TDE | PFAS in Earthworms

Bioindicator Protocol — Analytical and Decision Criteria | 8 November 2026

1. Purpose and Rationale

Earthworms are used as bioindicators of PFAS exposure in soil-dwelling organisms. TDE qualification does not require the complete analytical absence of PFAS, but rather an environmental burden consistent with the voluntary thresholds adopted by the project and with findings from other matrices.

To establish practical thresholds, the EU maximum levels for PFAS in crustaceans and bivalve molluscs laid down in Regulation (EU) 2023/915, Annex I, point 4.2.2.2, are used by analogy. These levels are not legally applicable limits for earthworms and do not represent natural background concentrations.

2. Table of TDE Limits

Parameter	TDE acceptance ≤ 50% EU	Integrated assessment > 50% and ≤ 100% EU	TDE exclusion > 100% EU
PFOS	≤ 1.50	> 1.50–3.00	> 3.00
PFOA	≤ 0.35	> 0.35–0.70	> 0.70
PFNA	≤ 0.50	> 0.50–1.00	> 1.00
PFHxS	≤ 0.75	> 0.75–1.50	> 1.50
Sum of 4 PFAS	≤ 2.50	> 2.50–5.00	> 5.00

Unit: µg/kg fresh weight (as received). The upper limits of the intermediate range include the EU reference value. The sum covers PFOS + PFOA + PFNA + PFHxS, using the lower-bound convention.

3. Decision Rule

- Acceptance: if each of the four PFAS and their sum are at or below 50% of the corresponding EU maximum level, the “PFAS in earthworms” requirement is met. Territorial qualification remains subject to all other TDE requirements.
- Integrated assessment: if at least one of the five indicators exceeds 50% but none exceeds 100%, assessment of this matrix is suspended pending comparison with concentrations in water and sentinel plants, and with other relevant environmental findings.
- Exclusion: if at least one of the five indicators exceeds 100% of the corresponding EU maximum level, the TDE criterion is not met. Failure to qualify, or loss of previously granted qualification, follows confirmation of the analytical result and the general TDE procedures.

- Other PFAS: unusual results for compounds included in the panel but not among the four regulated PFAS must be reported and considered in the integrated assessment; compliance with the sum of the four alone does not guarantee the absence of concerns.

4. Essential Analytical Conditions

The earthworm sampling and preparation method must be standardised, paying attention to material in the intestinal tract and moisture measurement. Results are expressed on a fresh-weight basis; if the laboratory reports concentrations on a dry-matter basis, conversion using the actual measured moisture content is required. The appointed laboratory must document the suitability of the LC-MS/MS method and the corresponding LOQs for the “earthworm” matrix.

The sum of the four PFAS is calculated using a lower-bound approach (results below the LOQ contribute zero); the regulatory concentrations also cover linear and branched isomers. A result below the LOQ is particularly favourable but is not a prerequisite for TDE recognition.

5. References and Rationale

1. European Commission. Regulation (EU) 2023/915, Annex I, Section 4.2, point 4.2.2.2 (“Crustaceans and bivalve molluscs”); consolidated text. EUR-Lex: https://eur-lex.europa.eu/eli/reg/2023/915/2026-08-19/ita
2. European studies on PFAS bioaccumulation in earthworms: Falk et al. (2019), German Environmental Specimen Bank; monitoring of earthworms at a reference site and a ski area near Trondheim (2019); Lasters et al. (2023), garden sampling in Flanders. Published values cover different PFAS panels and are used as qualitative evidence of bioaccumulation, not to derive TDE thresholds directly.

Methodological note: these are voluntary territorial qualification thresholds, not universal ecotoxicological limits or a health assessment of earthworms. Integrated assessment cannot offset a confirmed exceedance of a TDE criterion explicitly defined as exclusionary.