <http://bit.ly/1Ggx6UO>, or through [ecdc.europa.eu](http://ecdc.europa.eu) / publications.

# FWD-NEXT Expert Group members



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# Model process for analysing WGS data for FWD pathogens



Proposed model process to analyse whole genome sequence data at national level.

Per batch of new isolates:

1. Assign whole genome MLST nomenclature.
2. Assign taxonomical nomenclature.
3. Predict relevant phenotypes.
4. Match against rest of database.
5. Cluster new isolates and matches using core genome MLST.
6. Refine each cluster with SNPs or shared genome MLST.
7. Associate prior epi knowledge with each cluster.
8. Decide on follow-up actions per cluster, if any.
9. Submit to international databases to compare further.

# Future plans for WGS in EU/EEA-level FWD surveillance

- Continue supporting multi-country outbreak investigations with WGS
  - including also other pathogens (*STEC/VTEC*, *Listeria*)
- Comparison of different WGS data analysis pipelines used at the national level
- Collection and analysis of WGS data to fulfil other EU/EEA molecular surveillance objectives
  - Detection of multi-country clusters, identification of persistent strains likely originating from continuous sources, identification of new risk factors as strains can be linked more accurately to epidemiological data
- To be published soon: ECDC Strategy to harness WGS for strengthening EU outbreak investigations and public health surveillance





# Acknowledgements



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