DEPARTMENT INFECTIOUS DISEASES

European Union Reference Laboratory for Parasites
Unit of Foodborne and Neglected Parasitic Diseases
Department of Infectious Diseases
ISTITUTO SUPERIORE DI SANITÀ

XXI Workshop of National Reference Laboratories for Parasites
28th and 29th October 2026
Istituto Superiore di Sanità

PT-08

Molecular identification of *Echinococcus granulosus, Echinococcus multilocularis,* and *Taenia spp*



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ISTITUTO SUPERIORE DI SANITÀ (Rome, Italy)

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Aim of the PT-08

Evaluation of laboratory competence in molecular identification of

- *Echinococcus granulosus sensu lato*



- *Echinococcus multilocularis*



- *Taenia sp*



PT-08 since 2018

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PT timing 2026

February 20th

PTs REQUEST FORM 2026

https://www.iss.it/en/web/iss-en/eurlp-proficiency-testing

March 16th



April 17th



May 29th

Istituto Superiore di Sanità
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

Total number	30	PT items	fish fillet sandwiches	93
Public laboratories	0			
Private laboratories	2		PT panel composition	3 fish fillet sandwiches: one spiked with 1 larva, two spiked with 3 larvae.
NRL	28		Shipping	DHL
Shipping dates	15/03/2021			

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

Final Report PT-08. EMoI 1/2026

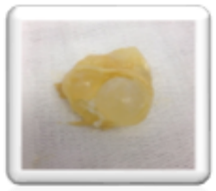
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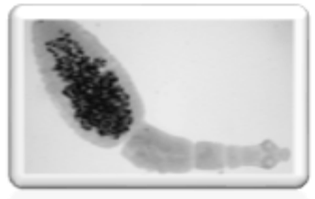
Preparation of samples PT08-2026

The panel consists of 4 tubes: each tube containing DNA extracted from canine faecal stool spiked with

- *E. granulosus s.s. cyst*



- *E. multilocularis worms*



- *Taenia Krabbei proglottid*



Negative: Canis lupus familiaris DNA



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Detection method PT08-2026

The samples were identified at species level through multiplex PCR

E.g. = *E. granulosus* (117 bp)

E.m. = *E. multilocularis* (395 bp)

Tae= Taenia spp (267bp)

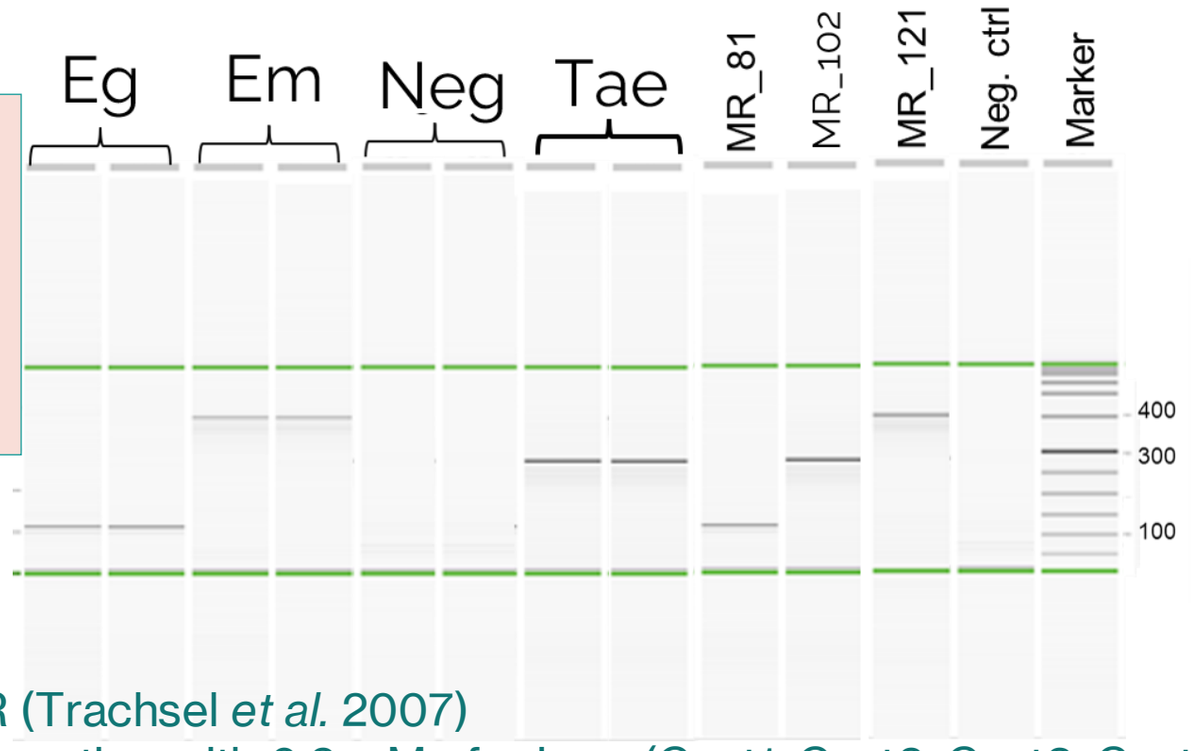
Negative = *Canis lupus familiaris* DNA

EURLP-Reference Material

MR_81 = *E. granulosus* (G3)

MR_102= Taenia spp

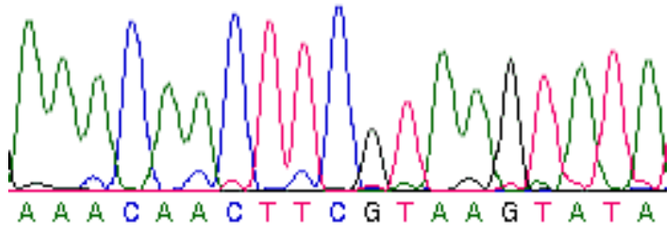
MR_121= *E. multilocularis*





Multiplex PCR (Trachsel *et al.* 2007)
In 30 µl total reaction with 0.2 mM of primer (Cest1, Cest2, Cest3, Cest4, Cest5)

PCR COX1 and NAD1 and sequencing



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Test material

Homogeneity was ensured by providing participants with aliquots of the same DNA preparation







The tubes were plugged and sealed using plastic parafilm, individually coded

Each PT panel was inserted in polystirene box with ice pack



All panels were delivered within 24-36 hours



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Evaluation criteria

The PT evaluation is qualitative and no statistical analysis of the results are applied

Lab code	Expected	Observed	Result (correct/incorrect)	Evaluation (positive/negative)
EMOLX	negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	negative E. multilocularis Taenia spp E. granulosus	correct correct correct correct	Positive
EMOL X	negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	*E. granulosus* *E. multilocularis* *Taenia spp* *E. granulosus*	incorrect correct correct correct	Negative
EMOL X	negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	negative *E. multilocularis* *E. granulosus* *E. granulosus*	correct correct incorrect correct	Negative

The result is “correct” if the PT items are correctly identified
The result is “incorrect” if PT items are incorrectly identified

The PT is “POSITIVE” if the results of all samples are “correct”
The PT is “NEGATIVE” if at least one result is “incorrect”

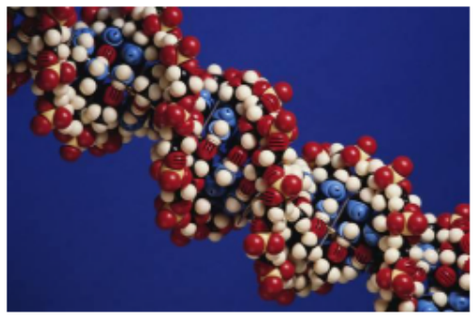
PT-08

European Union Reference Laboratory for Parasites (EURL-P)

Funded by the European Union

PT-08: Molecular identification of Echinococcus granulosus, Echinococcus multilocularis and Taenia spp.

Published 07/02/2022 - Edited 10/03/2026



Aim of the PT: Molecular identification of *Echinococcus granulosus*, *Echinococcus multilocularis* and *Taenia* spp.

Who can participate to the PT: all the National Reference Laboratories (NRLs) for parasites from EU member states or associated countries.

Procedure to analyze the PT items: the panel of items consists of four tubes containing: DNA extracted from faecal material contaminated with *Echinococcus granulosus sensu lato* (s.l.), DNA extracted from faecal material contaminated with *Echinococcus multilocularis*, DNA extracted from faecal material contaminated with *Taenia* spp. and a negative control. The EURL-P recommends the use of the multiplex PCR assay developed by Trachsel and colleagues (Parasitology, 2007, 134(Pt 6): 911-20). However, any other suitable molecular-based method will be accepted.

Criteria for the result evaluation: results evaluation is only qualitative. Participants have items, including the negative, are successfully identified.

If the PTP person in charge determines that the items distributed to participants or the will be notified immediately. The current scheme will be canceled, and a new one will be

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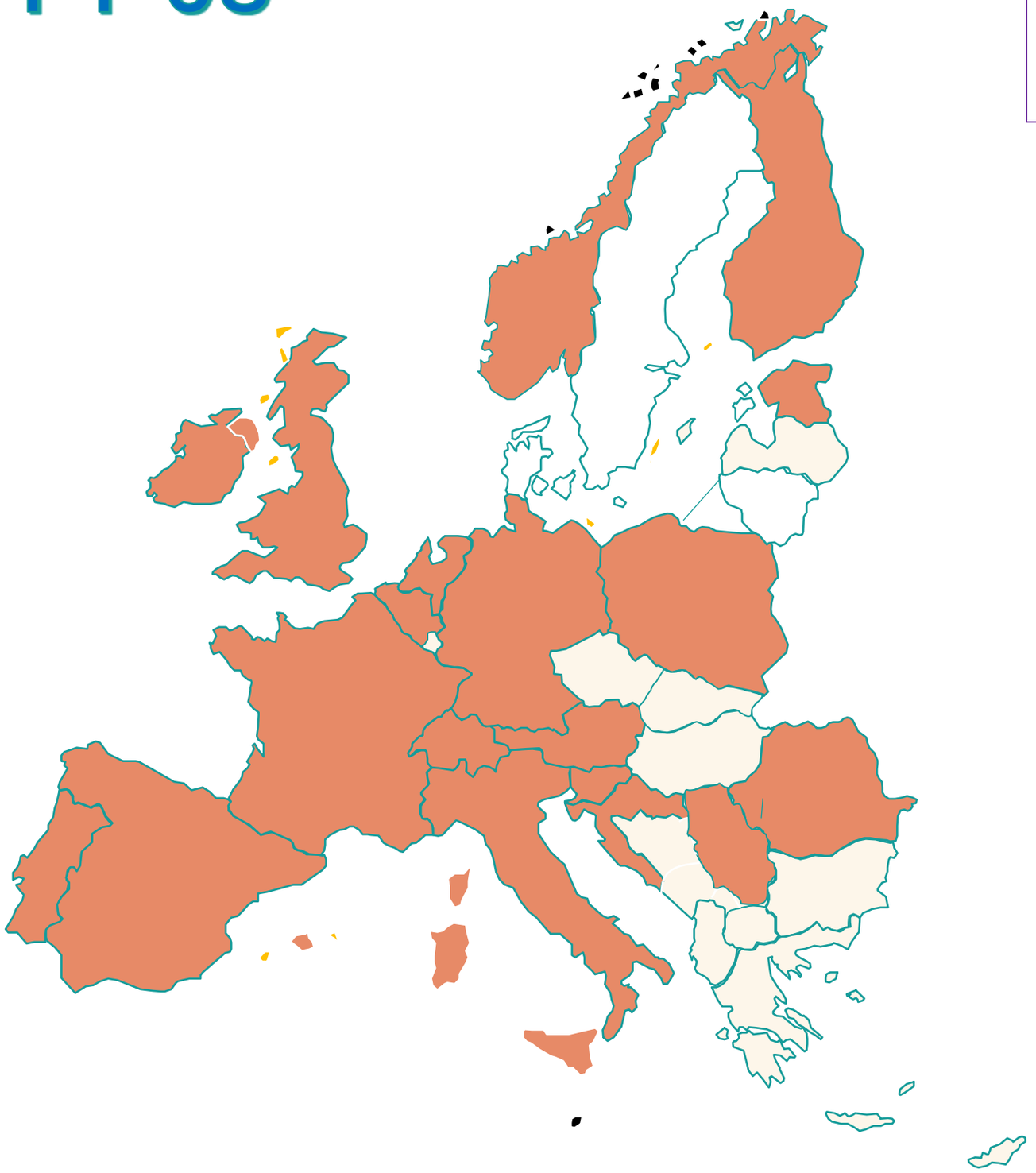
Instructions

EURLP

Procedure to analyze the PT items

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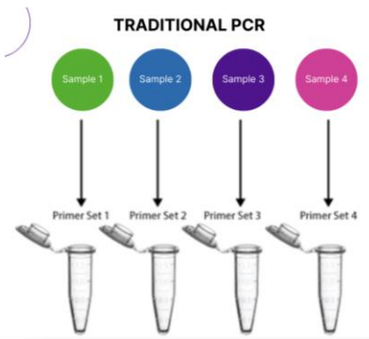
22 Participants

Austria
Belgium
Croatia
Denmark
Estonia
Finland
France
Germany
Ireland
Italy_1
Italy_2
Norway
Poland
Portugal
Republic of North Macedonia
Romania
Serbia
Slovenia
Spain
Switzerland
The Netherlands
United Kingdom

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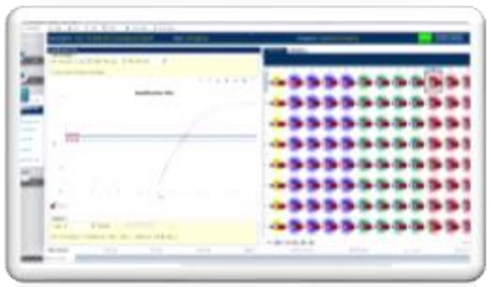
Detection method

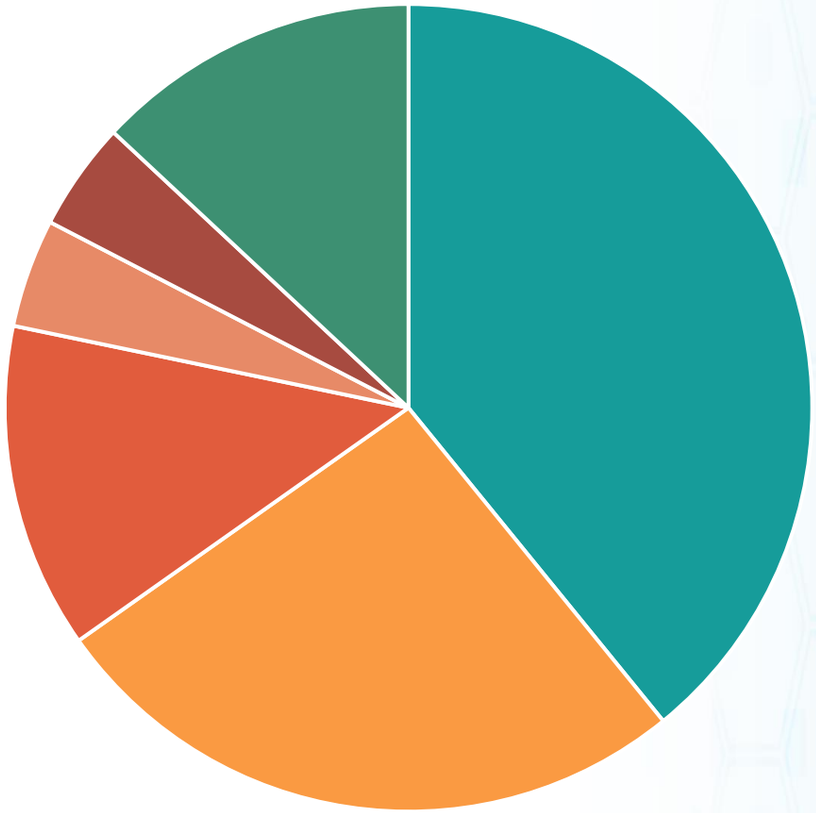
The EURLP recommends the use of the multiplex PCR by Trachsel *et al*.
however, any other suitable molecular-based method is accepted









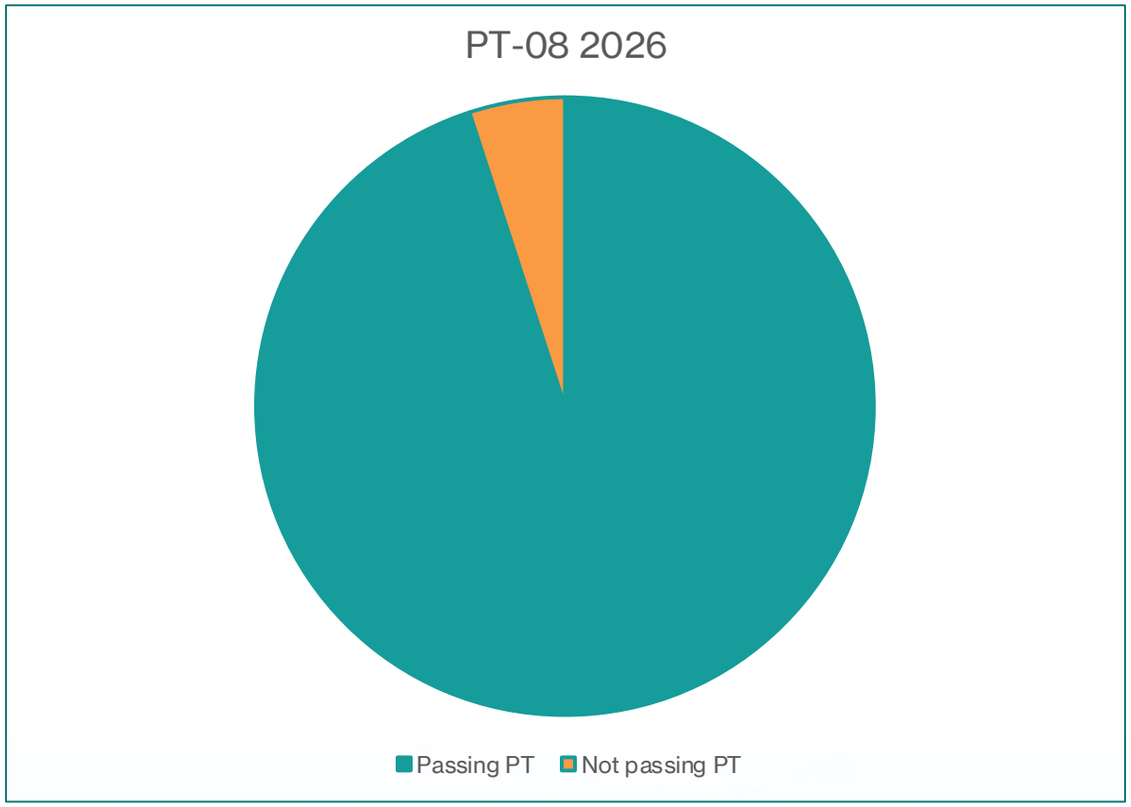


- Multiplex PCR by Trachsel et al, 2007
- Multiplex PCR by Trachsel et al, 2007 + other methods (Sequencing or Real Time PCR
- EURLP method (MI-15) + sequencing or + Real Time PCR (Øines et al, 2014)
- PCR (COI) + Sequencing
- In house method (PCR RFLP)
- Published method, not specified

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LAB CODE	RESULT
Emol_1	NEGATIVE
Emol_2	POSITIVE
Emol_3	POSITIVE
Emol_4	POSITIVE
Emol_5	POSITIVE
Emol_6	POSITIVE
Emol_7	POSITIVE
Emol_8	POSITIVE
Emol_9	POSITIVE
Emol_10	POSITIVE
Emol_11	POSITIVE
Emol_12	POSITIVE
Emol_13	POSITIVE
Emol_14	POSITIVE
Emol_15	POSITIVE
Emol_16	POSITIVE
Emol_17	POSITIVE
Emol_18	POSITIVE
Emol_19	POSITIVE
Emol_20	POSITIVE
Emol_21	POSITIVE
Emol_22	POSITIVE

PT-08 Results



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Results

Participants N=22

Participants passed N=21

Participants failed N=1

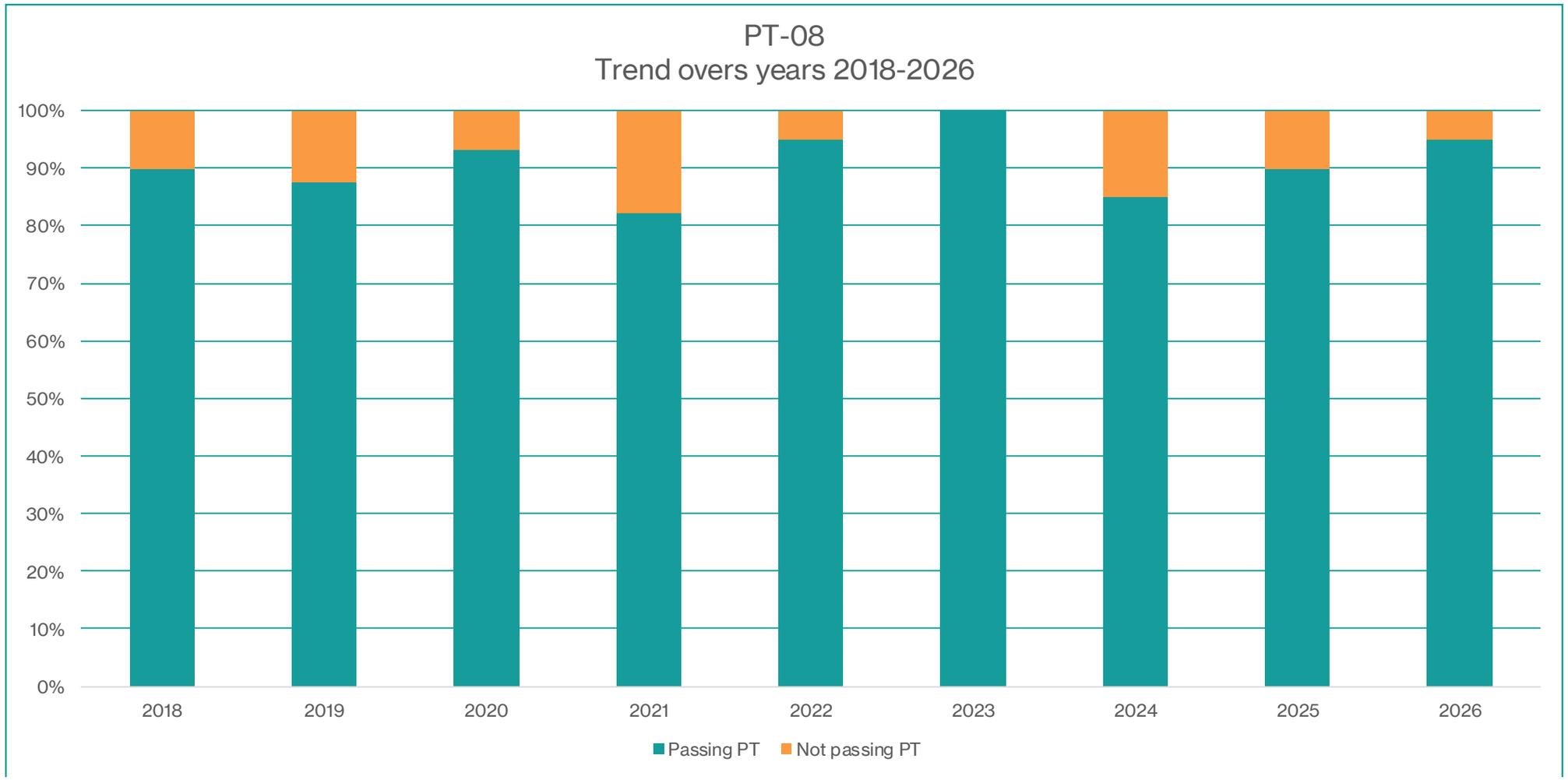
Observed	Assigned value	Outcome	Final evaluation
Negative *E. multilocularis* *Taenia spp.* *E. granulosus*	Negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	correct correct correct correct	POSITIVE
Negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	Negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	correct correct correct correct	POSITIVE
Taenia sp. *E. multilocularis* *Taenia spp.* *E. granulosus*	Negative *E. multilocularis* *Taenia krabbei* *E. granulosus*	incorrect correct correct correct	NEGATIVE

Four laboratories correctly identified the *Taenia* item as *Taenia krabbei*

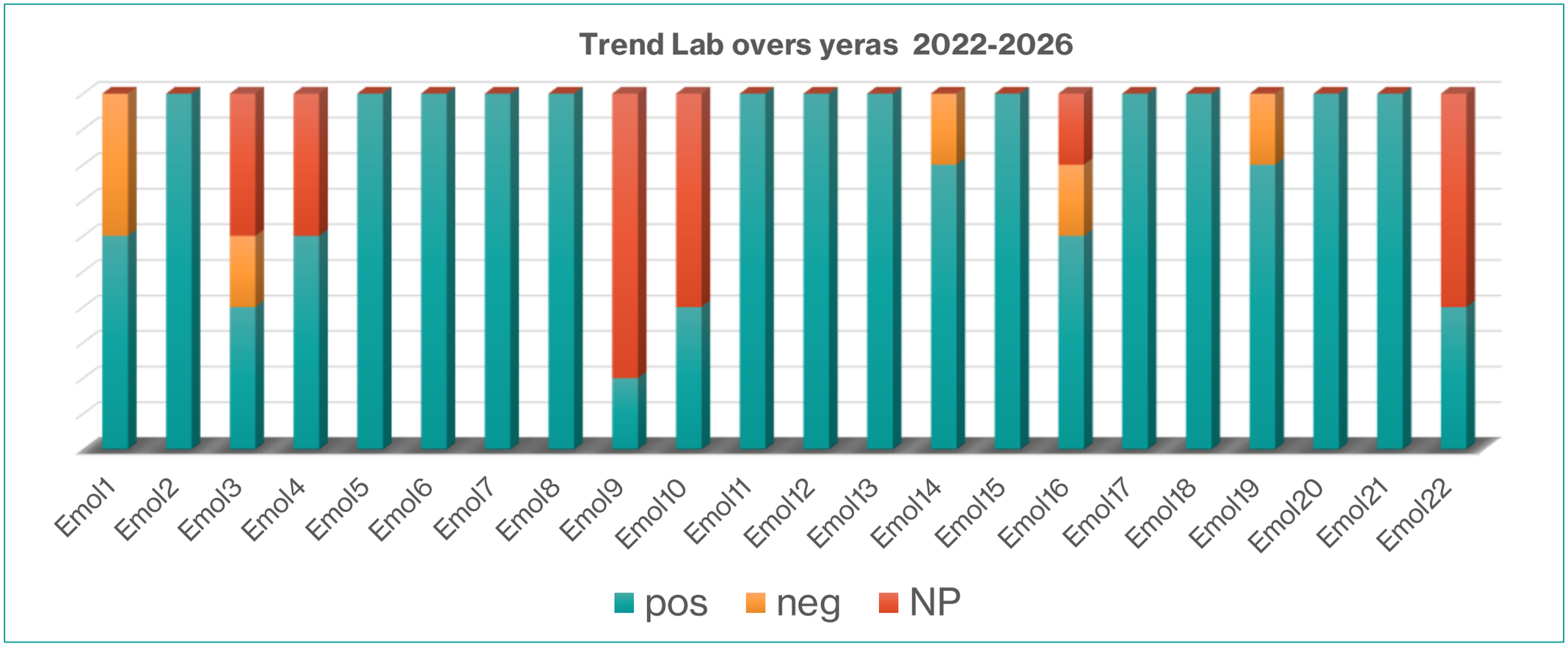
The negative item was misclassified as *Taenia sp.*

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Overtime comparison of results



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Trend Lab overs yeras 2022-2026
EmoI1
EmoI2
EmoI3
EmoI4
EmoI5
EmoI6
EmoI7
EmoI8
EmoI9
EmoI10
EmoI11
EmoI12
EmoI13
EmoI14
EmoI15
EmoI16
EmoI17
EmoI18
EmoI19
EmoI20
EmoI21
EmoI22
pos
neg
NP

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Conclusions

Out of 22 laboratories, 21 successfully passed the PT.

No specific errors were identified.

All the chosen methods appear to perform well, and NRL personnel demonstrate sufficient expertise to handle the PT-08

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Thanks for your attention



ACKNOWLEDGMENTS

Azzurra Santoro

and

EURLP TEAM