

## Complexity and challenges in agricultural water reuse monitoring from a German perspective

Pascal Hasselder <sup>a,\*</sup>, Manuela Helmecke<sup>b</sup>, Andreas Tiehm<sup>c</sup>, Benedikt M. Aumeier<sup>d</sup>, Christina Förster<sup>e</sup>, Daniel Zahn <sup>f</sup>, Johannes Ho<sup>c</sup>, Michael Stapf<sup>g</sup>, Nicole Zacharias<sup>h</sup>, Thomas Dockhorn<sup>i</sup>, Ulf Miehe<sup>g</sup> and Aki S. Ruhl<sup>a,j</sup>

<sup>a</sup> Section of Water Treatment (II 3.3), German Environment Agency, Schichauweg 58, 12307 Berlin, Germany

<sup>b</sup> Section of Water and Soil (II 2.1), German Environment Agency, Wörlitzer Platz 1, 06844 Dessau-Roßlau, Germany

<sup>c</sup> TZW: DVGW-Technologiezentrum Wasser, Karlsruher Straße 84, 76139 Karlsruhe, Germany

<sup>d</sup> Chair of Urban Water Systems Engineering, Technical University of Munich, Am Coulombwall 3, 85748 Garching, Germany

<sup>e</sup> Section of Microbiology of Drinking Water and Swimming Pool Water (II 3.5), German Environment Agency, Heinrich-Heine-Str. 12, 08645 Bad Elster, Germany

<sup>f</sup> Department of Environmental Analytical Chemistry, Helmholtz-Centre for Environmental Research – UFZ, Permoserstrasse 15, 04318 Leipzig, Germany

<sup>g</sup> Berlin Centre of Competence for Water, Grunewaldstraße 60/61, 10825 Berlin, Germany

<sup>h</sup> Institute for Hygiene and Public Health, University Hospital Bonn, Venusberg-Campus 1, 53127 Bonn, Germany

<sup>i</sup> Institute of Sanitary and Environmental Engineering, Technical University of Braunschweig, Pockelsstraße 2a, 38106 Braunschweig, Germany

<sup>j</sup> Chair of Water Treatment, Technische Universität Berlin, Straße des 17. Juni 135, 10623 Berlin, Germany

\*Corresponding author. E-mail: pascal.hasselder@uba.de

 PH, 0009-0004-4896-5976; DZ, 0000-0003-3038-9371

### ABSTRACT

Agricultural water reuse is one approach to mitigate water stress. In addition to the minimum requirements, the European water-reuse regulation 2020/741 mandates a risk management approach for agricultural water reuse. In contrast to the microbiological monitoring, the extent of the chemical risk assessment and monitoring is not clearly defined. The resulting complexity of a typical agricultural water-reuse scheme was analyzed. Potentially relevant parameters were identified based on European and German regulatory frameworks, concerning key subjects of protection. An interdisciplinary assessment of efforts and challenges, regarding required analyses, was accomplished, using expertise from recent research investigating agricultural water reuse in Germany. Suggestions were provided for disinfection validation, microbiological monitoring parameters and analytical methods. Additionally, chemical indicator parameters were suggested to address relevant processes during monitoring. Both microbiological and chemical parameters presented analytical challenges, which were described with future needs to support water-reuse implementation. Costs for analyses were estimated using available price information, highlighting the high costs of certain analyses, especially for organic micropollutants. Therefore, analyses need to be further facilitated by the application of process indicators and the implementation of cost-effective, multi-target methods tailored to the requirements of risk management for agricultural water reuse.

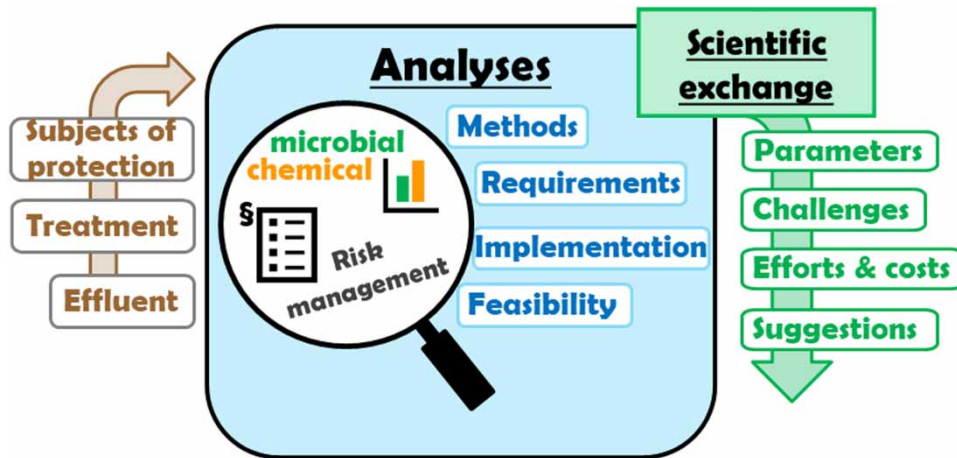
**Key words:** analyses, agriculture, costs, indicators, regulations, reuse

### HIGHLIGHTS

- Agricultural water reuse and its regulatory framework require complex analyses.
- Challenges exist regarding microbial and chemical analyses.
- Efforts and costs for analyses were estimated.
- Parameters exist for microbial and chemical monitoring.

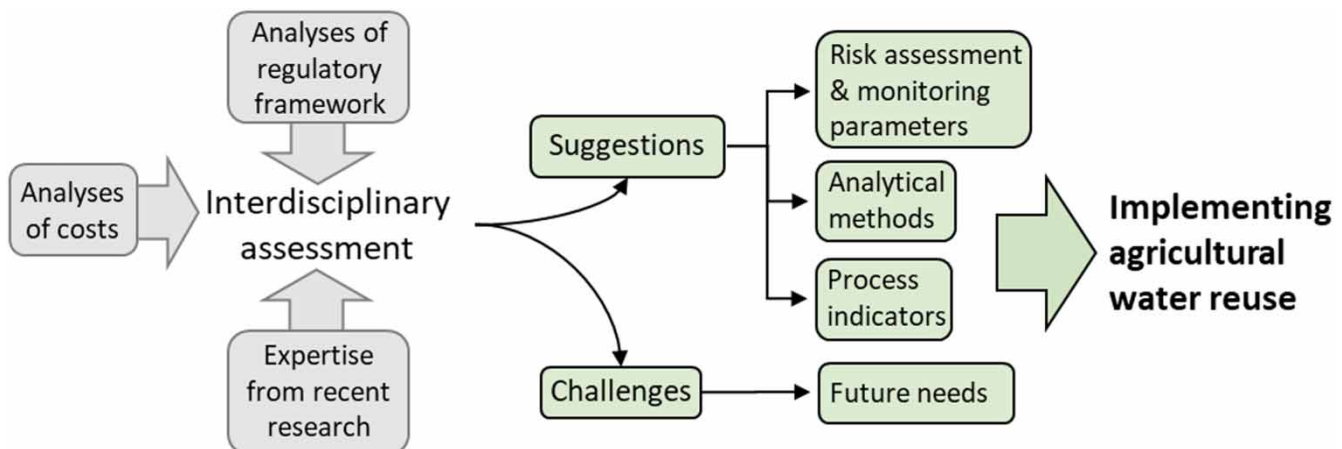
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## GRAPHICAL ABSTRACT



## 1. INTRODUCTION

Water scarcity and the increasing global demand for sustainable water management have highlighted the potential of water reuse as one important strategy worldwide (UN 2015; UNESCO 2017; Christou *et al.* 2024). In the European Union (EU), this is particularly critical for regions experiencing growing pressure on freshwater resources due to climate change, population growth and high agricultural demands (EU 2013a, 2018). In response, the EU adopted the water-reuse regulation (EU 2020a). This regulation not only defines minimum requirements for reclaimed water but also introduces a mandatory risk management approach, emphasizing the need to safeguard public health, the environment and agricultural production. A comprehensive investigation, bringing together the complex requirements of risk management, analytical capabilities, as well as related costs, is currently missing but is vital to successfully achieve implementation. As shown schematically in Figure 1, this contribution draws on an interdisciplinary assessment based on expertise from recent research projects, focusing on agricultural water reuse in Germany, including monitoring, sampling and analyses. This expertise served as a basis for informed discussions without delving into the details of each project. Additionally, the assessment involves an analysis of requirements considering the regulatory framework and the costs of analysis. These efforts have provided valuable insights into overcoming technical and operational challenges, ensuring the feasibility, safety and consideration of analysis-related costs of agricultural water reuse. More specifically, this paper aims to (1) discuss relevant parameters according to the subjects of protection, (2) identify challenges related to monitoring and analytical methods, (3) assess the associated efforts of monitoring and implementation and (4) suggest parameters for water-reuse monitoring.



**Figure 1** | Conceptual diagram showing the inputs to the interdisciplinary assessment (gray) and achieved outputs (green).

By sharing these findings, we hope to contribute to further facilitating the implementation of water-reuse projects, not only in Germany but also in other EU member states.

## 2. SURVEILLANCE OF AGRICULTURAL WATER REUSE

### 2.1. Regulatory framework

The EU water-reuse regulation (EU 2020a) distinguishes between four water quality classes A to D, covering the irrigation of fresh produce that is consumed raw (class A) to the irrigation of non-food crops (class D). Site-specific health and environmental risks need to be addressed in a risk management plan, as outlined in the EU regulation and the related delegated regulation (EU 2024a). While the EU water-reuse regulation (EU 2020a) applies directly in all participating member states, it is not the only relevant legislation but rather closes a gap between the existing environmental policies and those for food safety. The Urban Wastewater Treatment Directive (EU 1991) sets standards for the treatment process and the discharge of treated effluent. Its revised version (EU 2024b) also sets goals for advanced treatment, aiming at the elimination of micropollutants for wastewater treatment plants (WWTPs). Any water-reuse application needs to comply with the Water Framework Directive's (EU 2000) principle of non-deterioration, the Groundwater Directive's EU-wide groundwater quality standards (EU 2006), as well as the Environmental Quality Standards for surface waters (EU 2013b). Additional quality standards are expected as part of the revision of the EU water directives and are likely to include quality standards for pharmaceuticals in groundwater (EU 2022a). Further, the Nitrates Directive (EU 2008), the Bathing Water Directive (EU 2014) and the Drinking Water Directive (EU 2020b) may need to be considered in relation to the site-specific situation. In addition, there is relevant EU food (and feed) hygiene legislation as well as related documents. Depending on the type of irrigated produce, relevant requirements include those on the hygiene of foodstuff (EU 2004), feed hygiene (EU 2022b), microbiological criteria for foodstuff (EU 2020c), maximum contaminants in foodstuff (EU 2023), levels of pesticides in food and feed (EU 2005), use of sewage sludge (EU 1986) and the protection of animal health (EU 2009a, 2009b).

For the German context, there is corresponding national legislation; e.g. the provisions of the EU Groundwater Directive (EU 2006) are implemented in the German Groundwater Ordinance (DE 2010). Additionally, in Germany, the Federal Soil Protection and Contaminated Sites Ordinance (DE 2021) sets out precautionary values for inorganic and organic substances. Relevant norms relate to 'Hygienic concerns of irrigation water' (DIN 1999), 'Testing and evaluation of water for irrigation' (DIN 2009) and 'Guidelines for treated wastewater use for irrigation projects' (ISO 2020). The German Working Group on water issues of the Federal States and the Federal Government (LAWA) has published recommendations for a national regulatory framework to implement the EU regulation on water reuse (LAWA 2022). Respective legal provisions are currently under preparation. In addition, the German network of experts for water, wastewater and waste (DWA) is in the process of finalizing national guidelines that describe the technical requirements for agricultural and urban irrigation with reclaimed water (DWA in progress).

### 2.2. Water reuse system and analyses for risk management

The identification of hazards, exposure routes and the relevant subjects of protection enables one to initially assess risks before deciding on measures of risk management and a regular monitoring program. Risk management is connected to the entire reuse system, including the sampling points (SP) 1 to 7, schematically shown in Figure 2.

Conventional wastewater treatment is a key process for water reuse. Besides households, indirect industrial dischargers contribute to the WWTP inflow at SP 1. For the initial risk assessment, the evaluation of indirect dischargers, therefore, provides valuable information for sampling at SP 2 and the extent of the analyses. Advanced treatment (SP 3) can vary. Under the EU water-reuse regulation (EU 2020a), agricultural water reuse requires at least disinfection, while the highest quality class also mandates filtration. Additionally, the removal of chemical pollutants, such as organic micropollutants (OMPs), is required. An established solution is the use of activated carbon or ozonation (Jekel *et al.* 2015).

Subsequently, the reclaimed water is transported before being irrigated directly or stored *a priori*. The intermediate storage offers the possibility of balancing out fluctuations in irrigation demands and ensuring a continuous water supply for agriculture. Moreover, mixing in water from other sources could be optionally used to manage water quality. Such a process must be implemented in risk management accordingly and might require additional analyses. Although storage is not designated as a sampling point in the monitoring framework, analyzing water quality during storage can provide valuable insights into microbial dynamics, such as the potential regrowth of certain bacteria. Monitoring the point of use (SP 4) enables the assessment of the final reclaimed water quality and the prevention of the subsequent subjects of protection from potential hazards.



In Germany, there are recommendations to include *Enterococcus* spp. into the regular monitoring (based on the respective standard (DIN 1999)). Some stakeholders also suggest applying validation monitoring to water quality classes B and C for food crop irrigation (DWA in progress; LAWA 2022). Standard methods for the relevant parameters (partly requested by the EU regulation) are listed in Table 1.

Depending on the source water quality and the intended use of the reclaimed water, additional monitoring parameters may be relevant to assess long-term effects, including the presence of antibiotic-resistant bacteria (ARB) and antibiotic resistance genes (ARG). ARG can persist in treated water and spread through wastewater discharge (Stange *et al.* 2019; Sauter *et al.* 2021), potentially contaminating surface waters and agricultural environments. While some studies show that irrigation influences ARG levels in plants (Cerqueira *et al.* 2019), others show a minimal impact (Shamsizadeh *et al.* 2021; Liu *et al.* 2022), highlighting knowledge gaps. Insufficient treatment may allow antibiotic residues, ARB and ARG, to persist and spread via mobile genetic elements. The EU has recognized this issue, mandating in the revision of the Urban Wastewater Treatment Directive (EU 2024b) monitoring of resistance parameters in a large WWTP by 2025, emphasizing the need for improved assessment.

### 3.2. Chemical parameters

The EU water-reuse regulation (EU 2020a) and the associated delegated regulation (EU 2024a) do not have a standard list of relevant chemical parameters, which must be assessed according to site-specific conditions in the mandatory risk management and are more likely to represent chronic risks. The relevant parameters, specified in EU and German legislation relating to wastewater treatment, reclaimed water, groundwater and soil protection, are summarized in Table 2. Partly, substances that are included in the listed regulatory framework are excluded from Table 2 due to their lack of relevance for water reuse. For example, volatile hydrocarbons are rather important at contaminated sites. The explicit decisions on all excluded parameters are documented in the Supplementary information.

#### 3.2.1. Treated wastewater

The EU water-reuse regulation (EU 2020a) specifies minimum requirements for the quality of the reclaimed water, including biological oxygen demand (BOD), total suspended solids (TSS) and turbidity. Beyond that, typical operational parameters for

**Table 1** | List of microbiological parameters with respective methods of detection, besides the EU requirements with regard to SPs indicated in Figure 2

Group	Parameter	EU requirements and respective SPs in Figure 1	Standard methods
Bacteria	<i>E. coli</i>	Quality class A: removal exceeds 5 log units between SP 1 and SP 3 for the validation of the treatment train Quality classes A–D: $\leq 10$ , $\leq 100$ , $\leq 1,000$ and $\leq 10,000$ cfu/100 mL in weekly samples from SP 3 or 4	DIN EN ISO 9308-1 (2017) DIN EN ISO 9308-2 (2014) DIN EN ISO 9308-3 (1999)
	<i>Enterococci</i>	Not required in the EU but recommended in Germany (DIN 1999)	DIN EN ISO 7899-1 (1999) DIN EN ISO 7899-2 (2000)
	<i>Legionella</i> spp.	<1,000 cfu/L in biweekly samples from SP 4, and only in the case of aerosol formation	DIN EN ISO 11731 (2019)
Protozoa	<i>C. parvum</i> spores	Quality class A: Removal exceeds 4 log units between SP 1 and 3 for the validation of the treatment train	DIN EN ISO 14189 (2016)
Viruses	Somatic coliphages	Quality class A: Removal exceeds 6 log units between SP 1 and 3 for the validation of the treatment train	DIN EN ISO 10705-2 (2002)
	F-specific coliphages	Quality class A: Removal exceeds 6 log units for the validation between SP 1 and 3 for the validation of the treatment train	ISO 10705-1 (1995)
Parasites	Helminth oocysts	< 1 egg/L for the irrigation of pastures or forage	No standard available

The purpose of the analyses for risk management is described by either regularly sampling or validation for Quality class A.

**Table 2** | Chemical parameters from the regulatory framework that are potentially relevant for agricultural water reuse

	EU urban wastewater treatment directive (recast) (EU 2024b)	EU water reuse regulation (EU 2020a)	EU groundwater directive (EU 2006)	German groundwater ordinance (DE 2010)	German soil protection ordinance (DE 2021)	Testing and evaluation of water for irrigation (DIN 2009)	International standard for water reuse for irrigation (ISO 2020)
<b>Operational</b>							
BOD	L	L					L
Chemical oxygen demand (COD)	L						L
EC			L	L		L	L
pH				L	L	L	L
TOC <sup>a</sup>	L				L		
TSS	L	L		L		L	L
Turbidity <sup>b</sup>		L					
<b>Anions</b>							
Chloride			L	L		L	L
Sulfate			L	L			
Cyanide							
<b>Heavy metals and other elements</b>							
Antimony					L		
Arsenic			L	L	L	L	
Boron				L	L	L	L
Cadmium	L		L	L	L	L	L
Chromium				L	L	L	L
Cobalt				L	L		L
Copper				L	L	L	L
Lead	L		L	L	L	L	L
Mercury	L		L	L	L	L	L
Molybdenum					L	L	L
Nickel	L			L	L	L	L
Selenium					L		L
Sodium						L	L
Thallium				L	L		
Vanadium				L			L
Zinc				L	L	L	L
<b>Nutrients</b>							
Total nitrogen	L						
Total phosphorous	L						
<b>Corrosion inhibitors</b>							
1H-Benzotriazole	L						
4- and 5-Methylbenzotriazole	L						
<b>Industrial</b>							
PFAS <sub>4</sub>			P				

(Continued.)

Table 2 | Continued

	EU urban wastewater treatment directive (recast) (EU 2024b)	EU water reuse regulation (EU 2020a)	EU groundwater directive (EU 2006)	German groundwater ordinance (DE 2010)	German soil protection ordinance (DE 2021)	Testing and evaluation of water for irrigation (DIN 2009)	International standard for water reuse for irrigation (ISO 2020)
PFAS <sub>20</sub>			P				
PFAS <sub>24</sub>			P				
PAH <sub>16</sub>					L		
PCB <sub>6</sub> and PCB 118					L		
<b>Pharmaceuticals and personal care products</b>							
Amisulpride	L						
Candesartan	L						
Carbamazepine	L		P				
Citalopram	L						
Clarithromycin	L						
Diclofenac	L						
Hydrochlorothiazide	L						
Irbesartan	L						
Metoprolol	L						
Primidone			P				
Sulfamethoxazole			P				
Venlafaxine	L						

L indicates current legislation, P indicates the current official proposals for amending the GWD (EU 2022a, 2024c).

<sup>a</sup>TOC may replace COD in water-reuse contexts because the standard method is more sensitive. TOC and COD can be correlated in a specific water matrix.

<sup>b</sup>Turbidity may replace TSS in water-reuse contexts when filtration is applied. In this case, turbidity is more sensitive. TSS and turbidity can be correlated in a specific water matrix.

water quality surveillance and treatment process control are total organic carbon (TOC), UV absorbance and transmittance at 254 nm, pH and electrical conductivity (EC). Except for UV absorbance and transmittance, the operational parameters are widely covered in the regulatory context (not in the EU water-reuse regulation).

Among the inorganic parameters in Table 2, heavy metals are the most prominent. Different removal mechanisms in WWTPs leave low concentrations in WWTP effluents. Compared to lead, mercury and cadmium, lower but still moderate-to-high removals were reported for nickel in a German monitoring of the WWTP (UBA 2020). Irrigation with reclaimed water might be a potential source of the more mobile heavy metals. Nutrients, such as nitrogen and phosphorus, are common surveillance parameters for the discharge of treated wastewater, as it is obliged by the EU urban wastewater treatment directive (EU 1991).

OMP in wastewater originate from daily life applications and products, such as pharmaceuticals, personal care, material additives and many more (Loos *et al.* 2013; Finckh *et al.* 2022). Industrial discharge is another potential source of organic contaminants in wastewater and might lead to elevated concentrations of individual chemicals. Polyaromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs) are persistent and hydrophobic contaminants. The so-called 16 EPA-PAH (PAH<sub>16</sub>), a list proposed by the US Environmental Protection Agency (EPA), is typically analyzed as a representative PAH indicator. Likewise, six different congeners of PCB (PCB<sub>6</sub>) are commonly analyzed, besides the congener PCB 118, indicating dioxin-like PCB. In a Germany-wide monitoring of 49 conventional WWTPs, a good PAH elimination to a median effluent concentration below 10 ng/L was reported (UBA 2020). However, specific industries might discharge higher concentrations. In an industrial area in Italy, residual PAH and PCB concentrations up to several hundred ng/L were measured in WWTP effluents after advanced treatment, such as ozonation and activated carbon or after dilution of the treated wastewater with river water (Bruzzoniti *et al.* 2024). Pesticides, biocides and insecticides (in the following summarized as pesticides) in

wastewater originate from agricultural application or from urban run-off, for example, after release from construction materials (Jekel *et al.* 2015; UBA 2020). Although WWTP effluents may contain more pesticides than agricultural surface run-off, the latter contains higher loads (Masoner *et al.* 2023). Per- and polyfluoroalkyl substances (PFAS) originate from a wide range of applications in daily life products and industry (Glüge *et al.* 2020), are highly persistent and ubiquitously appear in the environment (Buck *et al.* 2011). Perfluorooctanesulfonic acid (PFOS), for example, partly adsorbs to activated sludge, while others, such as perfluorooctanoic acid (PFOA), persist in conventional wastewater treatment, causing relevant concentrations in WWTP effluents (Schultz *et al.* 2006; Sinclair & Kannan 2006; UBA 2020). The environmental relevance of trifluoroacetic acid (TFA), the final transformation product of many PFAS, is still debated (Arp *et al.* 2024). Secondary effluents or municipal WWTPs are key point sources for a large number of OMP used in health care and households, such as pharmaceuticals, corrosion inhibitors, sweeteners and many more (Loos *et al.* 2013; Finckh *et al.* 2022). While so far hardly addressed in the regulatory framework, the recast of the EU Urban wastewater treatment directive (EU 2024b) provides a list of 12 OMPs to evidence advanced treatment efficacy. This list mainly considers OMPs that originate from households but not from industry, such as PAH, PCB and PFAS. The German Centre for Micropollutants provides a list of 21 relevant OMPs based on the assessment of an expert committee (UBA 2025).

### 3.2.2. Soil

The German soil protection ordinance (DE 2021) provides precautionary values for the respective parameters that are listed in Table 2. In addition to the precautionary values, threshold values prescribe remediation measures if exceeded. Threshold values are provided for various exposure routes, such as from soil to groundwater, plants or humans. Since these thresholds primarily concern contaminated sites, they are not included in Table 2.

Heavy metals potentially cause long-term soil contamination. With respect to expected heavy metal concentrations in WWTP effluents, risks of soil contamination related to agricultural water reuse are assumed to be low, while nickel, lead, cadmium and mercury should be assessed depending on effluent characteristics (UBA 2016; LAWA 2022). EC of soil extracts is used as a surrogate parameter to monitor soil salinization according to technical guidelines for testing and evaluating water for irrigation (ISO 2009). Imbalance of sodium in relation to calcium and magnesium (sodium adsorption ratio) may cause disaggregation and slaking in respective soils.

Hydrophobic and persistent organic compounds are prone to adsorption and relevant parameters regarding long-term soil contamination. Examples are PAH and PCB with very high retardation factors (Estoppey *et al.* 2024). Elevated PAH and PCB soil concentrations were reported for a site in Tunisia, which is long term irrigated with secondary effluents from municipal and industrial wastewater (Haddaoui *et al.* 2016). Depositions of diffuse PAH releases from combustion processes are the main source of PAH accumulation in top soils, while releases from soils mainly occur with run-off and concomitant particle transport (Gocht *et al.* 2007). For PAH<sub>16</sub> and PCB<sub>6</sub>, precautionary values are provided in the German soil protection ordinance (DE 2021). From various PFAS, the long-chain PFAS are relevant regarding soil contamination (Rasmusson & Fagerlund 2024). The German soil protection ordinance (DE 2021) does not provide precautionary values or maximum yearly loads for PFAS. Different studies demonstrated the occurrence of various OMP in soils originating from agricultural water reuse (Paz *et al.* 2016; Ben Mordechay *et al.* 2021; Kodešová *et al.* 2024; Ahmadi *et al.* 2025). So far, no regulatory thresholds exist for maximum concentrations of such OMPs in soils or maximum yearly loads.

### 3.2.3. Groundwater

Vertical mass transport in soils occurs when mass input exceeds attenuation or the distribution equilibrium between the phases of the soil–water–air system leads to a transfer into the water phase, while the soil water content leads to water percolation. Generally, persistent and mobile substances bear a high potential to reach groundwaters and pass river bank filtration (Bautista *et al.* 2024; Muschket *et al.* 2024).

Besides several typical inorganic parameters and site characteristics such as pH, the German groundwater ordinance (DE 2010) considers the pesticides of substance classes and their metabolites, as well as nutrients. While pesticide concentrations are typically low in treated wastewater (UBA 2020; Finckh *et al.* 2022), many of them are used intentionally in agriculture. In the Supplementary Information, seven exemplary pesticides show that the application in a hypothetical scenario of 300 mm irrigation volume and a concentration of 1 µg/L in the irrigation water would be, in most cases, less than 1% compared to the instructions for use. Thus, monitoring pesticides in agricultural environments might be superimposed by factors other than water reuse. However, pesticides that are not used in agriculture but are present in WWTP effluents might be relevant.

The EU working group on groundwater published a voluntary watchlist for groundwater, which considers two PFAS (perfluorododecanoic acid and perfluoroundecanoic acid) and nine pharmaceutical substances (clopidol, crotamiton, diatrizoic acid, sulfadiazine, primidone, sotalol, ibuprofen, erythromycin, and clarithromycin) (EU 2019). In the EU, there is an ongoing negotiation on amending the groundwater directive. In detail, thresholds for the sums of 4, 20 and 24 PFAS (PFAS<sub>4</sub>, PFAS<sub>20</sub> and PFAS<sub>24</sub>), carbamazepine, sulfamethoxazole and primidone are considered (EU 2022a, 2024c).

### 3.2.4. Produce

Environmental factors, such as the level of exposure or soil organic matter, impact the amount of OMP uptake by plants; however, a general process description remains uncertain since the uptake varies from plant to plant and from substance to substance even in similar environments (Fernandes *et al.* 2024). Several studies reported high uptake of carbamazepine by plants (Ben Mordechay *et al.* 2021; García-Valverde *et al.* 2023; Fernandes *et al.* 2024). For children younger than 11 months, the estimated daily intake of carbamazepine may exceed the acceptable daily intake if secondary effluent was used for irrigation, but not if advanced water treatment was done (Benelhadj *et al.* 2024). Ahmadi *et al.* (2025) found detectable but, in general, low loads of 18 OMPs in agricultural produce irrigated with different treated waters, i.e. reclaimed water according to EU minimum requirements, advanced treated water and tap water. None of the measured OMP exceeded the acceptable daily intake for children or adults assuming a vegetarian diet, irrespective of irrigation water quality. The highest hazard quotients were found for hydrochlorothiazide and carbamazepine due to the very low acceptable daily intake and the elevated load in agricultural produce, respectively. Fernandes *et al.* (2024) reviewed the uptake of OMP by plants in soil-plant and hydroponic systems, considering that many OMPs form various substance classes and concluded that associated human health risks are below the recognized thresholds of acceptable daily intake.

## 4. CHALLENGES, EFFORTS AND COSTS RELATED TO ANALYSES

The implementation of monitoring programs itself might be challenging for sampling requirements in terms of time and techniques. Related efforts are described in the Supplementary Information.

### 4.1. Microbiological analyses

#### 4.1.1. Analytical challenges

During monitoring campaigns as part of the German research activities, the influents of 10 different WWTPs were compiled and analyzed for the parameters required for the validation monitoring, as mandated by the EU water-reuse regulation (Seis *et al.* 2024).

Coliphages in those WWTP influents occurred at concentrations that are too low for the validation since even near-complete elimination does not provide sufficient decimal logarithmic units (Seis *et al.* 2024). Therefore, large volumes of samples after the disinfection might be necessary in order to sufficiently lower the quantification limit. However, that may complicate the transportation and processing of the samples. Thus, on-site filtration methods have been suggested, e.g. the VIRADEL method (Fout *et al.* 2015; Betancourt 2023) or the hollow fiber ultrafiltration method (Hill *et al.* 2009), which is not routinely established and standardized in German laboratories. While dosing coliphages directly into the influent of a WWTP is not feasible, spiking MS2 and phiX174 phages before disinfection can serve as an alternative method to evaluate log removal credits for phages, as shown by Ho *et al.* (2024). These experiments allow for the controlled assessment of treatment performance by simulating pathogen concentrations under defined conditions.

Water constituents and associated flora in the irrigation system interfere with culture-based quantification methods of *Legionella* spp. A further developed method (LANUV 2019), therefore, provides a solution and performs very well in the research activities. It includes pretreatments, such as adding antibiotics, while using varying cultivation media and multiple batches help to achieve evaluable results.

Different methods for the detection of helminth oocysts were described (Mahapatra *et al.* 2022). However, no international standard method is available, and quantification is still challenging (Arthur 2021). Concerning commercial laboratories, an analytical gap for helminth oocysts in water was identified, leaving a discrepancy between the legal requirements and the available analytical capabilities.

#### 4.1.2. Efforts and costs

Alongside the German research activities, price information for the analyses was gathered and used to estimate costs of a realistic analysis program for agricultural water reuse. Considering the one-time process validation, the parameters *E. coli*, *C. perfringens* spores, as well as somatic and f-specific (total) coliphages, have to be analyzed. For example, analyzing 10 samples each from the two SPs at the WWTP influent and after the disinfection, considering these parameter costs between 4,800 and 11,000 Euros according to typical costs listed in Table 3. Total microbial monitoring costs for one irrigation season in Germany (20 weeks assumed) will range from 2,000 to 4,100 € (weekly: *E. coli*, *Enterococci*; biweekly: *Legionella* spp). The costs for the analyses of helminth oocysts could not be included because no laboratory service could be found. Unlike bacterial parameters, which are enriched using membrane filtration in standardized methods, analyzing coliphages becomes significantly more labor-intensive when larger sample volumes are required. Additional costs for services, such as sampling and logistics, as well as enrichment, are not included.

## 4.2. Chemical analyses

### 4.2.1. Analytical challenges

Very low concentrations of relevant compounds in reclaimed water can make enrichment necessary in order to reach sufficient quantification limits. In addition, separating target compounds from unwanted matrix, e.g. via solid phase extraction, is accompanied by extensive work, considering the extraction procedure itself and quality control of the applied method. If it is required for risk management, extraction from soil and plant samples, in particular, is accompanied by complex sample matrices that might negatively impact the quality of analyses. However, it is necessary to reduce disturbance by a matrix (Montemurro *et al.* 2019), and when the target compounds exhibit a polarity comparable to that of the unwanted matrix, separation becomes difficult. Regarding soil sample extraction, the time of soil sampling might also be relevant. Without leaching during irrigation, at the end of the irrigation season, salt contents in soils are the highest, which might negatively affect analyses, especially for polar analytes (Müller *et al.* 2020).

### 4.2.2. Efforts and costs

Based on price information gathered during the involved research activities, costs for analyses were estimated and provided in the Supplementary Information. It is noteworthy that analyses of requirements for agricultural water reuse are not always covered by typical parameter sets of laboratory services. Parameter sets, such as PAH<sub>16</sub>, PCB<sub>6</sub>, PFAS<sub>20</sub> and heavy metals, are often based on regulatory requirements, e.g. the drinking water ordinance (DE 2023) or soil protection ordinance (DE 2021). Analyses of individual OMPs are typically more expensive than those of parameter sets. However, for many of the OMP listed in Table 2, the only option was to estimate a price between 220 and 550 Euros based on price information for other OMPs and for individual analyses.

Readily available data from regular WWTP surveillance might be helpful to reduce the extent of analyses. At least data regarding nitrogen and phosphorus, many of the operational parameters and, to a certain extent, heavy metals are likely available from regular WWTP surveillance. Depending on the potential site-specific risks, the extent of analyses that are required

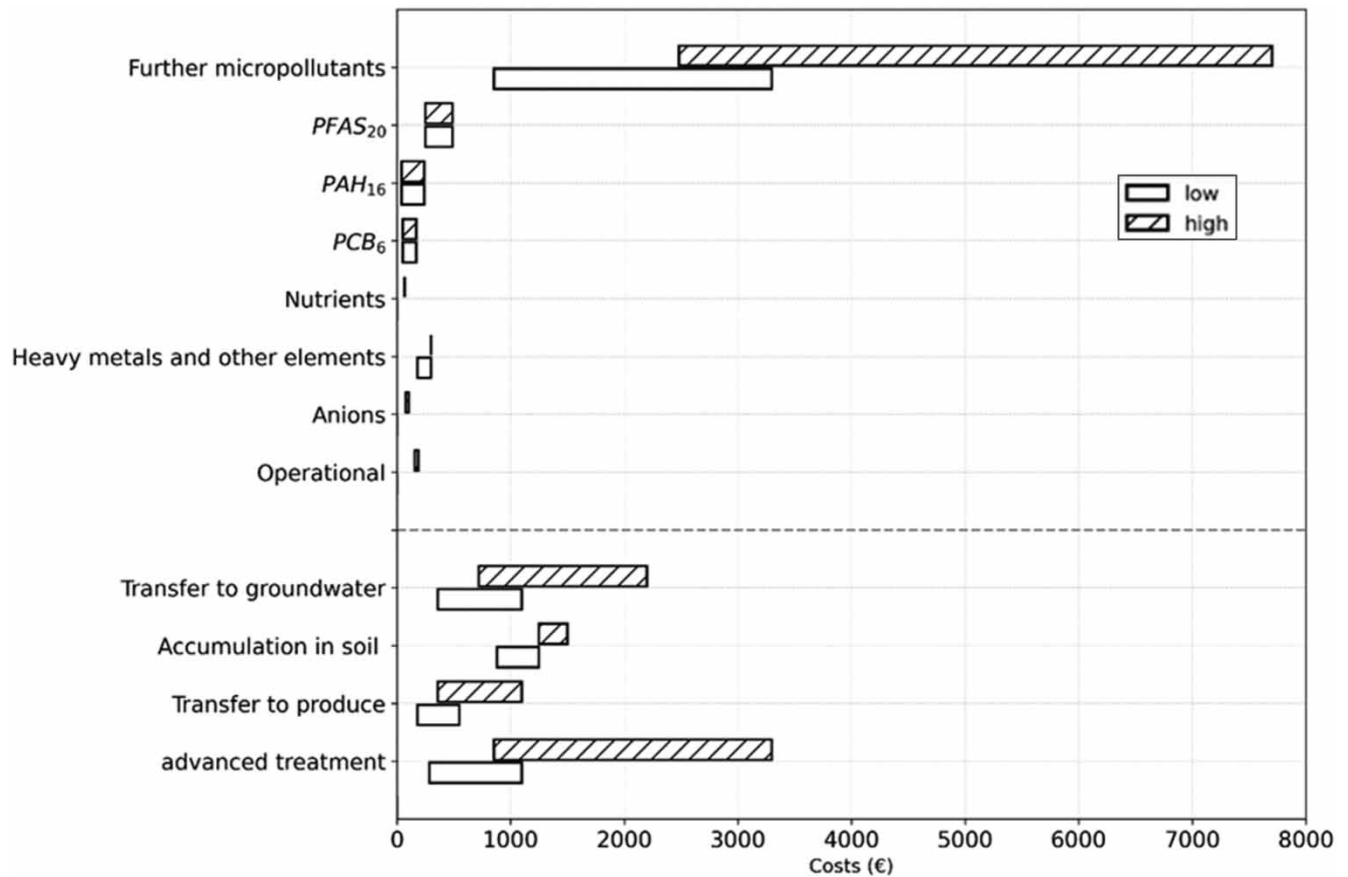
**Table 3** | Microbiological parameters and estimated related costs for one season with 20 weeks of irrigation

Parameter	Costs for analyses (€)		
	One sample	Complete validation	20 weeks of regular monitoring
<i>E. coli</i>	30–90	600–1,800	600–1,800
<i>Enterococci</i>	30–50	–	600–1,000
<i>Legionella</i> spp.	80–130	–	800–1,300
<i>C. perfringens</i> spores	30–60	600–1,200	–
Somatic coliphages	90–200	1,800–4,000	–
F-specific coliphages	90–200	1,800–4,000	–
Helminths-oocysts	Not available	–	Not available

Calculations include 10 samples each from SP 1 and SP 3 for the validation and weekly (*E. coli* and *Enterococci*) or biweekly (*Legionella*) samples from SP 4 as mandated by the EU water-reuse regulation (EU 2020a).

for risk management, as well as for regular monitoring, varies. The estimated costs for two theoretical scenarios with higher/lower parameters are shown in Figure 3. For the risk assessment (to be assessed at SP 2, WWTP effluent), the scenario 'high' covers all parameters as shown in Table 2. The scenario 'low' includes PAH<sub>16</sub>, PCB<sub>6</sub>, PFAS<sub>20</sub> and six OMPs while relying on available data regarding the operational parameters, nutrients and heavy metals. OMP analyses by far account for the biggest share of total costs. Considering both scenarios, the minimum and maximum costs for analyses of one sample, risk assessment analyses amount to approximately 1,600–3,400 and 4,500–9,300 Euros, respectively. However, actual costs may vary to a great extent. In consideration of the future requirements for wastewater treatment (EU 2024b), WWTPs above 150,000 p.e. (and those in areas at risk) will have to involve advanced treatment for OMP removal. In such cases, related costs would not contribute to the costs for water reuse, since they would be incurred, irrespective of whether the reclaimed water is used for agriculture. Depending on the WWTP influent and effluent quality, more or fewer parameters might be relevant for risk assessment. Since the risk assessment is carried out once, further aspects, such as the irrigated area and cultivated crops, will also determine how substantial the costs are for a specific project.

Similarly, two theoretical scenarios, 'low' and 'high', were applied for the regular monitoring. Each considers the four monitoring objects' OMP removal during advanced treatment, transfer to produce, accumulation in soil and transfer to groundwater (Table 4) with varying amounts of OMP contributing to the extent of analyses. Since price information was not available for the analyses of an appropriate set of PFAS in soil, the information for the analysis of PFAS<sub>20</sub> in water was applied. The total costs are highly dependent on how frequently analyses are done. The following assumptions are made to estimate the costs for one year based on data shown in Figure 3: analyses for advanced treatment are done monthly during 20 weeks of irrigation, transfer to produce and groundwater both are monitored yearly, and soil monitoring is done



**Figure 3** | Minimum and maximum estimated costs for one analysis each are shown as bars for two scenarios, 'low' and 'high'. Above the dashed line: analyses for the risk assessment whereby scenario 'low' includes PAH<sub>16</sub>, PCB<sub>6</sub>, PFAS<sub>20</sub> and six OMPs; scenario 'high' includes all parameters from Table 2. Below the dashed line: costs for regular monitoring according to the processes provided in Table 4.

**Table 4** | Proposed indicator parameters for the monitoring of processes related to risks associated with agricultural water reuse at SP indicated in Figure 2

Process	SP	Parameter requirements	Indicator parameters
OMP removal	Advanced treatment (SP 3) or water outlet (SP 4)	Different for moderate and high removals in WWTP and established treatment technologies, such as activated carbon and ozonation	UV absorbance at 254 nm, dissolved organic carbon Substances from the urban wastewater treatment directive (EU 2024b) listed in Table 2
Transfer to produce	Produce (SP 5)	High transfer to produce	Carbamazepine, hydrochlorothiazide, gabapentin
Accumulation in soil	Soil (SP 6)	High $K_d$ or $K_{oc}$ and persistence	Carbamazepine, heavy metals, PAH <sub>16</sub> , PCB <sub>6</sub> , PFOA, PFOS
Transfer to groundwater	Ground water (SP 7)	High mobility and persistence	Diatrizoic acid, primidone, gabapentin

every five years (costs divided by five). In that case, the minimum and maximum total costs for one year of monitoring amount to 2,100–7,400 and 5,600–20,100 Euros for the ‘low’ and ‘high’ scenarios, respectively. This discrepancy reveals that the most uncertain point of expensive OMP analyses is the main variable in these scenarios. A more detailed documentation on the methodology for the presented cost calculation is provided in the Supplementary Information. Since regular monitoring is an ongoing expense, a limited but at the same time meaningful set of parameters seems to be important.

## 5. SUGGESTED ANALYSES FOR RISK MANAGEMENT

### 5.1. Microbiological analyses

The parameter *E. coli* is widely used as the primary indicator for fecal contamination in water-reuse applications. It is easy to measure, comes in high concentrations in wastewater and has a well-established correlation with the removal of other enteric pathogens, making it a reliable and cost-effective parameter for validating the effectiveness of treatment processes and for routine monitoring. Coliform bacteria are typically measured alongside *E. coli* as they provide additional information without additional costs. The presence of coliforms can help identify issues with microbial control, as these bacteria are often more resilient. *Enterococcus* spp. exhibit even greater environmental persistence (Naclerio *et al.* 2008; World Health Organization 2008; Schneeberger *et al.* 2015). Therefore, *Enterococci*, which are not effectively eliminated during wastewater treatment, might pose a health threat to humans and animals. Thus, including these parameters in the validation process is important. Both *Enterococci* and coliforms might proliferate in water if conditions allow, signaling the potential for further contamination risks during storage. For routine monitoring, these parameters are useful for precautionary consumer health protection.

The concentration and detection frequency of *Salmonella* in municipal raw wastewater vary, depending on regional factors and specific conditions at WWTPs (Kinde *et al.* 1997; Espigares *et al.* 2006). *Salmonella* is the most common cause of food-borne-disease outbreaks in the EU (EFSA 2018). Liu *et al.* (2018) reported related disease outbreaks after irrigation with surface water, indicating the relevance. The reduction of *E. coli* in wastewater treatment is used as an indicator for the simultaneous reduction of other fecal pathogens, including *Salmonella* spp. However, the survival and removal rates of different microorganisms can vary; therefore, risk-based assessments should guide monitoring decisions, taking into account factors such as wastewater origin, treatment methods and intended water reuse. To optimize monitoring efforts while minimizing costs, we propose an adaptive analysis approach during validation. This would include testing *Salmonella* spp. as an additional parameter during the one-time process validation, which would not require additional sampling. If *Salmonella* is not detected in the validation phase, further testing can be omitted. However, if *Salmonella* is detected, further testing should be decided on a case-by-case basis.

*Legionella* is particularly important to monitor in water systems where aerosols may be generated. *Legionella* can regrow in water under certain conditions and the inhalation of contaminated aerosols can pose serious health risks. As stipulated by the EU regulation (EU 2020a), monitoring *Legionella* is essential when aerosols are a concern.

*C. perfringens* spore is a suitable and important parameter for the validation of disinfection as it is comparably resistant (Ho *et al.* 2024) while it likely occurs in concentrations high enough to achieve log removal values required by the EU water-reuse regulation (EU 2020a; Seis *et al.* 2024).

Coliphages are a relevant surrogate for human pathogenic viruses, can behave differently than *E. coli* during treatment processes and thus are also a valuable monitoring parameter. Influent concentrations to WWTPs are likely too low to evidence log removal values required by the EU water-reuse regulation (EU 2020a) unless enrichment is applied (Seis *et al.* 2024). According to the EU water-reuse regulation, which requires validation only for quality class A, validation is also considered successful when no coliphages are detected after disinfection following the standard methods listed in Table 1. In turn, any positive detection leads to failure of the validation when influent concentrations are too low to confirm the required log removal values, even if the treatment train could exceed the requirements. Relying solely on the absence of detectable coliphages to validate treatment efficacy requires careful consideration. For such an approach to be deemed valid, the limit of detection or the volume of water tested must be clearly defined and aligned with regulatory requirements. Otherwise, methods for enrichment from larger volumes must be implemented to comply with the sophisticated requirements. Therefore, Seis *et al.* (2024) suggest methods for enrichment by membrane filtration and provide statistical analysis to demonstrate that 90% of validation samples meet or exceed performance targets, as required by the EU water-reuse regulation (EU 2020a). With regard to validation, we emphasize the need to provide feasible and standardized methods, in particular, if sophisticated validation might be extended to water quality classes A, B and C, in Germany, despite the EU recommendations (EU 2020a).

The robust and reliable validation of uncensored log removal credits can be done by improving the limit of detection through sample enrichment or spiking to final disinfection, though this increases sampling effort. To balance this, it is suggested to document validated log removal credits for selected treatment trains in a public register, including operational conditions. These reference plants can then serve as models for replicate plants, where simplified validation monitoring, meeting EU minimum requirements, is sufficient due to the existing confidence in the treatment process (DWA In progress).

## 5.2. Chemical analyses

An initial risk assessment can be achieved by analyzing representative samples from the WWTP effluent (SP 2). On the basis of reliable site-specific data and in agreement with the risk management plan, the relevant parameters should be selected, as listed in Table 2, potentially complemented by other relevant OMPs. Coherence between risk assessment and monitoring parameters improves overall risk management.

We suggest a set of useful indicators listed in Table 4 for monitoring all the relevant processes and exposure routes indicated in Figure 2: advanced wastewater treatment for OMP elimination, accumulation in soil, transport to or migration with groundwater and plant uptake. However, it must be decided in complementary risk management plans which parameters are relevant in a site-specific scenario.

Monitoring the advanced treatment (SP 3) is crucial to sustain risk management: In case the WWTP already incorporates advanced treatment for OMP elimination, the revised urban wastewater treatment directive (EU 2024b) provides indicator parameters. Additionally, the reduction of UV absorbance at a wavelength of 254 nm and dissolved organic carbon are typical surveillance parameters during advanced treatment and OMP removal, while turbidity can signal anomalies in treatment performance.

Transfer and migration of groundwater matter regarding persistent and mobile substances: Persistent and mobile substances are important with regard to their potential transfer to groundwater and subsequent migration in groundwater bodies (Ternes *et al.* 2007; Dittmann *et al.* 2024). Thus, diatrizoic acid appears to be a useful indicator. Among PFAS, the short-chain and ultra-short-chain PFAS, in particular, tend to stay in the aqueous phase and thereby reach groundwater (Rasmusson & Fagerlund 2024). However, short-chain PFAS are suspected to be ubiquitously present in the atmosphere and deposited with rainfall (Björnsdotter *et al.* 2021). Trifluoroacetic acid is known to form after degradation of pesticides containing C-CF<sub>3</sub> moieties (Joerss *et al.* 2024), leaving it as a rather unprecise parameter for agricultural water-reuse monitoring. Gabapentin was proposed as an indicator because of its persistence, mobility and high abundance (Ahmadi *et al.* 2025; DWA in progress). Primidone was proposed to be amended to the groundwater directive as an indicator for anthropogenic impacts (EU 2022a).

Long-term accumulation in soil mainly concerns persistent substances with high  $K_d$  (or  $K_{OC}$ ): Meeting the criteria of the German soil protection ordinance (DE 2021), heavy metal loads to soil can be readily assessed from reclaimed water analyses. With regard to secondary effluent concentrations and expected irrigation volumes in Germany, risks emanating

from heavy metals were low but not negligible (UBA 2016; LAWA 2022). Other experiences regarding water reuse in a hydroponic system showed that heavy metal concentrations in the irrigation water were below the limit values of the German Drinking Water Ordinance (Bliedung *et al.* 2020). Based on reported PAH (UBA 2020) and PCB (Cybulski *et al.* 2022) concentrations in WWTP effluents, the related risk appears low, but not negligible in industrially influenced catchments (Bruzzoniti *et al.* 2024). Heavy metals, PAH and PCB, should be included in soil monitoring if there is evident concern of elevated concentrations in the reclaimed water. If relevant in the reclaimed water, monitoring PFOA and PFOS is useful since they potentially cause long-term contamination (Jahn *et al.* 2023; Rasmusson & Fagerlund 2024). Carbamazepine is a useful indicator that was often reported to occur in soils after water reuse (Ben Mordechay *et al.* 2021; García-Valverde *et al.* 2023; Kodešová *et al.* 2024).

OMP plant uptake follows complex mechanisms: Thus, it was challenging to propose a definite set of indicators. However, carbamazepine uptake was reported for agricultural water reuse and various types of produce and soils (Ben Mordechay *et al.* 2021; García-Valverde *et al.* 2023; Benelhadj *et al.* 2024; Dittmann *et al.* 2024). It, therefore, appears to be a meaningful indicator. Ultra-short chain PFAS like trifluoroacetic acid and perfluoropropionic acid show a high uptake potential in arugula (*Eruca sativa*) similar to carbamazepine (Seelig *et al.* 2025) but may have other sources than reclaimed water. Gabapentin can be a suitable indicator for water-rich agricultural produce due to its persistent and mobile nature (Ahmadi *et al.* 2025). Ahmadi *et al.* (2025) found that among 18 OMPs, hydrochlorothiazide and carbamazepine were the most critical with respect to acceptable daily intake via agricultural produce.

## 6. CONCLUSIONS

Agricultural water reuse is embedded in a complex regulatory framework, potentially requiring numerous microbiological and chemical parameters. This, and the requirement to decide on the extent of analyses according to site-specific conditions, pose a challenge to practitioners. For many parameters, analytical standard methods are available and services are provided by laboratories according to current legislative requirements, but, for some parameters, a lack could be identified. The use of chemical process indicators helps to representatively monitor exposure routes and reduce efforts.

With regard to the microbiological parameters, remaining analytical challenges need to be clarified and solutions must be implemented into standardized methods, such as the enrichment of coliphages from greater sample volumes and testing for helminth oocysts. A feasible and representative strategy for risk assessment and monitoring supports the work of the responsible authorities and practitioners. The findings are thus valuable input for the regulatory development. Further challenges arise in the assessment of those OMP that are not yet regulated. A better understanding of their eco- and human toxicology is needed. In light of uncertainty, the presence of OMP should be prevented or minimized following the precautionary principle.

The elaborated costs for extensive microbiological and chemical analyses are unique compared to conventional irrigation and contribute to complex economic constraints. Whenever conventional water is available at low costs, this circumstance will decrease the competitiveness of water reclamation. Thus, keeping costs for analyses at reasonable levels is one remaining challenge in order to make agricultural water reuse economically feasible. OMP analyses, in particular, may drastically increase costs as long as there is no standard list for which laboratories implement standard procedures. Confining to a few cost-efficient analytical multi-target methods tailored to the requirements of agricultural water reuse likely promotes the implementation of water-reuse projects.

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## DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

## CONFLICT OF INTEREST

The authors declare there is no conflict.

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