Final report PT-Tm 1/2016

PT "Identification of *Trichinella* larvae at the species level by a molecular method"

Design

Purpose	Evaluation of laboratories in charge for official control on food	
Scheme type	Single	
Participants	Public and private, European laboratories	
N. of participants	Depending on request	
Method	not regulated	
Test method	chosen by the participant	
PT items	Matrix	not applicable
	Item	*Trichinella* spp. larvae in 96% ethanol
	N. of samples	4 (10 larvae/each) or 12 (1 larva/each) 1.5 ml vials for each participant
	Distribution	Immediate shipment after preparation
Subcontracted activities	NA	
Results evaluation	Qualitative	

Implementation

N. of participants	21	PT items	PT panel 4 vials	10 larvae for each of the following species: *T. spiralis*, *T. britovi*, *T. pseudospiralis* and *T. murrelli*
Public laboratories	0			
Private laboratories	0		PT panel 12 vials	3 larvae for each of the following species: *T. spiralis*, *T. britovi*, *T. pseudospiralis* and *T. murrelli*
NRLs	21		Shipping	TNT Express
Shipping dates	March 14, 2016			

PT Provider
Unit of Gastro-enteric and Tissue Parasitic Diseases
Istituto Superiore di Sanità, Rome, Italy

PT Coordinator
Dr. Gianluca Marucci
e-mail: gianluca.marucci@iss.it; tel: +39 06 4990 2241

Viale Regina Elena, 299 – 00161 Rome, Italy

Qualitative results

The PT final evaluation was qualitative only. The PT was considered passed when all isolates or, in case of single larvae at least one of them for each isolate, are correctly identified.

Laboratory code	N. right identification	N. wrong identification	N. missed identification	Final evaluation
NRL1	**9**	**3**	**0**	**negative**
NRL3	12	0	0	positive
NRL4	12	0	0	positive
NRL6	4	0	0	positive
NRL7	12	0	0	positive
NRL8	4	0	0	positive
NRL10	12	0	0	positive
NRL12	4	0	0	positive
NRL14	**2**	**1**	**1**	**negative**
NRL16	4	0	0	positive
NRL17	**3**	**1**	**0**	**negative**
NRL18	The lab did not send results			NA
NRL21	**3**	**1**	**0**	**negative**
NRL22	12	0	0	positive
NRL23	4	0	0	positive
NRL24	12	0	0	positive
NRL25	**3**	**1**	**0**	**negative**
NRL34	**1**	**2**	**1**	**negative**
NRL35	**3**	**1**	**0**	**negative**
NRL42	The lab did not send results			NA
TLE6	4	0	0	positive

Legend: Laboratories that failed the PT are marked in bold.

Summary of qualitative results:

Total number of PT panels	21
Number of participant laboratories	21
Number of participants that passed the PT	12
Number of participants that failed the PT	7

Note: two laboratories did not send the results

Overtime comparison of results

Laboratory code	PT 2011	PT 2012	PT 2013	PT 2014	PT 2015	PT 2016
NRL1	positive	positive	positive	positive	positive	**negative**
NRL2	-	-	-	**negative**	-	-
NRL3	**negative**	-	positive	positive	positive	positive
NRL4	positive	positive	positive	positive	-	positive
NRL6	positive	positive	**negative**	positive	**negative**	positive
NRL7	-	**negative**	**negative**	positive	positive	positive
NRL8	positive	positive	positive	positive	positive	positive
NRL9	**negative**	-	**negative**	-	-	-
NRL10	**negative**	positive	-	positive	positive	positive
NRL11	positive	**negative**	positive	positive	-	-
NRL12	-	-	positive	-	positive	positive
NRL14	-	-	-	-	-	**negative**
NRL16	positive	positive	positive	positive	positive	positive
NRL17	-	-	-	positive	positive	**negative**
NRL18	-	-	-	-	-	NA
NRL21	positive	**negative**	positive	positive	positive	**negative**
NRL22	-	positive	**negative**	**negative**	positive	positive
NRL23	-	positive	positive	positive	positive	positive
NRL24	**negative**	-	positive	positive	positive	positive
NRL25	-	-	positive	positive	positive	**negative**
NRL34	**negative**	**negative**	positive	positive	positive	**negative**
NRL35	**negative**	**negative**	-	**negative**	positive	**negative**
NRL38	positive	-	-	-	-	-
NRL40	-	positive	-	-	-	-
NRL42	-	-	-	**negative**	-	NA
NRL44	-	-	-	**negative**	-	-
NRL45	-	-	-	-	**negative**	-
TLE6	-	-	-	-	-	positive

Comments:

Sixteen out of twenty one participants used the EURLP method (MI-02) or the method described by Pozio and La Rosa in 2003, both based on the Multiplex PCR technique originally described by Zarlenga et al. in 1999. Two labs used a modified version of the Pozio and La Rosa 2003 method, using only primers specific for the *Trichinella* species circulating in Europe. One lab used an alternative method (Rombout et al. 2001) based on PCR and sequencing of the 5S ribosomal DNA region. Seven out of twenty one participants failed the PT, they were invited to investigate the causes of the failure and to implement the appropriate corrective actions.

The reasons for the PT failure have been identified in: i) the use of an incomplete set of primers; ii) lack of experience in the application of the method leading to DNA amplification problems and iii) lack of experience in the interpretation of the bands pattern, in particular that from the *T. murrelli* non-European species. Only one lab asked, as corrective action, to analyse a new set of samples.

The Director
Dr. E. Pozio

Date 19/05/2016

PT Provider
Unit of Gastro-enteric and Tissue Parasitic Diseases
Istituto Superiore di Sanità, Rome, Italy

PT Coordinator
Dr. Gianluca Marucci
e-mail: gianluca.marucci@iss.it; tel: +39 06 4990 2241

Viale Regina Elena, 299 – 00161 Rome, Italy

Notes:

1. To guarantee confidentiality, participant laboratories are identified by alphanumeric codes. PT participant identity is kept confidential and bound by professional secrecy. If PT results have to be provided directly to a competent authority, the organizer shall send a written notice to inform the involved participants.
2. The organizer designates a qualified company for the transport and delivery of PT items.
3. Each participating laboratory receives a PT panel according to the PT scheme. Each PT item consists of 4 or 12 1.5 ml vials containing four different *Trichinella spp*. The homogeneity of PT items is ensured by an accurate control of the number of larvae spiked into each vial (item), made by two operators using a stereo microscope. PT items are stable for 5 years from the date of preparation (corresponding to the shipping date), provided that they are maintained in suitable conditions.
4. At the beginning of each year, the organizer draws up a PT program and makes it known by sending an email to the NRLs.
5. The final report issued for each PT round shows the PT program implementation.

End of the report

PT Provider
Unit of Gastro-enteric and Tissue Parasitic Diseases
Istituto Superiore di Sanità, Rome, Italy

PT Coordinator
Dr. Gianluca Marucci
e-mail: gianluca.marucci@iss.it; tel: +39 06 4990 2241

Viale Regina Elena, 299 – 00161 Rome, Italy