

PT: “Identification of *Trichinella* larvae at the species level by a molecular method”

Instructions

At arrival in the lab, the check of the package content and its condition of preservation has to be performed and reported in the **Form 1** (*MO/POPVI-03/04: Check package*)*

Test results have to be submitted within the date indicated in the PT request form. (use **Form 4** - *MO/POPVI-03/07: Results**- to submit the results); after this date, the form with the test results will not be accepted

Protocol to be used for the analysis of samples

The EURLP recommends performing the analysis using the EURLP method “Identification of *Trichinella* muscle stage larvae at species level by Multiplex-PCR”, however the participant can use any molecular method available in his/her laboratory, taking into account the following remarks:

- 1) if transfer of larvae from the original tube to a test tube is needed, it has to be done by taking larvae in a small volume of ethanol (2-5 µl), then is necessary to wait for ethanol evaporation before starting the DNA purification procedure;
- 2) to purify DNA from single larvae, we recommend to use kits based on magnetic beads that are more efficient compared to other systems. We suggest to use alternative DNA purification methods (based on silica-membrane), only on a pool of at least 6 larvae and to decrease the elution volume to 50-80 µl (if it is allowed by manufacturer) to obtain more concentrated DNA;
- 3) if the protocol used is multiplex PCR, every oligonucleotide must be present in the reaction mix at the same concentration;
- 4) the use of good quality Taq polymerase (e.g. hot start) reduces the risk to obtain non-specific PCR products.

You are requested to describe the method you applied in the **Form 3** (*MO/POPVI-03/06: Procedure**), and to list the instruments, reagents and materials used to perform the test in the **Form 2** (*MO/POPVI-03/05: List of instruments, reagents and materials used to perform the test*). In case you used a published method, indicate the reference and any variation from the original procedure in the dedicated column.

For any information or problem related to the PT participation, please address to:

Dr. Gianluca Marucci; e-mail: gianluca.marucci@iss.it

**Note: all forms will be sent by e-mail to the participants on the same day of PT panel shipping*