

Department of Infectious Diseases Unit of Foodborne and Neglected Parasitic Diseases European Union Reference Laboratory for Parasites



# Final report PT-04: An 1/2021

# PT-04: "Detection of Anisakidae L3 larvae in fish fillets"

## Design

Purpose	Evaluation of laboratories in charge of official control on food		
Scheme type	Single, simultaneous		
Participants	Public and private, European laboratories		
N. of participants	Depending on request		
Method	not regulated		
Test method	chosen by the participant		
PT items	Matrix	freshwater farmed fish fillet	
	Item	Anisakidae live larvae	
	N. of samples	3 for each participant	
	Distribution	Immediate shipment after preparation	
Subcontracted activities	NA		
Results evaluation	Qualitative		

## Implementation

N. of participants	30		Col. Cil. 4 con local	93 3 fish fillet sandwiches: one spiked with 1 larva, two spiked with 3 larvae.	
Public laboratories	0		fish fillet sandwiches		
Private laboratories	2	PT items	PT panel composition		
NRL	28		Shipping	DHL	
Shipping dates	15/03/20	21			

PT Provider Unit of Foodborne and Neglected Parasitic Diseases Istituto Superiore di Sanità

Viale Regina Elena, 299 - 00161 Rome, Italy

PT Person in charge: Dr. Marco Lalle

e-mail: marco.lalle@iss.it; tel: +39 0649902670



PTP N° 0005 P Membro degli Accordi di Mutuo Riconoscimento EA, IAF e ILAC Signatory of EA, IAF and ILAC Mutual Recognition Agreements

MO/MAQ/12.3 rev. 8 page 1 of 5



## Department of Infectious Diseases Unit of Foodborne and Neglected Parasitic Diseases European Union Reference Laboratory for Parasites



#### Results

The PT final evaluation was only qualitative. The PT was considered passed if the participant correctly identified all positive and all negative samples.

Laboratory code	N° of samples correctly identified	N° of samples NOT correctly identified	Final evaluation		
A1	3	0	Positive		
A2	3	0	Positive		
A3	3	0	Positive		
A5	3	0	Positive		
A6	3	0	Positive		
A7	3	0	Positive		
A9	3	0	Positive		
A10	3	0	Positive		
A11	3	0	Positive		
A12	3	0	Positive		
A13	3	0	Positive		
A15	3	0	Positive		
A16	3	0	Positive		
A17	2	1	Negative		
A18	3	0	Positive		
A19	3	0	Positive		
A20	3	0	Positive		
A21	3	0	Positive		
A25 (A23/A33)	3	0	Positive		
A26	3	0	Positive		
A28	3	0	Positive Positive		
A29	3	0			
A30	3	0	Positive		
A31	2	1	Negative		
A35	3	0	Positive		
A36	3	0	Positive		
A38	2	1	Negative		
A39	3	0	Positive		
A41	2	1	Negative		
A42	3	0	Positive		

### Legend:

• Laboratories that failed the PT are highlighted in black.

PT Provider Unit of Foodborne and Neglected Parasitic Diseases Istituto Superiore di Sanità

Viale Regina Elena, 299 - 00161 Rome, Italy

PT Person in charge: Dr. Marco Lalle

e-mail: marco.lalle@iss.it; tel: +39 0649902670



PTP N° 0005 P Membro degli Accordi di Mutuo Riconoscimento EA, IAF e ILAC Signatory of EA, IAF and ILAC Mutual Recognition Agreements

MO/MAQ/12.3 rev. 8 page 2 of 5



## Department of Infectious Diseases Unit of Foodborne and Neglected Parasitic Diseases European Union Reference Laboratory for Parasites



### Summary of results:

Total number of PT panels	30
Number of participant laboratories	30
Number of participants that passed the PT	26
Number of participants that failed the PT	4

### Overtime comparison of results

Laboratory code	2014	2015	2016	2017	2018	2019	2020	2021
A1	Р	Р	Р	Р	Р	Р	N	Р
A2	Р	Р	Р	N	Р	Р	Р	Р
A3	Ν	Р	Р	Р	Р	Р		Р
A5	Р	Р	Р	Р	Р	Р		Р
A6	Р	Р	Р	Р	Р	Р	Р	Р
A7	Р	Р	Р	Р	Р	Р	Р	Р
A8	Р	Р	Р		Р			
A9	Р		Р	N	Р		Р	Р
A10	Р	Р	N	Р	Р	Р	Р	Р
A11	Р	Ν	Р	N	Р	Р	N	Р
A12	Р	Ν	Р	Р	Р	Р	Р	Р
A13	Р	Р	Р	N	Р	Р	Р	Р
A15	Р	Р	Р	Р	Р	Р	Р	Р
A16	Ν	Р	Р	N	Р	Р	Р	Р
A17	NR		Р	Р	Р	Р	Р	N
A18	Р	Р	Р	Р	Р	Р	N	Р
A19	Р	Р	Р	Р	Р	N	Р	Р
A20	Р	Р	Р	Р	Р	Р	N	Р
A21	Р	Р	Р	Р	Р	Р	Р	Р
A23/A33/A25*	Ν	Р	Р	N	Р	Р	Р	Р
A25	Р	Р	Р	N				
A26	Р	Р	Р	Р	Р	Р	Р	Р
A28	Р		Р	Р	Р	Р	Р	Р
A29			Р		Р	Р	Р	Р
A30		Р	Р	Р	Р	Р	N	Р
A31		Р	Р	Р	Р	Р		N
A35					Р	N	Р	Р
A36				Р	Р	Р	Р	Р
A37					N			
A38					Р			N
A39						Р		Р
A41								N
A42								Р

PT Provider Unit of Foodborne and Neglected Parasitic Diseases Istituto Superiore di Sanità

Viale Regina Elena, 299 - 00161 Rome, Italy

PT Person in charge: Dr. Marco Lalle

e-mail: marco.lalle@iss.it; tel: +39 0649902670



PTP N° 0005 P Membro degli Accordi di Mutuo Riconoscimento EA, IAF e ILAC Signatory of EA, IAF and ILAC Mutual Recognition Agreements

MO/MAQ/12.3 rev. 8 page 3 of 5



Department of Infectious Diseases
Unit of Foodborne and Neglected Parasitic Diseases
European Union Reference Laboratory for Parasites



#### Comments:

In this PT round, 87% of the laboratories (26/30) passed the PT. Four laboratories failed the PT, and all reported false negative likely due to new unexperienced analysts or problems with the detection procedure. In particular, two used the Artificial Digestion (AD) method, one used the UV-press (UV) method, and one used the low sensitive candling. Noteworthy, 13 laboratories underestimated the number of spiked larvae in at least one sample and two laboratories overestimated the number of spiked larvae in one sample, reporting four larvae instead of three. In these cases, artificial digestion was applied as detection procedure (only one lab used UV method); the discrepancy could be explained by over-digestion or accidental breaking of the larvae during sample manipulation (e.g. sample mincing). Since the PT is only qualitative (presence/absence of the larvae), the PT results were considered positive but the laboratories were invited to consider the over-/underestimation and to take appropriate actions including further training of the analyst on morphological identification of the larvae or proper application of the used method.

Laboratories that failed the PT were invited to plan appropriate corrective actions (e.g. shipping of additional sample panel for an EQA).

This PT round highlighted a significant increase in both laboratory performance and a broader interest in the PT (two external private laboratories). Poor performance was observed in laboratories that joined the PT for the first time or that participated inconstantly. Lack of experience of technical personnel (either new or not familiar with the test methods) could explain the negative result.

After a constant decrease, we observed a net increase in PT participants (Figure 1) in comparison to the previous years (23 in 2020; 26 in 2019; 28 in 2018, 27 in 2017 and 30 in 2016). The relative percentage of detection methods adopted did not change substantially (Figure 1), although one laboratory moved from AD to UV method. Artificial digestion is still the prevalent method applied (23 laboratories), largely because it does not require special equipment. UV-press method, alone or in combination with digestion, was used in 7 laboratories. Except for one laboratory, candling was used in combination with artificial digestion and/or UV-press method.

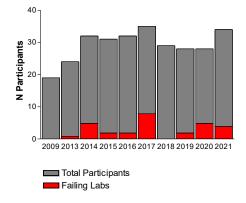


Figure 1. PT04 trend overtime

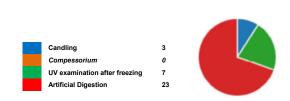


Figure 2. Summary of the detection methods applied

PT Provider Unit of Foodborne and Neglected Parasitic Diseases Istituto Superiore di Sanità

Viale Regina Elena, 299 - 00161 Rome, Italy

PT Person in charge: Dr. Marco Lalle

e-mail: marco.lalle@iss.it; tel: +39 0649902670



PTP N° 0005 P Membro degli Accordi di Mutuo Riconoscimento EA, IAF e ILAC Signatory of EA, IAF and ILAC Mutual Recognition Agreements

MO/MAQ/12.3 rev. 8 page 4 of 5



Department of Infectious Diseases Unit of Foodborne and Neglected Parasitic Diseases European Union Reference Laboratory for Parasites



PTP person in charge

Dr. N

The Director

Dr. S.M. Cacció

Date 29/04/2021

#### Notes:

- To guarantee confidentiality, participant laboratories are identified by alphanumeric codes. PT participant identity is kept confidential and bound by professional secrecy. The PTP reserves itself the right to provide the laboratory PT result to the competent authority upon request.
- 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 a fish fillet sandwich spiked or not with live Anisakidae larvae. The homogeneity of PT items is ensured by an accurate control of the number of larvae spiked into each sample (item) made by two operators. PT items are stable for 7 days from the date of preparation (corresponding to the shipping date), provided that they are maintained in suitable conditions.
- At the beginning of each year, the organizer draws up a PT program and inform the NRLs by email.
- 5. The final report issue of each PT round shows the PT program implementation.

End of the report

PT Provider Unit of Foodborne and Neglected Parasitic Diseases Istituto Superiore di Sanità

Viale Regina Elena, 299 – 00161 Rome, Italy

PT Person in charge: Dr. Marco Lalle e-mail: marco.lalle@iss.it; tel: +39 0649902670



PTP N° 0005 P
Membro degli Accordi di Mutuo Riconoscimento
EA, IAF e ILAC
Signatory of EA, IAF and ILAC
Mutual Recognition Agreements