Proposals for NORMAN Joint Programme of Activities 2026

Title	**Evaluation of the performance boundaries and applicability of high-throughput effect-directed analysis (HT-EDA) in the evaluation of receiving water quality.**
Type of activity	Pilot Study
Leader	EHU (Iker Álvarez Mora), VU (Maria Margalef, and Frederic Béen)
Topic / activities	***Background / Justification for the proposed activity:*** The applicability of high-throughput EDA has been demonstrated in multiple previous studies within the work package. In most successful cases, investigations have focused on matrices with high contaminant loads, such as wastewater. Cleaner samples, such as surface waters or drinking waters, represent a greater challenge for EDA studies, as contaminant concentrations often approach the sensitivity limits of both analytical techniques and bioassays. This project aims to explore the boundaries of EDA by evaluating the biological activity of receiving water samples through various bioassays, to determine the sensitivity thresholds required for each assay before and after fractionation. In later stages of the project, we will address the analytical limits by implementing techniques that reduce detection limits and expand the detectable chemical space (i.e., complementary ionization approaches and chromatographic separations using different columns). The work will aim to address key questions, including: (i) which bioassays are most suitable for EDA studies in receiving waters; (ii) what levels of pre-concentration can be achieved for each assay; (iii) whether effect drivers can be detected at such low levels; and (iv) for which bioassays toxicity can be reliably predicted using the modelling tools developed by other WPs. ***Description of the proposed activity and expected outcomes for 2026:*** The present project aims to explore the boundaries of EDA by performing activities in two different stages: **Stage 1: Sample extraction and bioassay testing. Evaluation of the bioassay limits of detection.** 1. Sample collection, extraction, and distribution (using large volume devices preferably). 2. Development of an inter-laboratory EDA-QAQC for fractionation alignment. 3. Toxicity testing using novel and established bioassays. 4. Chemical monitoring of all samples + toxicity prediction based on MS2 signal (MLinvitroTox). 5. Design tailored fractionation methods to capture toxic chromatographic regions. **Stage 2: Fractionation of most active samples and bioassay testing** 1. Fractionation of selected extracts for bioassay testing, and future chemical evaluation. 2. First prioritization of effect drivers using routine analysis (ESI). After stages 1 and 2 we expect to obtain a number of active fractions that will allow us to move to the follow-up project (Stage 3), where the chemical composition of these active fractions will be analysed using different analytical tools allowing to increase the chemical space coverage. This follow-up project will be in close collaboration with the cross-working group activity non-target screening (NTS). ***Added value / Link with other NORMAN activities and / or other projects*** The development of QAQC for EDA will allow different participants to perform collaborative EDA projects without the need to centralize all activities in one lab. This QAQC will facilitate for example the alignment of retention times between instruments performing fractionation, and chemical analysis, and application of non-target screening workflows for toxicity driver prioritization. Even the follow-up project (Stage 3) is not an activity included in this JPA, it is already planned for the following year, in close collaboration with the cross-working group activity non-target screening (NTS).
Participants	University of the Basque Country, Vrije Universiteit Amsterdam, Het Water Laboratorium, RECETOX, IDAEA-CSIC.
Proposed in-kind contribution	Person months
Contribution needed from NORMAN Association[1]	Budget for consumables for extraction and bioassay testing, and shipment of extracts. In total 15K.

[1] Please, provide here a transparent justification of the requested resources and of the in-kind contribution, thereby distinguishing between the costs associated with "person-months" for the organisation, the "travelling costs" for invited speakers and the costs for the logistics (e.g. meals, room rental etc.)