18th Annual Worksop of the National Reference Laboratories for *E. coli* – 05-06 October 2023

The training program of the EURL for *E. coli* 2022-2023



Image from: https://accademiadomani.it/pacchetto/





Image from: https://viaggi.corriere.it/weekend/roma-segreta-gliitinerari-insoliti-e-i-classici-come-non-li-avete-mai-visti/



Istituto Superiore di Sanità, Food Safety, Nutrition and Veterinary Public Health Department European Reference Laboratory for *Escherichia coli*



Training activities of the EURL for *E. coli* in 2023

- Training course on characterization of pathogenic *E. coli* through NGS (two-days)
- STEC detection in food matrices based on the ISO TS 13136:2012 (five days stage)
- Identification and characterization of the different groups of pathogenic *E. coli* (four days stage)

The call was launched at the beginning of 2023, with the deadline for responding in mid-February

NRLs were informed that, in addition to the satisfaction survey, a final questionnaiere for evaluating the achievement of the training objectives would be administered to participants

Training course on characterization of pathogenic *E. coli* through NGS (two-days)

This course was held online in the days 11-12 July 2023

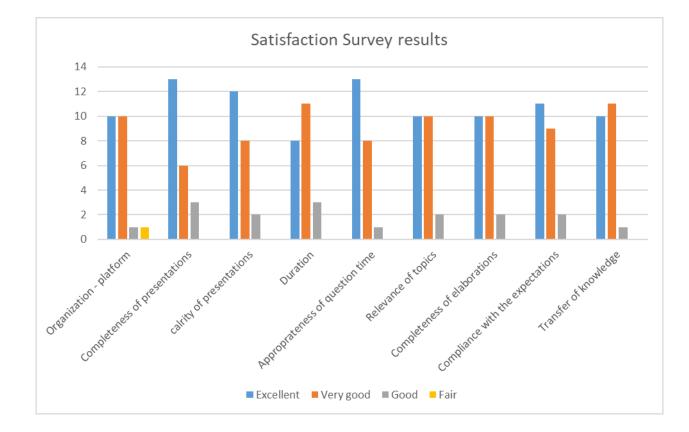
Presentations from EURL for E. coli staff

Assignments of excercises

Live-chat to provide assistance

22 participants – 16 from EU and non-EU NRLs and 6 from Italian OLs

86,4% of participants successfully met the training objectives reaching >80% compliance assessed through a dedicated questionnaire



Starting from 2022...

Scientists for training prgrams on laboratory procedures have been hosted in person at ISS ③



Training on the Identification and characterization of the different groups of pathogenic *E. coli* 17-21 April 2023



EU Reference Laboratory for E. coli Department of Food Safety, Nutrition and Veterinary Public Health Unit of Food Microbiology and Foodbome Diseases Istituto Superiore di Sanità



Program for a 4-days training at the EURL-VTEC, Istituto Superiore di Sanità, Rome, on the identification and characterization of the different groups of pathogenic *E. coli* by Real Time PCR amplification of their virulence genes

Day 1 (14:30-17:30)

- Overview of the activities and procedures in place at the EURL.
- Opening discussion on the work-plan and overview of the activities to be done during the stage.
- Preparation of the cultures of test and control strains.

Day 2 (9:30-17:30)

- Preparation of DNA samples for Real Time PCR from test and control strains.
- Real Time PCR for the identification of the E. coli pathogroups STEC and EAEC.

Day 3 (9:30-17:30)

- Real Time PCR for the identification of the E. coli pathogroups EIEC and ETEC.
- DNA template preparation for the identification of stx gene subtypes by conventional PCR.
- stx-subtyping by conventional PCR (EU-RL VTEC_Method_006_Rev 1).

Day 4 (9:30-17:30)

- Agarose gel electrophoresis to visualise the stx-subtyping PCR results.
- Analysis of the results obtained.
- General discussion on the activities carried out.
- Questionnaire on the trainee satisfaction toward the stage

All the three days include explanatory discussions driven by the EURL-VTEC experts and practical sessions carried out by the trainee with an hands-on approach, under the supervision of the EURL-VTEC staff.

Two scientists from EU NRLs participated



Both participants successfully met the training oblectives

Training on STEC detection in food matrices based on the ISO TS 13136:2012

6-10 November 2023 13-17 November 2023



EU Reference Laboratory for E. coll Department of Facel Safety, Number and Veterlany Public Nearth Unit of Facel Infractionary and Facebase Isofitutio Superiore di Sanita



Program for a 5-days training at the EU-RL VTEC, latitute, superiors of sanita, Rome, on the detection of STEC in food matrices according to the ISO T\$ 13136:2012 and the characterization of the isolated STEC strains

Day 1 (14:30-17:30)

- Overview on the activities and procedures of the EURL.
- Opening discussion on the work-plan and overview on the activities to be dope during the stage.
- Enrichment of food samples according to the ISO TS 13136:2012
- Introduction to the EURL Laboratory procedure for STEC O104 identification

Day 2 (9:30-17:30)

- DNA purification from food enrichment cultures.
- Real Time PCR for VTEC identification according to the ISO TS 13136:2012
- Real Time PCR for O104 identification
- Isolation of STEC from Real Time PCR-positive food enrichment cultures: streaking the plates

Day 3 (9:30-16:30)

- Picking up and pooling of colonies and backup onto solid media (nutrient agar)
- Confirmation of suspected colonies as STEC by conventional PCR (Annex C of the ISO TS 13136:2012)

Day 4 (5:30-17:30)

- Molecular serogrouping by conventional PCR (EU-RLVTEC_Method_003_Rev1 25/03/2014)

EU-RL VIEC-Judoka Program (SOT513136 Ray 3

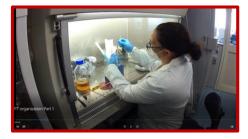
17/12/2018

Day 5 (9:30-13:30)

- Agarose gel electrophoresis to visualise the stx-subtyping PCR results
- Analysis of the results obtained
- General discussion on the activities carried out
- Questionnaire on the trainee satisfaction toward the stage

All the five days include explanatory discussions driven by the EURL-VTEC experts and practical sessions carried out by the trainee with an hands on approach, under the supervision of the EURL-VTEC staff.

Hands-on



Seven scientists will be attending

17/12/2018

2nd joint training – June 20th-21st 2023 RIVM

Introduction to Bioinformatics for genomic data mining

TUESDAY 20 JUNE 2023

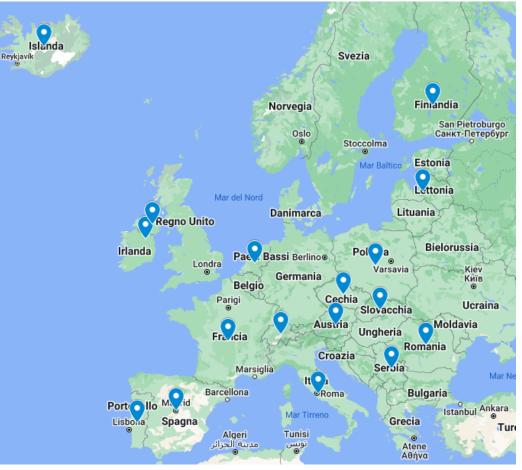
9.15	Registration (for entering the premises of RIVM)
9.45	Welcome and general overview on the joint training activities (Wilma Jacobs, EURL- Salmonella)
10.00	 Introduction to WGS Sequencing platforms & output data (20') (Adrien Asséré, EURL-Listeria monocytogenes) Verification of the integrity of the raw data files (like md5sum) (20') (Angela van Hoek, EURL-Salmonella) Bioinformatics analysis of NGS data: approaches and opportunities (command-line tools, commercial software, webservers) (20') (Joakim Skarin, EURL-Viruses)
11.00	Coffee break
11.20	Introduction on quality check and trimming (Valeria Michelacci, EURL-VTEC)
11.40	Hands-on exercises – Quality check and trimming Quality check and trimming using different tools and platforms. Results interpretation. FastQC and Positional and Quality Trimming on ARIES (Valeria Michelacci, EURL- VTEC) Quality check and trimming with Seqsphere (Bo Segerman, EURL-Campylobacter)
12.45	Lunch break
13.45	Introduction on assembly and assembly statistics (Lauge Holm Sørensen, EURL-AMR)
14.00	Hands-on exercises – Assembly statistics Assembly statistics using different tools and platforms. Results interpretation. Hands-on SPAdes and Quast from ARIES (Federica Gigliucci, EURL-VTEC) Demo: Seqsphere Assembly Quality Check (Angela van Hoek, EURL-Salmonella)
15.00	From BAM to BCF and beyond, making sense of aligned data (Paolo Vatta, EURL- Parasites)
15.30	Hands-on exercises – Mapping approach Demonstration of mapping through different platforms. Hands-on E. coli virulotyping using a mapping approach (Federica Gigliucci,EURL- VTEC) Demo: Mapping with Seqsphere (Déborah Merda, EURL-Listeria monocytogenes)

WEDNESDAY 21 JUNE 2023

8.45	Registration (for entering the premises of RIVM)
9.15	Introduction to gene detection using BLAST approach (Marina Cavaiuolo, EURL- CPS)
9.30	Hands-on exercises – Search of genetic features on contigs Identification of virulence and AMR genes using different tools and platforms. Results interpretation. Demonstration of genes identification on contigs through different platforms. Hands-on ResFinder on CGE webserver (Lauge Holm Sørensen, EURL-AMR) Demo: Salmonella virulotyping with Seqsphere (Angela van Hoek, EURL- Salmonella)
10.15	Parasites WGS: opportunities and challenges (Simone Cacciò, EURL-Parasites)
10.45	Coffee break
11.00	Introduction to genome comparisons: gene-by-gene VS SNPs (Guidance document for cluster analysis) (Bo Segerman, EURL-Campylobacter)
11.30	Demonstration of gene-by-gene approach through different platforms: ARIES (EURL-VTEC) Seqsphere (Angela van Hoek, EURL-Salmonella) Starflow (Déborah Merda, EURL-Listeria monocytogenes and Marina Cavaiuolo, EURL-CPS)
12.15	Hands-on exercises – Visualisation of clustering data Demonstration via Grapetree (Déborah Merda, EURL-Listeria monocytogenes)
12:45	Lunch break
13.45	Update on the EFSA OneHealth WGS database (Mirko Rossi, EFSA)
14:15	Information on activities of inter EURLs Working Group on NGS and guidance documents released (Valeria Michelacci, EURL-VTEC)
14:45	Wrap up (Wilma Jacobs-Reitsma, EURL-Salmonella)
15.00	Closure



Participants – geographic distribution



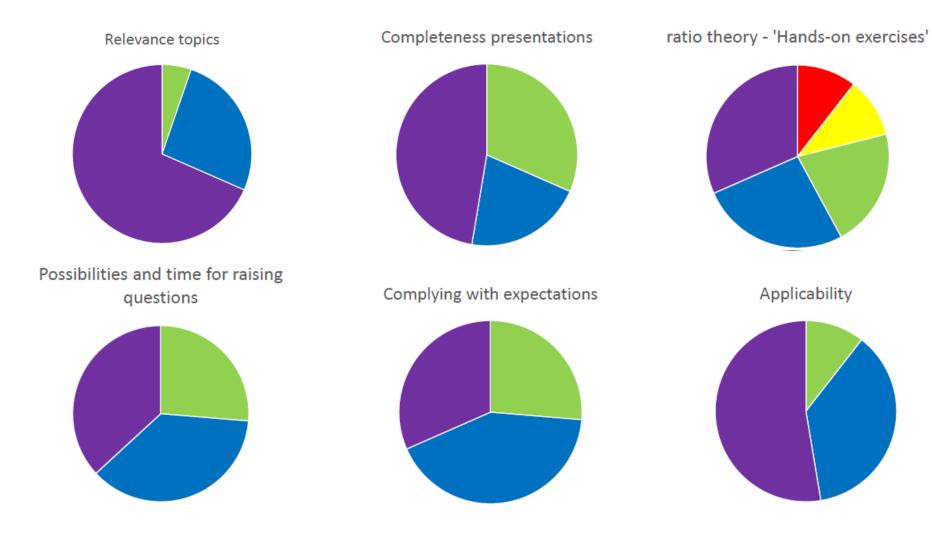
24 participants from different NRLs4 at own costs2 supported by EURL VTEC

17 different countries (13 Member States + 2 EFTA/EEA + Northern Ireland + 1 Candidate Country)

Many participants came from labs appointed as NRLs for multiple pathogens

2023 - Training satisfaction survey

■ Poor ■ Fair ■ Good ■ Very good ■ Excellent



Thank you for your attention!