

EU Reference Laboratory for E. coli

Department of Food Safety, Nutrition and Veterinary Public Health
Unit of Food Microbiology and Foodborne Diseases
Istituto Superiore di Sanità



Program for a 5-days training at the EURL-VTEC, Istituto Superiore di Sanità, Rome, on the detection of STEC in food matrices according to the ISO TS 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 done 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 (9:30-17:30)

- Molecular serogrouping by conventional PCR (EU-RLVTEC_Method_003_Rev1 25/03/2014).
- Identification of stx subtypes by conventional PCR (EU-RL VTEC_Method_006_Rev 1 18/06/2013).

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.