

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



# Final report PT-An 1/2020

# PT report on "Detection of Anisakidae L3 larvae in fish fillets"

### 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	fresh water 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	23		fish fillet	78		
Public laboratories	-		sandwiches	76		
Private laboratories	-	PT items	PT panel composition	3 fish fillet sandwiches: one spiked with 0 larvae, two spiked with 1 larva.		
NRL	23		Shipping	TNT Express		
Shipping dates	09/03/2	.020				

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#### Results

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

Laboratory code	N° of samples correctly identified	N° of samples NOT correctly identified	Final evaluation	
A1	2	1	Negative	
A2	3	0	Positive	
A6	3	0	Positive	
A7	3	0	Positive	
A9	3	0	Positive	
A10	3	0	Positive	
A11	2	1	Negative	
A12	3	0	Positive	
A13	3	0	Positive	
A15	3	0	Positive	
A16	3	0	Positive	
A17	3	0	Positive	
A18	2	1	Negative	
A19	3	1	Positive	
A20	2	1	Negative	
A21	3	0	Positive	
A25	3	0	Positive	
A26	3	0	Positive	
A28	3	0	Positive	
A29	3	0	Positive	
A30	2	1	Negative	
A35	3	0	Positive	
A36	3	0	Positive	

### Legend:

• Laboratories that failed the PT are marked in bold.

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### Summary of results:

Total number of PT panels	23
Number of participant laboratories	23
Number of participants that passed the PT	18
Number of participants that failed the PT	5

#### Overtime comparison of results

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

Note: \*Lab code A25 re assigned in 2018 and 2019; P, positive; N, negative; NR, no result received; gray boxed, no participation

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#### Comments:

This PT round resulted in 78% of the laboratories (18/23) that passed the PT. Five laboratories failed the PT all reporting false negative likely due to new unexperienced analysts or troubleshoots with the detection procedure (all applied artificial digestion). Noteworthy, two laboratories overestimated the number of spiked larvae in one sample, reporting two larvae instead of one. Since the PT is only qualitative, presence/absence of the larvae, the PT results were considered positive but the laboratories were invited to take into consideration the over-overestimation and take appropriate actions including further training of the analyst on morphological identification of the larvae. Two laboratories reported fragmentation of the larvae. Also in these cases artificial digestion was used as detection procedure and likely over-digestion or accidental breaking of the larvae during the sample manipulation (e.g. sample mincing) could explain the discrepancy. No further corrective action was required. Due to restriction applied following the novel Coronavirus emergence EURLP was not able to organize any EQA scheme for laboratories that failed the PT before the the writing of this Final Report.

This PT round highlighted a significant decrease in the performance of the participant laboratories; thisoutcome likely is the consequence of: i) spiking of fish fillets with a low number of larvae; ii) inesperience of new technical personnel in some of the labs; ii) troubleshoots in the test procedure.

Over the last years, the number of PT participants decreasing in comparison to the previous years (26 in 2019; 28 in 2018, 27 in 2017 and 30 in 2016) as well as the relative percentage of detection methods adopted (Figure 1). Artificial digestion is still the prevalent method applied (19 Laboratories), largely because it does not require any special equipment. UV-press method, alone or in combination with digestion, was used in six laboratories. Candling was used exclusively combined with artificial digestion and/or UV-press method.

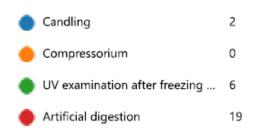


Figure 1. Summary of the detection methods applied



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The Director Dr. S.M. Cacciò

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Date 27/03/2020

#### Notes:

- 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 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.
- 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 issue of each PT round shows the PT program implementation.

End of the report

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