Responsible Manufacturing Effluent Management

Technical Guidance Document

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Contents

Executive Summary

This paper is intended as Technical Guidance for the Pharmaceutical Industry to help identify and mitigate the potential impacts of active pharmaceutical ingredients (API) in wastewater from manufacturing operations. The approaches it describes are intended as general guidelines for carrying out risk assessments in different exposure scenarios for APIs in manufacturing effluent and for implementing such programs. All elements of this guidance may not be applicable at all sites, and the degree of rigor should vary with the risk posed by the APIs to the environment. It envisions a flexible approach in implementing these guidelines based on company-specific procedures.

This guidance focuses on risk management for API in manufacturing effluent. Wastewater risk management is the process by which risk-based discharge targets for API are set, implemented, and monitored. Wastewater targets are met firstly by preventative measures during area cleaning or material transfer and secondly by providing effective wastewater treatment either on-site or off-site.

This guidance is not intended to override regulatory requirements, nor should it be considered as a substitute for a clear understanding of, and compliance with, regulatory requirements. Compliance with laws, regulations and environmental permits is a mandatory requirement for all API and drug product manufacturing operations.

However, because regulations often do not specifically address API discharges, except through general protective clauses, this guidance was developed to help manufacturers implement the following principles for responsible effluent management; first for their own manufacturing facilities, and second, for their suppliers' manufacturing sites:

- Compliance with applicable company standards,
- Implementation of defined, sound wastewater management programs that are based on risk management and good engineering principles,
- Definition of site and API specific discharge targets based on safe concentrations (Predicted No Effect Concentration – PNEC) in the receiving surface waters,
- Discharge of manufacturing wastewater containing API must have an environmental risk assessment; if a risk is identified, appropriate additional controls will be implemented to mitigate the risk to an acceptable level.

Specific guidance for implementing environmental risk assessments (ERA) for APIs is provided. The elements of a predictive ERA such as hazard definition, exposure assessment, effects assessment, risk characterization and risk mitigation and management are discussed.

1 Introduction

This paper is intended as Technical Guidance for the Pharmaceutical Industry to help identify and mitigate the potential impacts of active pharmaceutical ingredients (API) in wastewater from manufacturing operations. The approaches detailed herein describe general guidelines for carrying out risk assessments in different exposure scenarios for APIs in manufacturing effluent and for implementing such programs. All elements of the guidance may not be applicable and the intent is that the industry maintains a flexible approach in implementation based on company-specific procedures. This guidance is not intended to override regulatory requirements, nor should it be considered as a substitute for a clear understanding of, and compliance with regulatory requirements. Rather, because regulations often do not specifically address API discharges, except through general protective clauses, this document is intended to help manufacturers implement the following principles for responsible effluent management; first for their own manufacturing facilities, and second, for their supplier's manufacturing sites:

Compliance with laws, regulations and environmental permits is a mandatory requirement for all API and drug product manufacturing operations

- ✓ *Compliance with applicable company standards,*
- ✓ *Implementation of defined, sound wastewater management programs that are based on risk management and good engineering principles,*
- ✓ *Definition of site and API specific discharge targets based on safe concentrations in the receiving surface waters,*
- ✓ *Discharge of manufacturing wastewater containing API must have an environmental risk assessment; if a risk is identified, appropriate additional controls will be implemented to mitigate the risk to an acceptable level.*

Additionally, the member companies of AESGP, EFPIA and Medicines for Europe – as part of the Inter Associations Initiative (IAI) Pharmaceuticals in the Environment (PiE) Task Force – have developed a set of principles for responsible effluent management for their own, and supplier, manufacturing sites:
Ensuring compliance with environmental laws, regulations, permits, or other internal obligations that the company has determined to be necessary, requires a systematic management approach. While compliance management is not a focus of this guidance, it specifies some key process steps that enable understanding and ensures and demonstrates environmental compliance.

Understanding and ensuring compliance with laws, regulations and permits as well as other internal standards or guidelines that the company has determined to be necessary, typically requires the following steps. These steps generally apply to many EHS responsibilities. In the context of this guidance, practices specifically related to the operation's wastewater (and resulting waste) management are covered:

Implementation of defined, sound wastewater management programs that are based on risk management and good engineering principles is proposed. However, concepts such as waste minimisation should also be considered.

This technical guidance is focussing on responsible manufacturing effluent management for API. It applies to all API with the exception of naturally occurring substances such as vitamins, electrolytes, amino acids, peptides, proteins, nucleotides, carbohydrates and lipids. Antimicrobial substances are also included in the scope, although reference is given to the activities of the Antimicrobial Resistance (AMR) Industry Alliance.

Whereas traditional wastewater effluent parameters such as pH, BOD, COD, TOC, N_{total}, P_{total}, etc. are included in the first principle ("compliance with laws, regulations and environmental permits is a mandatory requirement for all API and drug product manufacturing operations") it has to be pointed out that other substances that may not be locally regulated, e.g. starting materials, process intermediates, solvents, should be considered case-by-case.

2 Wastewater management programs

Principle: Implementation of defined, sound wastewater management programs that are based on risk management and good engineering practices

Key elements of a sound wastewater management include:

(1) Possession of a valid authorization/license/permit for water discharge
(2) Controlling or minimising wastewater at the source; from an environmental perspective, mass loads of API are relevant
(3) Characterization of wastewaters that cannot be avoided or recycled; measurements and calculations (balancing) could both be used to characterise wastewater emissions depending on circumstances (e.g. safety margins vs. targets)
(4) Identification and setting of targets for wastewater discharge and disposal, considering legal, permit and company requirements
(5) Meeting wastewater targets
- Firstly by preventative measures during area cleaning or material transfer
- Secondly by providing, where necessary, effective wastewater treatment either on-site of off-site. (see Caldwell *et al.*, 2016 [1] for more details)

(6) Monitoring wastewater emissions as well as the proper functioning of control measures
(7) Acting in case of irregularities related to wastewater
(8) Controlling spills and calamities relevant to wastewater according to implemented procedures
(9) Management of change related to wastewater
(10) Information, documentation, communication:
- Availability of information relevant to assessing environmental impact of wastewater pollutants and to the rationale for the choice of any necessary control systems,
- Integration of wastewater disposal process with production planning; manufacturing units should be involved and supported as early as possible in identifying critical wastewater streams (good communication between EHS experts and manufacturing organisations is key)

(11) Training
- Appropriate training of operational staff

(12) Auditing:
- Internal audits include the wastewater management program and address the proper functioning of the wastewater management process. Following-up audit results and defining corrective and preventive action (CAPA) is a part of the audit process where applicable.

This guidance focuses on risk management for API in manufacturing effluent. Wastewater risk management is the process by which risk-based discharge targets for API are set,

implemented, and monitored. Thus, the following chapters detail guidance on the key elements (4), (5), and (6) of wastewater management programs.

3 Setting, meeting and monitoring API discharge targets for wastewater

Principle: Definition of site and API specific discharge targets based on safe concentrations in the receiving surface waters

Principle: Discharge of manufacturing wastewater containing API must have an environmental risk assessment; if a risk is identified, appropriate additional controls will be implemented to mitigate the risk to an acceptable level.

While the prevention of waste generation during area cleaning or material transfer is a fundamental step, the discharge of wastewaters with residual chemicals must be evaluated. Internal company procedures are in place to manage risks from API in wastewater. Procedures should include a scope definition that considers the potential for API in process wastewater to be discharged. The procedures include risk assessment, target setting, developing and implementing an action plan, and monitoring (see Figure 1). The process description specifies triggers for the assessment. If the risk assessment identifies unacceptable risks, internal discharge targets should be created to mitigate the risk to an acceptable level. Targets are risk-based and relate to safe concentrations (Predicted No Effect Concentration – PNEC) in the receiving surface waters and/or other relevant environmental compartments. API discharge targets are a mass-loading or upstream concentration that are back calculated from the environmentally safe concentration and the particular environmental scenario at their facility and at the receiving water.

Figure 1 Process for wastewater management

An action plan focusing on technical and organizational measures is established and subsequently implemented to ensure that API discharge targets will be met. The selection and design of such measures is situationally dependent, such as e.g. site location, site configuration, product profile.

A monitoring program can be established to evaluate the effectiveness of the implemented controls. It can include parameters to be quantified, methods, sampling plans and the frequency for monitoring.

4 Environmental risk assessment (ERA)

4.1 Fundamentals

Principle: Discharge of manufacturing wastewater containing API must have an environmental risk assessment; if a risk is identified, appropriate additional controls will be implemented to mitigate the risk to an acceptable level.

Specific guidance for implementing ERAs for API is provided in this section. Figure 2 illustrates the predictive ERA. This section 4 focuses on Steps 1 to 3 (hazard definition, exposure assessment, effects assessment and risk characterization), whereas Step 4, risk mitigation and management, is covered in section 5.

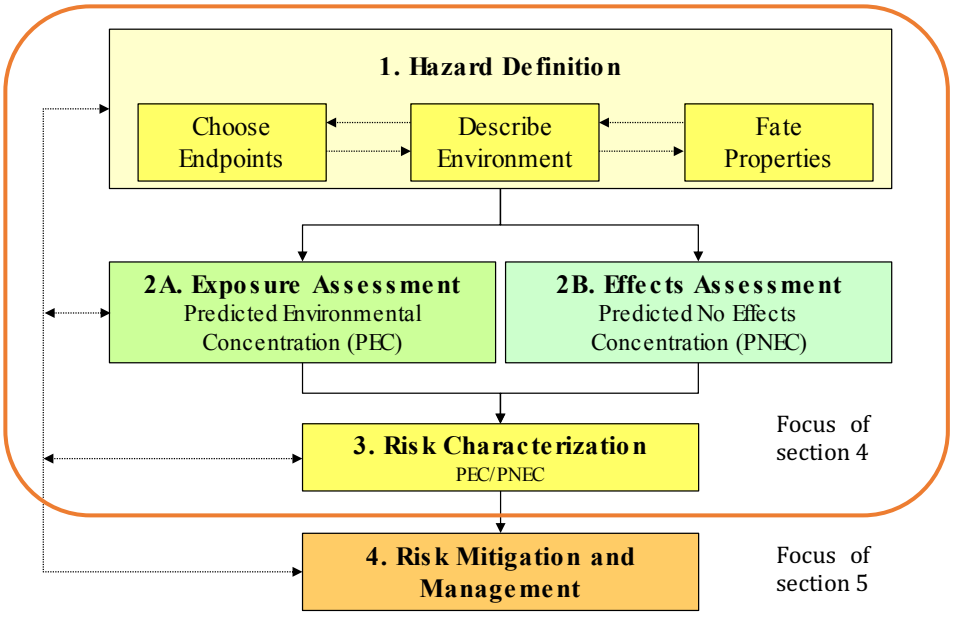

Figure 2 Predictive environmental risk assessment

Step 1 (hazard definition) includes an understanding of the environmental situation, API fate properties, and species in the environmental compartment. While the primary focus and basic requirement of this guidance is on environmental organisms in surface water (the "most likely exposure scenario", see section 4.2), other potential exposure scenarios

include human drinking water, terrestrial organisms following land application of biosolids or irrigation with wastewater, or effects on biological treatment plants. See Table 1 in section 4.3 for a list of exposure scenarios and related protection goals.

Step 2 (exposure assessment / effects assessment) involves a calculation of predicted environmental concentrations (PEC = exposure) and predicted no effect concentrations (PNEC = effects). Derivation of appropriate values for effects (PNEC) and exposure (PEC) to be used under exposure scenarios selected is critical to the process of risk assessment. Derivation of PNECs is the subject matter of section 4.3. Calculation of the PEC is covered in section 4.4. The PEC is the sum of the background concentration and the process contribution (PC) from the manufacturing operation. When the background concentration is 0, then PEC is equal to PC.

In **Step 3** (risk characterization), a comparison of PEC and PNEC provides a qualitative measure of risk, which is defined as the risk quotient: RQ = PEC ÷ PNEC. Generally, an RQ ≥1 indicates a potentially unacceptable risk to organisms in a specific environmental compartment (note: RQ ≥1 is not a bright line limit/trigger for controls, but it may be used for risk prioritization). An RQ <1 indicates that the risks are generally acceptable. Risk ratios inform understanding the level at which an individual API can be discharged safely into the environment and/or the level of treatment of manufacturing wastes that will be required to achieve that safe discharge concentration.

If multiple exposure scenarios apply, the Risk Quotient (PEC ÷ PNEC) is calculated for each scenario. The exposure scenario driving overall environmental risk should be determined ("limiting scenario") to address risk mitigation appropriately.

4.2 Exposure scenarios

The most likely environmental compartment to be exposed to manufacturing effluent is surface waters (rivers, streams, lakes, oceans) through either direct discharge or indirect after first going through a wastewater treatment plant (WWTP). Hence, this Technical Guidance primarily describes the approach for this environmental compartment.

The exposure in WWTP can also be relevant, in particular for antimicrobials (high API concentrations plus high bacterial density). Also, highly toxic APIs could affect a WWTP directly.

Another important exposure consideration is the time profile of emissions resulting from batch-wise manufacturing. Batch production may potentially result in intermittent, transient peak concentrations in the environment ("intermittent release" is the technical term used in EU guidance) if production wastewater is not metered out over time. Longer-term batch production campaigns or continuous manufacturing may result in chronic discharges over longer time periods. Depending on the manufacturing method, either or both of these scenarios may need to be evaluated.

There are other environmental exposure scenarios and protection goals that can be considered dependent on the local situation. Various discharge scenarios may require evaluation to identify adequate strategies for mitigation and management of API-

containing wastewater prior to discharge at a given facility. Table 1 in section 4.3 offers users a non-exhaustive list of exposure scenarios and related protection goals.

To start with, an understanding of the amounts of API that are discharged via a site's wastewater is required to determine the level of exposure from an API in the environment. The first question to be answered by a facility is whether API may be lost to the aqueous effluent through process activity or equipment cleaning.

Aqueous wastes will typically undergo some form of treatment (either on-site or municipal) before discharge. In this case, the fate of the API during wastewater treatment should be considered. API may be removed via hydrolysis, oxidation, biodegradation, or adsorption to (activated) sludge.

Downstream of the facilities' point of discharge, predicting environmental concentrations of pharmaceutical compounds requires an understanding of how pharmaceuticals may enter the environment, the chemical form in which they occur in the environment, and the various chemical, biological and transport processes which will influence the behaviour of the API in the environment and its distribution among different environmental compartments. This analysis should include the behaviour of the API in treatment systems external to the discharging facility through which it is processed.

The basic scenario of discharging wastewater to surface waters requires characterization of the receiving water and the available mixing zone for the discharge to estimate exposure from the amount of API that is discharged.

4.3 Effects assessment: establishing criteria (PNECs)

Deriving PNEC values involves considerable specialized professional judgment. An API PNEC for an environmental compartment protects the species in that compartment from harmful effects from that API.

For aqueous discharge from pharmaceutical manufacturing, the most likely environmental compartment impacted is surface water since the facility may directly or indirectly discharge to surface water. Therefore, a chronic $PNEC_{surface\ water}$ is the primary criterion to be established.

When setting PNECs to protect species from acute or chronic effects, account should be taken as to whether emissions of an API are infrequent (such as from batch-wise manufacturing) or regular.

There are some scenarios in which species living predominantly in other compartments may be exposed to an API through the surface water or where the API is transported to a different environmental compartment. These scenarios may be dependent on the mechanism of action of the API (e.g. antibiotics), the method of discharge (e.g. land application of wastewater or sewage sludge), or chemical characteristics of the API (e.g. hydrophobicity).

A list of such scenarios, the PNEC that may be derived, and examples of the organism groups that the PNEC protects are presented in Table 1. Whether additional PNECs may

be derived depends on site-specific discharge, API-specific characteristics, and company-specific procedures.

Table 1 Scenarios, protection goals and the respective criteria (PNECs) (green: the most likely exposure scenario; yellow: further exposure scenarios for surface waters)

Scenario	Protection goals	Criteria (PNECs)
Effluent discharge (directly or indirectly) to surface water	Aquatic species that live in the surface water	Chronic $PNEC_{surface\ water}$
Effluent discharge involves mixing zone with more concentrated zone compared to chronic exposure (e.g. very large dilution factor in surface water); or short term (pulse) concentrations expected	Aquatic organisms transiently exposed (acute exposure due to travel through mixing zone or intermittent discharge)	Acute $PNEC_{surface\ water}$
Effluent discharge to ocean or sea	Aquatic organisms in saltwater from chronic exposure	Chronic $PNEC_{marine\ water\ bodies}$
Effluent discharge to ocean or sea involves mixing zone with more concentrated zone compared to chronic exposure (e.g., very large dilution factor or pulse concentrations expected	Aquatic organisms in saltwater transiently exposed (acute exposure due to travel through mixing zone or intermittent discharge)	Acute $PNEC_{marine\ water\ bodies}$
Drinking water inlet close to the effluent stream, areas where surface water may be used as a drinking water source, or areas with recreational use of surface water	Humans exposed through drinking water	$PNEC_{drinking\ water}$
Subsistence fishing downstream of effluent discharge containing API with potential for bioaccumulation (high K_{OW} or BCF)	Humans exposed through eating fish	$PNEC_{human\ use}$
Effluent discharge containing API with potential for bioaccumulation (high K_{OW} or BCF)	Fish-eating predators such as birds and mammals	$PNEC_{secondary\ poisoning}$
Effluent discharge containing API with potential for partitioning to sediment (high K_{OC})	Sediment-dwelling species	$PNEC_{sediment}$
Effluent discharge to soil, either via irrigation or partitioning to biosolids (high K_{OC}) and application of biosolids to soil	Terrestrial organisms	$PNEC_{soil}$
When toxicity to sewage microorganisms is high (e.g. antibiotics)	Sewage treatment microorganisms	$PNEC_{stp}$
Effluent discharge containing antibiotic compound(s)	Prevent emergence of antimicrobial resistance	$PNEC_{MIC}$
Land-applied effluent (irrigation) containing API that will move through soil easily (low K_{OC}) or highly hydrophilic or by bank filtration	Groundwater species	$PNEC_{groundwater}$

4.3.1 Derivation of chronic and acute PNECs for surface waters

Dataset

PNEC values for aquatic organisms are normally derived from studies with only a few species that are considered representative models for other organisms. Typically, the dataset should preferably include at least one study in species from each of the three trophic levels (e.g. algae, invertebrates, fish). However, for some classes of compounds a more tailored testing strategy is needed. For example, the preferred data set for antibiotics includes additional data derived from tests with cyanobacteria. For endocrine active compounds, tests that evaluate reproduction and development in fish and or frogs should be considered.

Studies should be conducted using standardized methods (e.g., OECD) and employing Good Laboratory Practices (GLP). Studies from the peer-reviewed literature may also be used, but only with great care given concerns regarding data quality to ensure that the methods and results are relevant to the ecosystem in question and reliable. Typically, data should give a good indication of the impact of the API on survival, growth and/or reproduction of aquatic life. Studies considering genomic, cellular, and/or organ effects should only be considered 'supportive' of other data on population-relevant endpoints. Assessment criteria may be useful to help judge the reliability of non-standardised tests (e.g. Moermond *et al.*, 2017 [2]; Klimisch *et al.*, 1997 [3]) but expert judgment is often required.

Once all available data have been gathered, the generally-accepted approach is to use the most conservative result to derive the PNEC. If data are available, lowest concentration for 10% mortality or effect ($L(E)C_{10}$) or no-observed-effect concentration (NOEC) from chronic/reproductive studies are preferred. Otherwise concentrations for 50% mortality or effect ($L(E)C_{50}$) from acute studies may be used.

Typically, assessment factors (AF) are applied to the lowest toxicity value to take into account uncertainties associated with the test species and measured endpoint. The magnitude of the AF is reduced with increasing confidence in the data set. The lowest toxicity value in the available dataset is divided by the assessment factor and the result is the PNEC.

Methods

There are several available written methodologies for determining PNEC values for APIs in environmental compartments. The most broadly applicable is the ECHA REACH guidance [5] in combination – for intermittent release – with the Technical Guidance for Deriving Environmental Quality Standards (EQS) in the context of the EU Water Framework Directive (WFD) [6] (see Table 2 for the surface and Table 3 for marine water bodies), but other methods may be appropriate.

Where the risk assessment is being performed for marine waters, the ECHA REACH recommends that an additional order of magnitude be applied where freshwater species results are used. The lowest applicable PNEC value is used in the risk assessment. PNECs may define long (chronic exposure) and short (acute exposure) term concentrations of APIs that are protective of the environment.

Table 2 General rules for assessment factor selection in the EU for surface waters (ECHA REACH guidance and TGD EQS)

Chronic $PNEC_{surface\ water}$

Available data	Assessment factor
At least one short-term L(E)C_{50} from each of three trophic levels (fish, invertebrates (preferred *Daphnia*) and algae)	1000
One long-term EC_{10} or NOEC (either fish or *Daphnia*)	100
Two long-term results (e.g. EC_{10} or NOECs) from species representing two trophic levels (fish and/or *Daphnia* and/or algae)	50
Long-term results (e.g. EC_{10} or NOECs) from at least three species (normally fish, *Daphnia* and algae) representing three trophic levels	10

Acute $PNEC_{surface\ water}$ (intermittent release)

Available data	Assessment factor
At least one short-term L(E)C_{50} from each of three trophic levels (fish, invertebrates (preferred *Daphnia*) and algae)	100
At least one short-term L(E)C_{50} from each of three trophic levels (fish, invertebrates (preferred *Daphnia*) and algae)	10[a)]

[a)] Acute toxicity data for different species do not have a higher standard deviation than a factor of 3 in both directions OR known mode of toxic action and representative species for the most sensitive taxonomic group included in the data set.

Table 3 General rules for assessment factor selection in the EU for marine water bodies (ECHA REACH guidance and TGD EQS)

Chronic $PNEC_{marine\ water\ bodies}$

Available data	Assessment factor
Lowest short-term L(E)C_{50} from freshwater or saltwater representatives of three taxonomic groups (algae, crustaceans and fish) of three trophic levels	10000
Lowest short-term L(E)C_{50} from freshwater or saltwater representatives of three taxonomic groups (algae, crustaceans and fish) of three trophic levels, + two additional marine taxonomic groups (e.g. echinoderms, molluscs)	1000
One long-term result (e.g. EC_{10} or NOEC) (from freshwater or saltwater crustacean reproduction or fish growth studies)	1000
Two long-term results (e.g. EC_{10} or NOEC) from freshwater or saltwater species representing two trophic levels (algae and/or crustaceans and/or fish)	500
Lowest long-term results (e.g. EC_{10} or NOEC) from three freshwater or saltwater species (normally algae and/or crustaceans and/or fish) representing three trophic levels	100
Two long-term results (e.g. EC_{10} or NOEC) from freshwater or saltwater species representing two trophic levels (algae and/or crustaceans and/or fish) + one long-term result from an additional marine taxonomic group (e.g. echinoderms, molluscs)	50
Lowest long-term results (e.g. EC_{10} or NOEC) from three freshwater or saltwater species (normally algae and/or crustaceans and/or fish) representing three trophic levels + two long-term results from additional marine taxonomic groups (e.g. echinoderms, molluscs)	10

Acute $PNEC_{marine\ water\ bodies}$ (intermittent release)

Available data	Assessment factor
At least one short-term L(E)C_{50} from each of the three trophic levels of the base set (fish, crustaceans and algae)	1000
At least one short-term L(E)C_{50} from each of the three trophic levels of the base set (fish, crustaceans and algae)	100[a)]
At least one short-term L(E)C_{50} from each of three trophic levels of the base set (fish, crustaceans and algae) + one short-term L(E)C_{50} from an additional specific saltwater taxonomic group	500
At least one short-term L(E)C_{50} from each of three trophic levels of the base set (fish, crustaceans and algae) + one short-term L(E)C_{50} from an additional specific saltwater taxonomic group	50[a)]
At least one short-term L(E)C_{50} from each of three trophic levels of the base set (fish, crustaceans and algae) + two or more short-term L(E)C_{50}s from additional specific saltwater taxonomic groups	100
At least one short-term L(E)C_{50} from each of three trophic levels of the base set (fish, crustaceans and algae) + two or more short-term L(E)C_{50}s from additional specific saltwater taxonomic groups	10[a)]

[a)] Acute toxicity data for different species do not have a higher standard deviation than a factor of 3 in both directions OR known mode of toxic action and representative species for the most sensitive taxonomic group included in the data set.

For antibiotics, cyanobacteria data should be included. Proposed rules for the derivation of PNEC values are described in Tell *et al.*, 2019 [4] and the draft EMA ERA Guidance of 2018 [10]. Tell *et al.*, 2019 [4] describe the derivation of two PNECs for antibiotics: environmental ($PNEC_{ENV}$) are derived from toxicity endpoint data and antibiotic resistance ($PNEC_{MIC}$) are derived from MIC data from the EUCAST database and that are published by Bengtsson-Palme and Larsson, 2016 [12]. The lower of the two values applies.

There are many other methodologies that could be used for deriving PNEC-type values. A non-exhaustive list of different methodologies with PNECs and comments is presented in Table 4. One reason to choose one method over another may be local regulatory expectations. Different methodologies may use different assessment factors and may assume different dilution factors.

Methodology for other PNEC values based on scientific judgment
The following considerations only apply when it is deemed necessary because of the local situation. API PNEC values for humans and wild mammals can normally be derived from data collected to support the registration of pharmaceuticals for use in humans. An expert in human health risk assessment should evaluate the data available for an API considering: available results from non-clinical pharmacology data; acute and chronic mammalian toxicity studies; mutagenicity and carcinogenicity studies; reproductive and developmental studies; and clinical pharmacology and safety information. The anticipated exposure (PEC) is also useful in determining if such efforts are necessary. In order to calculate a PNEC in drinking water for human populations, an acceptable exposure to API should be identified, which is considered to result in no appreciable risks to individuals in sensitive sub-populations of humans, such as children or individuals with organ system impairment. This acceptable exposure can be converted into a concentration by determining the amount of water consumed by individuals in the sensitive population. The type and size of uncertainty factors used to determine an acceptable daily exposure can depend on the quality and completeness of the data set and examples of methodology can be found in the regulatory literature e.g. EMA, 2014 [16] and ASTM, 2020 [17] (see Table 4).

Table 4 Published methodology for determining PNEC Values

Method	PNEC calculations described	Comments/Specific Use	Ref.
ECHA REACH	Chronic surface water Marine water Surface water - intermittent STP Freshwater sediment Marine sediment Soil Secondary poisoning	Uses chronic and/or acute ecotoxicity data	[5]
Water Framework Directive (WFD)	Chronic surface water ($QS_{fw,eco}$) Chronic marine water ($QS_{sw,eco}$) Acute freshwater ($MAC\text{-}QS_{fw,eco}$) Acute marine water ($MAC\text{-}QS_{sw,eco}$)		[6]

Method	PNEC calculations described	Comments/Specific Use	Ref.
	Sediment ($QS_{sediment}$) Human drinking water ($QS_{dw,hh}$) Humans eating fish ($QS_{biota,\ hhfood}$) Secondary poisoning (QSsec pois)		
US EPA Great Lakes Guidance	Acute surface water Chronic surface water	Calculates both Tier I (full ecotoxicity dataset available) and Tier II (subset available) criteria	[7]
US FDA	Acute surface water Chronic surface water		[8]
EMA ERA Guidance	Chronic surface water Microorganism Groundwater Sediment Terrestrial	Uses only chronic data	[9] [10]
AMR Industry Alliance	$PNEC_{ENV}$ $PNEC_{MIC}$ (according to Bengtsson-Palme & Larsson, 2016 [12])	Specifically for antibiotics	[4] [11]
VICH	$PNEC_{surfacewater}$ $PNEC_{sediment}$ $PNEC_{oral}$ $PNEC_{oral,\ predator}$ $PNEC_{dung}$ $PNEC_{micro\text{-}organisms}$ $PNEC_{earthworms}$ $PNEC_{plants}$	In support of the VICH guidelines GL6 and GL38	[13]
WHO, 2011 [14] US EPA, 2000 [15] WFD, 2018 [6]	PNECs in drinking and surface water to protect humans drinking water taken from surface water and eating fish taken from surface water	These guidances use an acceptable oral intake * to calculate PNECs in drinking and surface water that protect humans.	
EMA, 2014 [16] ASTM E3219 – 20 [17] PIC/S, 2018 [18] ECHA, 2012 [19]	-	These contain methods of deriving acceptable oral intake for humans. ASTM, EMA and PIC/S are specific for pharmaceuticals.	

* Acceptable oral intake can be obtained from concepts like ADI (acceptable daily intake), ADE (acceptable daily exposure), HBEL (health based exposure limit), PDE (permitted daily exposure), TTC (threshold of toxicological concern), TDI (tolerable daily intake), DNEL (derived no-effect level), DMEL (derived minimal effect level), RfD (reference dose), BMD (benchmark dose) – all of these are referenced in the sources in the table.

4.3.2 Where to find ecotoxicity data and PNECs

Ecotoxicity data and PNEC values can be found in various sources, such as European public assessment reports (EPAR) [20], in public databases such as iPiE Sum [22], FASS.se [21] or the AMR Industry Alliance [24], in peer-reviewed literature review or in Safety Data Sheets and ERA summaries published by some companies. Some of these sources are listed in Table 5.

Table 5 Selected public sources for ecotoxicity data and PNECs

Source	Comments	Ref.
European public assessment report (EPAR)	The European Medicines Agency (EMA) publishes detailed information on the medicines assessed by the Committee for Medicinal Products for Human Use (CHMP) and Committee for Medicinal Products for Veterinary Use (CVMP).	[20]
FASS.se	IVL Swedish Environmental Research Institute (IVL) has since 2005, with the launch of the system of self-declarations of environmental classification, conducted a project focused on review of the self-declarations financed by LIF - the Research-Based Pharmaceutical Industry in Sweden and the Foundation for IVL Swedish Environmental Research Institute (SIVL)	[21]
iPiE-Sum	The iPiE Summary Database Search provides high level summarized access to the properties, environmental fate characteristics and ecotoxicity of APIs which are collected during the course of the iPiE project (Intelligence-led Assessment of Pharmaceuticals in the Environment) from 2016 to 2019.	[22]
WikiPharma database	The WikiPharma database, provided by MistraPharma contains publicly available ecotoxicity data for pharmaceutical substances, focusing on human pharmaceuticals available on the Swedish market. MistraPharma was funded by the Swedish Foundation for Strategic Environmental Research (Mistra) (not all data are quality assessed).	[23]
AMR Industry Alliance	AMR Industry Alliance Antibiotic Discharge Targets; PNEC-ENV and PNEC-MIC data for antibiotics covering both chronic surface water toxicity and antibiotic resistance selection (incl. supplemental data with ecotoxicity data)	[24]
WET Center Pharmaceutical PNEC list	Pharma PNEC Lists. Contains link to antibiotic PNECs that are maintained by the AMR Industry Alliance (see above). Pharmaceutical PNECs are provided to facilitate environmental risk assessment of pharmaceuticals in surface water downstream of the mixing zone (i.e., not at the point of discharge or entry into the environment). The PNECs were derived from guideline studies (e.g., OECD, USFDA, USEPA) conducted under Good Laboratory Practices (GLP). An appropriate assessment factor was applied to the lowest chronic toxicity endpoint unless otherwise indicated as derived from acute data.	[25]

Source	Comments	Ref.
Vestel *et al.*, 2016	Use of acute and chronic ecotoxicity data in environmental risk assessment of pharmaceuticals.	[26]
Tell *et al.*, 2019	Science-based targets for antibiotics in receiving waters from pharmaceutical manufacturing operations.	[4]
Gunnarsson *et al.*, 2019	Pharmacology beyond the patient – the environmental risks of human drugs.	[30]
Roos *et al.*, 2012	Prioritising pharmaceuticals for ERA: Towards adequate and feasible first-tier selection.	[32]
Le Page *et al.*, 2017	Integrating human and environmental health in antibiotic risk assessment	[33]
Bengtsson-Palme and Larsson, 2016	Concentrations of antibiotics predicted to select for resistant bacteria: Proposed limits for environmental regulation.	[12]

4.3.3 PNEC values in the absence of ecotoxicity data

In some cases, insufficient environmental toxicity data exist to derive a robust PNEC value, e.g. for some older established pharmaceuticals which pre-date current regulatory ERA requirements or for newer pharmaceuticals in the development pipeline. In such cases, scientific judgment can be used to either estimate a PNEC value using a read-across approach or to determine whether using a *de minimis* PNEC is appropriate.

Read-across approaches

- A read-across approach can be considered if ecotoxicity data are available for molecules with a similar chemical structure or with a similar mechanism of action. The use of ecotoxicity data from another molecule should consider differences that may exist for uptake and clearance and potency at the target. Additional exposure factors may be warranted.
- In some cases, environmental species models have been used in pharmaceutical discovery and development to investigate pharmacology or to predict toxicology. Zebrafish (typically embryos and larvae) are a common model that has been used to investigate the effects of molecules using aqueous exposures. While not conforming to standard ecotoxicity protocols and not covering all trophic levels, data from these models may be useful in identifying concentrations that have pharmacological or toxicological effects in fish. Appropriate assessment factors would need to be applied to protect other trophic levels.
- Another potential approach is employment of the fish plasma model as described by Huggett *et al.*, 2003 [27] and Rand-Weaver *et al.*, 2013 [28]. This model uses the plasma concentration in humans or mammals following a pharmacologically or toxicologically effective dose in humans or mammals. It is assumed that the same internal concentration in fish will have a similar pharmacological or toxicological effect. Then, a water-to-blood partitioning model for fish (e.g. Fitzsimmons *et al.*, 2001 [29]) is applied to calculate the concentration in water that will result in that internal concentration. As with the discovery and development models, appropriate assessment factors would be applied to protect other trophic levels and pharmacodynamic differences between mammals and fish.
- In a study with various steroid estrogens Caldwell et al., 2012 [30] used *in vivo* vitellogenin (VTG) induction studies to determine the relative potency of the

steroid estrogens to induce VTG and to construct a species sensitivity distribution (SSD) based PNEC also for compounds with insufficient data.

De minimis or "default" approaches

- A *de minimis* or "default" approach uses a single value for the PNEC for most pharmaceuticals. One possibility is to align default values with regulatory expectations for pharmaceutical registration. The US FDA (1993) considers 1 µg/L ("at the end of the pipe", that is, prior to dilution into surface water) to be the threshold for concern for pharmaceuticals. The EMA (2006) considers 0.01 µg/L (in surface water) to be the threshold of concern. Concentrations below these levels are considered to be safe for environmental species by these regulatory agencies. However, both agencies stipulate that there are pharmaceuticals with certain mechanisms (e.g. interaction with reproductive hormone and thyroid receptors, antimicrobial activity) for which these concentrations are not protective.
- De *minimis* PNECs could also be adopted from recent retrospective analyses of available aquatic toxicity data with pharmaceuticals. Gunnarsson *et al.*, 2019 [31] reviewed the range of chronic $PNEC_{surface\ water}$ for 133 compounds and found that for more than 90% these PNECs were >0.01 µg/L and for all hydrophilic ($logD_{OW}$ <3) substances the PNECs were >0.1 µg/L. When endocrine active substances (EASs) were removed from the analysis more than 90% had PNECs >0.1 µg/L irrespective of hydrophobicity. A similar analysis was carried out on 195 APIs using PNECs from the Swedish FASS.se database of pharmaceuticals (available at http://www.fass.se/LIF/startpage and reported in Roos *et al.*, 2012 [32]). These data demonstrate that in more 90% of cases the PNECs reported in FASS.se were ≥0.1 µg/L (Supplemental Table S3 – Roos *et al.*, 2012 [32]). The data reported in these two analyses can be used to guide *de minimis* levels of concern as appropriate based on mechanism and hydrophobicity.
- The current PNEC list of the AMR Industry Alliance now stands at 125 antibiotics; however, it is recognized that this list does not encompass all manufactured antibiotics. Therefore, Vestel *et al.*, 2022 [34] conducted a statistical evaluation of currently available data and a default PNEC of 0.05 µg/L for antibiotics in the absence of other data was derived.

Without data collected with the pharmaceutical of interest, all approaches will necessarily be conservative to reduce potential risks. Whichever approach is selected, whether one of those discussed above or something different, scientific expertise should be employed to justify the PNEC used.

4.4 Exposure assessment: calculating PECs

This section addresses the main steps needed and relevant considerations pertaining to the PEC calculation for discharges to surface water as the most likely exposure scenario. Other exposure scenarios may be considered depending on the local situation or company-specific procedures.

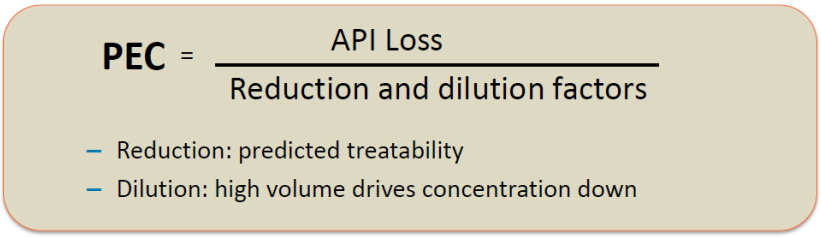

Figure 3 Determination of predicted environmental concentration (PEC)

The PEC is the concentration that results when the final API mass flow entering the receiving water is distributed in the water volume. The "API Loss" as denoted in Figure 3 is the mass of API lost "at source" in the operating facility. Reduction and dilution factors are numbers factoring in (1) reduction processes and (2) dilution in a given water volume to arrive at a concentration. The PEC is the sum of the background concentration and the process contribution (PC) from the manufacturing operation. When the background concentration is 0, then PEC is equal to PC.

In some countries such as e.g. Switzerland, the environmental protection law prohibits active dilution of emissions in order to reach threshold values. From an environmental perspective (API loads) active dilution should be omitted.

This simple equation is best understood when taking the perspective of API mass flow emanating from the facility as a wastewater discharge and flowing through a sequence of steps designed to treat the effluent before it reaches the receiving water. Starting with the API loss at source (in the operating facility), the final API mass flow entering the receiving water is determined by physical, chemical and/or biological reduction (removal) processes occurring within these treatment steps. The above description can be denoted using mass balance nomenclature as depicted in Figure 4, where WWTP denotes Wastewater Treatment Plant, and POTW denotes Publicly Owned Treatment Works (off-site).

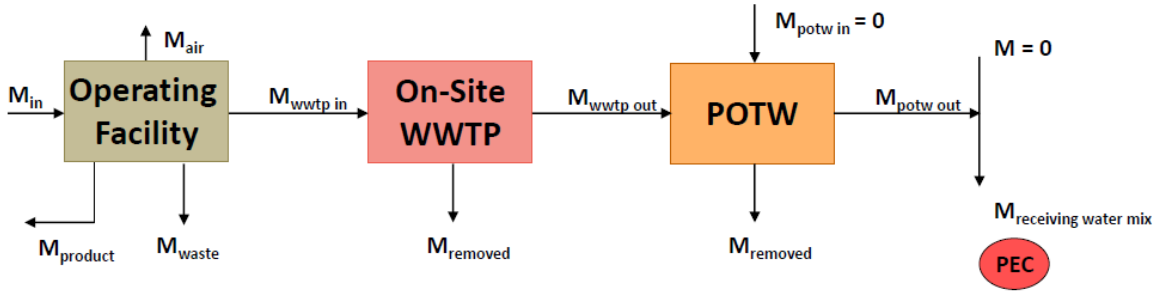

Figure 4 *PEC calculation based on API loss in aqueous waste*
M = Mass API; this calculation assumes $M_{potw\ in}$ = 0 and M receiving water (upstream from discharge point) = 0, when in reality, they can be >0 due to other (non-manufacturing) sources (e.g., human use/excretion).

4.4.1 Determining API losses from the facility

The first step in PEC calculation is to determine API losses from manufacturing facilities. This is usually performed by estimating losses in process aqueous waste and/or measuring the API concentration in the site's aqueous effluent.

Mass balances

Mass balances are used to estimate losses in process aqueous waste. They are an inventory of waste streams (solid, liquid, or gaseous) that may contain the API including estimates of the concentrations of API in each waste stream as well as its volume [1], as in Equation 1:

Equation 1 *Mass (M) = Flow (Q) × Concentration (C)*

Information about waste streams can be found in process descriptions, batch records, and other documentation. Initially, concentration estimates can be calculated from the mass of API and volume involved (e.g., mass in lot/batch, number of batches/year) and information on API losses, for example, from cleaning operations [1]. Guidances of the Pharmaceutical Supply Chain Initiative (PSCI) to calculate mass balances are listed in Appendix A1 in section 8.

Measuring the API concentration in the site's aqueous effluent

Chemical analysis of waste stream samples to determine the actual concentrations may be conducted to remove uncertainty. When measuring API concentrations in effluent, the limit of quantification (LoQ) of the chosen analytical method must be sensitive enough to measure anticipated effluent concentrations or risk-derived targets at the sampling point.

An important consideration is the choice of the sampling point: Samples of wastewater taken at or close to the point of generation (POG) typically have a much higher API concentration, thus requiring less analytical sensitivity. Wastewater testing at the site end of pipe typically requires a much lower LoQ, thus higher analytical effort (because of higher dilution and prior contaminant removal by wastewater treatment); however, testing wastewater at the site end of the pipe provides more representative data for actual

contaminant concentrations, factoring in potential on-site wastewater treatment, in the site's discharge.

Ensure that samples are taken during a typical manufacturing campaign and that sampling covers any cleaning (except any cleaning materials that are collected and disposed outside of the wastewater system) that occurs after the campaign. This requires an understanding of the manufacturing and WWTP operations (for example, batch vs. continuous manufacturing schedules and WWTP residence times, etc.). It has to be ensured that the samples are taken during a typical manufacturing campaign and that the samples are collected during API discharge. Further guidance on sampling and analytical measurement of APIs in wastewater is provided in Appendix A2 in section 9.

4.4.2 Factoring in reduction in biological wastewater treatment

The following guidance considers a biological WWTP's API reduction in the PEC calculation. It may be applied for plants on-site or those external to the facility. The final API mass flow entering the environment is determined by physical, chemical and/or biological reduction (removal) processes occurring within these treatment steps. These processes can be described using mass balance nomenclature as depicted in Figure 5, where WWTP denotes Wastewater Treatment Plant, and POTW denotes Publicly Owned Treatment Works (off-site).

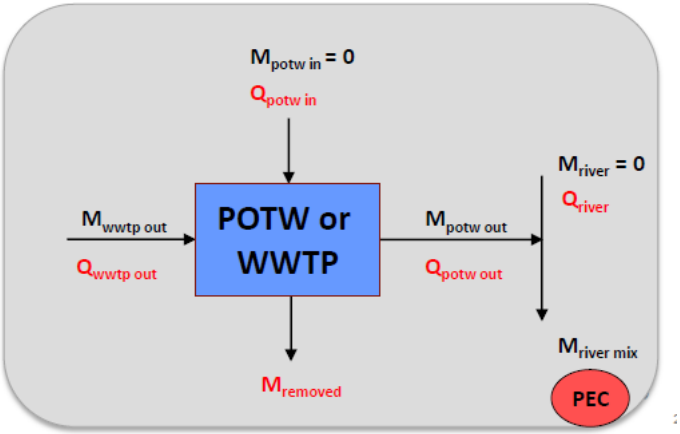

Figure 5 Mass balance of wastewater treatment steps
M = Mass; Q = Flow

It should be noted that the potential for toxicity to WWTP microorganisms is a recommended screening to assure an acceptable discharge rate to the WWTP. This screening can be implemented using the "Sewage Treatment Plant" (STP) exposure scenario described in Table 1 in section 4.3. This screening is important because impact to the wastewater treatment plant performance will reduce its effectiveness, and, therefore, its ability to remove API and other waste. If the predicted concentration of an API discharged to a WWTP exceeds a concentration that could affect the performance of the WWTP, actions are needed to either reduce the concentration of the API (e.g., by stream segregation or equalization) to an acceptable level or to render it less toxic to the WWTP biota. Such considerations may be particularly relevant for antibiotics, for example, which are designed to be toxic to microorganisms.

Once the influent to the WWTP is at a concentration that will not harm the WWTP microorganisms, the removal of API by WWTP processes can be factored into the PEC calculation. It must be noted that the degree of removal of APIs (if any) in a biological treatment process depends on the API's physical and chemical characteristics, WWTP technology and operational efficiency. The key processes that characterize the rate of transformation of organic contaminants in WWTPs are: hydrolysis rate – k_{hydrol}; biotransformation rate – k_{bio} in water, sludge; oxidation rate (via a specific oxidant); reduction rate (via a specific reductant); and photolysis rate – k_{photo}. Also sorption to sludge may be an important elimination process depending on the physical and chemical characteristics of the API.

Many APIs are hardly removed in conventional biological treatment because of their physical and chemical characteristics. Other technologies for enhanced treatment of these recalcitrant compounds are mentioned in section 5.3.

Based on this background, a conservative estimate, initially assumes 0% removal. Where biodegradability and sorption data for the API in question are available, this can be used to estimate a more refined removal efficiency. Predictive models such as SimpleTreat 4.0 (Struijs 2014) ([35] [36]) or removal data from the scientific literature may be used to establish removal efficiencies for specific APIs. These models allow the input of the parameters relevant to a specific WWTP so that various scenarios can be explored. A list of characteristics that are needed for the operation of a specific WWTP in order to be able to calculate removal using SimpleTreat is available in Struijs, 2014 [35], in an accompanying UBA document of 2015 [36] and in the ECHA Chapter R.16 guidance (2012) for environmental exposure assessment [37]. The facility should have a clear understanding of the range within those parameters vary with their real-world treatment plant and potential consequences of parameter variations for the risk assessment.

In addition to modelling, empirical influent and effluent measurements of API for an on-site treatment plant can give a good removal rate specific to that plant.

4.4.3 Factoring in dilution

For calculating the PEC based on API discharged through aqueous effluent, determine the dilution factor from the final effluent discharge into the receiving environment (e.g., river, lake, estuary, ocean).

Referring to Figure 5 above, the basic calculation of the dilution factor (DF) is straightforward, see Equation 2:

Equation 2 $$DF = (Q_{effluent} + Q_{upstream}) \div Q_{effluent}$$

where:

- $Q_{upstream}$ is the river flow rate upstream of Point of Discharge
- $Q_{effluent}$ is the discharge flow rate at the Point of Discharge. In an indirect discharge scenario, this flow rate equals $Q_{POTW\ out}$ in Figure 5.

Depending on the environmental situation, $Q_{effluent}$ is determined by the facility's discharge flow only (direct discharge situation), or, it is the discharge flow of an external treatment plant to which the facility discharges its effluent (indirect discharge situation).

Consider whether variations in river flow rates significantly impact overall risk. Typically, low flow conditions for streams should be used as a conservative starting point and different regulatory authorities provide guidance on how this is derived, e.g.:

- In the European Union (EU), the low-flow rate or 10^{th} percentile flow rate from the previous 7 years should be used if available (ECHA REACH, 2016 [37]). This calculation applies only to rivers, not estuaries or lakes. Where only average flows are available, the flow for dilution purposes should be estimated as one third of this average. The ECHA REACH guidance [37] requires that a maximum dilution factor (DF) of 1000 should not be exceeded. However, in reality, there are many situations where a DF >1000 is achievable and supported by river flow rates and discharge rates.
- In the United States, a 7Q10 flow is used when calculating surface water concentrations for regulated chemicals which is the smallest value of mean discharge computed over any 7-consecutive days over a 10 year period (USGS, 2009 [38]). These 7Q10 flow values are typically considerably lower compared to mean flow and would provide much more conservative estimates of environmental exposure.
- In Switzerland, the flow rate Q_{347} means the flow rate which, averaged over ten years, is reached or exceeded on an average of 347 days per year and which is not substantially affected by damming, withdrawal or supply of water (Swiss Waters Protection Act [39]).

Some local regulators may place a limit on the proportion of the channel width or the stream flow that can be used for diluting a given contaminant or API in this case. This is technically equivalent to defining a "mixing zone" around the Point of Discharge outside of which contaminant concentrations must meet acute or chronic limits. These mixing zones are often defined by the local environmental regulations.

To account for effluent discharges to water bodies with mixing zones, appropriate adjustment factors can be used. Further guidance on using and calculating dilution factors considering mixing zones is provided in Annex A3 in section 10.

4.5 Determining risk (risk characterization)

Calculating a risk quotient (RQ) from established measures of exposure (PEC) and effect (PNEC) is straightforward. Uncertainty with PEC and PNEC values and consequently for RQ should be evaluated. If needed, several iteration cycles for calculating and refining RQ must be run. If RQ after adequate refinement indicates a potentially unacceptable risk, measures for risk mitigation and management following the establishment of internal discharge targets might be needed. Further guidance on risk mitigation and management is presented in section 5.

Examples for (external) risk characterization guidances are listed in Annex 4 in section 11.

One of the two factors driving the RQ is the PEC. A tiered approach for refining the PEC based on different levels of environmental information is presented. The basic idea behind the tiered approach is that discharges that do not have a significant impact on a water body are deselected. In the following paragraphs this tiered approach is explained for the different types of receiving water bodies: rivers, lakes, transitional waters and coastal waters.

The flow diagram in Figure 6 illustrates how a user can apply the tiered assessment approach to assess its products.

Tier 0
At this Tier 0 irrespective of the receiving water body, if the concentration in the effluent is below the chronic PNEC value no further evaluation is needed because this discharge will not lead to an exceedance of the chronic surface water PNEC. If the effluent concentration is greater than the chronic PNEC, then higher tiers need to be completed.

Tier 1
In Tier 1, the concentration in the receiving water is calculated using site-specific hydraulics and some default assumptions about dilution. If the PEC is less than the chronic PNEC then the evaluation is complete. If the calculated PEC is greater than the PNEC, then a higher tier needs to be completed. The PECs can also be calculated for locations where acute PNEC and drinking water PNEC values apply, if those have been developed and if it is deemed to be required by the local situation.

Tier 2
Tier 2 uses more site-specific knowledge of both the effluent and the receiving water to determine dilution factors. Simple models and mathematical equations can be utilized to describe mixing in this area of the receiving water body. As in Tier 1, concentrations in an acute mixing zone and at a drinking water intake should be calculated if PNECs for those compartments have been determined and if it is deemed to be required by the local situation.

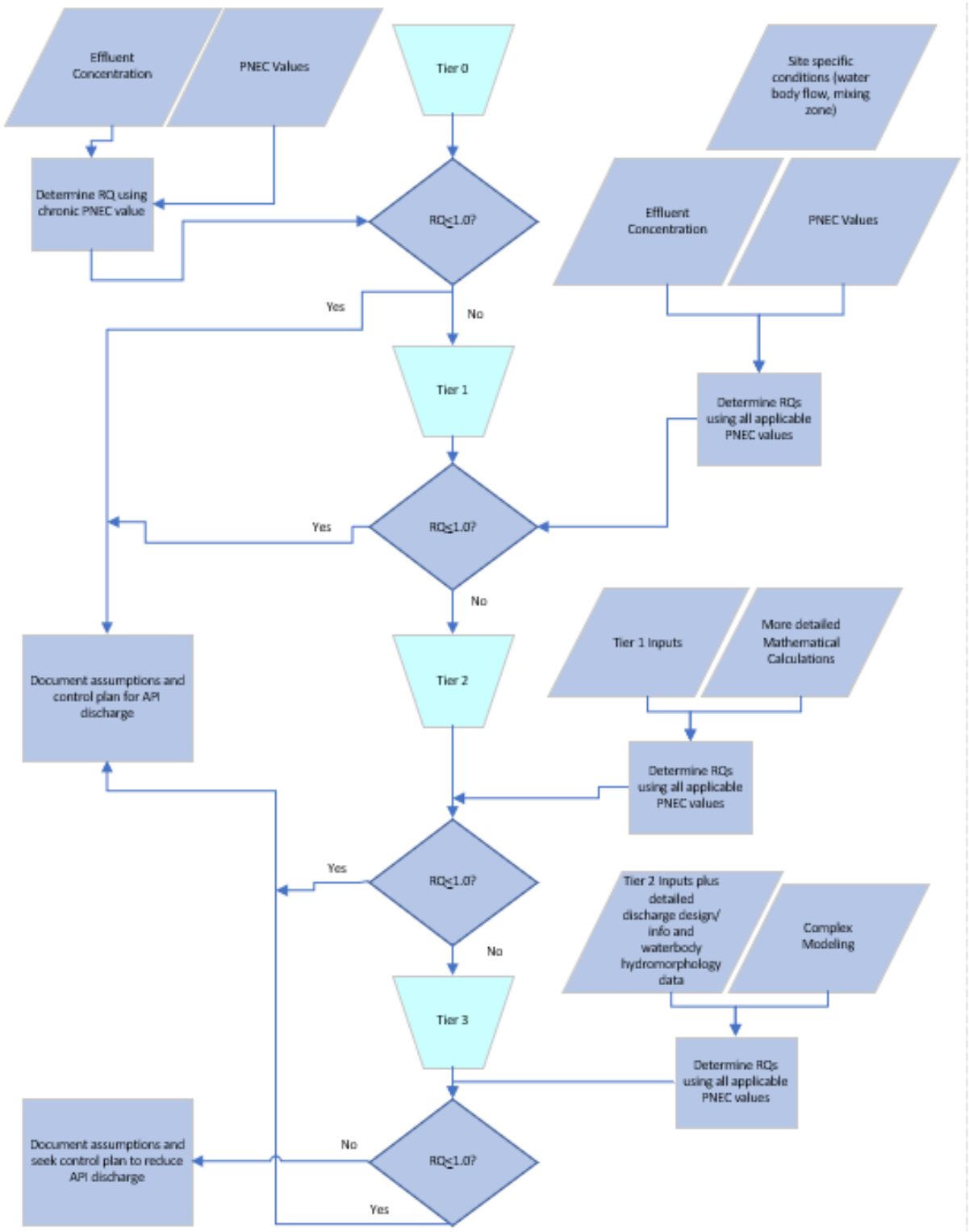

Figure 6 Tiered risk assessment approach

Tier 3

In Tier 3, more complex models of the mixing zones are used, varying from 2-dimensional approaches based on the Fisher equations to complex 3-dimensional or empirical models such as CORMIX[1]. These calculate the dilution of the discharged effluent as a function of the distance from the discharge point. The choice of approach depends on the individual situation. Clearly complex 3D models will require extensive input data to describe the situation in a reliable way (effluent discharge design, effluent velocity, bed topography, river flow, interactions with tributaries etc.). In order to model complex situations, data requirements may be demanding with data broken down into a network of individual small area units or grids, i.e. the bed topography, has to be gathered grid by grid as input for calculations to be performed.

The dimensions of the model-area are important, as at the boundary of the model area the influence of the emission has to be negligible. In modelling terms, the area in the vicinity of the discharge point is often described in great detail with a fine grid while at a greater distance from the outfall a more general representation may be adequate. More than one model may be needed if the influence of the discharge is not negligible at the boundary of the modelled area. This has consequences for the necessary computer time and costs as modelling in this way can become a complex exercise. The principles described above hold for the modelling of mixing zones in all kinds of water types, such as rivers, tidal rivers, lakes and estuaries.

[1] CORMIX is a USEPA-supported mixing zone model and decision support system for environmental impact assessment of regulatory mixing zones resulting from continuous point source discharges. The system emphasizes the role of boundary interaction to predict steady-state mixing behaviour and plume geometry (see http://www.cormix.info for more information).

5 Risk mitigation and management

5.1 Risk reduction hierarchy

Understanding potential emissions of APIs at the point of generation (POG) allows for prioritization and decisions to be made about segregating and controlling waste streams which could have an adverse environmental effect if released. To avoid high loads of APIs entering a site's wastewater effluent, a good understanding of the content of APIs in waste streams is important. Waste stream analysis can allow manufacturers to potentially optimize and implement the most effective pollution prevention and control measures (see also Caldwell *et al.* (2016) [1]) from where part of the following sections are cited).

The following risk reduction hierarchy can be proposed:

1. Reduce overall API through process improvements (i.e. yield improvement)
2. Minimise API losses to wastewater by equipment cleaning (i.e. dry clean before wet clean)
3. Segregate and collect concentrated waste streams at point of generation or "POG" (i.e. first equipment rinse water)
4. Assess alternatives for POG waste streams (destruction through off-site incineration, volume reduction through on-site evaporation, destruction through on-site treatment)
5. WWTP modifications/improvements (i.e. advanced oxidation, membrane separation)

Process improvements

To increase/optimize the process yield, modernization of the process could be a preferred option to prevent or minimise upstream the API load of a wastewater stream. However, this may not be an alternative because of good manufacturing practice (GMP) requirements.

Equipment cleaning

Cleaning procedures can be optimized to reduce the API loading and to lower disposal costs by performing a thorough initial dry cleaning and by reducing the volume of high-strength rinses being generated. An additional separate cleaning step (pre-rinsing) can remove large portions of APIs from large-volume wash waters. The high-load pre-rinse streams can be separated and addressed subsequently by a selective technology or incineration/thermal oxidation.

If dry cleaning is performed, workplace safety has to be carefully monitored. Dry cleaning may not be an option, if cleaning-in-place (CIP) is mandatory according to the company's standard operating procedure (SOP).

Segregation of waste streams

Mass balances, as introduced above, can also aid to identify wastewater stream(s) that could be segregated for disposal at an off-site facility, waste streams suitable for effective on-site treatment prior to disposal, and waste streams that will require specific pre-treatment prior to disposal to a wastewater treatment system.

Analyses need to be conducted to determine whether any residuals could pose a risk either to a subsequent WWTP (i.e., inhibition or interference) or to a receiving

environment (i.e., lake, river, or ocean) after discharge. To avoid high loads of APIs entering a site's wastewater influent, a good understanding of the content of APIs in waste streams is important. Waste stream analysis can allow manufacturers to potentially optimize and implement the most effective pollution prevention and control measures.

Alternatives for POG waste streams

Active pharmaceutical ingredient removal is compound-specific and should be addressed on a case-by-case basis. Removal efficiencies of different treatments vary with different APIs, depending on the suitability of the treatment for the API and on the specific wastewater composition in each case (e.g., salinity, turbidity, organic load). Mass transfer processes (API trapping) may be employed to remove APIs from solution into the solid phase, thereby concentrating the volume of waste for treatment. Activated carbon adsorption, chemical precipitation or flocculation, membrane separation or thermal processes (evaporation) generate either concentrated liquids or solids (for incineration). Further, advanced oxidation at POG such as ozonation or electrochemical oxidation such as Fenton's reagent is effective.

WWTP *modifications*/improvements

Many facilities in API production and final dosage production in the pharmaceutical industry rely on the use of neutralization, equalization, and biological (primarily activated sludge) treatment technologies for their wastewater treatment. However, many APIs are hardly removed in conventional biological treatment because of their physical and chemical characteristics. More advanced technologies such as ozonation or electrochemical oxidation such as Fenton's reagent are applied at manufacturing sites to remove specific compounds for which conventional treatment approaches do not work.

End-of-pipe treatment can also be considered as an alternative, although this option is not preferred because of higher volumes, mixing with other chemicals, and lower concentrations of the compound to be treated.

5.2 Wastewater testing and assessment

As discussed above, the decision on whether a particular wastewater stream can be discharged directly to a biological WWTP is an important production factor for any site. In order to properly reach this decision, a discharger must evaluate whether a wastewater has the potential to cause a toxic effect in an activated sludge system at the concentrations expected to be present (with a presumed safety factor), and then assess the biodegradation/removability of the API.

5.2.1 Initial Wastewater Testing and Assessment

To assess the removability of the individual API, consideration should be given to an appropriate test method, e.g., OECD 302 B, or equivalent, to characterize the total effect of all elimination mechanisms in a biological treatment plant [48]. Additionally, sludge respiration inhibition testing (e.g. according to OECD test guideline no. 209 or equivalent) should be applied to assess a potential toxicity to the activated sludge microorganisms.

OECD 302 B tests can be complemented with toxicity controls and the analysis of oxidised nitrogen compounds (N_{ox}) such as nitrate and nitrite in order that they can be used as an overall assessment tool, giving results not only for the elimination of compounds (or the fraction of refractory carbon), but also on the toxicity of the heterotrophic microorganisms (that degrade the carbon substrates) and even on the toxicity to nitrifying microorganisms. However, the OECD 302 B test has the drawback of a long incubation time of up to 28 days to give reliable results (although modifications with a shortened incubation time of 7 days exist).

Whole effluent toxicity (WET) testing may be used to assess the combined effects of all constituents of a complex effluent rather than assessing the toxicity of single chemicals or constituents and could be a predictor of the toxicity potential of effluents. Advantages and disadvantages of using WET testing are discussed in Caldwell et al., 2016 [1].

5.2.2 Site-specific evaluations

Removability through physical-chemical pretreatment or in a WWTP relates to the specific properties of the substance(s) involved. Results from a lab test do not refer to the specific conditions at a given production site, where the availability of an industrial or a municipal/mixed WWTP, with different substance concentrations, flow rates, adaptation of the activated sludge (AS), AS concentration, hydraulic and AS retention times, or possibly precipitation, flocculation, denitrification, dephosphatation, filtering or other additional treatment steps, may have a strong influence on removal rates [48]. Therefore, it is recommended to perform pilot testing on-site or in a laboratory environment if more information is needed from the OECD Tests.

Strategies for the management of wastewater streams on a multi-purpose site can be ineffective if individual wastewater stream management cannot be ensured. Management of wastewater streams should be automated whenever possible. In some cases, facilities should consider writing local procedures to ensure appropriate wastewater segregation. Within the manufacturing of a single API, and/or different API production campaigns, the destination of wastewater streams may change frequently.

5.3 Pretreatment options

In certain cases, particularly from formulation single recalcitrant or potentially ecotoxic wastewaters are investigated in more detail for the possibility of physico-chemical pretreatment. In order to ensure the destruction or removal of highly active pharmaceutical ingredients (APIs), such investigations may encompass physical removal through precipitation, flocculation, or adsorption to activated charcoal or other substrates, possibly furthering hydrolysis through raising or lowering the pH, with or without additionally heating the wastewater, or ozonation. Additionally, treatment with UV radiation, or advanced oxidation processes (AOPs) using UV with photosensitisers or oxidisers, may be tested [48].

Best Available Techniques (BAT) Reference Documents (BREFs) from the EU can be consulted for pretreatment options for wastewaters from the chemical sector ([49] [50]). Pretreatment options and case studies are also found in the literature, see e.g. Deegan et al., 2011 [51], Martz, 2012 [52], Caldwell *et al.*, 2016 [1], Pal, 2018 [53] and Straub *et al.*, 2020 [48].

Examples for pretreatment options were also presented by the Pharmaceutical Supply Chain Initiative (PSCI):

- In the course of a PSCI sponsored webinar on how to manage APIs in manufacturing effluent (Part 3) which took place on 25th October 2016 (https://pscinitiative.org/resource?resource=297)
- In the course of the PIE/AMR Deep Dive training seminar held on 17th September 2019 in Hyderabad, India (https://pscinitiative.org/resource?resource=482)

However, one should be conscious that any kind of pretreatment will generate costs, including environmental costs, from investments made, over increased energy consumption, additional raw materials needed, more CO_2 produced, or other kinds of wastes generated. Wastewater incineration in general is the last option, as an inordinate amount of energy is needed to evaporate water, often constituting well over 98% of a wastewater, to eventually combust the minor residues of recalcitrant or (eco)toxic organics. Therefore, a careful comparison between available pretreatment options should be made, to identify the optimal under the given circumstances [48].

6 Glossary

AMR	Antimicrobial resistance
AOP	Advanced oxidation processes
API	Active Pharmaceutical Ingredient
AS	Activated sludge
BAT	Best available technique
BCF	Bioconcentration factor
BREF	Best available technique reference document
CAPA	Corrective and preventive action
CIP	Cleaning-In-Place
CORMIX	CORnell MIXing Zone Expert System supported by the USEPA
DF	Dilution factor
DMEL	Derived Minimal Effect Level
DNEL	Derived No-Effect Level
DQO	Data quality objective
EAS	Endocrine active substance
EC10	Effective concentration that causes 10% of the maximum response
EC50	Effective concentration that causes 50% of the maximum response
ECHA	European Chemicals Agency
EE2	Ethinylestradiol
EHS	Environment, Health & Safety
EMA	European Medicines Agency
EPA	US Environmental Protection Agency
EPAR	European public assessment reports
EQS	Environmental Quality Standards
ERA	Environmental Risk Assessment
EU	European Union
EUCAST	The European Committee on Antimicrobial Susceptibility Testing
FDA	U.S. Food and Drug Administration
GLP	Good Laboratory Practices
GREAT-ER	Geo-referenced Regional environmental Exposure Assessment Tool for European Rivers
IBC	Intermediate Bulk Container
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
iPiE	IMI Project Intelligence-led Assessment of Pharmaceuticals in the Environment
K_{OC}	Soil adsorption coefficient
K_{OW}	Octanol/water partition coefficient
LC10	Lethal concentration that causes 10% of the maximum response
LC50	Lethal concentration that causes 50% of the maximum response
LoQ	Limit of quantification
M	Mass
MDL	Method detection limit
NOEC	No effect concentration
OECD	Organisation for Economic Co-operation and Development
PEC	Predicted environmental concentration
PhATE	Pharmaceutical Assessment and Transport Evaluation model

PIC/S	Pharmaceutical Inspection Convention
PNEC	Predicted no effect concentration
POG	Point of generation
POTW	Publicly owned treatment works
PSCI	Pharmaceutical Supply Chain Initiative
Q	Flow
QA	Quality assurance
QC	Quality control
QS	Quality standard
REACh	Regulation (EC) concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals
RQ	Risk quotient
SOP	Standard operating procedure
STP	Sewage treatment plant
TGD	Technical Guidance Document
TSD	Technical support document
VICH	Trilateral (EU-Japan-USA) programme aimed at harmonising technical requirements for veterinary product registration
WET	Whole effluent toxicity (testing)
WFD	EU Water Framework Directive
WHO	World Health Organization
WWTP	Wastewater treatment plant

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8 Appendix A1: Guidance for calculating mass balances

Mass balances are an inventory of waste streams (solid, liquid, or gaseous) that may contain the API including estimates of the concentrations of API in each waste stream as well as its volume, as in Equation 1:

Equation 3: Mass (M) = Flow (Q) x Concentration (C)

Figure 7 below depicts the principle of accounting for known mass inputs and outputs of an operating facility to estimate the unaccountable losses. In the simplest case all yield losses of API from a production process are present in one single aqueous waste. However, in reality losses are often distributed over different pathways, with losses in solid forms, semi-solids, as well as with aqueous process wastes. If all losses in solid or semi-solid forms are known with the required precision, then the loss through the aqueous process waste can be deducted from the mass balance. Some API losses in solid forms, such as with tablet waste, are precisely quantifiable while other losses are not (e.g. filter residues). These complications limit the applicability and / or precision of mass balances for estimating API loss to wastewater discharge.

Information about waste streams can be sourced from process descriptions, batch records, technical service reports, etc. Preliminary concentration estimates can be derived from the masses of API and volumes involved (e.g., mass in lot/batch, maximum daily losses based on number of batches/day and cleanings/day, etc.) using known chemical, physical and biological properties of the compound and information on API losses, e.g., from cleaning operations. In some cases, these estimates may be confirmed analytically.

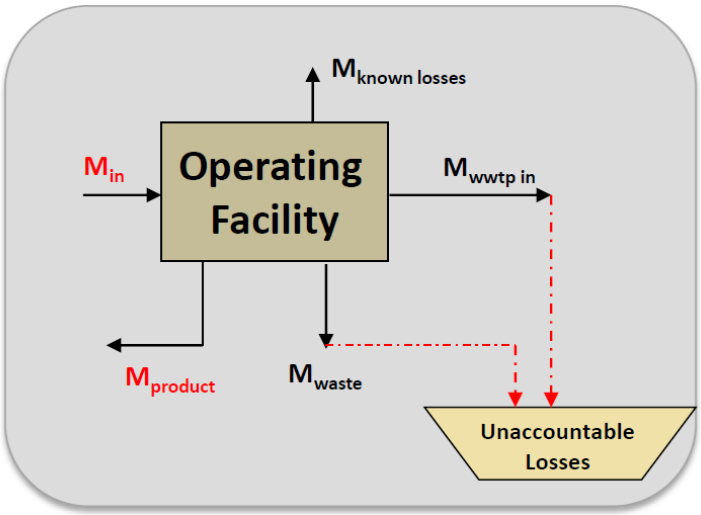

Figure 7 Mass balance of the operating facility
M = Mass API

Losses should be quantified as daily loss (e.g. in kg/day). To cover the short term peaks, a *maximum* of API predicted to be lost within a 24 h processing period could be used in the equation.

In order to use an *average* daily API loss as used for chronic exposures, the estimated API loss/year can be calculated according to the following steps:

i. Estimate or measure the mass of API lost during a typical batch.
ii. Determine the total mass of API lost during all manufacturing campaigns in one year.
iii. Determine the number of days of manufacturing activities in one year.
iv. This is used to calculate an average loss during the manufacturing period.

However, caution needs to be done when considering to do this. Depending on the mode of action and/or the dose response curve of the toxicity data, the calculated average may not be appropriate.

Examples for mass balances were presented by the Pharmaceutical Supply Chain Initiative (PSCI):

- In the course of a PSCI sponsored webinar on how to manage APIs in manufacturing effluent (Part 2) which took place on 15th June 2016 (https://pscinitiative.org/resource?resource=295)
- In the course of the PIE/AMR Deep Dive training seminar held on 17th September 2019 in Hyderabad, India (https://pscinitiative.org/resource?resource=482)

9 Appendix A2: Sampling and analysis of pharmaceutical industry wastewater for APIs

Section 1. Introduction

Measuring the Active Pharmaceutical Ingredient (API) content in pharmaceutical industry wastewater is another risk assessment tool for evaluating discharges from manufacturing. The data can be used to supplement API losses estimated by mass balance methods (see section 4.4.1 and Appendix A1 in section 8).

There are some challenges with generating meaningful data when sampling pharmaceutical wastewater, so this document is designed to give the user some practical guidance when developing sampling and analytical plans, and it offers guidance for evaluating analytical results.

The principals and procedures described in this guidance are not substitutes for any of the specific sampling and analysis provisions required by regulatory authorities.

Section 2. Sampling Plan

A well-designed wastewater sampling plan will ensure that representative samples are collected. Choosing the sample location, sample type/sampling equipment, number of samples and sample dates will depend on several factors, including:

- API of interest
- Sampling objectives
- Analytical target and Data Quality Objectives
- Site production schedule
- Wastewater discharge temporal variations and residence times in collection/treatment systems

2.1 Pre-Planning

Understanding the general production process flow for the API of interest is paramount. A process flow diagram showing the point of generation (POG) and fate (wastewater treatment, off-site incineration, recovery, etc.) for all liquid losses will help determine what, where, how and when to sample.

The sampling objective should be clearly understood. Wastewater samples are typically collected to help quantify API losses associated with a production line or specific unit operation. When quantifying API losses for an entire production line, sampling the total wastewater discharged from the site or a building is most common. Although not always practical, sampling a dedicated process wastewater line (no sanitary or utility wastewater) is desired to minimise analytical matrix interferences. Sampling a specific unit operation at the POG is useful in distinguishing high API waste streams from low API waste streams. Understanding the relative strength of API-containing waste streams can help drive targeted control strategies.

The analytical target (detection limit) and data quality objectives (DQOs) should be established with the contract laboratory well in advance of sampling. For wastewater

sampling purposes, the DQOs and analytical detection limits will help determine how much (volume) and how many samples should be collected for analysis. Guidance on setting analytical detection limits and DQOs is provided in more detail in Section 3.

Given the typical campaign operation of pharmaceutical manufacturing, aligning wastewater sampling with the production schedule can be challenging. Know the schedule well in advance and be prepared for changes. Special emphasis may be needed on short-run campaigns where opportunities to sample may be limited.

Typical batch operations in pharmaceutical production usually generate wastewater discharges that are temporal in nature (i.e. equipment cleaning). Understanding when and where in the process the target API wastewater discharges occur will help establish a sampling timeframe. Include the residence times of wastewater collection and wastewater treatment systems when establishing sample dates.

2.2 Choosing a Sample Location

Choosing the sample location will depend on the sampling objective. Targeting specific production processes at or near the point of generation (POG) or quantifying API in the total wastewater discharged from the site are the two basic scenarios.

A challenge when sampling at POGs is accessibility. There may be access restrictions due to GMP protocol or there may be no simple means to divert targeted waste streams to a sample collection point. Use of totes/IBCs and temporary piping may be necessary, and this could be disruptive to the normal production process. Careful planning with production personnel is necessary.

When sampling at the POG does not fit the sampling objective or when it is not practical, sampling the total wastewater discharge from the site is an option. Accessibility becomes less of an issue, particularly for sites required to collect routine samples required by permit or license. The challenge with this scenario is with the laboratory analysis because a total wastewater effluent sample is more complex, and it can present more matrix interferences that could impact analytical detection limits (see section 3). On the other hand, dilution means you may need a more sensitive analytical method.

Choose a location that meets the sampling objective and is the least intrusive to production operations.

2.3 Choosing Sample Type/Sampling Equipment

There are two types of wastewater samples: grab or composite.

A grab sample is a single sample collected over a short period of time (usually instantaneously). Analysis of a grab sample will indicate the characteristics of the wastewater sampled at a location and point in time. It cannot usually be extrapolated to longer averaging times.

Grab samples are useful and typically most practical when collecting at the point of generation (POG) in the production process, and when a wastewater discharge occurs

over a short period of time (i.e. equipment cleaning), or where routine discharges have quality criteria (i.e. an aqueous mother liquor). However, it is important to ensure that a grab sample is representative of the entire discharge. In cases where it is not, collecting a series of grab samples to form a composite is acceptable.

Composite samples are intended to represent the composition of a wastewater over a specified averaging period (e.g., typically 24-hours). There are two types of composite sample:

- Flow-weighted – the composite consists of multiple grab samples collected during the averaging period and whose volume added to the composite sample is calculated based on the wastewater flow at the time that the grab sample was collected.
- Time-weighted – grab samples collected at specified time intervals during the averaging period are added in equal volume to the composite sample.

In general, flow-weighted composites are the preferred method for sampling continuous wastewater discharges because they usually provide the most representative sample. However, flow-weighted composite samples require special equipment that may not be readily available or practical, unless there is an existing permit or license requirement. In cases where it is possible to collect a flow-weighted composite sample, collecting a time-weighted composite sample is acceptable.

Sample collection can be performed either manually or automatically. The decision as to the type of sampling equipment to use is generally site-specific and depends upon the type of samples required to meet the project objectives and the planned duration of the sampling program (e.g., routine monitoring versus one-time sampling). Programmable automatic samplers that can collect either grab samples or composite samples simplify sample collection and minimise the amount of manual intervention.

The following principles should be considered when selecting sampling equipment:

1. Physical conditions for obtaining the samples – Accessibility to the sampling point is typically more challenging when sampling at the POG, especially when working in GMP areas or where there is no convenient means to divert targeted waste streams to a sample collection point. The use of totes/IBCs and temporary piping may be necessary
2. Volume of sample required for all specified analyses – The analytical lab requirement is typically small (<1 litre), but it is important to collect a large enough sample that is representative of the entire discharge; size collection equipment appropriately.
3. Compatibility of sample containers with the type of analyses to be performed – Glass amber bottles are preferred to minimise API adherence to sample container walls and to minimise photolysis; establish specific requirements with the contract laboratory.
4. Requirements for preservative addition and holding times – Most samples must be preserved to prevent changes in chemical composition between the times of sampling and analysis. Maximum holding times for preserved samples should also be established to assure that the analysis is conducted before chemical composition changes occur in the stored samples. The required preservation and holding times for samples collected will either be supplied by the approved analytical method or determined when a method is developed using your sample matrix.
5. Sample refrigeration – provisions should be made to keep samples cool during collection (composite samplers), during interim storage and during shipping.
6. Programming capabilities of automatic samplers – ideally capable of collecting flow-weighted samples when flow monitoring can be integrated.

In summary, when choosing sample type and sampling equipment:

- Collecting representative samples is required
- Choose sample type depending on the project objective and wastewater discharge duration
- Use grab samples typically at POG and/or when discharge duration is short
- Use composite samples typically when total wastewater effluent from the site is sampled
- Composite samples are usually preferred (most representative)
- Automatic samplers offer the most flexibility and minimal manual intervention
- Establish sample container type, sample volume, preservatives, holding times and packaging/shipping requirements with the contract laboratory

2.4 Determining Number of Samples

The number of samples to collect will depend on several factors: project objectives, the nature and frequency of the wastewater discharge, the duration of the discharge and the residence time through the wastewater collection system and treatment plant, where applicable.

When sampling at or near the point of generation, sample sets should be defined based on the nature and discharge frequency of the process operations. A typical POG sampling scenario involves collecting equipment cleaning samples. Wastewater discharges from equipment cleaning will vary widely in volume and duration depending on equipment size and cleaning methodology (manual vs. clean-in-place). Also, the discharges may vary from batch-to-batch, so consider collecting multiple samples.

When sampling wastewater effluent, collecting a minimum of three consecutive daily composite samples is recommended. In some cases, consider extending sampling to more than three days if discharges to wastewater occur over an extended period. Conversely, smaller sample sets may be justified given site-specific conditions, such as batch operations where all activities (including equipment cleaning) may occur on one day. However, a larger dataset is usually more desirable because it can capture variability and peak discharges.

2.5 Establishing Sample Dates

Establish sample dates based on the production schedule for each API of interest, the process knowledge predicting process steps with API losses, the nature and duration of the discharge (batch vs. continuous), and the residence time through wastewater collection systems and treatment plants, where applicable.

Sampling dates should be directly linked to the discharge activity and its duration. Collecting samples at the POG should be straightforward. Often these discharges are of a batch nature and short duration, so timing the collection with production personnel is critical. Otherwise a sampling opportunity could be missed or samples not representative of the actual discharge could be collected.

Sampling dates at the site wastewater effluent involves a little more planning. It is still important to understand the timing of API-containing discharges associated with the process, but when to start and stop collecting daily composites depends on the residence time through wastewater collection system and treatment plant if sampling downstream from the POG. For process operations where there is a primary source of API-containing wastewater (equipment cleaning), start sampling when the activity is expected to occur and ensure that the sampling event is long enough to account for any residence time. For example, if cleaning starts on Monday and the residence time is 24 hours, the recommended 3-consecutive day sample period would be adequate. Consider extending sampling to more than 3 days if equipment cleaning activities or other discharges to wastewater occur over a period of several days.

2.6 Recordkeeping Considerations

Maintaining complete records is very important. When samples are collected and how they are handled until receipt by the lab is typically captured on a chain-of-custody form provided by the contract lab. It will typically provide:

- The sample identification number.
- The container description (material, volume).
- The analyses to be performed on the sample.
- Any preservatives added to the sample.
- Any special instructions for sample handling or analysis.
- The date, time, and signature of everyone that is responsible for and has possession of the sample, beginning with the individual collecting the sample and ending with the individual at the laboratory that takes custody of the sample.

If not using a standard chain-of-custody form, maintain a sampling log that captures the information above. In the sampling log, it is also recommended that production activities generating wastewater for the API of concern are recorded, and that wastewater flow rates (either at the POG or total site discharge) are recorded so that mass discharge rates can be calculated.

Section 3. Analytical Plan

The combination of low PNECs and lack of standard analytical methods to measure APIs in wastewater presents a challenge.

It is not typical to monitor APIs in wastewater unless there is a permit or license requirement. The limited regulatory framework means that few commercial labs have the capacity or expertise to test API in a complex wastewater matrix at the low concentrations typically needed (ng/L) to make meaningful risk assessments. Internal Quality Control labs can test for API but typically in a clean matrix and at a high method detection limit (mg/L).

Given these limitations, it is often necessary to partner with a lab (commercial or academic) to develop analytical methods sensitive enough to measure an API concentration that would result in a PEC lower than the PNEC based on site specific flow rates and receiving water dilution factors. It doesn't necessarily mean that the analytical method detection limit must be less than the PNEC.

Determining which analytical method is most appropriate should be discussed with the laboratory. It is also important to understand the quality assurance/quality control (QA/QC) specifications for analytical methods that will be used, detection and quantitation limits, and matrix interferences.

3.1 Analytical method selection

Analytical method selection involves, at a minimum, selecting methods that meet the following:

1. The quality control (QC) tests in the method must be an integral part of the method;
2. The QC acceptance criteria in the method must be part of the method; and
3. The method detection limit (MDL) should be at least one third (1/3) the concentration limit being targeted.

The concentration limit being targeted can be calculated from the PNEC value and dilution factors. For example, if measuring API in total site wastewater effluent discharging directly to a surface water with a dilution factor of 50 and a PNEC of 0.01 µg/L, an MDL of 0.2 µg/L would be appropriate (0.01 µg/L × 50 × 0.33). Note that there may be instances where an MDL of 1/3 of the concentration limit is not achievable. These instances should be handled on a case by case basis.

3.2 Detection and Quantitation Limits

Detection and quantitation limits are essential components of an analytical method. Many different names are given to detection and quantitation limits by the different organizations that develop analytical methods. However, all of them can be simplified into two basic definitions:

- A *detection limit* is the lowest concentration of a substance that can be identified in a sample matrix. The concentration is so low that the concentration of the chemical present in the sample is uncertain and cannot be reported with acceptable accuracy.
- A *quantitation limit* (also called quantification limit, LoQ) is the lowest concentration of a substance in a sample matrix that can be measured at a specified level of precision (e.g., ± 30%). The quantitation limit for a substance in a sample matrix is always greater than the detection limit for that substance in the same matrix.

The difference between detection limits and quantitation limits is very important. At a quantitation limit, there is a much lower chance of a false positive measurement (i.e., reporting a substance as present when it is not) than there is at a detection limit. While not always possible, it is best to assure that the quantitation limit is enough for determining whether a PNEC value is being met.

3.3 Quality Assurance/Quality Control

Discuss Data Quality Objectives (DQOs) with the lab so that they can integrate quality assurance/quality control (QA/QC) measures into the analysis. There are several QA/QC measures that can be used to interpret the quality of the laboratory data.

3.3.1 Analysis of Spikes

The term spike refers to a known quantity of a target analyte that is added to a sample before analysis. The recovery of a spike from a sample (expressed as a percent of the spike concentration) is a measurement of the accuracy of the analysis. Accuracy is defined as how close a measurement is to the true concentration of the target analyte in a sample. The lab should establish a range of recoveries that is acceptable. Sometimes, spikes before sampling to cover the whole process (preservation, sampling, cooling, etc.) make sense. Also field blanks should be taken.

3.3.2 Analysis of Duplicates

Duplicate analyses are used to evaluate precision, which is the variance in measured concentrations. Discuss with the lab what duplicates analysis is appropriate. If field duplicates are desired, then that should be programmed into the sampling plan. Otherwise, the lab can perform method or instrument duplicates on random samples that are received provided that enough sample volume is collected. The lab should establish a precision range that is acceptable.

3.3.3 Analysis of Blanks

A blank is a sample that should be completely free of the target API. The objective of the blank is to detect contamination and/or interference problems, or to document their absence. As with duplicate samples, blanks can be introduced at various points in the sampling and analytical process. There are several types of blanks commonly used: trip, field, equipment, method, instrument. If field or equipment blanks are desired, then that

should be programmed into the sampling plan. At the very least, field blanks should be considered for analysis.

3.3.4 Analysis of Standards

Standards are used to assess instrument calibration and method performance. Most instrumental test methods require analysis of calibration standards every day the instrument is used, and one or more check standards are processed with every batch of samples analysed. The lab will typically prepare the standards and it will establish acceptance criteria.

3.3.5 Matrix interferences

Both the physical properties and chemical composition of a sample can influence the ability of an analytical method to measure a target analyte. Typically, matrix interferences will cause poor precision, poor recovery, and/or elevated MDLs and quantitation levels in a sample. If the interference is severe, the method may be unable to achieve the method performance requirements.

Matrix interferences most often occur in complex samples, and particularly in untreated and/or partially treated process wastewaters. Dilution of the sample is one approach to remove high concentration interferences, but it can elevate detection limits to concentrations that exceed target values. Most analytical methods include procedures that laboratories can implement to try to reduce matrix interferences. Be sure to identify samples where there is a higher risk of matrix interferences.

Section 4. Data Evaluation

How the data will be used will vary depending on the project objective. Are total API losses from the site being quantified? Are select processes being targeted to isolate and control a part of the API loss?

No matter the case, and as a first step, use the maximum measured API concentration in your risk analysis. If this worst-case condition indicates that the PEC is less than the PNEC, generally no further action is required. If the PEC is greater than the PNEC under these worst-case conditions, additional statistical analysis of the sample results should be performed to determine the appropriate indicator value for the risk assessment. For example, it may be appropriate to average the sample results. Also, if additional treatment occurs downstream of the discharge, it may be appropriate to perform modelling of the treatment system.

It is not uncommon to see API concentrations measured in wastewater that result in Predicted Environmental Concentrations (PECs) lower than those derived from mass balances, especially when conservative assumptions are made in the mass balance analysis. An order of magnitude difference should not be an alarm. When there is a big difference between the two methodologies, re-examine mass balances, validate the representativeness of the samples collected and consider additional wastewater testing.

Wastewater Sampling & Analytical Plan Checklist

For recordkeeping purposes, consider using this checklist to capture details for each sampling event

Wastewater Sampling Plan	
API of Interest	Click here to enter text.
Sampling Objective	Click here to enter text.
Process Flow Diagram	Click here to enter text.
Sample Location	Click here to enter text.
Sample Type	Click here to enter text.
Sample Equipment	Click here to enter text.
Number of Samples	Click here to enter text.
Sample Dates	Click here to enter text.
Production Activity Log	Click here to enter text.
Wastewater Flow/Volume	Click here to enter text.
Analytical Plan	
Lab Name	Click here to enter text.
Analytical Method	Click here to enter text.
Method Detection Limit	Click here to enter text.
Method Quantification Limit	Click here to enter text.
Sample Volume Required	Click here to enter text.
Sample Container Type	Click here to enter text.
Sample Preservative	Click here to enter text.
Holding Time	Click here to enter text.
Quality Control Measures	☐ Spikes ☐ Duplicates ☐ Blanks ☐ Standards
Quality Control Details	Click here to enter text.

10 Appendix A3: Guidance for calculating dilution factors considering mixing zones

There are many local situations where a receiving water body is only a narrow channel or is subject to low flows. In these situations, there is a risk that the mixing zone will occupy a major part of the cross- section which can have adverse consequences for the passage of aquatic life and could impact a large percentage of sessile organisms downstream of a discharge. To prevent such problems, some local regulators may place a limit on the proportion of the channel width occupied by the mixing zone. For example, the Netherlands limits the mixing zone to 25% of the cross-section of the water body. Therefore, it is important for a facility to understand if there is local guidance on assumed mixing. Also, where shellfisheries, drinking water abstractions, or other areas of special ecological significance are the discharge point, it is important to consider these features when determining the mixing zone. The distance between such features and the discharge point can be of great importance, especially when the distance is less than 10 times the width of the water body.

Several of the modelling and estimating principles used for rivers can be used for lakes. However, the definition of the dimensions of the mixing zone can significantly differ from the definition used for rivers. A major difference between lakes and rivers is the streaming velocity. In general, lakes are much less free-flowing than rivers. The mixing zone can be represented by a half a circle. In most cases the width of a lake is large. Making the length of the mixing zone proportional to the dimensions of the water body, i.e. the area of the water body, length and width of the water body, seems to be logic used by the Water Frame Directive implementation procedures. However, the European Chemicals Agency and U.S. EPA implementation procedures recommend more conservative ways of estimating dilutions for lake discharges. More information on the procedures can be found in Table 6.

Table 6 provides a comparison of the assumptions and inputs for calculating appropriate dilution factors while considering mixing zones in rivers, lakes and ocean receiving waters as recommended by the European Commission (Water Framework Directive), the European Chemicals Agency (REACH), and the US and Canadian EPAs. Assumptions and inputs useful for comparison of PECs to chronic, acute and drinking water PNEC values are included. The choice of which calculation methods are used may be driven in part by local regulator expectations. Table 6 does not include any calculation factors for estuary/tidal waters as they are case specific.

An overview for selecting the appropriate hydraulics for meeting PNEC values was also presented by the Pharmaceutical Supply Chain Initiative (PSCI) in the course of the PIE/AMR Deep Dive training seminar held on 17th September 2019 in Hyderabad, India (https://pscinitiative.org/resource?resource=482).

Table 6 Comparison of European Commission, European Chemicals Agency and United States Environmental Protection Agency surface water quality assessment factors used to assess compliance with Environmental Quality Standards

Guidance	**WFD**	**REACH**	**US EPA / Canada**
Agency	European Commission	European Chemicals Agency	US Environmental Protection Agency / Environment Canada
Guidance Reference	Water Framework Directive's Technical Background Document on Identification of Mixing Zones (2010) ([54] [55])	REACH Guidance on information requirements and Chemical Safety Assessment Chapter R.16: Environmental exposure assessment Version 3.0 (2016) [37]	Technical Support Document for Water Quality-based Toxics Control (1991) ([56] [57] [58] [59])
RIVERS - Chronic			
Default dilution factor for chronic PNEC values	Assume no dilution (i.e. no mixing)	A standard dilution of 10 is used when releasing to a freshwater environment.	Not provided. However, you could assume no dilution (i.e. no mixing)
Stream flow used for dilution analysis to meet chronic PNEC values	Q90 (The flow which is exceeded during 90% of the time. Sometimes also called a 10th percentile flow)	When carrying out a site-specific assessment, specific data on the receiving water may be used with regard to the dilution capacity of the environment (site-specific data should be justified and explained). However, it should be noted that a dilution factor higher than 1000 should not be used in any case.	7Q10 (The lowest 7 consecutive day flow that occurs once every 10 years)
Mixing zone size allowed for chronic PNEC	Up to 100% Q90. The WFD guidance notes that In some countries a limit is placed upon the proportion of the channel width occupied by the mixing zone. For example, in the Netherlands the mixing zone is limited to 25% of the cross-section of the water body. In the discharge test criteria are chosen in such a way that when mixing zone criteria can be met in streaming water bodies at distance (L), the cross section taken	Based on defaults only unless computer modelled or dye tested	While the TSD does not prescribe mixing zones for chronic PNEC values, EPA regulations require each state to adopt mixing zone rules and EPA approve them. The most common general guideline is that the chronic mixing zone should be limited to no more than ¼ (25%) to ½ (50%) of the cross-sectional area and/or volume of flow of the stream, leaving at least ½ to ¾ (75%) free as a zone of passage for aquatic biota, nor should it extend

Guidance	WFD	REACH	US EPA / Canada
	by in the mixing zone, bounded by EQS, (in general) will not be greater than 25%.		over ½ of the width of the stream. Higher allowances are allowed if complete mixing can be demonstrated.
Recommended averaging period to meet chronic PNEC values	Not specified. PNEC values appear to be treated as daily maximum values.	Not specified. PNEC values appear to be treated as daily maximum values.	Recommended to not exceed a 4 day average unless basis for PNEC was based on longer term testing (i.e. 7 or 21 days). Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met.
RIVERS - Acute			
Mixing zone allowed to meet acute PNEC values	Q90 (The flow which is exceeded during 90% of the time. Sometimes also called a 10th percentile flow); and the acute PNEC value must be met at 0.25 the stream width and downstream at the edge of mixing zone at distance of 10 times width of the water body of the discharge. The maximum allowed downstream distance is 1000 meters. (CORMIX modelling is usually used to demonstrate dilution factors)	Modelling (such as CORMIX) can be used to demonstrate dilution factors.	1:1 dilution of effluent assumed. 1Q10 (The lowest day flow that occurs once every 10 years). Zones can be expanded if discharge velocity is >3 m/s, limited to 50 times the discharge length scale and must show that the acute PNEC value is met within a distance of 5 times the local water depth in any horizontal directions. (CORMIX modelling is usually used to demonstrate dilution factors)
Recommended averaging period to meet acute PNEC values.	Not specified. PNEC values are treated as daily maximum values.	Not specified. PNEC values are treated as daily maximum values.	1-day maximum. Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met.

Guidance	WFD	REACH	US EPA / Canada
RIVERS Drinking Water			
Default dilution factor for drinking water PNEC values	No default dilution factor specified.	No default dilution factor specified.	30Q5 for non-carcinogens; and harmonic mean flow for carcinogens.
Site specific dilution analysis to meet drinking water PNEC values	Where drinking water is located in the vicinity of the discharge point, it is important to ensure that these form part of the overall appraisal process when determining the mixing zone. The distance between such features and the discharge point can be of great importance, especially when the distance is less then L or 10 times the width of the water body.	Not specified.	Advanced computer simulations may be allowed to refine loading capacity.
Recommended averaging period to meet drinking water PNEC values	Not Specified.	Not Specified.	1-day maximum. Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met.
LAKES – Chronic	WFD	REACH	US EPA/Canada
Default dilution factor for chronic PNEC values	Where there is no flow or rainfall (the ultimate worst-case scenario) the effluent concentration thus has to meet EQS because in theory the concentration reaches EQS due to lack of dilution by other streams, not taking into account processes such as partition, degradation and evaporation.	Not specified.	Discharges to lakes are not entitled to a default mixing zone. Effluents shall meet chronic PNEC values at the point of discharge.
Site specific dilution	Difficult to identify	Modelling (such as	Ambient mixing is minor

Guidance	WFD	REACH	US EPA / Canada
analysis to meet chronic PNEC values	simple criteria for lakes. One of the most determining factors in this context is the type of initial mixing. Two types of mixing can be identified PLUME-mixing and JET-mixing. Mixing in the near vicinity of the point of discharge (the first few meters) can be described by either jet-mixing or plume-mixing. The mixing pattern with the highest calculated mixing-factor is used to describe the mixing in the first few m from the point of discharge. Use of mixing models can produce dilution factors of 10 or greater.	CORMIX) can be used to demonstrate dilution factors.	for lakes and reservoirs because flow velocity is assumed to be minimal and mixing is accomplished by means of the discharge momentum and buoyancy. While EPA has not nationally set mixing zones for lakes, it has approved default dilution factors of 10:1 or up to 10% of a lake surface area, whichever is less, in many states.
Recommended averaging period to meet chronic PNEC values	Not specified. PNEC values appear to be treated as daily maximum values.	Not specified. PNEC values appear to be treated as daily maximum values.	Recommended to not exceed a 4 day average unless basis for PNEC was based on longer term testing (i.e. 7 or 21 days). Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met. See TSD for more details
LAKES – Acute	WFD	REACH	US EPA/Canada
Default Mixing zone allowed to meet acute PNEC values	No default mixing zones for acute PNEC values.	No default mixing zones for acute PNEC values.	No default mixing zones for acute PNEC values.
Site Specific Acute mixing zones to meet acute PNEC values	Acute mixing zones shall be sized on a case-by-case basis. Computer modelling or dye testing can be used.	Acute mixing zones shall be sized on a case-by-case basis. Computer modelling or dye testing can be used.	Acute mixing zones shall be sized on a case-by-case basis. Computer modelling or dye testing can be used.
Recommended averaging period to	Not specified. PNEC values are treated as	Not specified. PNEC values are treated as daily	1-day maximum. Averaging allowed based

Guidance	WFD	REACH	US EPA / Canada
meet acute PNEC values.	daily maximum values.	maximum values.	on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met. See TSD for more details.
LAKES – Drinking Water	WFD	REACH	US EPA/Canada
Default dilution factor for drinking water PNEC values	No default dilution factor specified.	No default dilution factor specified.	Same as chronic PNEC value default mixing zone (see above).
Site specific dilution analysis to meet drinking water PNEC values	Where drinking water is located in the vicinity of the discharge point, it is important to ensure that these form part of the overall appraisal process when determining the mixing zone. The distance between such features and the discharge point can be of great importance, especially when the distance is less then L or 10 times the width of the water body.	Not specified.	Same as chronic PNEC value site-specific mixing zone (see above). Advanced computer simulations may be allowed to refine loading capacity.
Recommended averaging period to meet drinking water PNEC values	Not Specified.	Not Specified.	1-day maximum. Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met. See TSD for more details.
OCEAN Chronic	WFD	REACH	US EPA/Canada
Default dilution factor for chronic PNEC values	For emissions along the shoreline or open waters, the total length of the mixing zone is L m. Equation is used that leads to a mixing zone positioned between a point L/2 m	A standard dilution of 100 is used when releasing to a marine environment.	No uniform default value from EPA. However, EPA recommends simple single port and multiple port discharge calculations for chronic mixing (i.e. far-field) zones. See Appendix A for

Guidance	WFD	REACH	US EPA / Canada
	downstream and a point L/2 m upstream of the point of discharge. When using a maximum length of 1000 m for the mixing zone this leads to a mixing zone defined as half a circle with a radius of 500 m. For the average depth at the shoreline a value of 5 m is assumed. This results in a maximum volume of the mixing zone: $V_{mixing-zone} = \pi \div 2 \times (500)^2 \times 5 = 1.96 \times 10^6 [m^3]$		calculations and examples.
Ocean dilution used for site specific dilution analysis to meet chronic PNEC values	For emissions along the shoreline or open waters, the total length of the mixing zone is L m. Equation is used that leads to a mixing zone positioned between a point L/2 m downstream and a point L/2 m upstream of the point of discharge. When using a maximum length of 1000 m for the mixing zone this leads to a mixing zone defined as half a circle with a radius of 500 m. For the average depth at the shoreline a value of 5 m is assumed. This results in a maximum volume of the mixing zone: $V_{mixing-zone} = \pi \div 2 \times (500)^2 \times D [m^3]$ D = specific depth	When carrying out a site-specific assessment, specific data on the receiving water may be used with regard to the dilution capacity of the environment (site-specific data should be justified and explained). However, it should be noted that a dilution factor higher than 1000 should not be used in any case.	Computer modelling or dye testing can be used.
Mixing zone size allowed for chronic	Computer modelling or dye testing can be used.	Computer modelling or dye testing can be used.	Computer modelling or dye testing can be used.

Guidance	WFD	REACH	US EPA / Canada
PNEC			
Recommended averaging period to meet chronic PNEC values	Not specified. PNEC values appear to be treated as daily maximum values.	Not specified. PNEC values appear to be treated as daily maximum values.	Recommended to not exceed a 4 day average unless basis for PNEC was based on longer term testing (i.e. 7 or 21 days). Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily maximum is met. See TSD for more details.
OCEAN Acute	WFD	REACH	US EPA/Canada
Mixing zone allowed to meet acute PNEC values	Initial mixing adjacent to the point of discharge jet-mixing or plume-mixing can be estimated. See pages 17-18 in the		The TSD recommends a simplistic screening equation be used to estimate the initial dilution available in the vicinity of a discharge using the following equation: S = 0.3 (x/d) S = flux-averaged dilution x = distance from outlet d = diameter of discharge outlet The equation provides a minimum estimate of mixing because it is based on the assumptions that outlet velocity is zero and the discharge is neutrally buoyant. See TSD for more details.
Recommended averaging period to meet acute PNEC values	Not specified. PNEC values are treated as daily maximum values.	Not specified. PNEC values are treated as daily maximum values.	1-day maximum. Averaging allowed based on long term average statistical calculation guidance for a monthly average that assures 95% confidence that the daily

Guidance	WFD	REACH	US EPA / Canada
			maximum is met. See TSD for more details.

Example Calculations of Allowable Discharges Based on Receiving Water Body and Guidance Followed

Table 7 demonstrates how the size of the receiving water body (e.g. low, medium, high and very high rivers; shallow, medium, and deep oceans) and guidance followed impacts the calculated allowable mass discharges to meet chronic and acute PNEC values.

The following assumptions were made for the comparisons of the three guidances summarized:

- River Example Calculations used an assumed upstream concentration of 0.
- No high rate effluent diffuser
- The chronic PNEC is 1 µg/L and the acute PNEC is 10 µg/L;
- The upstream flow values used were obtained from gaging station on a river in Spain. The average, Q90 and 7Q10 flow values were calculated from an 11 year data set:
  - Average flow = 510,037 m^3/day
  - Q90 flow = 28,944 m^3/day
  - 7Q10 flow = 8,220 m^3/day

For illustration purposes, the Q90 and 7Q10 values were simply progressively increased by factors of 10, 100 and 1,000 to show medium, large and very large dilution ratios.

Ocean Examples

A single discharge port was assumed for 1,000 m^3/day, 10,000 m^3/day and 100,000 m^3/day discharges into a shallow depth (5 meter), a medium depth (30 meter) and a deep depth (60 meter) in the ocean. Effluent discharge velocity at port equals 1 m^3/sec (not a high velocity diffuser – no acute mixing zone).

Table 7 Results for the different Guidances

A. WFD Guidance Results

Water type	Effluent Discharge Volume	Upstream Flow	Effluent Discharge to River Dilution Ratio	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value (100% stream dilution)	Allowable Mass discharge for Chronic PNEC value (25% stream dilution)	Allowable Mass discharge for Acute PNEC value (25% stream dilution)
River	(m^3/day)	(m^3/day)		(µg/L)	(µg/L)	kg/day	kg/day	kg/day

Water type	Effluent Discharge Volume	Upstream Flow	Effluent Discharge to River Dilution Ratio	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value (100% stream dilution)	Allowable Mass discharge for Chronic PNEC value (25% stream dilution)	Allowable Mass discharge for Acute PNEC value (25% stream dilution)
Small River	10,000	28,944	2.8944	1	10	0.04	0.02	0.17
Medium River	10,000	289,440	28.944	1	10	0.30	0.08	0.82
Large River	10,000	2,894,400	289.44	1	10	2.90	0.73	7.34
Very Large	10,000	28,944,000	2894.4	1	10	28.95	7.25	72.46

Water type	Port Depth	Effluent Discharge Volume	Effluent Discharge Velocity	Effluent Discharge to Ocean Dilution Ratio	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value	Allowable Mass discharge for Acute PNEC value
Ocean	m	(m^3/day)	m/sec		(µg/L)	(µg/L)	kg/day	kg/day
Shallow	5	10,000	1.0	196	1	10	1.96	0.10
Medium	25	10,000	1.0	980	1	10	9.80	0.10
Deep	50	10,000	1.0	1,960	1	10	19.60	0.10

B. REACH Guidance Results

Water type	Effluent Dis-charge Volume	Default Dilution of 10:1	Upstream Flow Q90	Effluent Dis-charge to River Dilution Ratio	Chro-nic PNEC	Acute PNEC	Allowable Mass discharge for chronic PNEC value (default stream dilution)	Allowable Mass discharge for chronic PNEC value (100% stream dilution)	Allowable Mass discharge for acute PNEC value (no stream dilution)
River	(m^3/day)	(m^3/day)	(m^3/day)		(µg/L)	(µg/L)	kg/day	kg/day	kg/day
Small River	10,000	100,000	28,944	2.8944	1	10	0.35	0.04	0.10
Medium River	10,000	100,000	289,440	28.944	1	10	0.35	0.30	0.10
Large River	10,000	100,000	2,894,400	289.44	1	10	0.35	2.90	0.10
Very Large River	10,000	100,000	28,944,000	2894.4	1	10	0.35	28.95	0.10

Water type	Port Depth	Effluent Discharge Volume	Effluent Discharge Velocity	Default Mixing Dilution Ratio	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value	Allowable Mass discharge for Acute PNEC value
Ocean	m	(m^3/day)	m/sec		(µg/L)	(µg/L)	kg/day	kg/day
Shallow	5	10,000	1	100:1	1	10	1.00	0.10
Medium	25	10,000	1	100:1	1	10	1.00	0.10
Deep	50	10,000	1	!00:1	1	10	1.00	0.10

C. US EPA/Canada Guidance Results

Water type	Effluent Discharge Volume	7Q10 Upstream Flow	Effluent Discharge to River Dilution Ration	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value (50% stream dilution)	Allowable Mass discharge for Acute PNEC value (1:1 dilution)
River	(m^3/day)	(m^3/day)		(µg/L)	(µg/L)	kg/day	kg/day
Small River	10,000	8,220	0.822	1	10	0.01	0.20
Medium River	10,000	82,200	8.220	1	10	0.05	0.20
Large River	10,000	822,000	82.200	1	10	0.42	0.20
Very Large	10,000	8,220,000	822.000	1	10	4.12	0.20

Water type	Port Depth	Effluent Discharge Volume	Effluent Discharge Velocity	Effluent Discharge to Ocean Dilution Ratio	Chronic PNEC	Acute PNEC	Allowable Mass discharge for Chronic PNEC value	Allowable Mass discharge for Acute PNEC value
Ocean	m	(m^3/day)	m/sec		(µg/L)	(µg/L)	kg/day	kg/day
Shallow	5	10,000	1	13.6	1	10	0.14	0.10
Medium	25	10,000	1	23.1	1	10	0.23	0.10
Deep	50	10,000	1	61.9	1	10	0.62	0.10

From the results in Table 7A, Table 7B and Table 7C, the following observations can be seen:

- The WFD methods for evaluating chronic and acute toxicity are the least restrictive, especially at higher dilution volumes.
- The ECHA default for chronic mixing will not be protective in situations where there is less than 10 to 1 mixing.
- The ECHA and EPA methods for application of ocean mixing zones for chronic toxicity are more closely aligned than the WFD method. This is because the WFD method relies on a method based a volume of dilution in 500 m radius of the discharge port.

Models such as e.g., the U.S. PhATE or EU GREAT-ER river models may be used to revise the crude PEC values, and to explore the spatial and temporal variability to better understand the risks to humans and biota, to evaluate risk mitigation and management options. References that used the PhATE and GREAT-ER models to refine pharmaceutical risk assessments are amongst others Anderson et al., 2004 [40] and Caldwell et al., 2019 [41].

11 Appendix A4: Examples for external guidance documents for risk characterization

The following (external) guidance documents can be consulted in the performance of risk characterizations:

REACH guidance	European Chemicals Agency (2016): Guidance on information requirements and Chemical Safety Assessment Chapter R.16: Environmental exposure assessment Version 3.0 February 2016. https://echa.europa.eu/documents/10162/13632/information_requirements_r16_en.pdf [37]
EMA guidance (human drugs)	EMA ERA Guideline, EMEA/CHMP/SWP/4447/00 corr 2. https://www.ema.europa.eu/en/documents/scientific-guideline/guideline-environmental-risk-assessment-medicinal-products-human-use-first-version_en.pdf [9] EMA ERA Guideline, EMEA/CHMP/SWP/4447/00 Rev. 1. https://www.ema.europa.eu/en/documents/scientific-guideline/draft-guideline-environmental-risk-assessment-medicinal-products-human-use-revision-1_en.pdf [10]
EMA guidance (veterinary drugs)	EMA VICH Topic GL6. CVMP/VICH/592/98-Final. https://www.ema.europa.eu/en/documents/scientific-guideline/vich-gl6-environmental-impact-assessment-eias-veterinary-medicinal-products-phase-i-step-7_en.pdf [42] EMA VICH GL38. CVMP/VICH/790/03-Final. https://www.ema.europa.eu/en/documents/scientific-guideline/vich-gl38-environmental-impact-assessments-veterinary-medicinal-products-vmps-phase-ii_en.pdf [43]
FDA guidance	US FDA Guidance for Industry Environmental Assessment of Human Drug and Biologics Applications. https://www.fda.gov/media/70809/download [8]
US EPA	The US EPA published a series of Risk Assessment Guidelines for human health and the environment (https://www.epa.gov/risk/risk-assessment-guidelines). The conduct of an ERA from planning, problem formulation to analysis and risk characterization is described (https://www.epa.gov/risk/conducting-ecological-risk-assessment). A Risk Characterization Handbook was published in 2000 [44].
Australia	Environmental risk assessment guidance manual for agricultural and veterinary chemicals. http://www.nepc.gov.au/resource/chemical-risk-assessment-guidance-manuals [45]
Japan	Various ERA guidance documents have been published in the course of the Chemical Substances Control Act

	(https://www.env.go.jp/en/chemi/cs_control_act.html; e.g. Methods for the Risk Assessment of Priority Assessment Chemical Substances [46].
Korea (case study)	Environmental Risk Assessment of Pharmaceuticals: Model Application for Estimating Pharmaceutical Exposures in the Han River Basin [47]
EU project	DANTES: Demonstrate and Assess New Tools for Environmental Sustainability. Methods and Tools for Assessment of Environmental Risk. https://dantes.info/Publications/Publication-doc/An%20overview%20of%20ERA%20-methods%20and%20tools.pdf
ECETOC	The organization ECETOC provides a collaborative space for scientists from industry, academia and governments to develop and promote practical, trusted and sustainable solutions to scientific challenges which are valuable to industry, as well as to the regulatory community and society in general. In the course of these activities many Technical Reports to various subjects in the area of ERA were published (http://www.ecetoc.org/publications/technical-reports/).