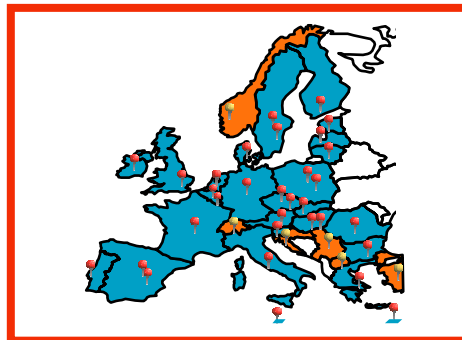


Development of a central repository for molecular typing data on VTEC strains isolated from food and animal sources

Stefano Morabito & Antonella Maugliani

EU Reference Laboratory for *E. coli*



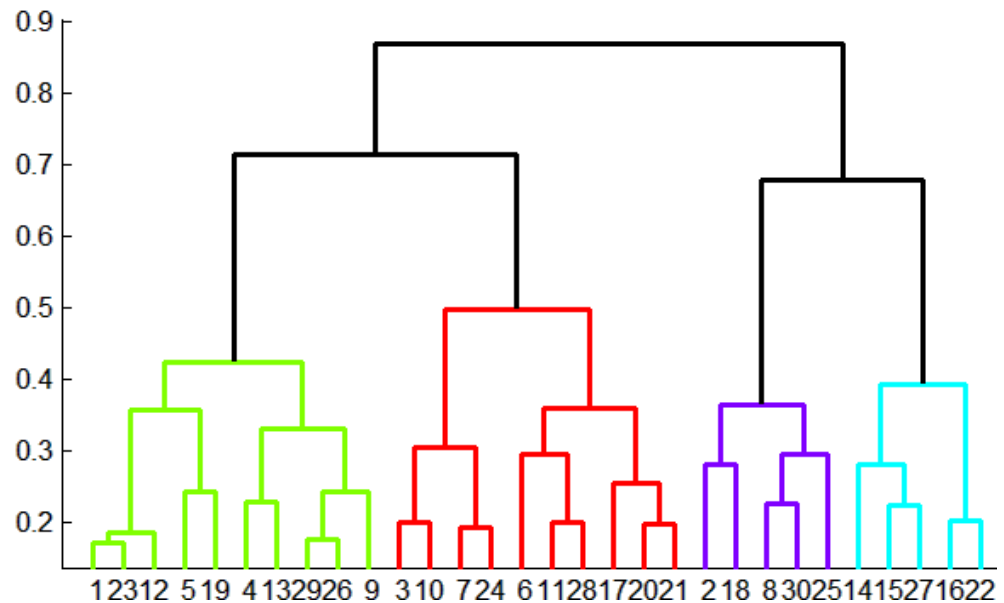
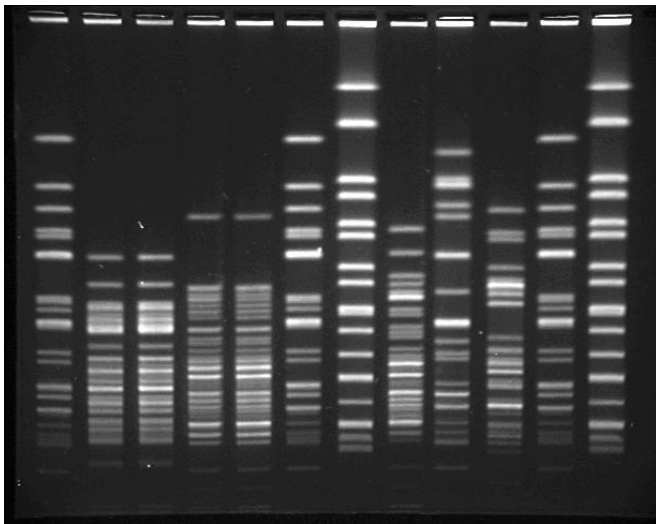
EU RL VTEC mandate - 2006-2011

Preparedness for VTEC identification, detection and characterisation

- ✓ Development and distribution of methods
- ✓ Ad hoc training (24 visits up to today)
- ✓ Ring trials (6+1 on seed testing)
- ✓ Capacity building of the NRLs network for molecular epidemiology in support to epidemiology of VTEC infections (1 PT on *vtx* typing)

Step forward

- ✓ Capacity building of the NRLs network for molecular epidemiology, to support the epidemiology of VTEC infections in the EU



Molecular surveillance



The International Molecular Subtyping Network for Foodborne Disease Surveillance

Africa, Asia Pacific, Canada, Europe, Latin America & Caribbean, Middle East, USA

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Key Supporters

Niid Logo

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Links that may provide useful information

[Reports and Newsletters](#)

A series of updates and hints from the PulseNet method development laboratory.



The technique in a nutshell



[Learn more](#) (much more)...

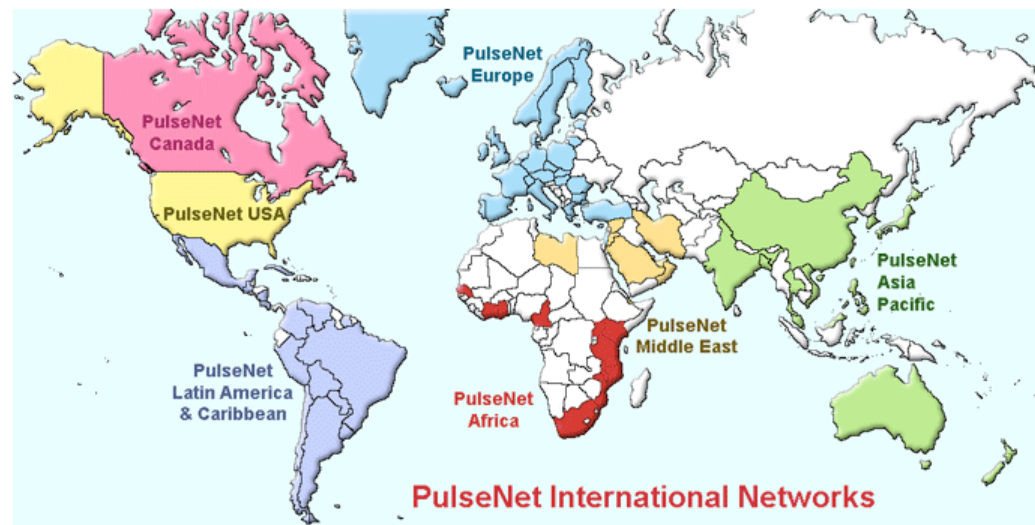
Welcome to PulseNet International.

Foodborne illnesses do not respect any borders. As a result of increasing international trade, food produced in one country may be consumed in a different part of the world and cause disease if contaminated with a foodborne pathogen.

Similarly, international travel is increasing and it is possible to get to almost any destination from almost any place in the world in a matter of hours. Therefore, a disease contracted in one part of the world may first become apparent thousands of miles away.

PulseNet International is a network of National and regional laboratory networks dedicated to tracking foodborne infections world-wide. Each laboratory utilizes standardized genotyping methods, sharing information in real-time.

The resulting surveillance provides early warning of food and waterborne disease outbreaks, emerging pathogens, and acts of bioterrorism.



What's available on this site?

Molecular typing of VTEC in Europe



The International Molecular Subtyping Network for Foodborne Disease Surveillance

Africa, Asia Pacific, Canada, Europe, Latin America & Caribbean, Middle East, USA

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Networks

PulseNet Europe

PulseNet Europe is the molecular surveillance network for food-borne infections in Europe. It is an internationally unique network that - in addition to public health laboratories - has institutions from the veterinary and food sector as equal participants.

PN Europe was funded from September 2004 until November 2006 by the FP network of excellence Med-Vet-Net, a virtual European Zoonosis Centre (www.medvetnet.org). The main aim was to establish real-time linked surveillance database system to detect infection clusters and investigate outbreaks of *Salmonella*, verocytotoxigenic *E. coli* (VTEC) and *Listeria monocytogenes* in Europe.

[Visit website](#)

A PN Europe communication system called [PNE forum](#), is used to exchange of active information and alert PN Europe partners when an international cluster is detected at the central database or partner has detected a cluster at the national level. The PFGE image of the profiles can also be attached to the alert email.

The quality of the PFGE profiles, which will be uploaded to the central database, and uniform naming are very important. Therefore, six PN Europe curators have been chosen. They take care of the central database, perform the naming and confirmation of the PFGE profiles submitted by partners and perform central cluster detection of all pathogens at regular intervals and alerts PN Europe partners of detected clusters through communication system.

A European Quality Assurance System (EQAS) system was created and certification of partners has been started to ensure the comparability of the data uploaded from different laboratories. The certified partners, who have signed the memorandum of understanding, which defines the rules for exchange of molecular typing data, have been given passwords to the central database.

A memorandum of understanding (MoU) between PN Europe partners has been signed by all partners to define the rules for collaboration allowing exchange of information and molecular typing data between PN Europe partners. A MoU with PN USA has also been drafted.

Why establishing a molecular typing repository at the EU-RL VTEC

Need for availability of molecular data on **non human** strains

The NRLs Network for *E. coli* is established and managed continuously

Skills, equipment and bioinformatic software are available at the EU_RL and in several NRLs

Expertise available on:

- **PFGE**
- ***Vtx genes-typing***
- **MLVA (O157)**
- **Phagetyping (O157)**

Why establishing a molecular typing repository at the EU-RL VTEC

The data collected and stored will be accessible by EC institutions involved in VTEC surveillance, monitoring and control, such as EFSA and ECDC. The database itself will remain property of the EC and the Member States.

A repository for VTEC O157 is held at the EU RL VTEC

BioNumerics

File Edit Database Subsets Experiments Comparison Identification Scripts VNTR Help Antibiotics Window

Complete view

Database entries

Key	ISS ID	Source	Phagetype	vtx subtype	1	2
G@images@PFGE028@007	ED 472	HUMAN	PT8	vtx1a; vtx2c		
G@images@PFGE028@008	ED 479	HUMAN	PT21/28	vtx1a; vtx2c		
G@images@PFGE#21@004	ED 515	HUMAN		vtx1a; vtx2c		
G@images@PFGE#21@008	ED 517	HUMAN		vtx1a; vtx2c		
G@images@PFGE#22@003	ED 593	HUMAN		vtx1a; vtx2c		
G@images@PFGE#22@004	ED 594			vtx1a; vtx2c		
G@images@PFGE024@002	ED 659	HUMAN		vtx1a; vtx2c		
G@images@PFGE031@003	ED 667	HUMAN	PT8	vtx1a; vtx2c		
G@images@PFGE030@007	ED 687	HUMAN	PT14	vtx1a; vtx2c		
G@images@PFGE030@009	ED 704	HUMAN	PT8	vtx1a; vtx2c		
G@images@PFGE025@003	ED 164B	HUMAN	PT2	vtx2a		
G@images@PFGE026@008	ED 308	HUMAN	RDNC	vtx2a		
G@images@PFGE#21@009	ED 518	HUMAN		vtx2a		
G@images@PFGE030@004	ED 665	HUMAN	RDNC	vtx2a		
G@images@PFGE031@002	ED 666	HUMAN	PT23	vtx2a		
G@images@PFGE031@004	ED 668	HUMAN	PT23	vtx2a		
G@images@PFGE031@007	ED 670	HUMAN	PT23	vtx2a		
G@images@PFGE024@007	ED 698	HUMAN		vtx2a		
G@images@PFGE025@004	ED 179	HUMAN	PT43	vtx2a; vtx2		
G@images@PFGE025@007	ED 212	HUMAN	PT14(CR)	vtx2a; vtx2		
G@images@PFGE026@003	ED 280	HUMAN	PT49	vtx2a; vtx2		
G@images@PFGE026@004	ED 297	HUMAN	PT20	vtx2a; vtx2		
G@images@PFGE028@002	ED 449	HUMAN	PT49	vtx2a; vtx2		
G@images@PFGE#22@008	ED 601	HUMAN		vtx2a; vtx2		
G@images@PFGE#22@009	ED 604	HUMAN		vtx2a; vtx2		
G@images@PFGE#23@004	ED 607	HUMAN		vtx2a; vtx2		
G@images@PFGE024@003	ED 679	HUMAN		vtx2a; vtx2		
G@images@PFGE031@009	ED 680	HUMAN	PT2	vtx2a; vtx2		
G@images@PFGE027@003	ED 13			vtx2c		

Experiments

Name	Created
1 PFGE Xbal	
2 MLVA	

Comparisons

Name	Created
GELPFGE26Lug2011	2011-(-)
PFGE 24	2011-(-)
PFGE 25	2011-(-)
PFGE 26	2011-(-)
PFGE 27	2011-(-)
PFGE 28	2011-(-)

Analyses

Name	Created
analisi 28_12	2010-(-)
analisi 8_3_11	2011-(-)
ANALISI26_8_11	2011-(-)
analisi27Lug	2011-(-)
BRAENDERUP 29AG...	2011-(-)

Database: EU RL VTEC O157 Multi (connected) 352 entries 1 experiments D:\trasferimento vecchio P 157 Multi

A repository for VTEC O157 is held at the EU RL VTEC

Entry edit

File Edit VNTR Attachments Window

Database fields

Exp...

Key: G@images@PFGE031@003

ISS ID: ED 667

Source: HUMAN

Phagetype: PT8

vtx subtype: vtx1a; vtx2c

year of isolation:

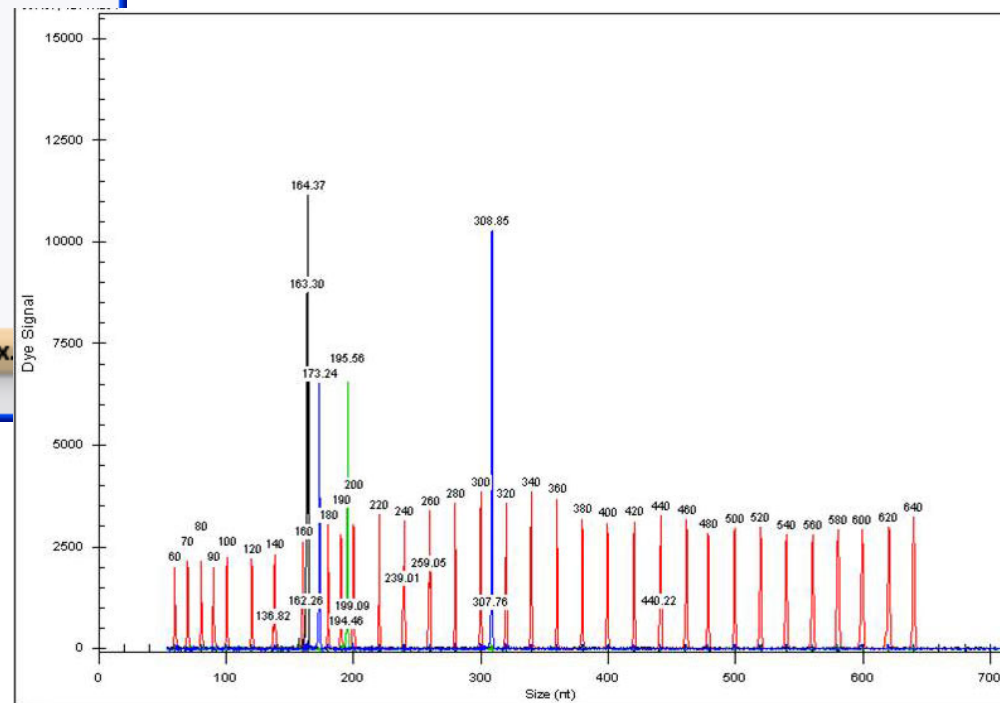
Sample type: sample

PFGE-Xbal-pattern:

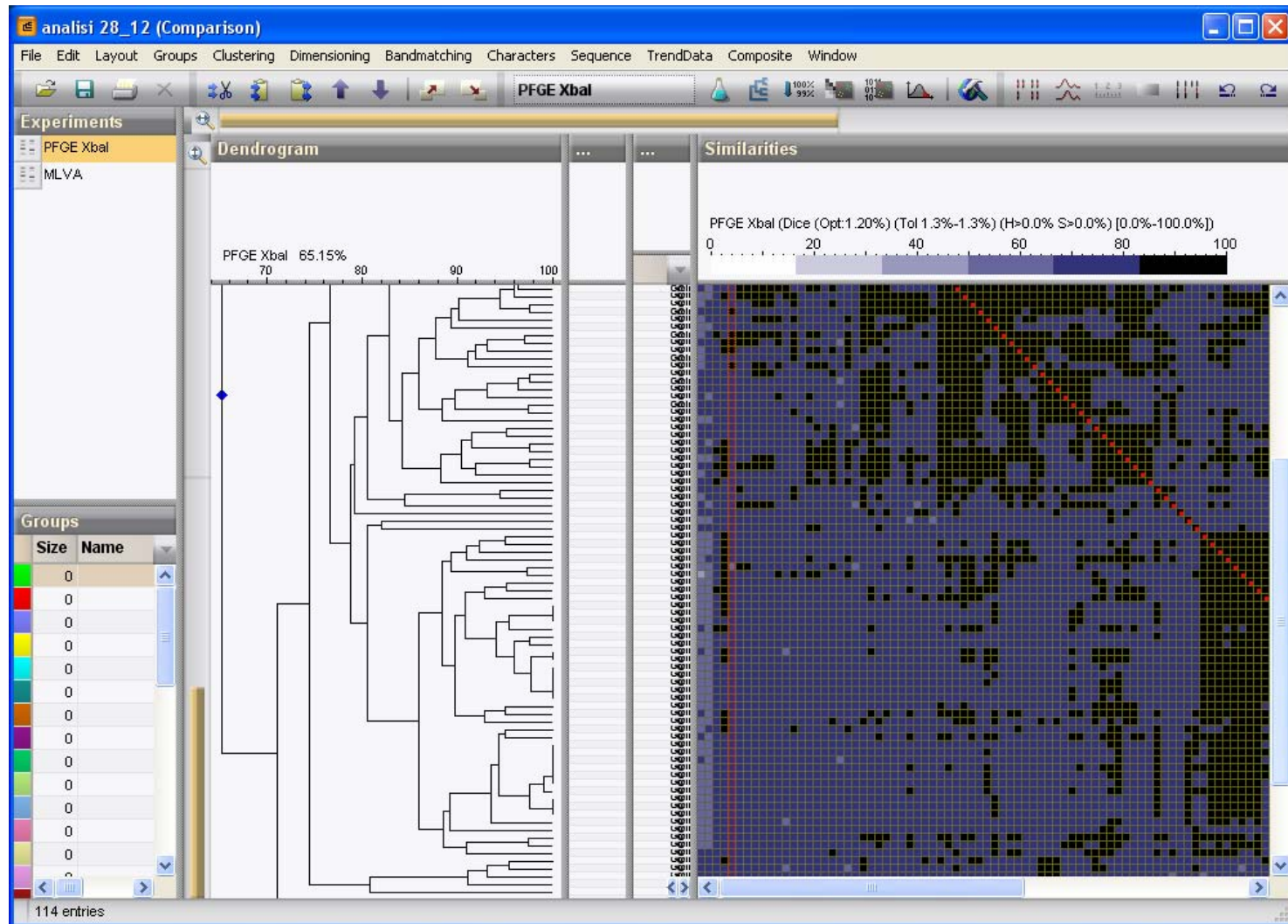
PFGE-BlnI-pattern:

Database fields Relations Ex...

Level:



A repository for VTEC O157 is held at the EU RL VTEC



Road Map (Months 0-6)

Liaison and link with the ongoing initiatives of ECDC on the molecular typing of strains from human infections (TESSy 3.0)

New inventory on the availability of equipment and technical skill on molecular typing (PFGE and MLVA) among the NRLs

Kick off meeting with 5 - 6 NRLs (ECDC will be invited) selected on the basis of the inventory to define and launch the initiative

Memorandum of Understanding with PulseNet International for comparison and assignment of PFGE profiles

**Development of standard training programs on:
PFGE**

MLVA (O157)

Vtx typing

BioNumerics analysis of profiles

Road Map (Months 7 - 12)

An extended protocol for Vtx-typing is distributed the NRLs which includes all the Vtx Sub types

Training stages on PFGE/MLVA/Vtx typing based on the standard training programs

A ring trial on PFGE/MLVA/Vtx-typing is carried out (certification of labs to submit profiles)

Objectives

The PFGE and MLVA protocols are standardized

The PFGE and MLVA profiles management is harmonized

The number of NRLs carrying out the PFGE/MLVA is increased

The number of labs performing the Vtx typing is increased and the scheme extended

Meanwhile . . .

The possibility is already in place for NRLs to:

Send VTEC strains to EU RL VTEC for PFGE typing

Send VTEC O157 strains to EU RL VTEC for MLVA and Phage typing

then . . .

The possibility for NRLs to:

Send PFGE profiles of VTEC strains to EU RL VTEC for cluster analysis

Send MLVA profiles of VTEC O157 strains to EU RL VTEC for cluster analysis

The EU RL VTEC Database

The database will be held in local at the beginning- NRLs will:

- send strains for typing. The generated profiles will be included in the database
- send the profiles generated in their own labs (E-mail) for uploading and analysis (Run by EU RL VTEC)
- ECDC and PulseNet intl. can make enquiries

The database will be open for uploads analyses from the external sources in phase two (2013?)- The NRLs will:

- upload the profiles generated in their own labs
- compare their profiles with those existing in the repository
- ECDC and PulseNet intl. can interrogate the DB directly

Future

Full availability of the repository for comparison with data from human strains (ECDC network and PulseNet international)

Full compatibility with TESSy 3.0

Harmonized typing schemes and EQA following the same approach than the previous collaboration between EU RL VTEC and FWD (SSI)