# Update on the STEC Network preparedness for molecular typing data collection (PT-PFGE6)

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www.iss.it/vtec





#### The EURL-VTEC activities for the preparedness on molecular typing

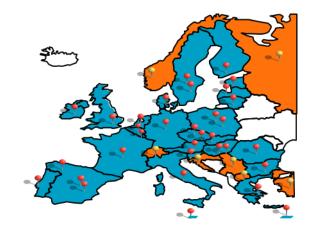
 $\sqrt{\phantom{0}}$  Proficiency tests (EQA) on PFGE (6 rounds in 2012-2018)

#### THE PT-PFGE7 STRAINS WILL BE SENT NEXT MONTH



√ Training sessions on PFGE (2012-2018)11 sessions

**27 TRAINEES** 



√ Basic courses on BioNumerics software (2014-2018)

4 sessions (3 joint with other EURLs)

**45 TRAINEES** 





#### **AIMS & OBJECTIVES**



EQA ON PFGE: for the assessment of the capability to produce good quality profiles

TRAINING: for the transfer of knowledge on the use of the correct protocols





To provide high quality PFGE profiles
For the non human database for
molecular typing





#### The EURL-VTEC Proficiency testing activity on PFGE

#### PT-PFGE1 (with PT10, January-March 2013)

- 16 EU NRLs + Norway, Switzerland and Egypt
- 11 E.coli strains
- Conducted jointly with the ECDC-FWD network

#### PT-PFGE2 (with PT11, June-September 2013)

- 23 EU NRLs + Norway and Switzerland
- 6 E.coli strains

#### PT-PFGE3 (with PT13, April-June 2014)

- 25 EU NRLs + Norway and Switzerland
- 7 E.coli strains

#### PT-PFGE4 (April-June 2015)

- 24 EU NRLs + Norway, Switzerland, Macedonia and Russia
- 10 E. coli strains
- Conducted jointly with the ECDC-FWD network

#### PT-PFGE5 (November-December 2016)

- 17 EU NRLs + Norway and Turkey
- 10 E. coli strains, in the framework of the **PT18**

#### 6. PT-PFGE6 (with PT20, November 2017-**January 2018)**

- 19 EU NRLs + Norway
- 6 E. coli strains, together with the PT-WGS1

### 7. PT-PFGE7 (with PT23 COMING SOON)

- 18 EU NRLs + Norway, Turkey and Russia
  - 6 E. coli strains





#### **EQA** on molecular typing: evaluation NRL's Proficiency





√ PT-PFGE1 and PT-PFGE2: PulseNet Procedure PNQ01(visual assessment)

√ From the PT-PFGE3: EFSA SOP for PFGE profile interpretation (BioNumerics assisted)



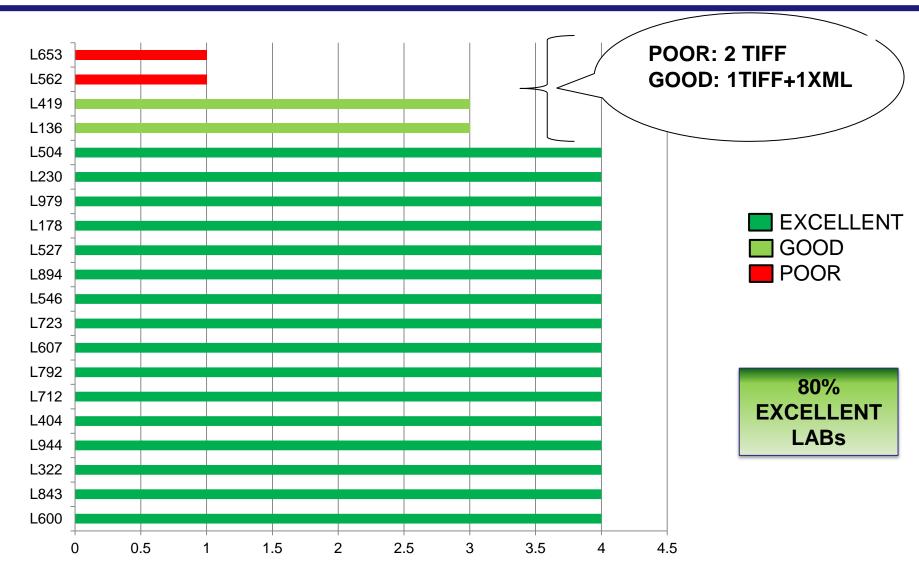
From the PT-PFGE4: we have included the possibility to submit the XML files, generated by the BioNumerics software







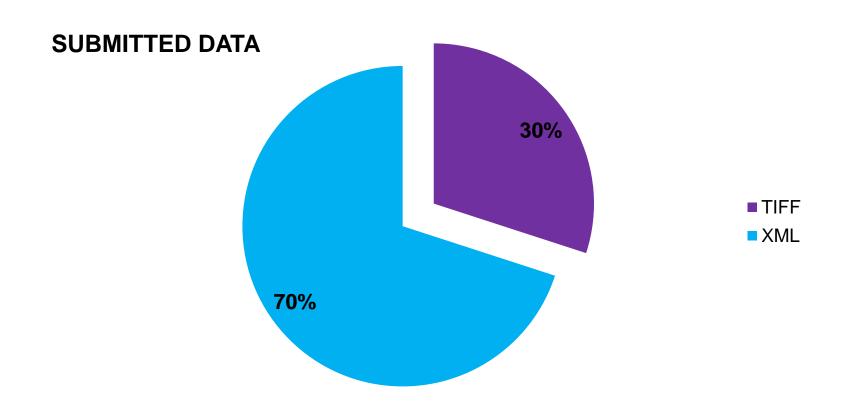
#### PT-PFGE6 RESULTS - 1







#### PT-PFGE6 RESULTS - 2



- TIFF: GEL IMAGE FILE + EXCEL FILE
- XML: FILE GENERATED BY THE SOFTWARE BIONUMERICS AFTER THE ANALISYS PERFORMED BY THE NRL





#### BAND ASSIGNMENT EVALUATION

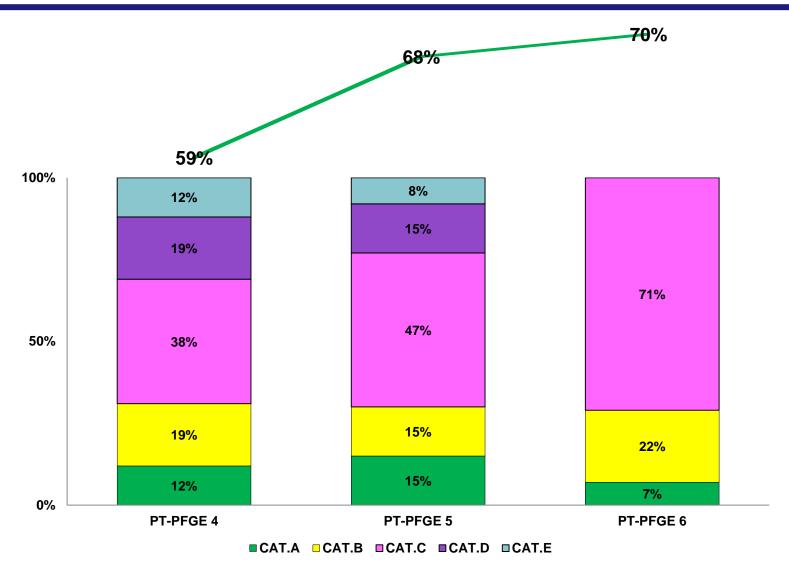
THE LABORATORIES WERE ASSIGNED TO CATEGORIES FROM A TO E, ACCORDING TO THE FOLLOWING CRITERIA:

- A: NO MODIFICATIONS OF THE BAND ASSIGNMENT IN THE XML FILES WERE NEEDED.
- B: ONLY SOME MODIFICATIONS OF THE BAND ASSIGNMENT WERE NEEDED.
- C: MAJOR MODIFICATIONS OF THE BAND ASSIGNMENT OR COMPLETE RE-ASSIGNMENT WERE NEEDED.
- D: BOTH NORMALIZATION AND BAND ASSIGNMENT HAD TO BE REPEATED.
- E: THE XML FILE WAS NOT USABLE FOR THE CLUSTER ANALYSIS.





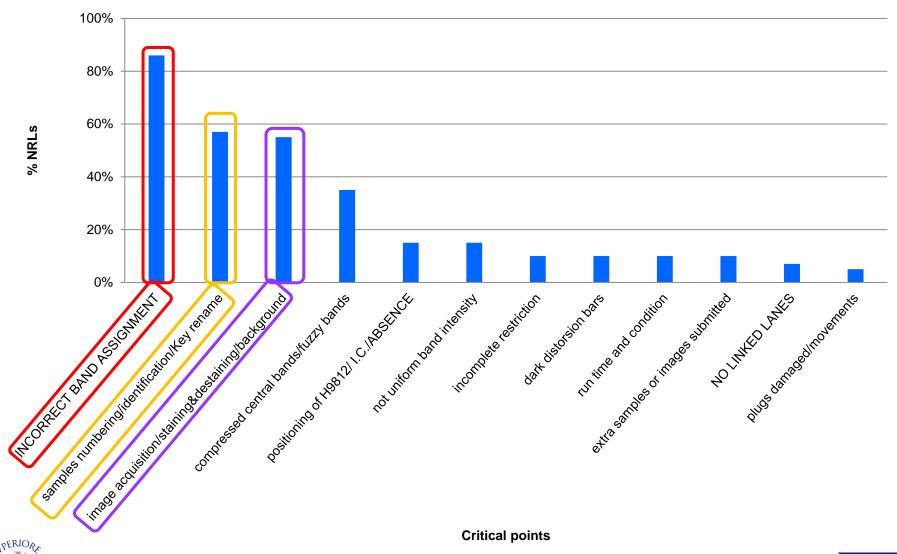
#### TREND FOR THE XML SUBMISSION







#### **CRITICAL POINTS PT-PFGE6**





#### **NEW FINDINGS**



#### √ RECURRING ERRORS

**50% NRLS** (10 PARTICIPANTS) PRESENTED 15 RECURRING ERRORS:

- 11 VISUAL ASSESSMENT, RELATED TO GEL QUALITY
- 4 IN DEPTH ANALYSIS, LINKED WITH BN (ALWAYS KEY RENAMED)

#### √ 2 NEW ENTRIES NRLs

ALREADY TRAINED IN 2017 AND 2018 FOR PFGE AND ONE NRL TRAINED ALSO FOR BN

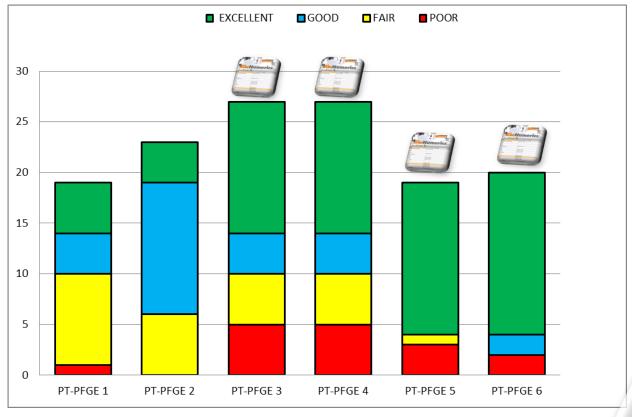
BOTH EXCELLENT WITH TIFF AND XML SUBMITTED





#### PFGE Proficiency testing (EQA): Trend from 2012...

#### NRLs performance in producing PFGE profiles



No. of NRLs

**PFGE PT rounds** 







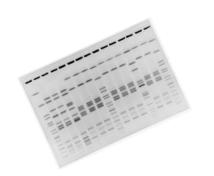




#### **Training Activities for molecular typing**

On molecular typing of VTEC by PFGE @ISS Rome





60% of NRLs trained

 Training Course on the Use of BioNumerics Software to analyse PFGE data of Shiga-toxin producing *Escherichia coli*, Salmonella and *Listeria* monocytogenes (1 EURL-VTEC edition +3 Joint Editions)



37% of NRLs trained







- Importance of the participation in the PT after the training session ( see the 2
   NEW ENTRIES)
- Proficiency test activity confirm its utility for the preparedness of the network for the EFSA-DB
- 80% of the participants in the PT-PFGE6 accomplishing an excellent result
- A consolidate network on Joint analysis has been created- one more Joint Course is planned for molecular typing in 2019
- The introduction of the analysis of the recurring errors will be used by the EURL VTEC to find the areas needing dedicated training sessions
- The number of Participants in the PT-PFGE rounds demonstrate the interest of the NRLs to the molecular typing (60% of Labs continue the application)







## PLEASE DON'T FORGET TO REPORT THE PRESENCE OF THE PROBLEMS ON TIME BEFORE THE DEADLINE FOR THE SUBMISSION IN ORDER TO FIND A SOLUTION TOGETHER



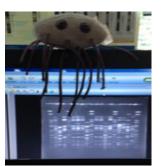




#### **THANK YOU...**



































Vision paper on the development of data bases for molecular testing of foodborne pathogens in view of outbreak preparedness.

 PFGE SOPs produced by EURLs for STEC, Listeria monocytogenes and Salmonella describes all the details for the production, interpretation and curation of the PFGE profiles

http://www.efsa.europa.eu/sites/default/files/scientific\_output/files/main\_docume nts/704e.pdf; /702e.pdf; / 703e.pdf

- Technical Specification on molecular typing data collection, on metadata and on the ownership of the data
- 11 MS nominated their rapresentative for submitting molecular typing data (for 2016)
- √ EFSA DATA-BASE exist and some PFGE profiles are present
- √ The curators have already start the activity (end of January 2018)
- $\sqrt{\ }$  Just 2 MS uploaded the profiles



**SETTING-UP** 



