Twenty rounds of proficiency test activity organized by the European Union Reference Laboratory for *Escherichia coli* (EURL-VTEC)

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EURL-VTEC
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The EU Reference Laboratory for *E. coli* (EURL-VTEC), established at the Istituto Superiore di Sanità in 2006, according to the **Regulation (EC) No. 882/2004**, coordinates a network of 34 EU National Reference Laboratories (NRLs) plus many non-EU NRLs **(Fig. 1)**. One of the main objectives of its mandate is to ensure that the methods used by the NRLs for the identification and typing of pathogenic *E. coli* strains and their detection in food and animal samples are harmonized.

The EURL develops and evaluates methods, distributes reference materials, hosts scientists from NRLs for training stages, collaborates with other EU structures (EFSA, ECDC) and organizes proficiency tests (PT).

Fig. 1: EURL-VTEC's network

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Microbiology of food and animal feed — Real-time polymerase chain reaction (PCR)-based method for the detection of food-borne pathogens — Horizontal method for the detection of Shiga toxin-producing *Escherichia coli* (STEC) and the determination of O157, O111, O26, O103 and O145 serogroups

Fig. 2: ISO/TS 13136

Since 2006, the EURL has developed and evaluated standard operating procedures for the identification and typing of STEC and for their detection in food, mainly based on PCR detection of virulence genes. In particular, it coordinated the development of the **ISO/TS 13136:2012** on the detection of STEC in food and animal feed, based on the Real Time PCR screening of food enrichment cultures **(Fig. 2)**.

To evaluate both the methods and the performance of the NRL network in their application, the EURL organized so far **20 rounds** of proficiency tests (PT) conducted in compliance with the International Standard **ISO/IEC 17043:2010 (Table 1)**.

Eight PTs were dedicated to bacterial typing and included the detection of STEC virulence genes by PCR and the identification of the serogroups most involved in human disease in Europe. **Twelve PTs** were dedicated to the detection of STEC in different matrices (carcass swabs, milk, spinach, water, seeds, sprouts, spent irrigation water, ground beef meat and rocket salad) using the ISO/TS 13136:2012.

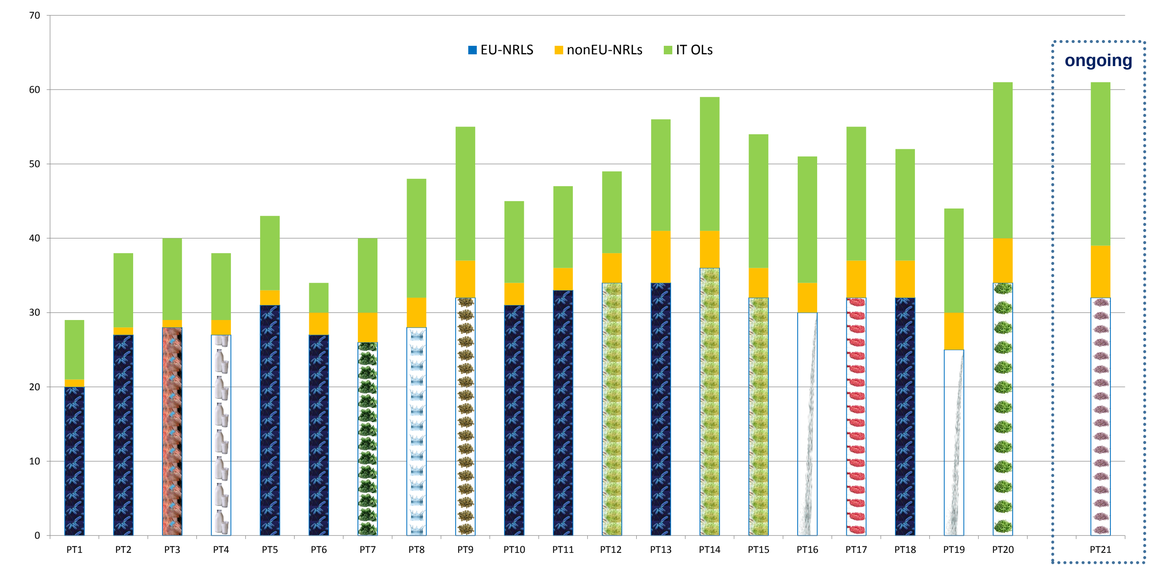

Fig. 3: Trend of the participation in the EURL-VTEC PT program

		Nr. of participants	Matrix	Year	Nr. of samples
Identification and typing of pathogenic *E. coli* strains	PT1	29	strains	2007	5
	PT2	38		2008	7
	PT5	43		2010	5
	PT6	34		2010-11	5
	PT10	45		2012-13	15
	PT11	47		2013	6
	PT13	56		2014	7
	PT18	52		2016	10
Detection of STEC in different matrices	PT3	40	carcass swabs	2009	5
	PT4	38	milk	2010	2
	PT7	40	spinach	2011	3
	PT8	48	water	2012	2
	PT9	55	seeds	2012	2
	PT12	49	sprouts	2013	3
	PT14	59		2014	3
	PT15	54		2015	3
	PT16	51	spent irrigation water	2015	3
	PT17	55	ground beef meat	2016	3
	PT19	44	spent irrigation water	2017	3
	PT20	61	rocket salad	2017	3
	PT21	*61*	*sprouts*	*ongoing*	*3*

Table 1: PTs organized by the EURL-VTEC

Considering EU NRLs, non-EU NRLs and Italian official laboratories **(Fig. 3)**, a positive trend was observed in both the number of participating laboratories (from 29 in 2007 up to 61 in 2017) and their performance (up to 97.2 % of EU/non-EU NRLs correctly identified the presence of virulence genes in the test samples). The overall evaluation highlighted that an excellent preparedness has been built in the EU towards the ability to identify the main virulence genes of STEC, while the capacity to detect other *E. coli* pathotypes and their most represented serogroups, although to a lesser extent, is also present with a good performance.

The control of pathogenic *E. coli* in food and animals represents a challenge for the development of specific detection methods and requires a network of skilled and trained laboratories throughout the EU for their detection in the vehicles of infection. The EURL-VTEC is working to consolidate such a network, in order to gather harmonized data and develop standardized operative procedures and tools to face possible emergencies.