

WEBINAIRE

29 - 30 septembre 2022

SOLUTIONS BASÉES SUR LA NATURE – NBS – POUR LE TRAITEMENT ET LA RÉUTILISATION DES EAUX USÉES ET DES BOUES D'ÉPURATION EN MÉDITERRANÉE

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Constructed wetlands systems and emerging contaminants



Pollutants

<i>Conventional pollutants</i>	SS, BOD, COD, COT, ammonia, nitrates, nitrites, total N, TKN, organic N, phosphorus, bacteria, viruses
<i>Non conventional pollutants</i>	recalcitrant organic substances, VOCs, surfactants, heavy metals, total dissolved solids
<i>Emerging contaminants (Ecs) Contaminants of emerging concern (CECs)</i>	??? Known or unknown???

- **ECs** are in general *unregulated* compounds, which may be candidate for future regulation depending on research on their potential health effects and monitoring data regarding their occurrence.

A few words to clarify...

- **Contaminants** are defined as inputs of alien and potentially toxic substances into the environment; not all contaminants cause pollution, as their concentrations may be too low.
- **'Pollutants'** are defined as anthropogenically-introduced substances that have harmful effects on the environment.

Sometimes the distinction between *contaminants* and *pollutants* is not simple:

- concentrations at which contaminants become pollutants cannot always be defined;
- long-term damage to organisms or systems may occur that is not evident initially.

In the following we will assume that all the micropollutants may pose negative effects to the environment and thus

microcontaminants = micropollutants

What is «emergent»?



Emerging Pollutants in
Water and Wastewater



What is an
emerging
contaminant?

in a broad sense any **synthetic or naturally-occurring chemical** or any **microorganism** that is **not commonly monitored or regulated** in the environment **with potentially known or suspected adverse ecological and human health effects**



Their names...

Chemicals found in pharmaceuticals, personal care products, pesticides, industrial and household products, metals, surfactants, industrial additives and solvents.

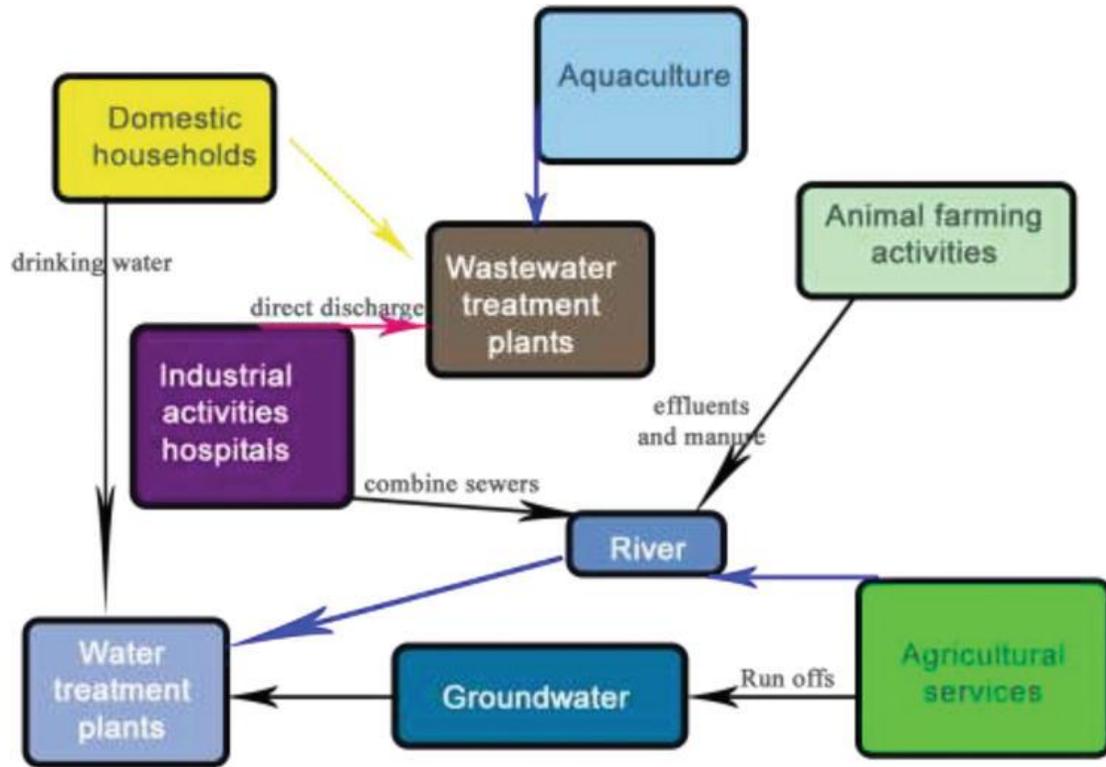


Their quantities...

Many of them are used and released continuously into the environment even in very low quantities and some may cause chronic toxicity, endocrine disruption in humans and aquatic wildlife and the development of bacterial pathogen resistance

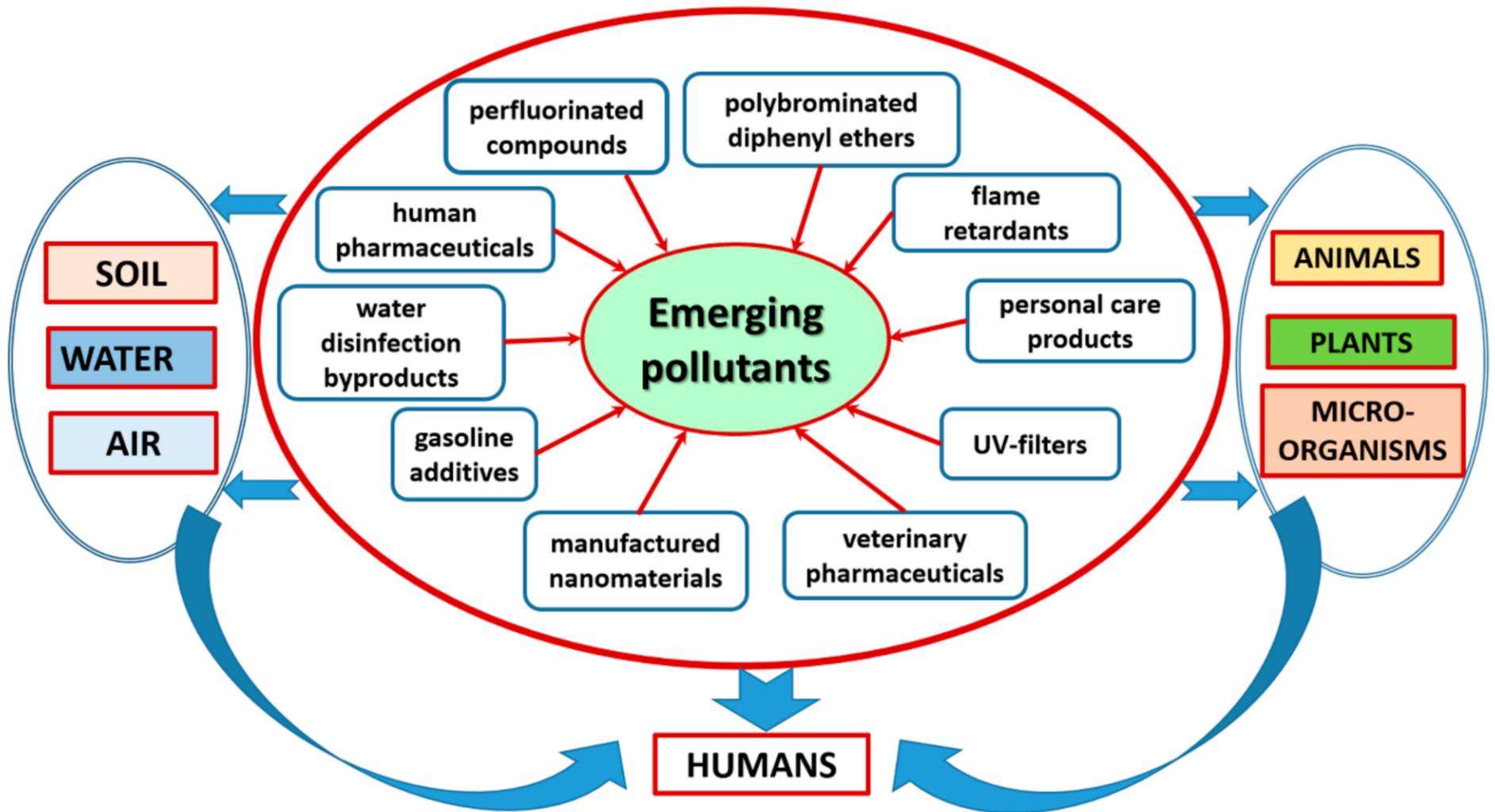


Emerging contaminants: Sources, Pathways, Receptors

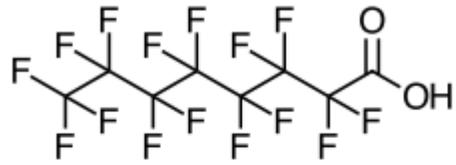


Rasheed et al., 2019
<https://doi.org/10.1016/j.envint.2018.11.038>

Common classes of emerging contaminants



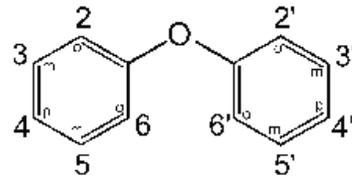
Examples of emerging contaminants and their structure



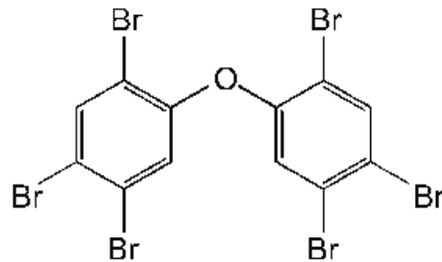
Perfluorooctanoic acid (PFOA)



Perfluorooctane sulfonate (PFOS)



Generalized structure of PBDE



2,2',4,4',5,5'-Hexabromodiphenyl ether (PBDE-153)

Flame retardants



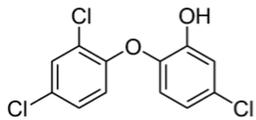
(a)

(b)

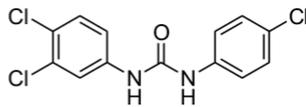
(c)

- (a) Pentabromo chloro cyclohexane
- (b) Tetrabromo phthalic anhydride
- (c) Hexabromo benzene

Personal care products

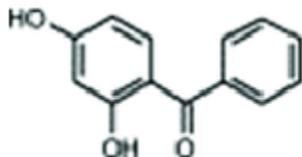


Triclosan



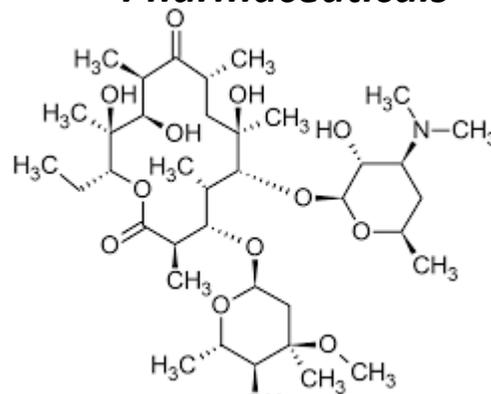
Triclocarban

UV filter



benzophenone 1

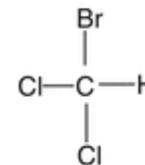
Pharmaceuticals



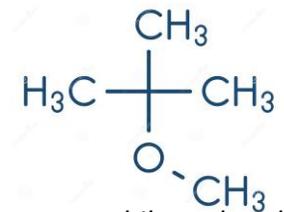
Erythromycin

DBPs

Bromodichloro-
methane



Gasoline additives



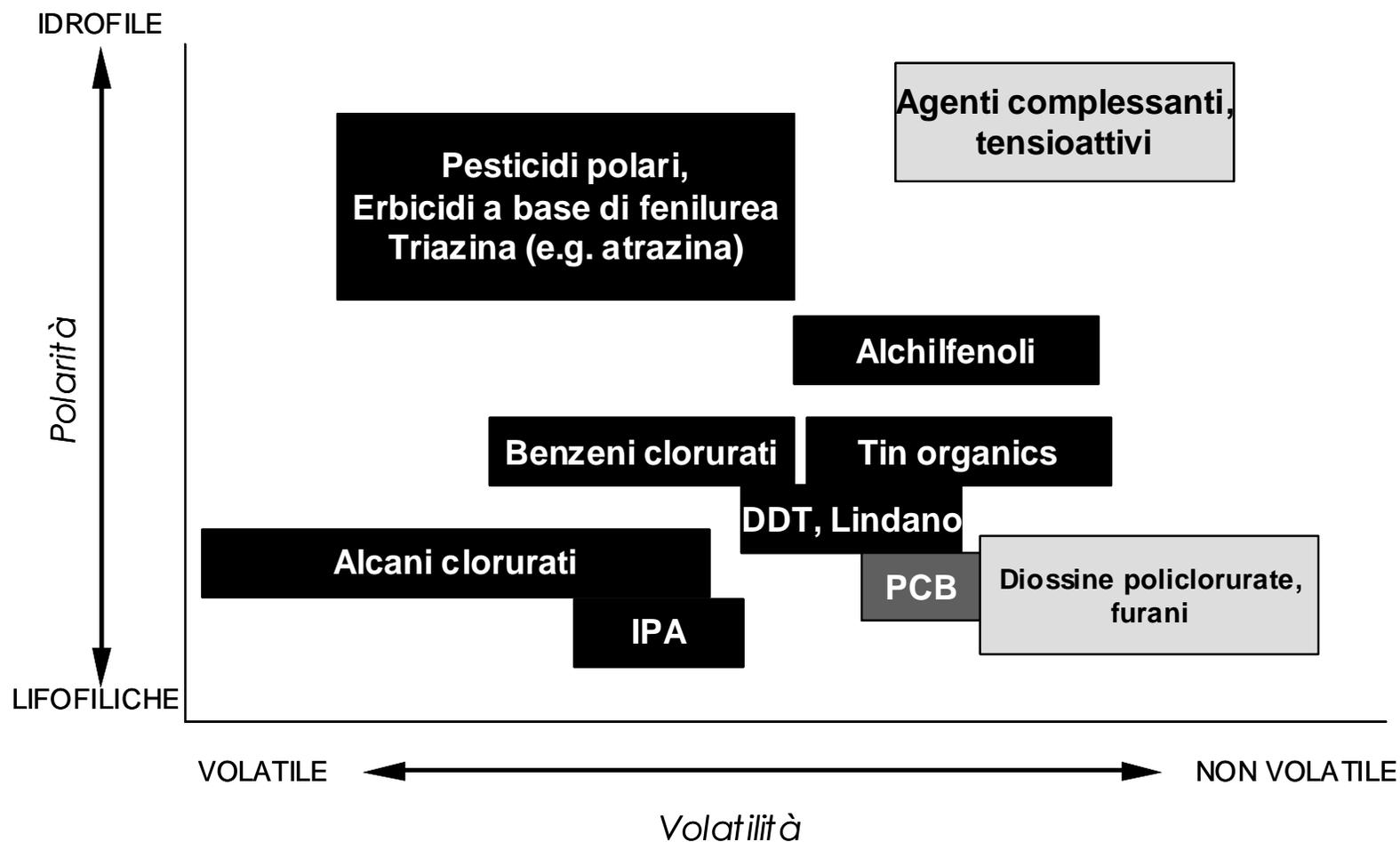
methyl tert-butyl ether

Recently added among the emerging contaminants

- **Microplastics** (particles whose size is in the range 0,1 - 5.000 μm) and **nanoplastiche** (particles whose size is in the range 0,001 a 0,1 μm)
- **Antibiotic resistant bacteria**, ARB
- **Antibiotic resistant genes**, ARG

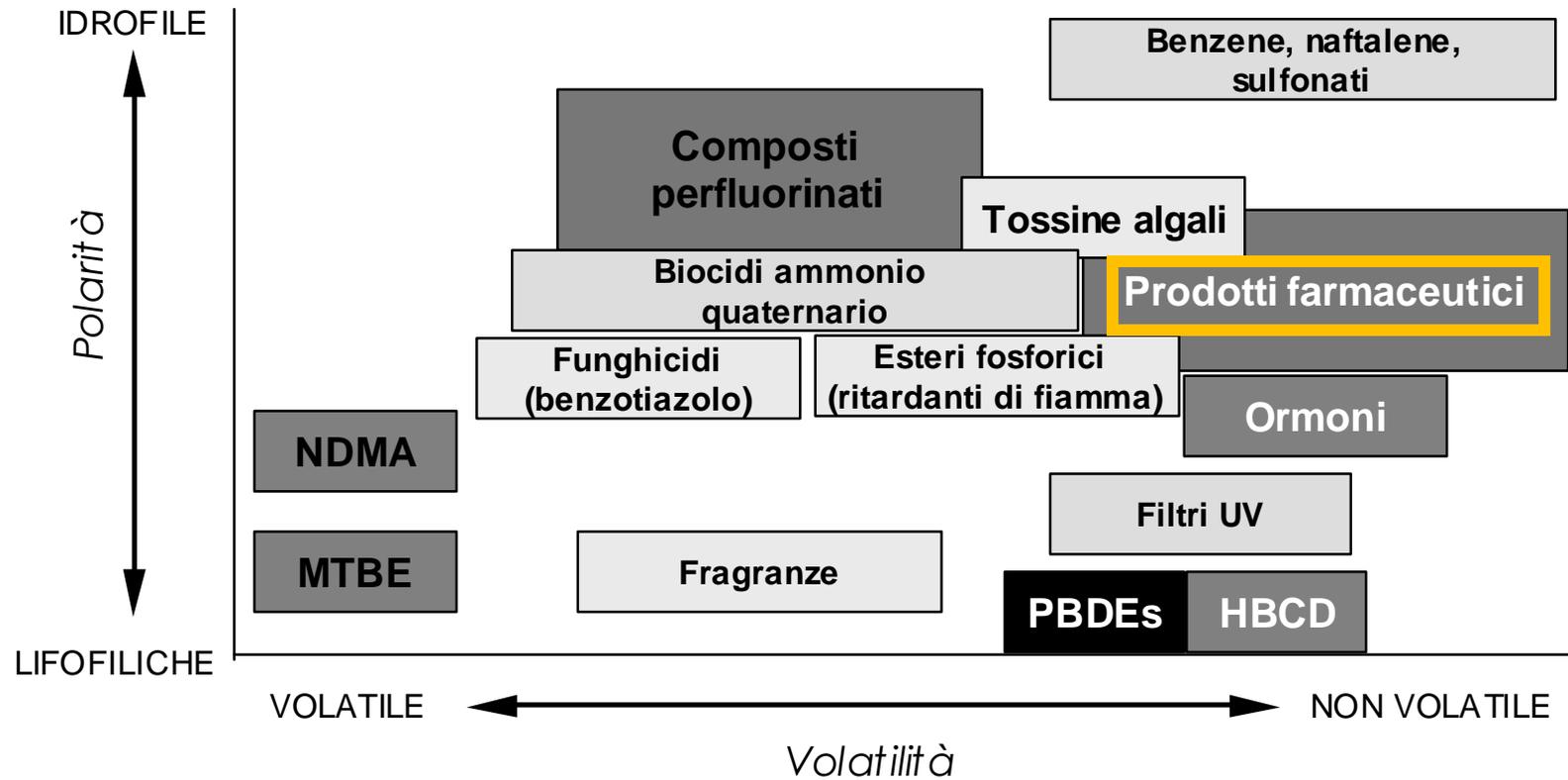
**Most studied in the water compartment:
Pharmaceutical compounds
(still unregulated)**

Regulated micropollutants at EU level



Others emerging contaminants

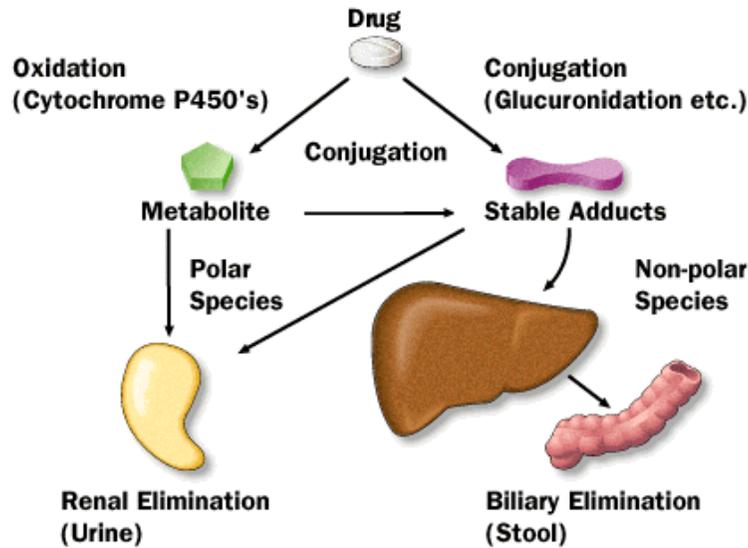
Ternes and Joss, 2006



Focus on PHARMACEUTICAL compounds

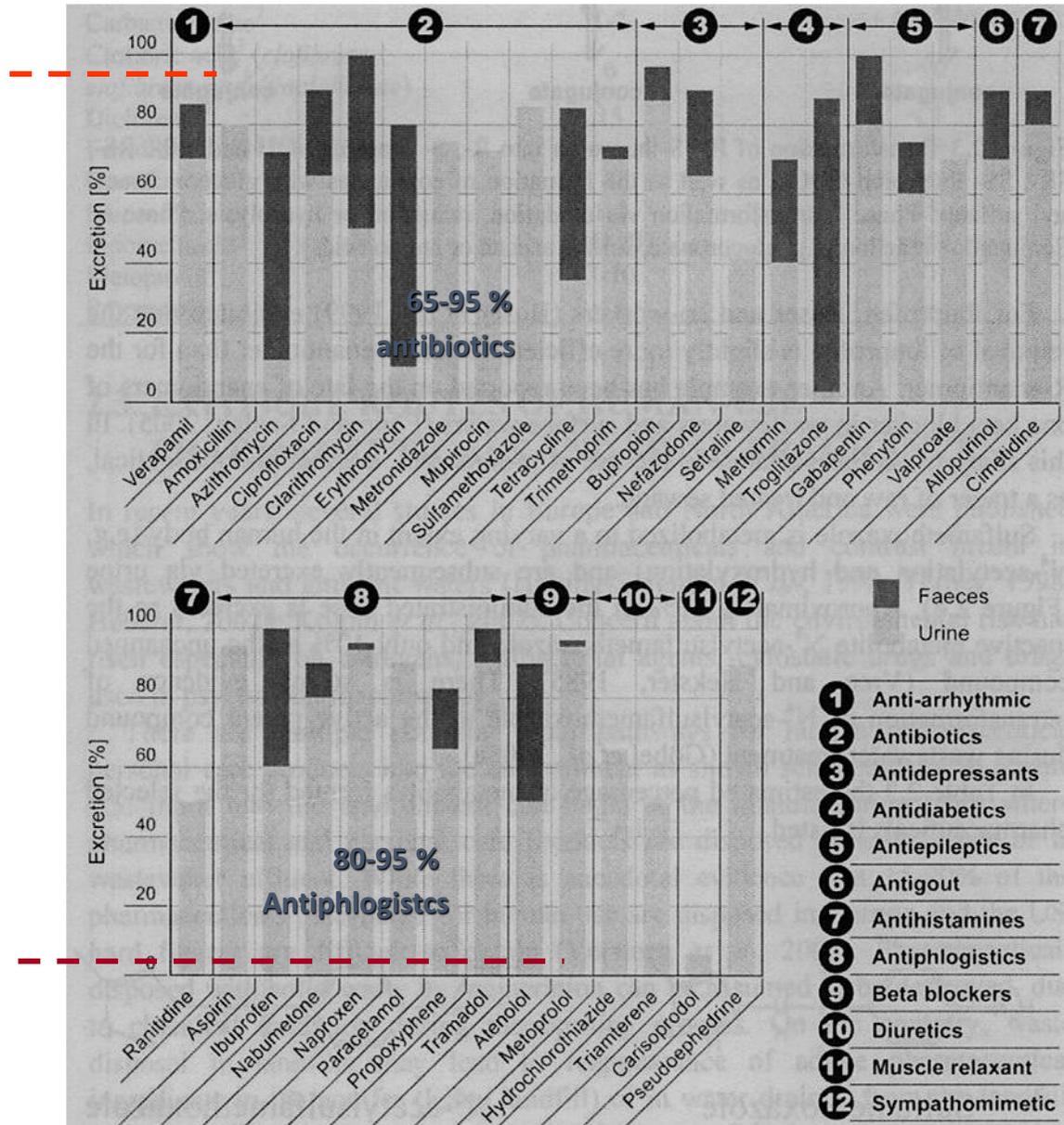


Pharmaceutical administration → Excretion



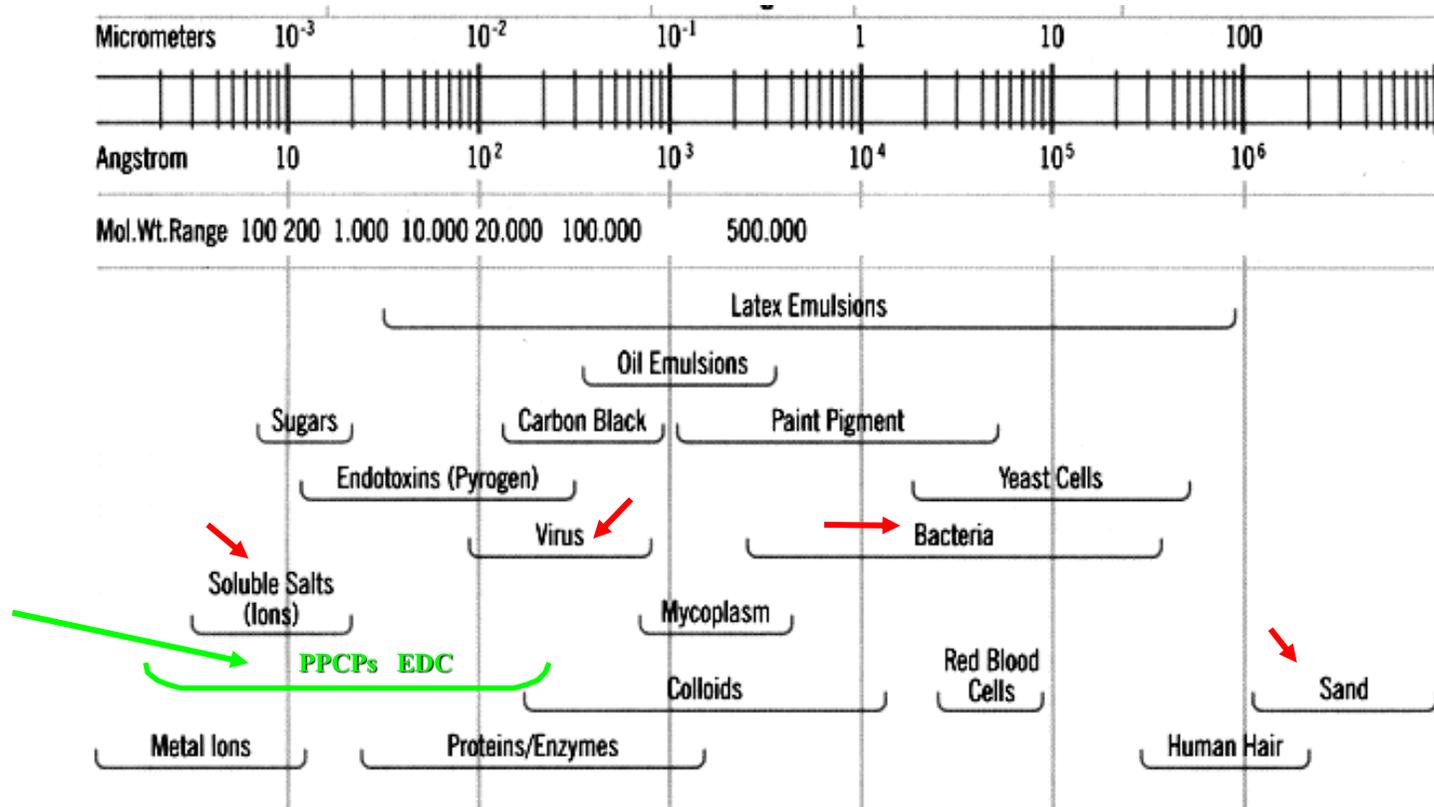
% excretion: 2-95%

Ternes and Joss, 2006



Pharmaceutical compounds differ for....

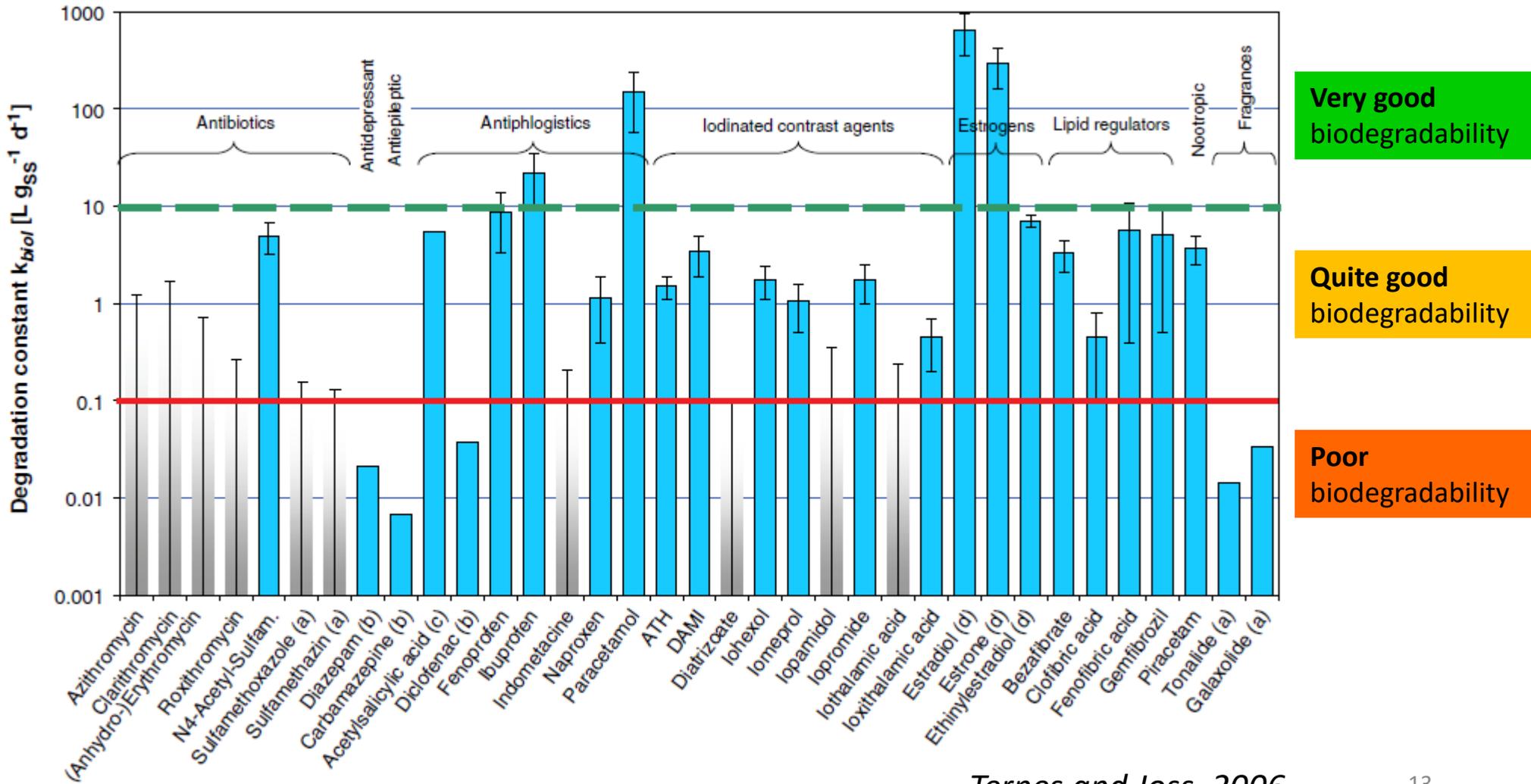
- Dimension and molecular weight
- Percentage of excretion
- Biodegradability
- Tendency to adsorb onto a solid phase
- Volatility
- Photodegradability, persistence in the environment, stability



Biodegradability

$$\frac{dc_A}{dt} = -k_{biol} c_A^n$$

c_A = concentration of compound A, t = time, k_{biol} = degradation constant, n = reaction order



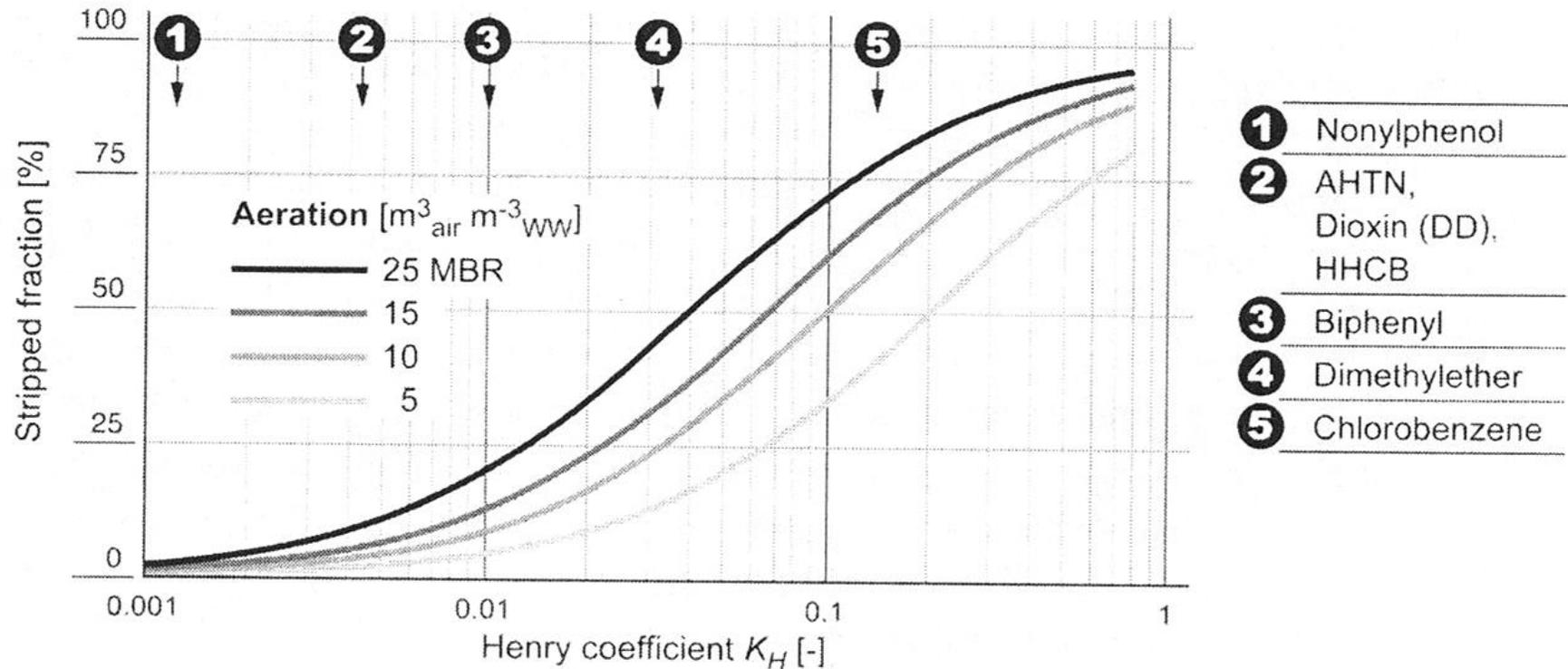
Sorption onto a solid phase (sludge, activated carbons, particles...)

K_{ow} = water octanol partition, k_d sorption coefficient

$$\text{Log } k_d = 1.14 + 0.58 \text{ Log } K_{ow}$$

Analytes	Use	MW (g/mol)	Log K_{ow}	
Gemfibrozil	Anti-cholesterol	250.2	4.77	Excellent adsorption
Triclosan	Antibiotic	289.6	4.76	
Estradiol	Steroid	272.2	4.01	
Ibuprofen	Pain reliever	206.1	3.97	
Progesterone	Steroid	314.2	3.87	Good adsorption
Oxybenzone	Sunscreen	228.1	3.79	
Ethinylestradiol	Birth control	296.2	3.67	
Testosterone	Steroid	288.2	3.32	
Naproxen	Analgesic	230.1	3.18	
Estrone	Steroid	270.4	3.13	
Erythromycin-H ₂ O	Antibiotic	733.9	3.06	
Diazepam	Anti-anxiety	284.8	2.82	
Androstenedione	Steroid	286.2	2.75	
Atrazine	Herbicide	215.1	2.61	
Dilantin	Anti-convulsant	252.3	2.47	
Carbamazepine	Analgesic	236.3	2.45	
Estriol	Steroid	288.4	2.45	
DEET	Insect repellent	191.3	2.18	Low adsorption
TCEP	Fire retardant	285.5	1.44	
Trimethoprim	Antibiotic	290.1	0.91	
Sulfamethoxazole	Antibiotic	253.1	0.89	
Diclofenac	Arthritis	318.1	0.70	
Meprobamate	Anti-anxiety	218.3	0.70	
Acetaminophen	Analgesic	151.2	0.46	
Pentoxifylline	Blood viscosity control	278.1	0.29	
Caffeine	Stimulant	194.2	-0.07	
Iopromide	X-ray contrast media	790.9	-2.1	

Stripping/volatilization



CONTRIBUTION TO REMOVAL DUE TO SOLAR EXPOSURE

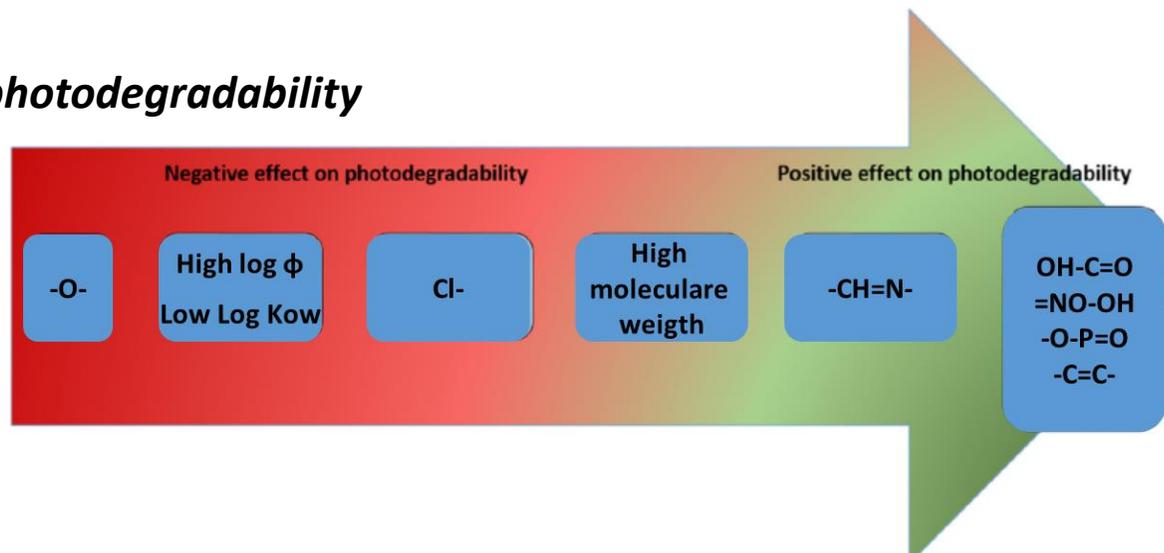
According to Mathon et al. 2016

CECs are classified into three classes:

- FAST-PHOTODEGRADABLE (half-life < 8h)
- MEDIUM-PHOTODEGRADABLE (8h < half-life < 168h = 7 days)
- SLOW-PHOTODEGRADABLE (half-life > 168h)

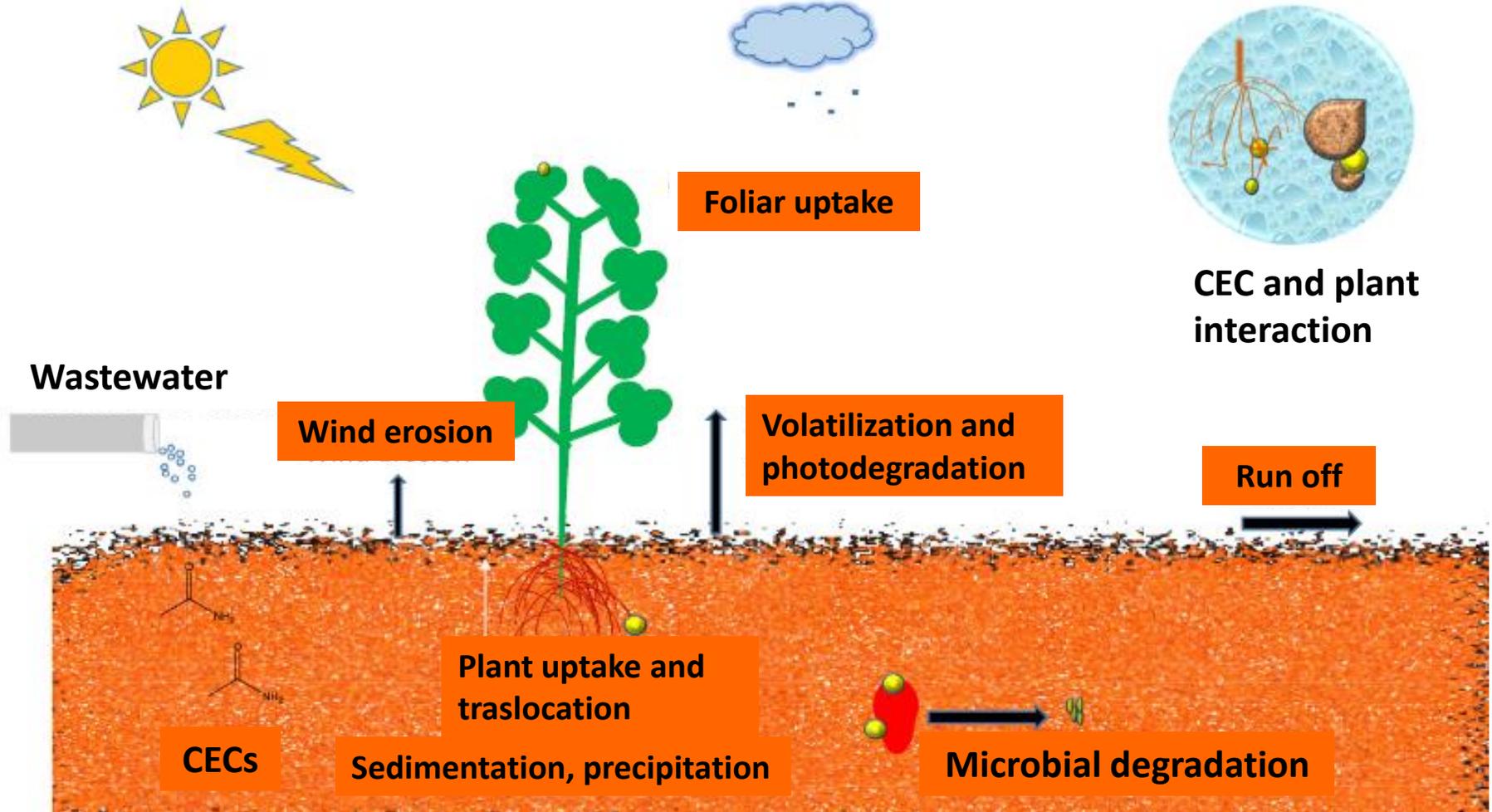
Influence of the functional groups on the photodegradability

Effect on photodegradability



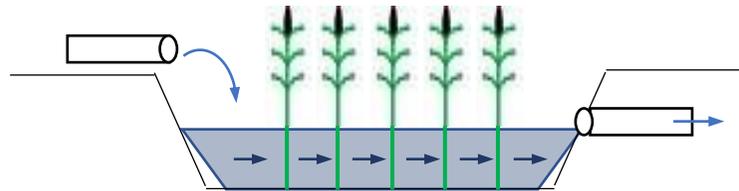
How can we apply these considerations to CW systems?

To understand/predict EC pathways in the bed or basin

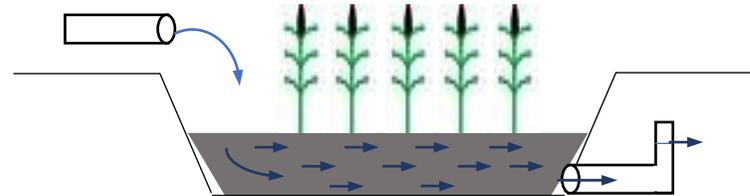


Different types of Constructed wetlands

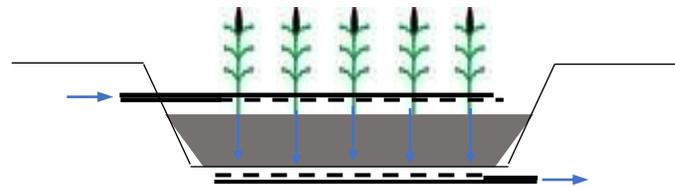
Surface flow system



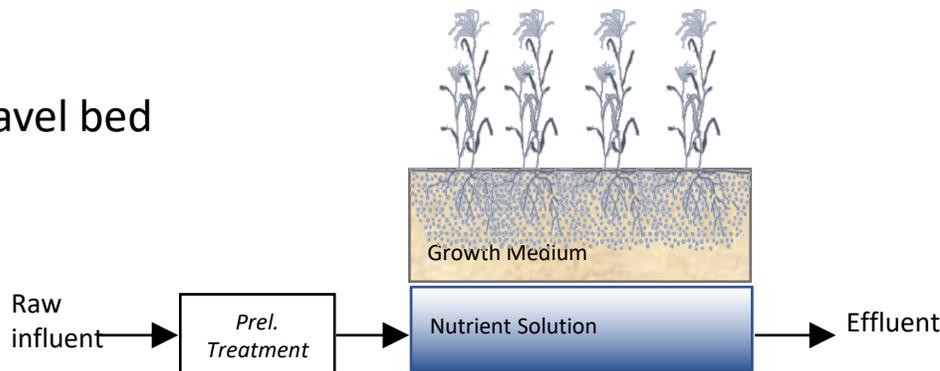
Horizontal subsurface flow system



Vertical subsurface flow system



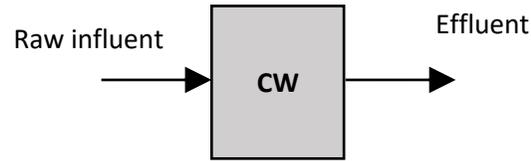
Hydroponic gravel bed



Different operational conditions...
How do they affect the
removal mechanisms?

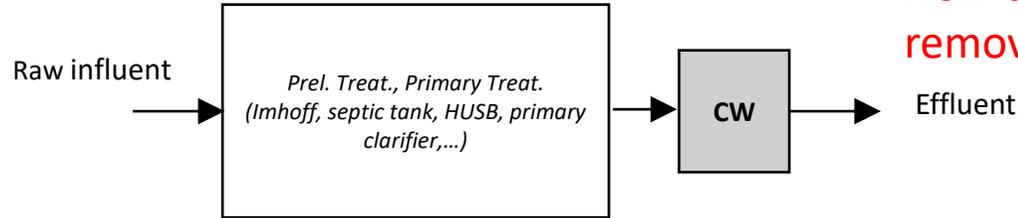
Different *role* in the Treatment = different STEP

Primary step



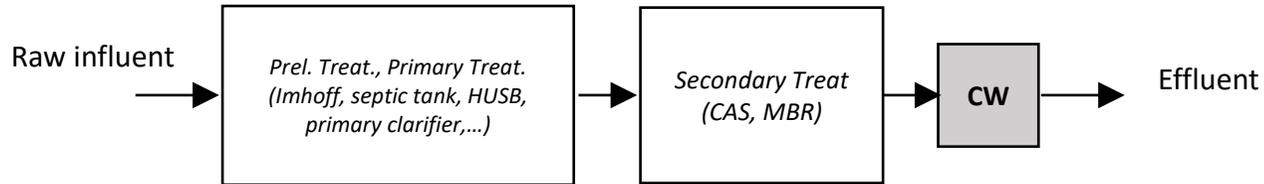
Different influent concentrations
Different matrix effects..

Secondary step

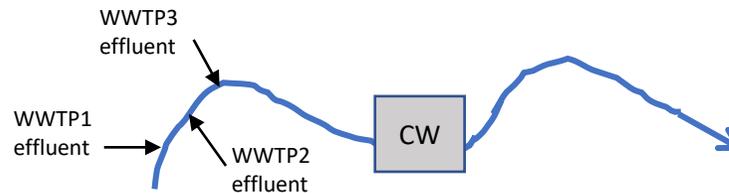


How do they affect the
removal mechanisms?

Tertiary step

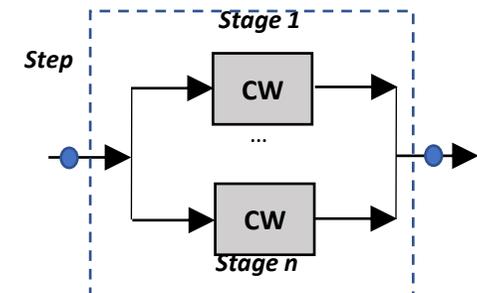
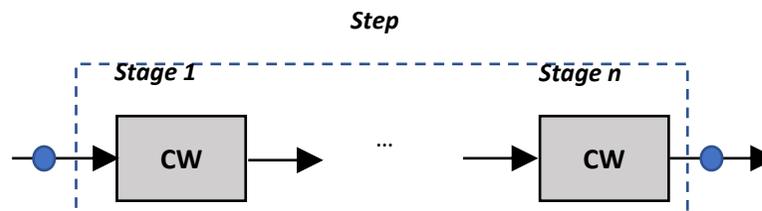


Restoration Wetland



Multistage steps

● Sampling point

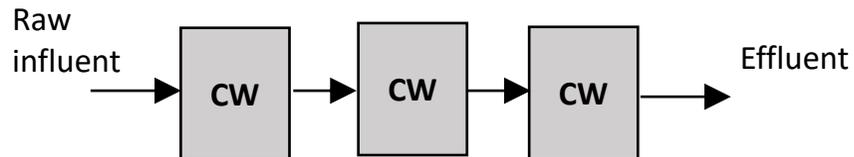
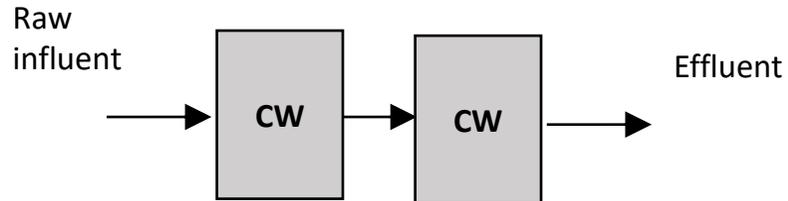
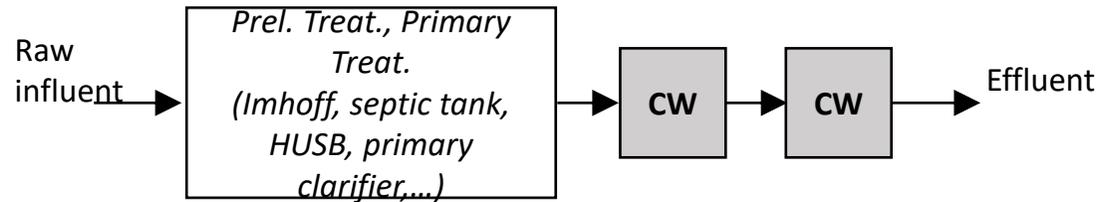


Treatment stages

Different influent concentrations
Different matrix effects..

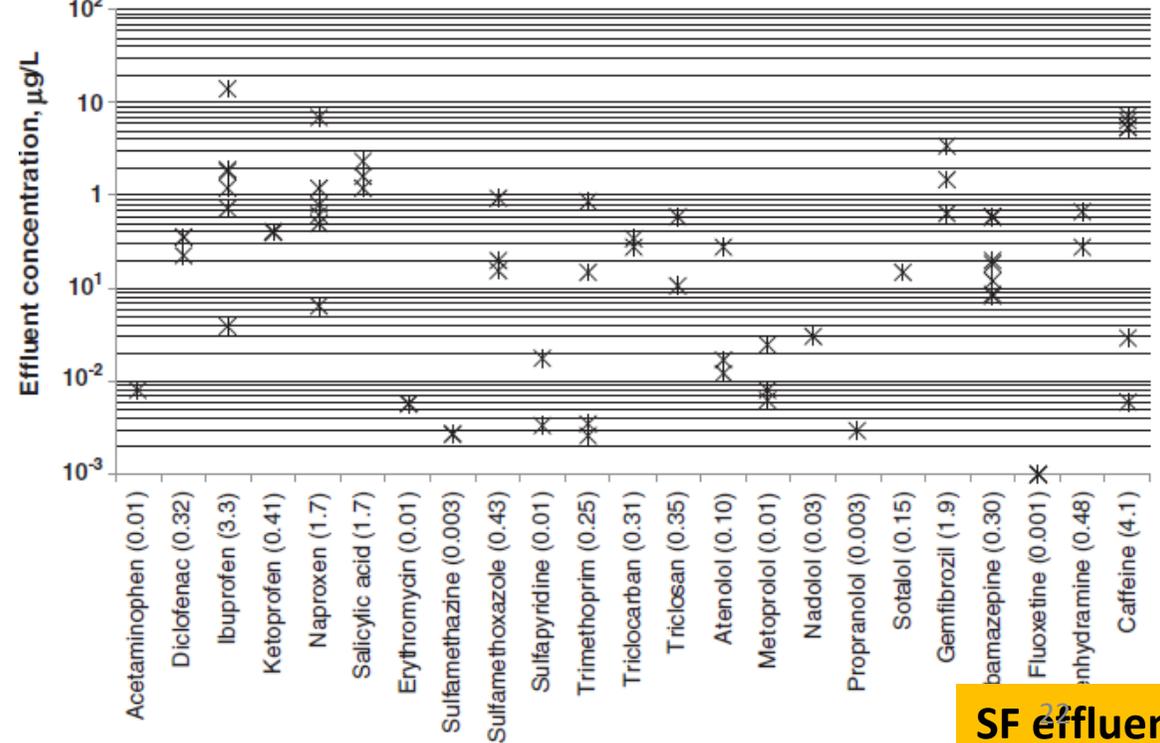
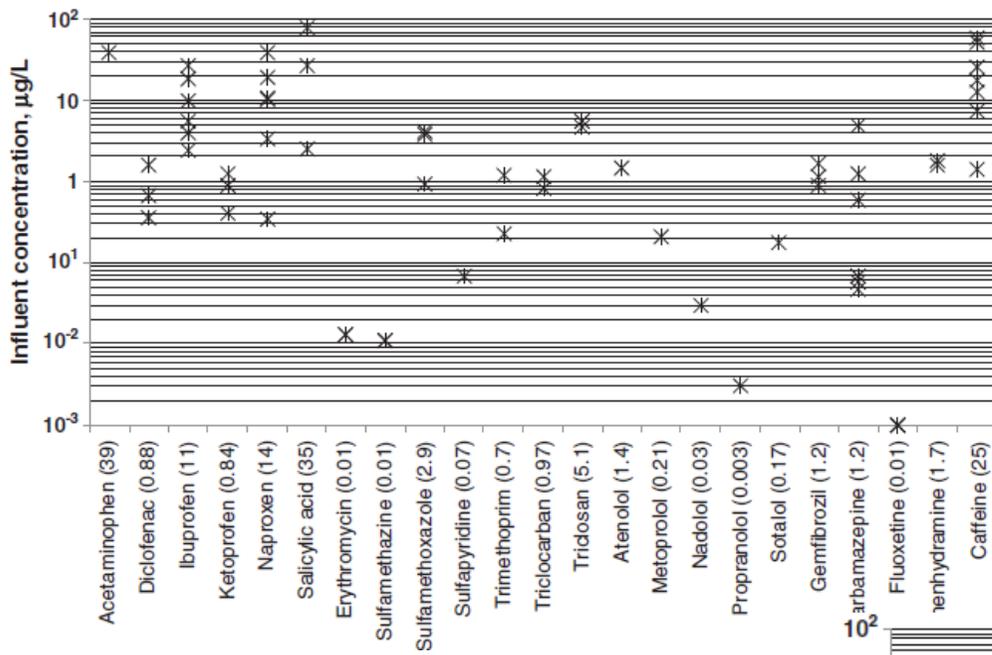
Hybrid **systems**

How do they affect the
removal mechanisms?



Lessons learned from the literature

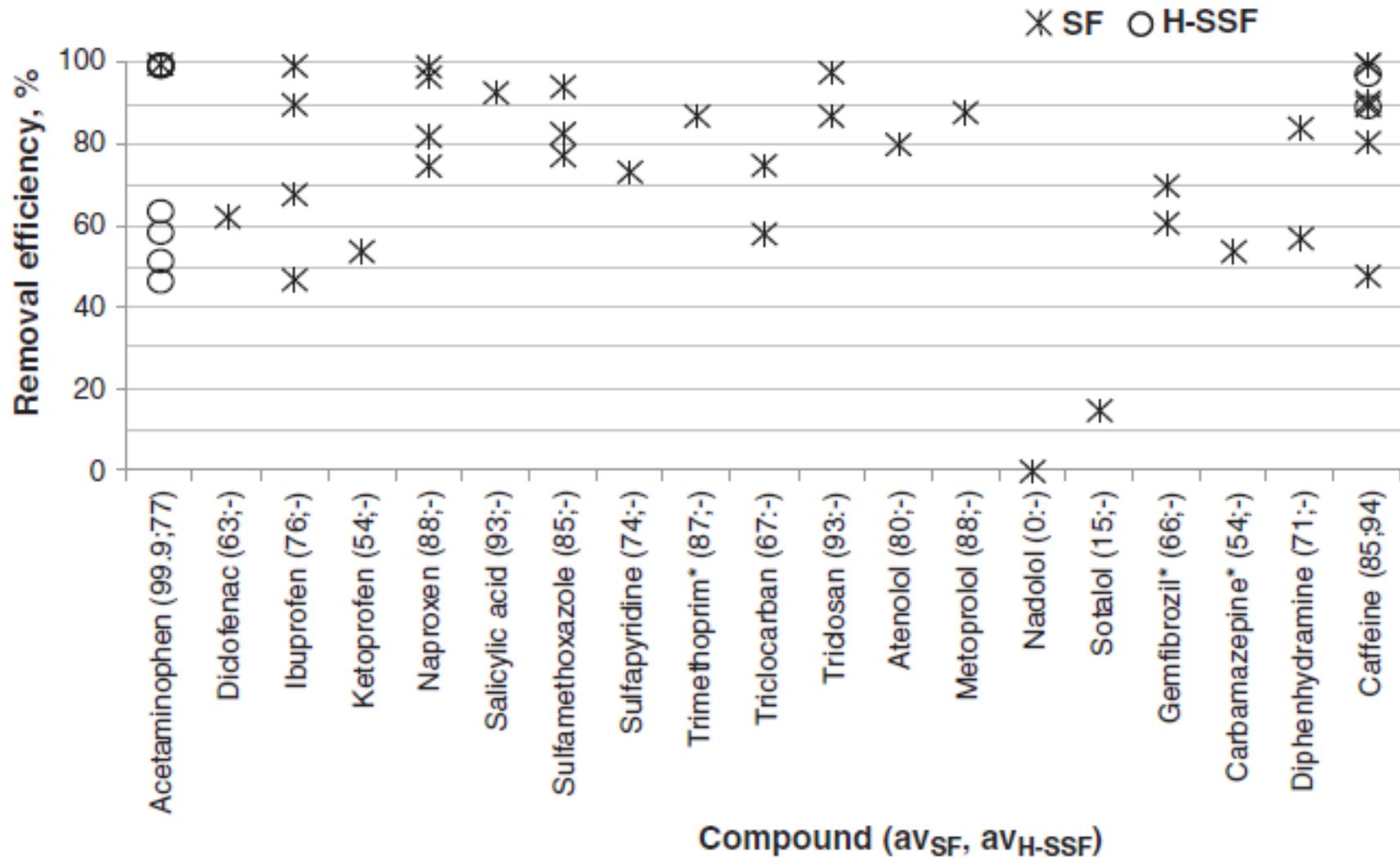
CW acting as primary step:
influent and effluent



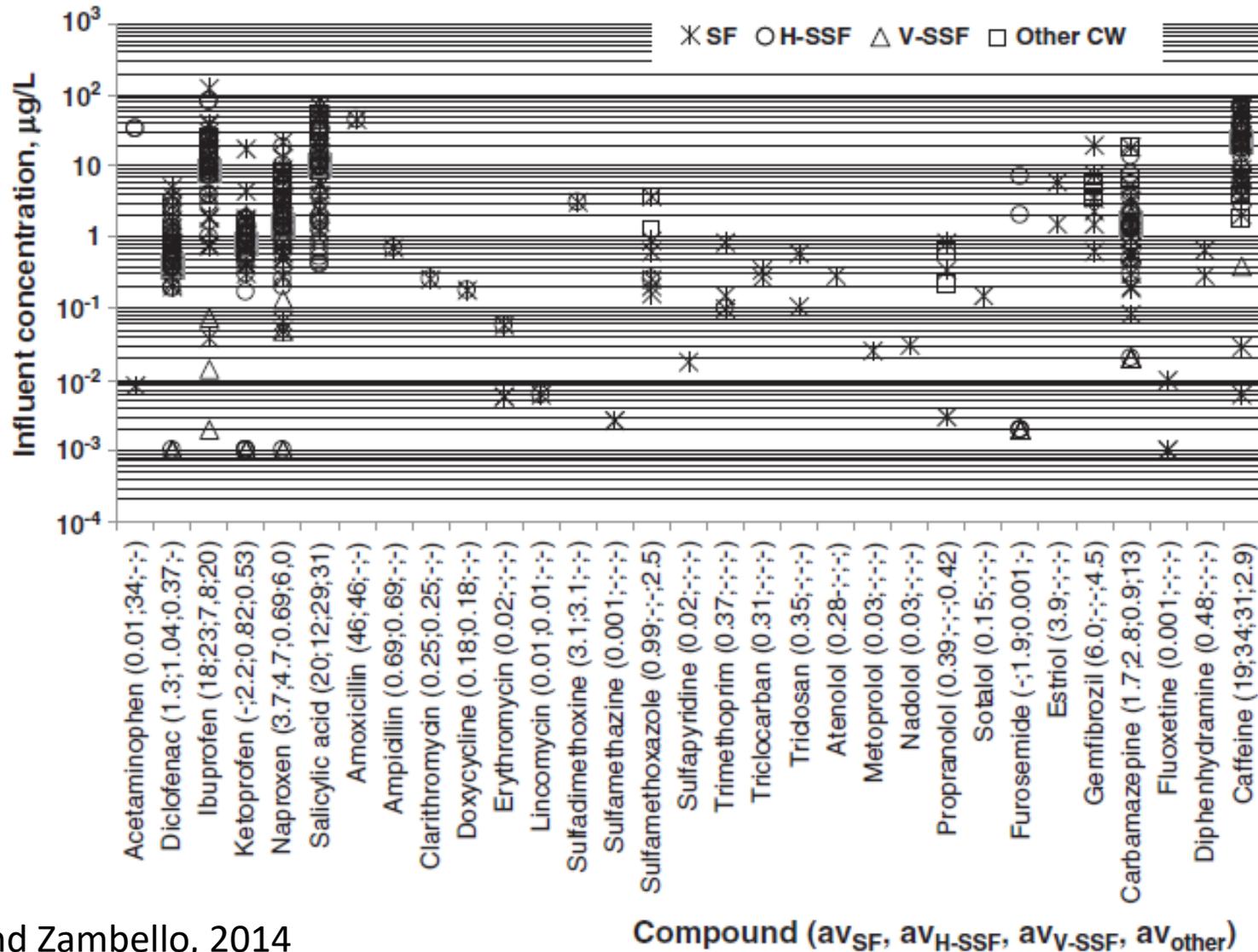
Verlicchi and Zambello, 2014

SF effluent

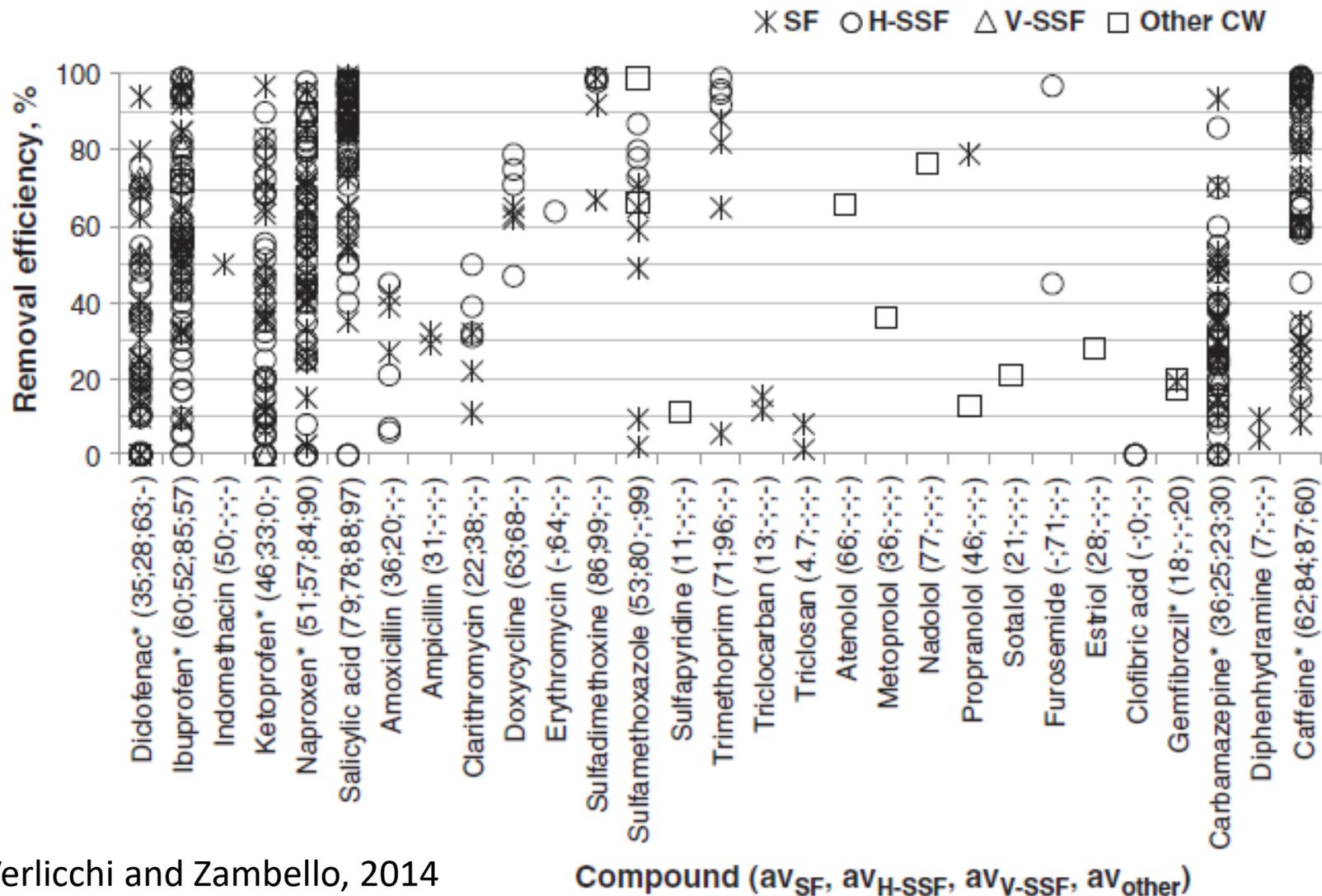
CW acting as a primary step: average removal efficiencies



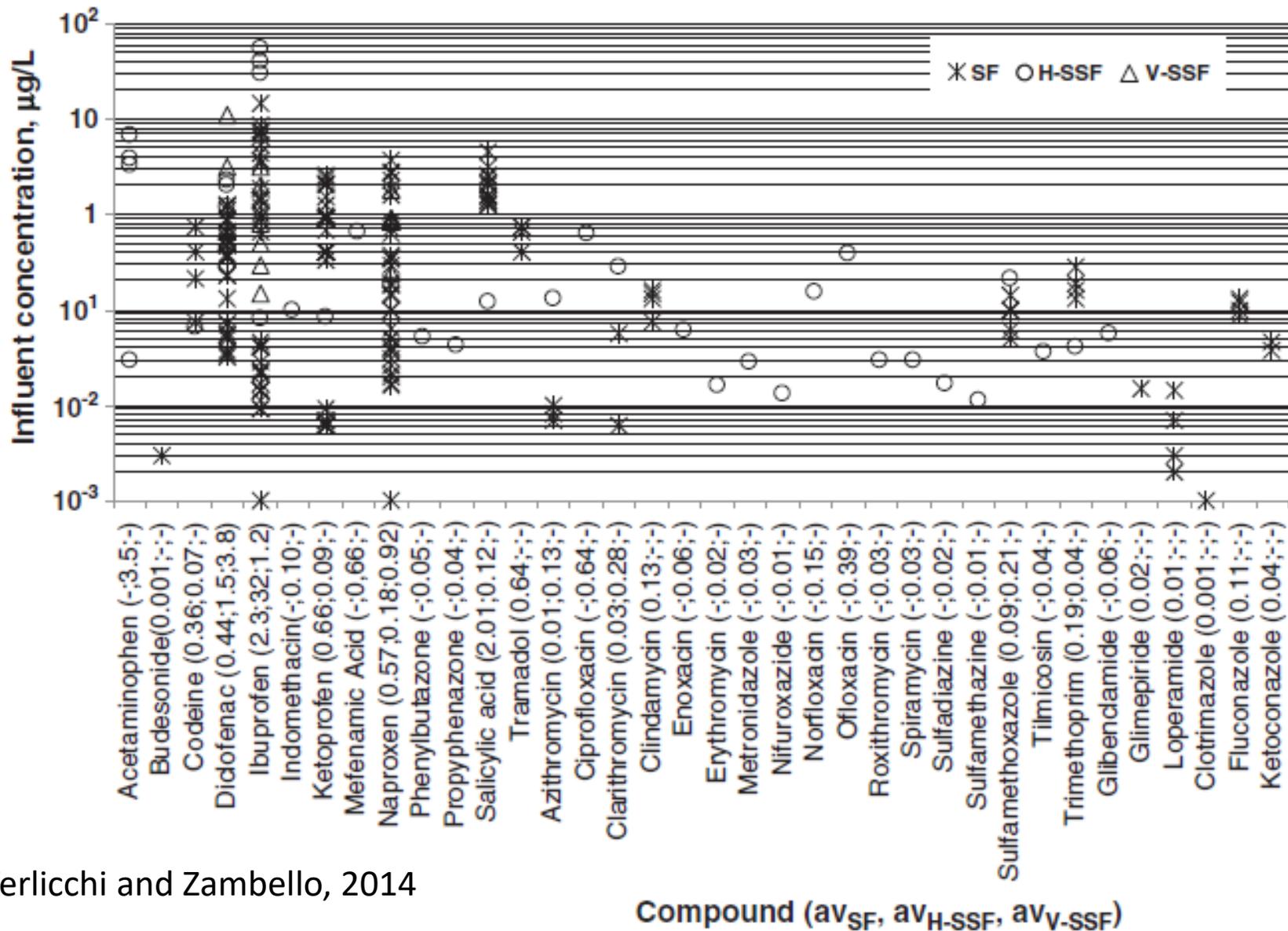
CW acting as a secondary step: **influent concentrations**



CW acting as a secondary step: removal efficiencies

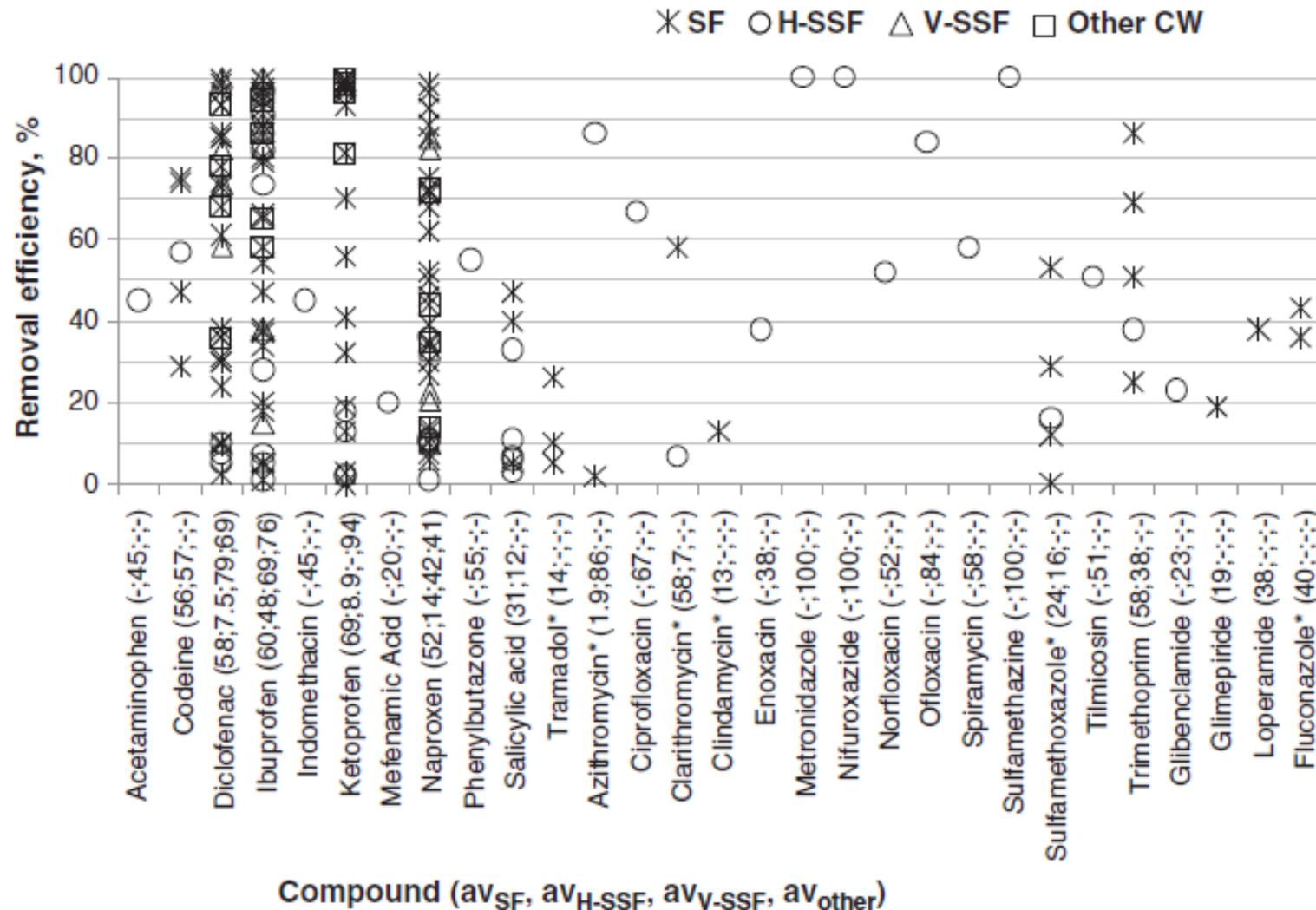


CW acting as a tertiary step: influent concentrations

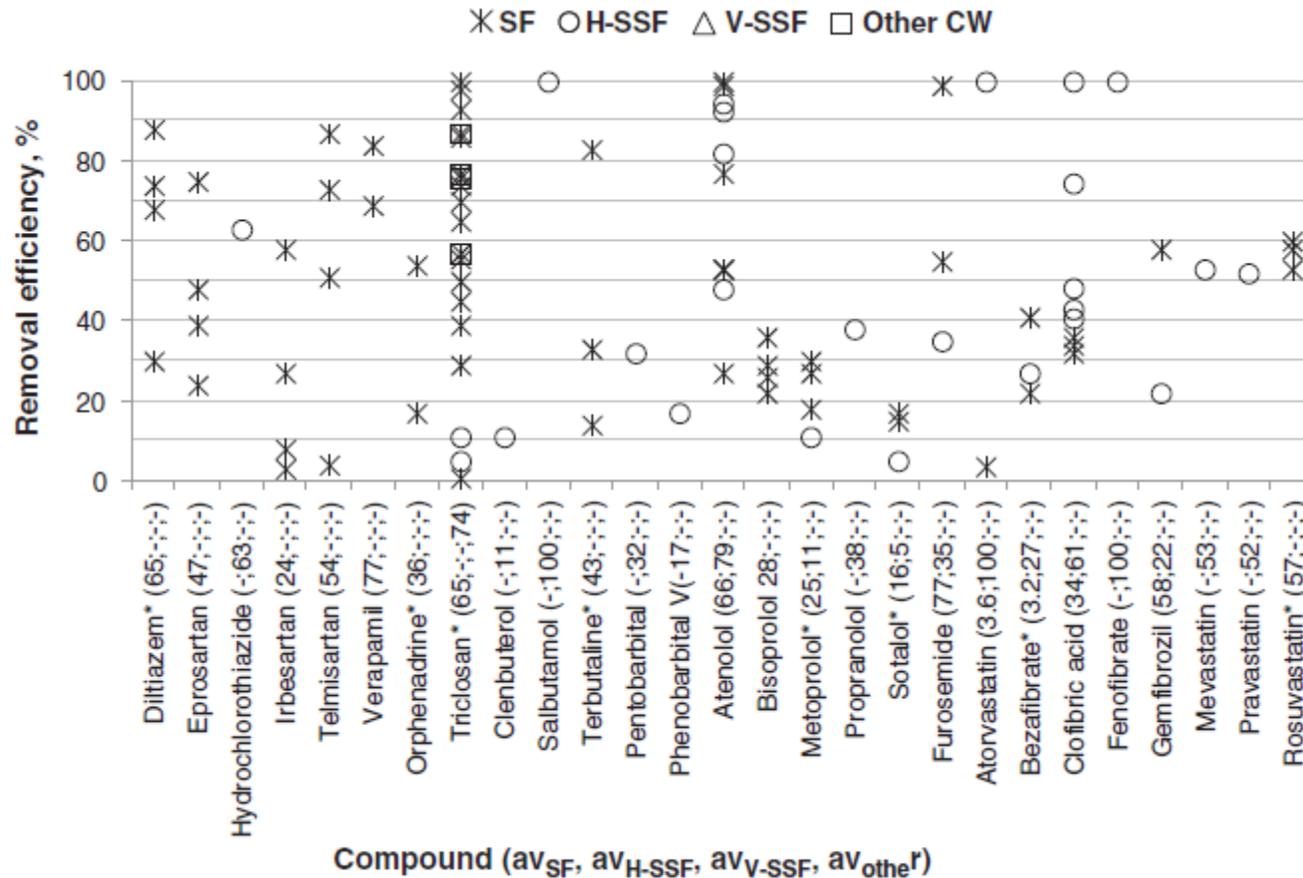


Verlicchi and Zambello, 2014

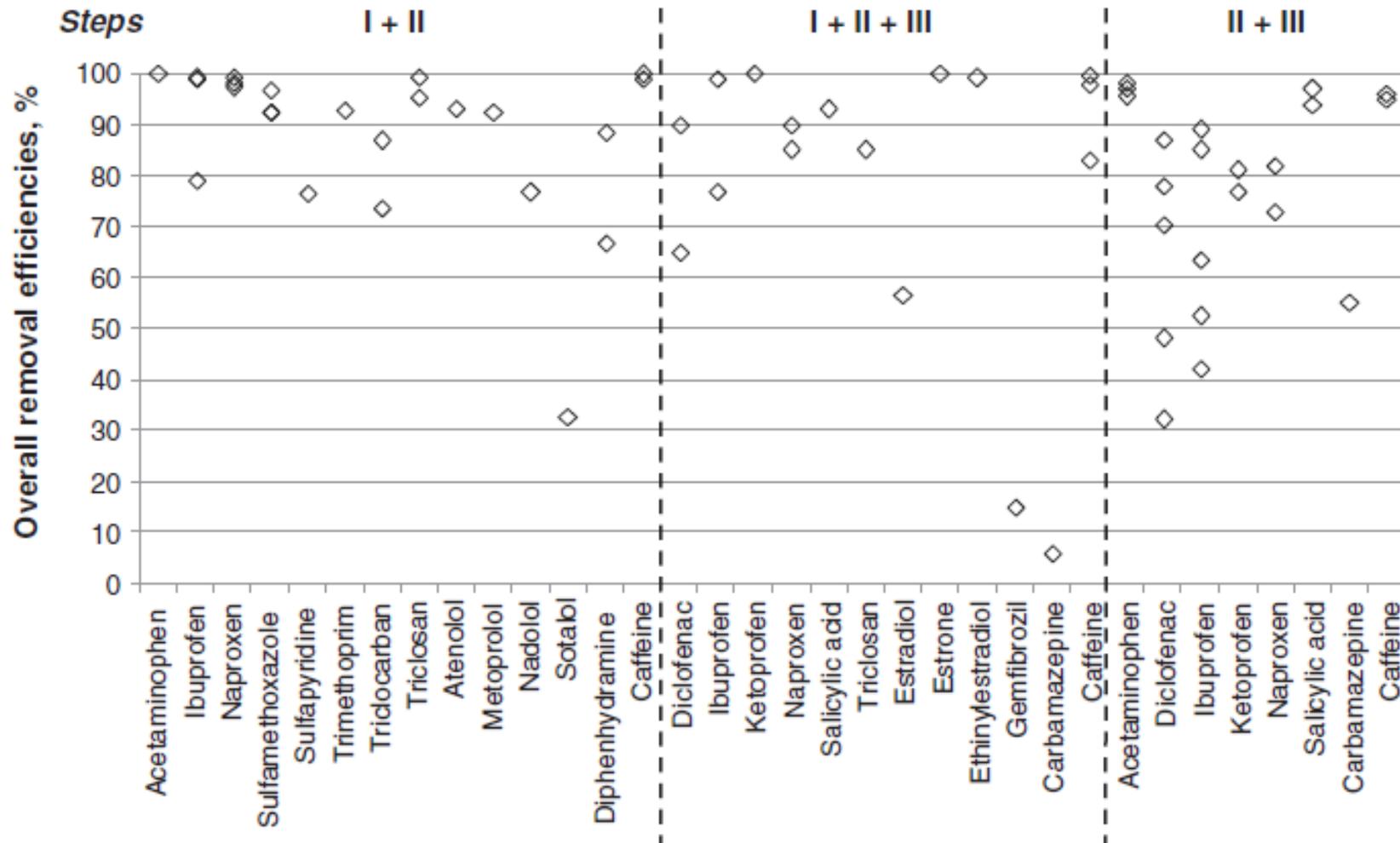
CW acting as a tertiary step: removal efficiencies



CW acting as a tertiary step: removal efficiencies



Hybrid CWs: removal efficiencies



Verlicchi and Zambello, 2014

HSSF acting as a polishing step

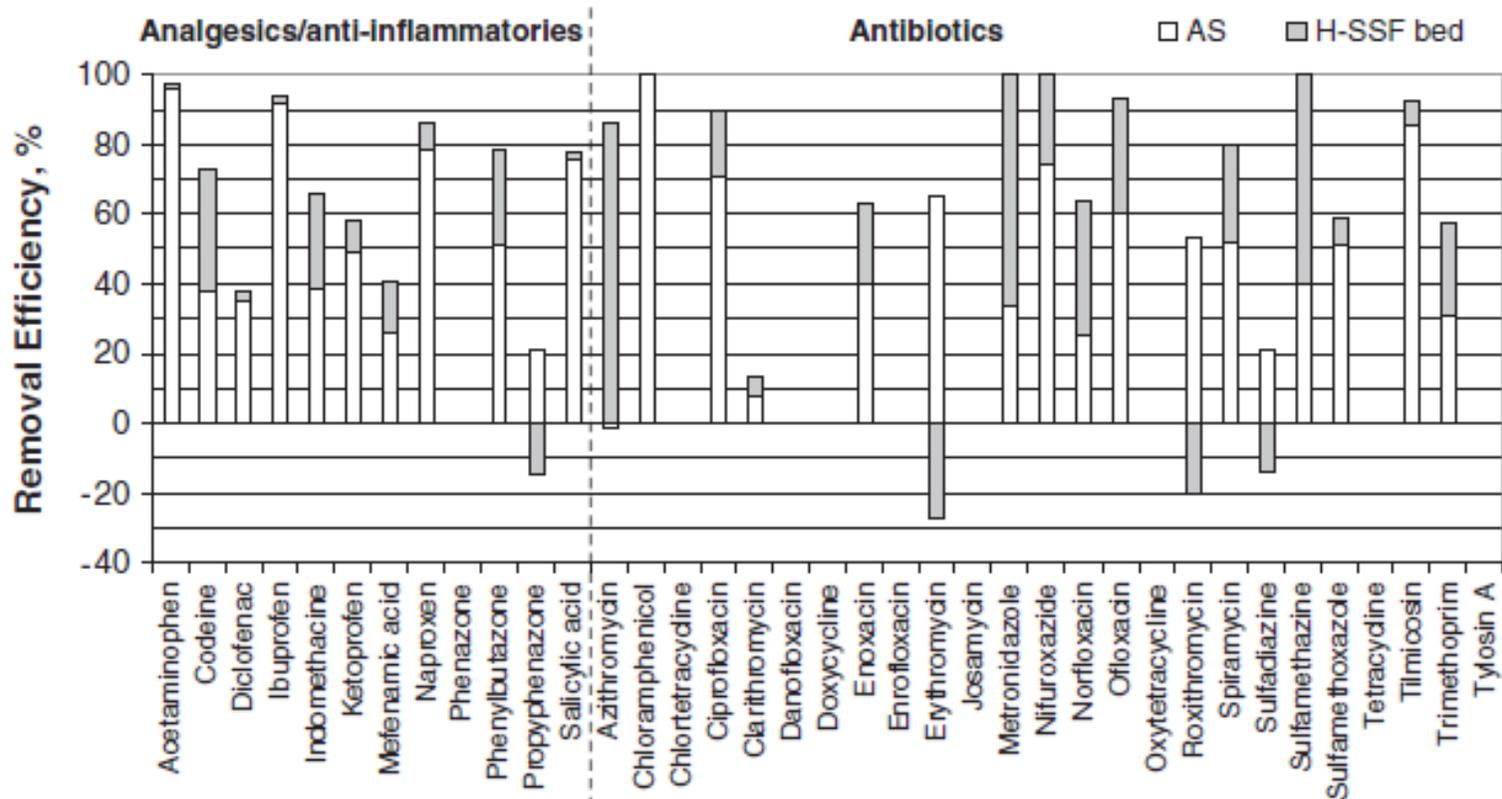
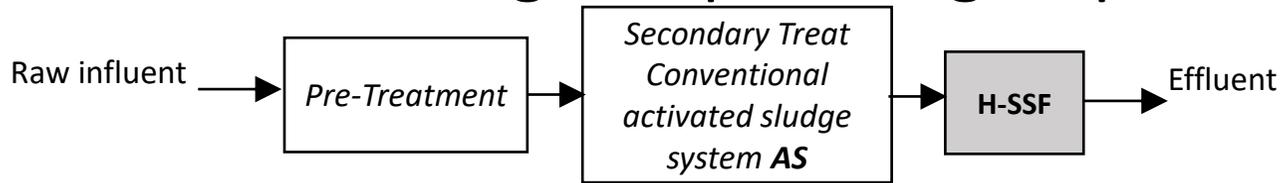
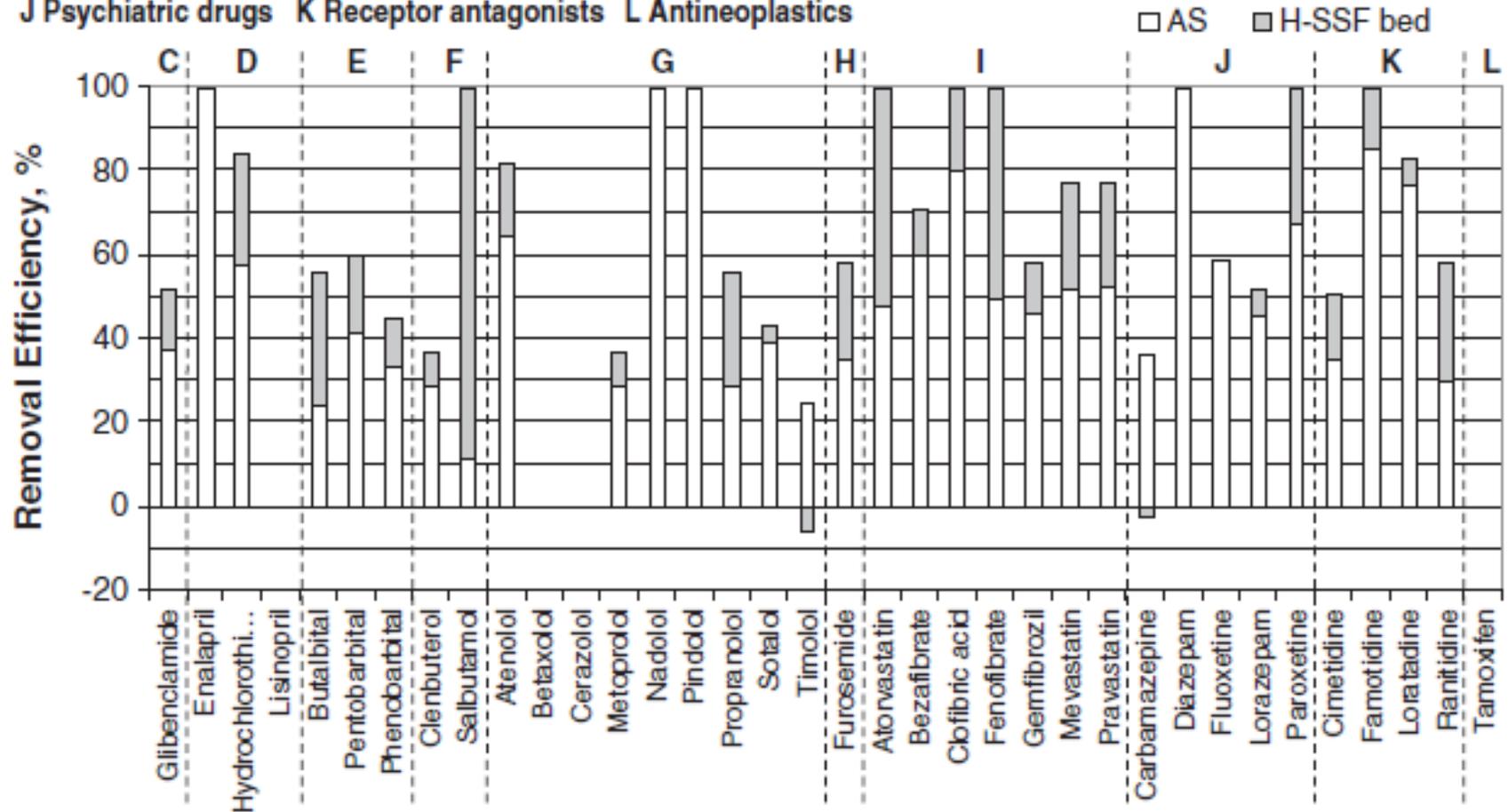


Fig. 10. Global average removal efficiencies for the selected analgesics/anti-inflammatories and antibiotics and relative contributions of each step.

HSSF acting as a polishing step

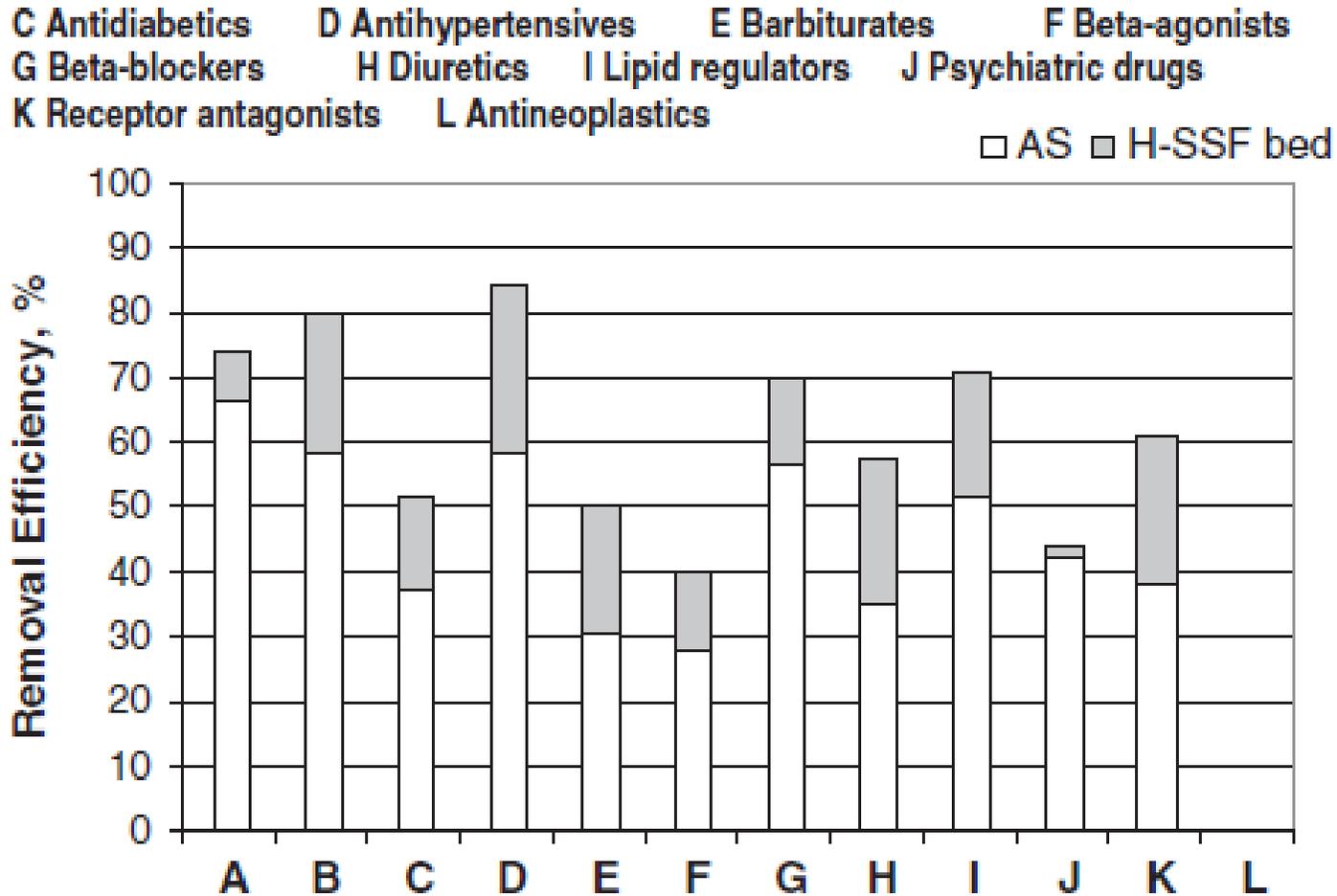
C Antidiabetics D Antihypertensives E Barbiturates F Beta-agonists G Beta-blockers H Diuretics I Lipid regulators
 J Psychiatric drugs K Receptor antagonists L Antineoplastics



AS= conventional activated sludge

Verlicchi et al., 2013

HSSF acting as a polishing step



AS= conventional activated sludge

Verlicchi et al., 2013

Warning: Micropollutants occurrence: high variability over time (day, week...)

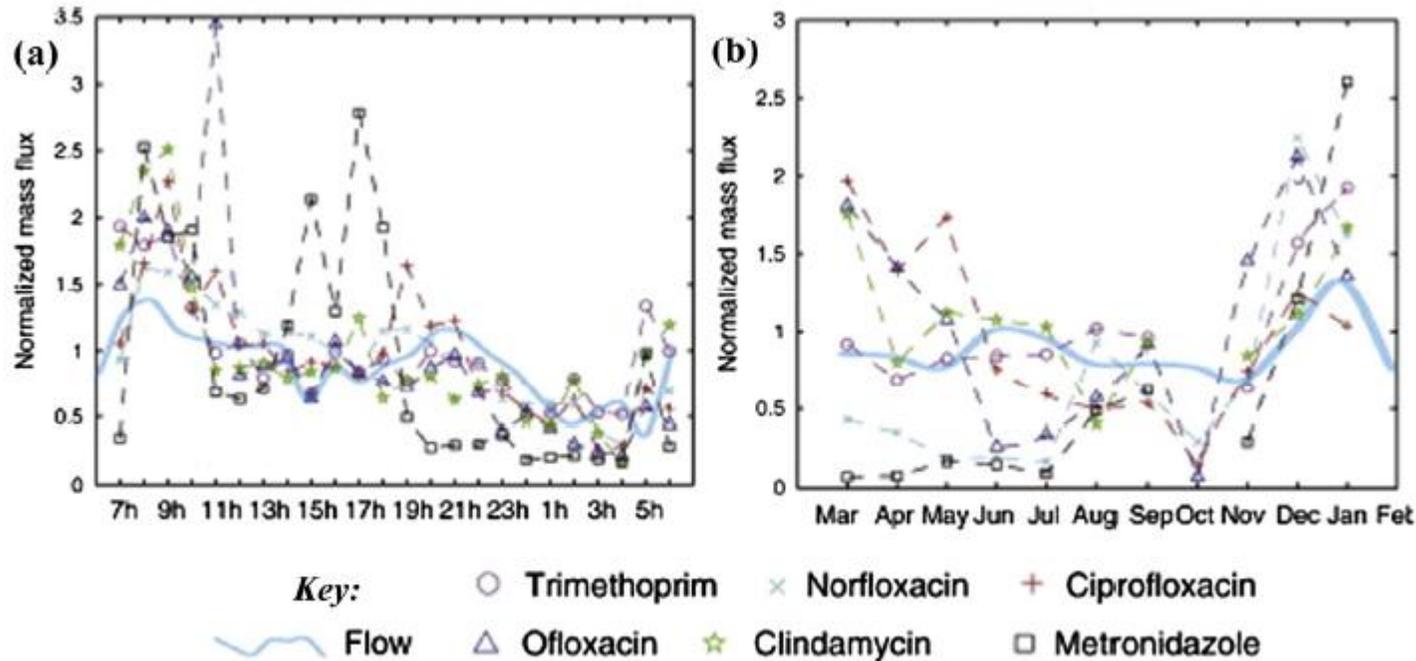


Fig. 1 – Fluctuations of mass flux for selected antibiotics in wastewater throughout a one day period (a) and during the course of a year (b) – adapted from [Coutu et al. \(2013\)](#).

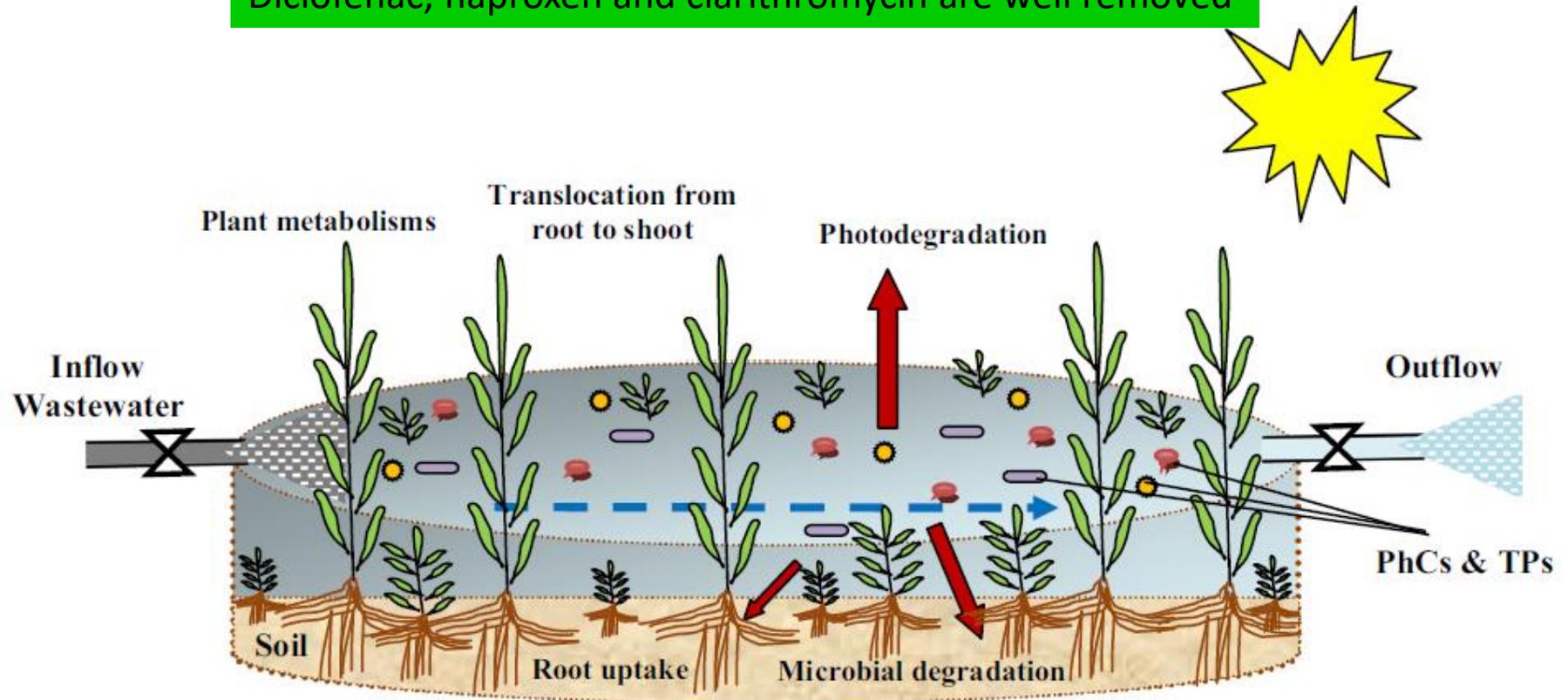
Important to adopt the most pertinent sampling mode and frequency and to clearly report them

Composite samples...flow proportional... sufficient number of samples...
removal evaluated on the influent and effluent load... → **check the quality and assure data reliability**

Free water surface systems

main PhC removal mechanism is photodegradation, while microbial degradation and plant uptake also contribute to some extent in the removal

Diclofenac, naproxen and clarithromycin are well removed

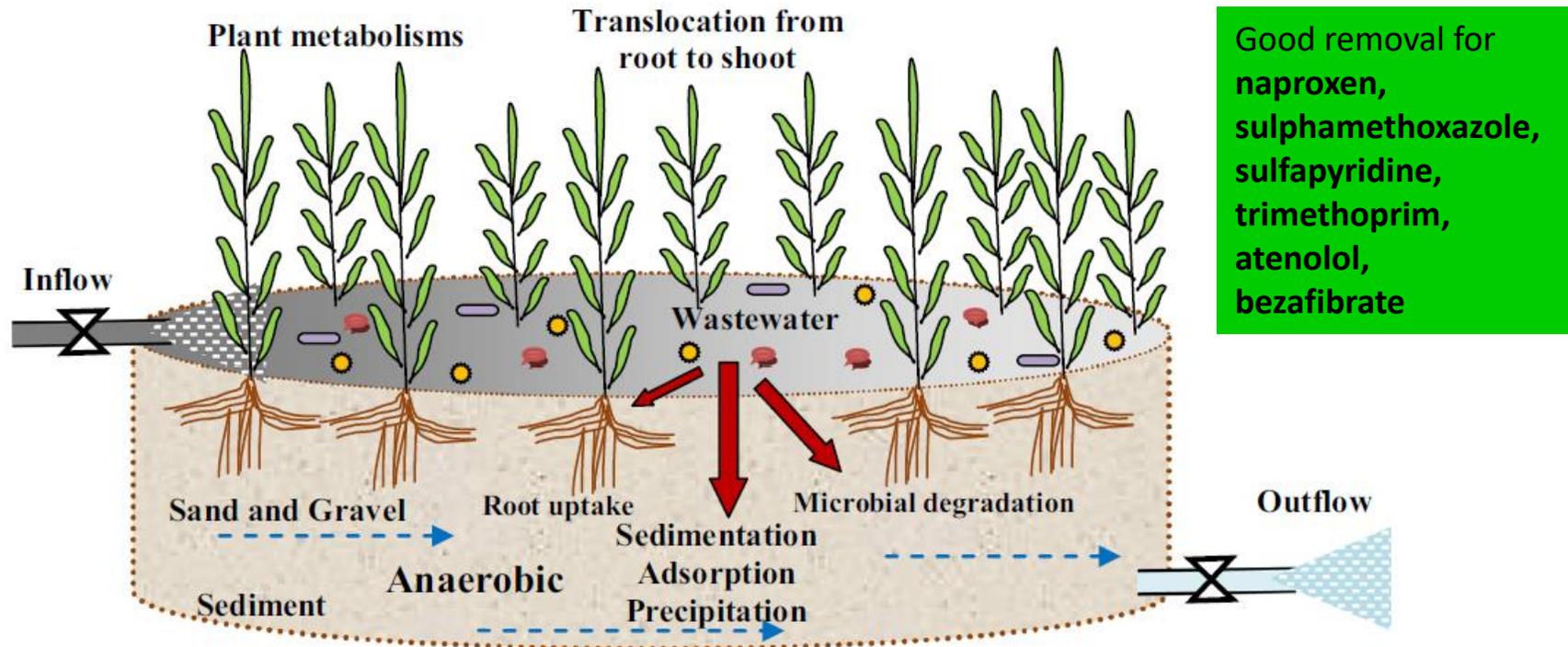


Ilyas and Van Hullebush (2020)



Horizontal subsurface flow systems H-SSF

anaerobic biodegradation is an important removal mechanism of PhCs besides their removal by the filter media (through **sedimentation, adsorption, and precipitation**) and **plant uptake**



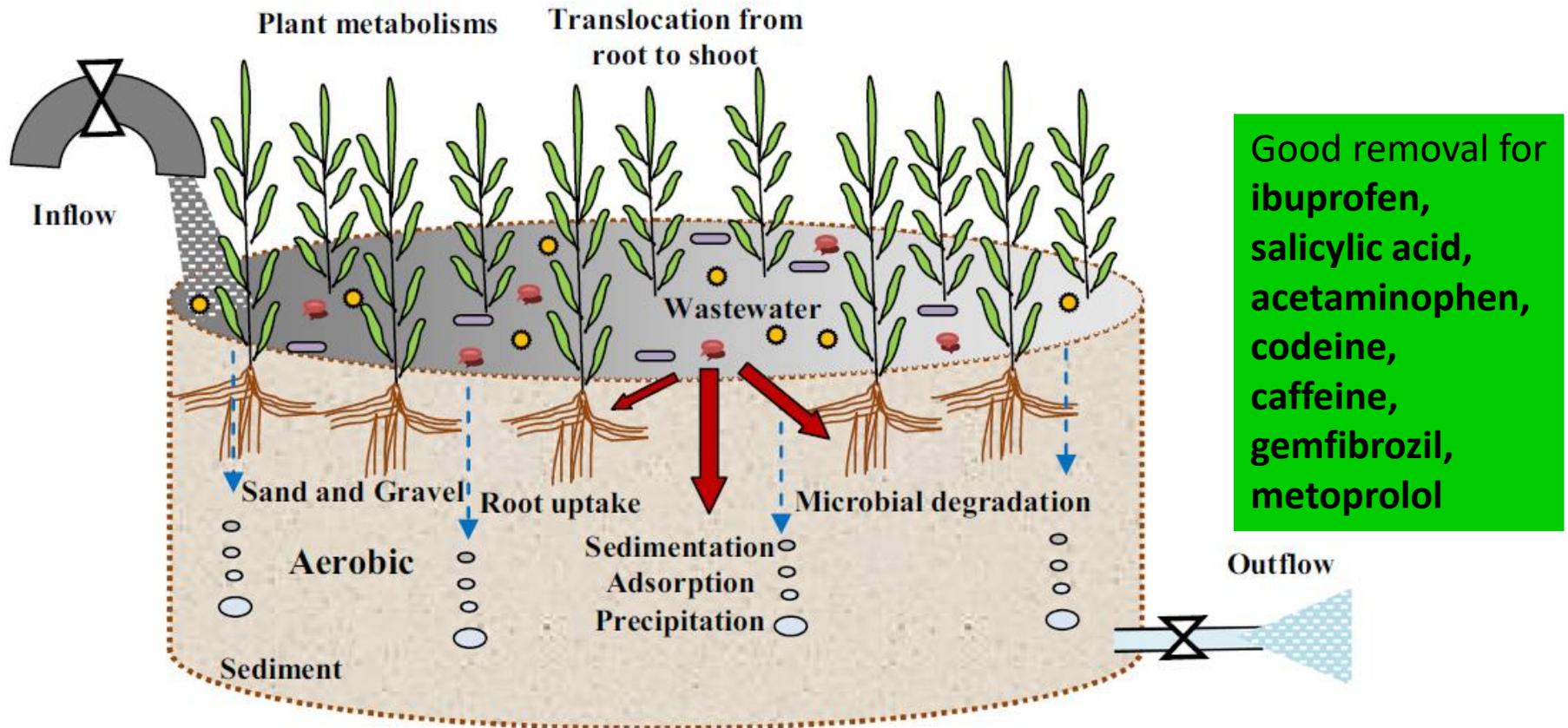
Anaerobic biodegradation is slower than aerobic one, thus longer HRT are necessary to achieve the removal efficiencies in aerobic conditions

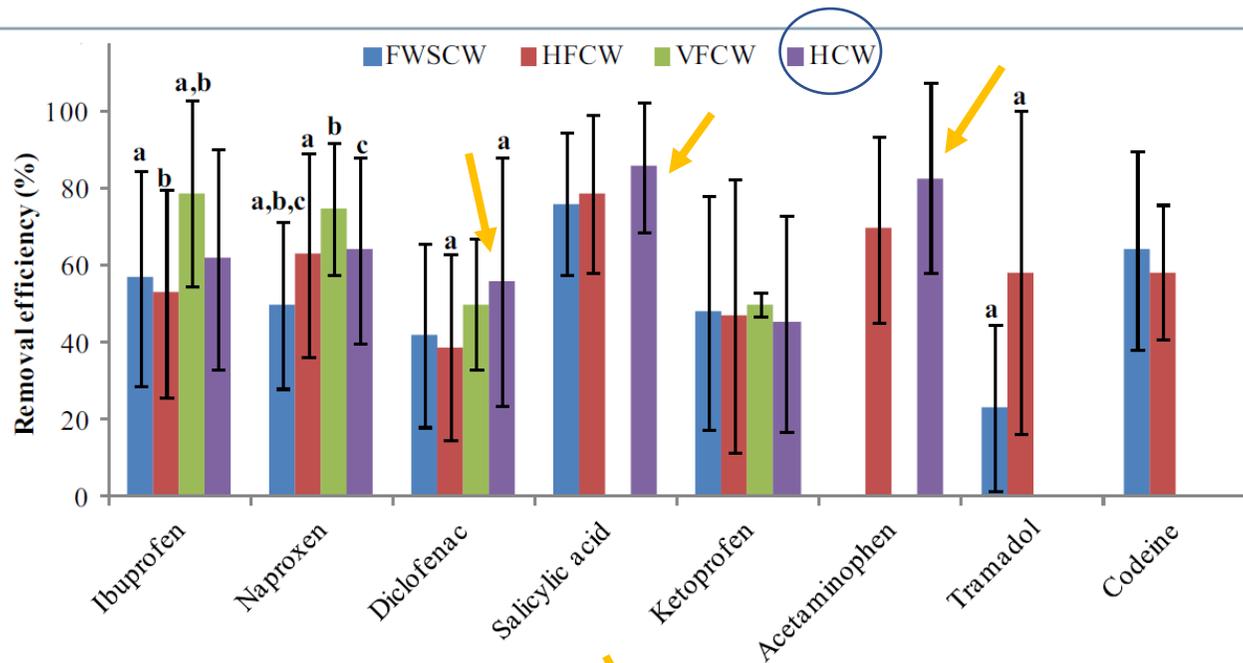
Ilyas and Van Hullebush (2020)



Vertical subsurface flow systems (V-SSF)

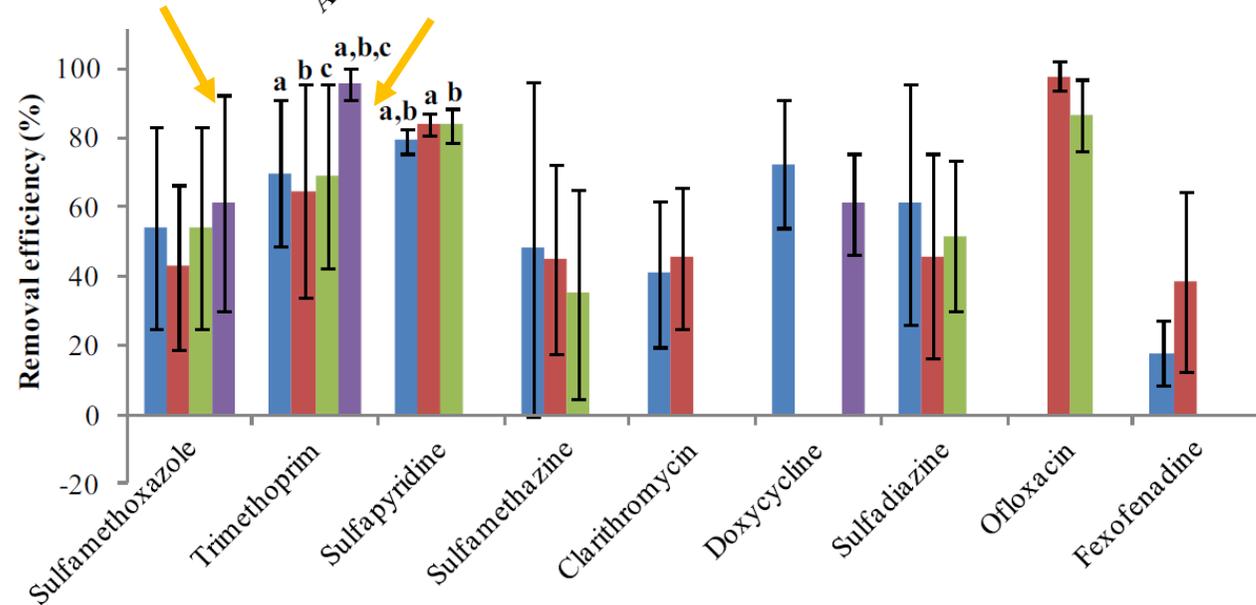
The **aerobic biodegradation** is responsible for the removal of PhCs by V-SSF among other dominant processes (e.g., sedimentation, adsorption, and plant uptake)





Sometimes, the combination of different conditions may enhance the removal of many PhCs.

This may occur in the hybrid systems HCWs (violet in the pictures)



Hydraulic retention time HRT – This is an important parameter for the empirical design and operation of H-SSF beds. According to Zhang et al. (2012), the removal efficiencies for salicylic acid, ketoprofen, clofibric acid were linearly proportional to the influent mass loading rate at HRTs ranging between 2 and 6 days, making it possible to describe the removal of these substances by a constant first-order kinetic decay.

HRT

Temperature – High temperatures improve the removal of some compounds. Zhang et al. (2012) found that at tropical temperatures, ketoprofen was better removed than in temperate climates; Similarly, Hijosa-Valsero et al. (2010b) found higher removal efficiencies in the summer than in the winter.

Temperature

Redox potential RP – It seems that anoxic ($-100 \text{ mV} < \text{RP} < 100 \text{ mV}$) and aerobic ($\text{RP} > 100 \text{ mV}$) conditions favour the biodegradation of organic micropollutants through the promotion of biogeochemical reactions (Matamoros et al., 2008a).

Redox potential

Attempts to predict the behavior

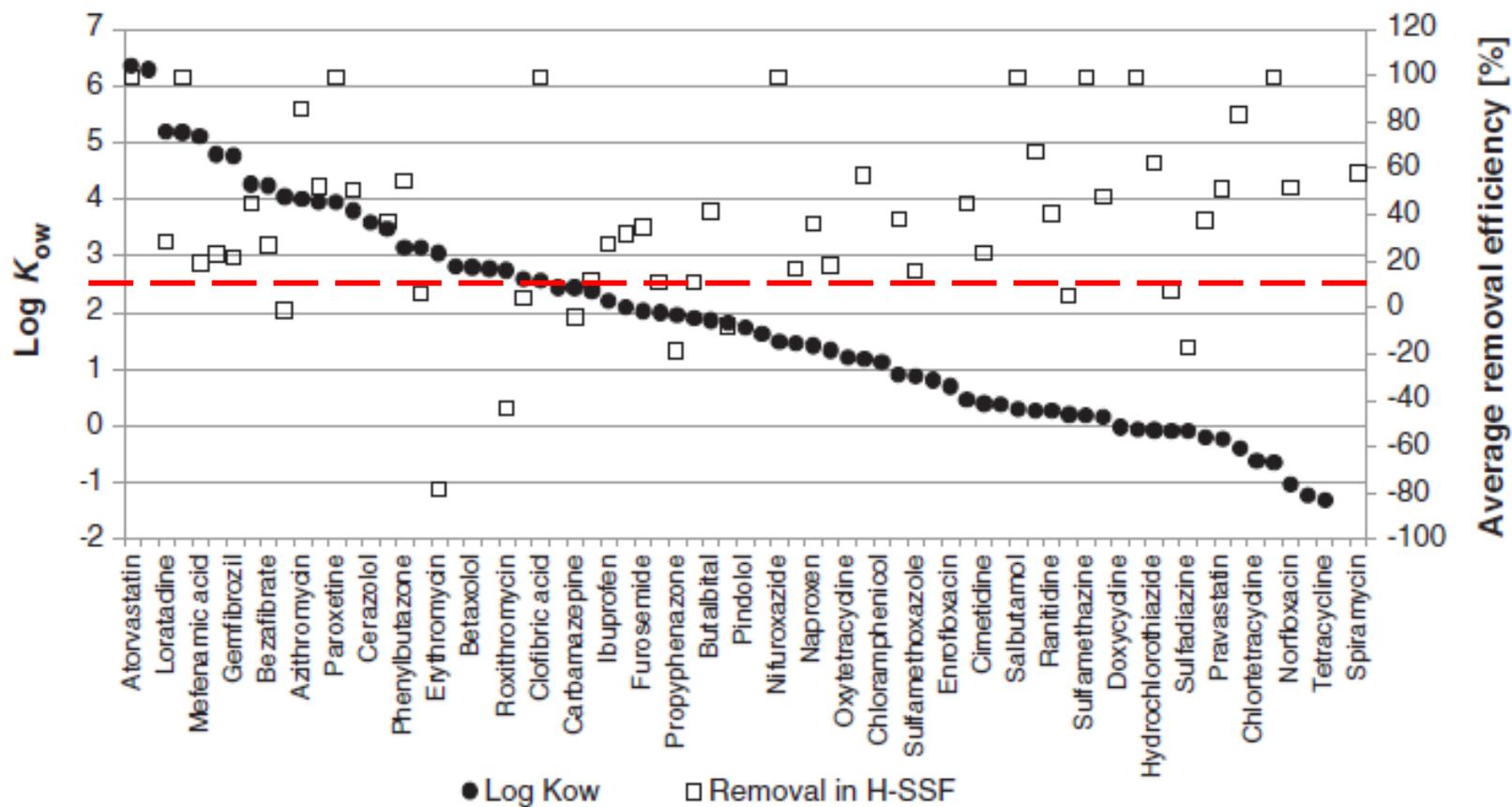


Fig 9. Percentage average removal efficiency profiles and hydrophobicity (log K_{ow}) of the selected compounds.

Removal of the most prescribed classes of antibiotics in different types of CWs

Mechanisms

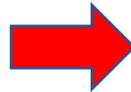
- Volatilization
- Photodegradation
- Precipitation
- Substrate adsorption
- Plant uptake and accumulation
- Microbial degradation

Design and Configuration

Operational conditions

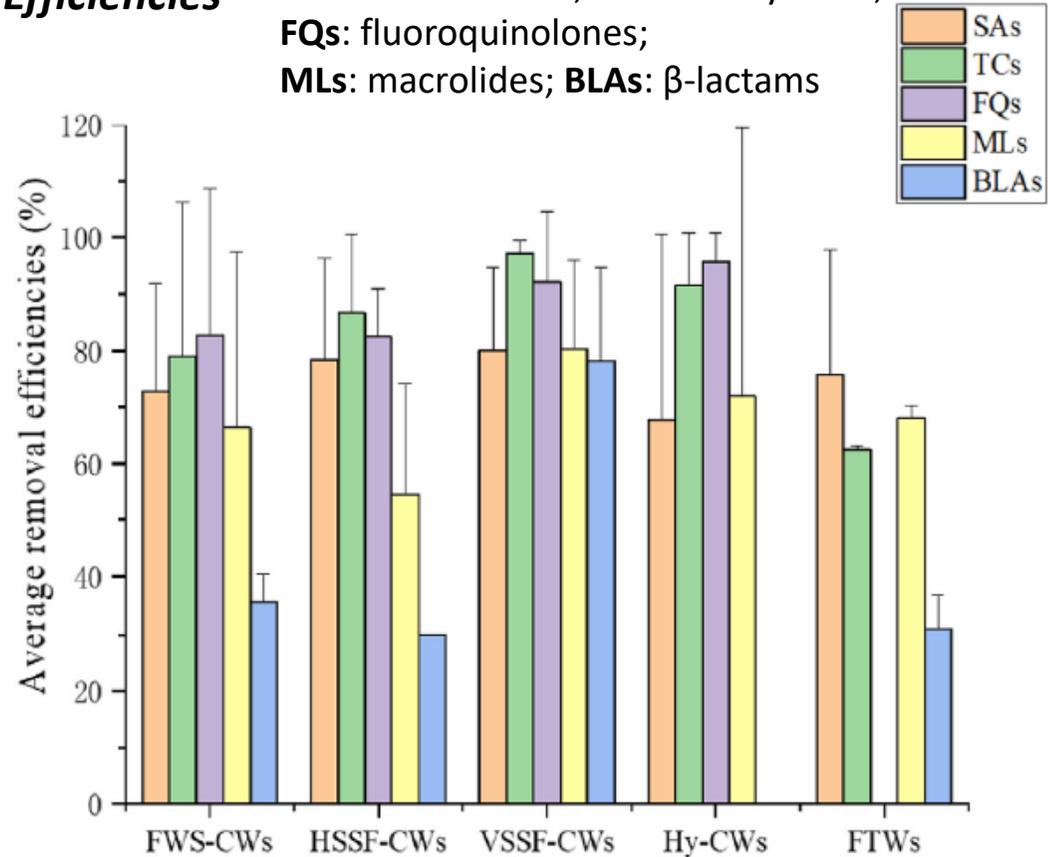
Plant species

Hydraulic load



Efficiencies

SAs: sulfonamides; TCs: tetracyclines;
 FQs: fluoroquinolones;
 MLs: macrolides; BLAs: β -lactams



(FTW= floating treatment wetlands)

Removal pathways

H bondings among ionic functional groups (*aniline and amide*) and polar groups (i.e., hydroxyl and phenolic hydroxyl) present in organic materials;

Electrostatic interactions between the negatively charged soil, clay or other filling material and the positively charged antibiotics

Cation bridging responsible for the sorption of anions by natural sorbents in wetland systems

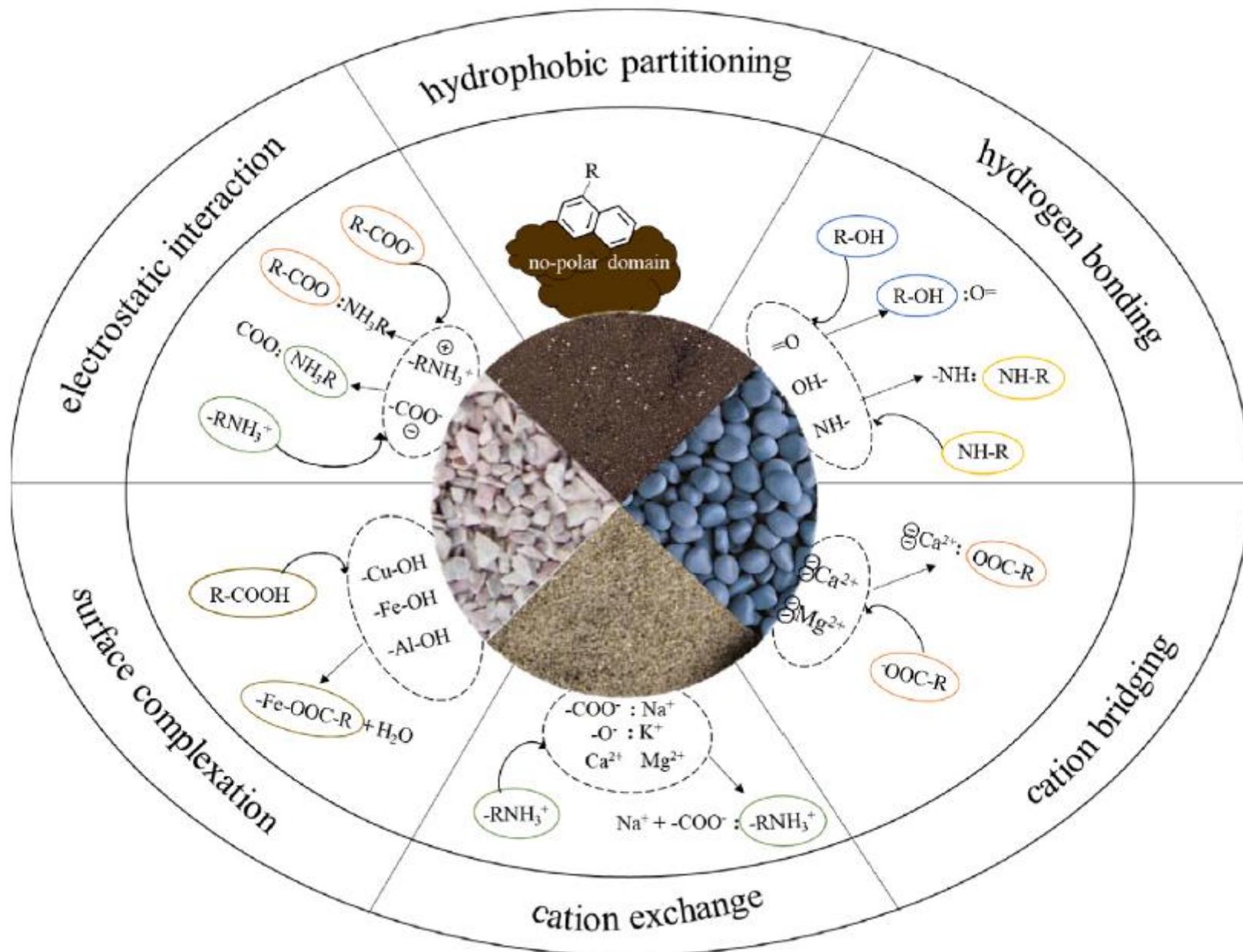
Cation exchange between the cationic amine group and the negatively charged surface sites

Complex formation with metal ions on the substrate surface, such as Ca^{2+} , Mg^{2+} , Fe^{3+} , Al^{3+} , for antibiotic molecules containing polar/ion functional groups

Most antibiotics are ionizable molecules. At pH= 6-8 macrolides are cationic and are efficiently adsorbed, sulfonamides are in anionic form and their removal is mainly due to degradation.

Different responses to HLR changes: they may favor/disturb sorption process





Role of macrophytes

Higher removal in presence of macrophytes

Root uptake: difficult to evaluate and predict. It is widely accepted that for organic compounds with moderate hydrophobicity ($1.0 < \log K_{ow} < 3.5$) and/or with low MW (MW < 500) can easily penetrate cell membranes and be taken up by plant roots (Le-Minh et al., 2010; Yan et al., 2016) through different mechanisms, such as passive uptake via protein channels, protein-mediated energy-dependent active uptake, etc.

By contrast, antibiotics with strong hydrophobicity ($\log K_{ow} > 3.5$) and/or with high MW (>500) tend to bind tightly to the substrate and/or root surface, and be precluded by cell membranes of plant root.

Therefore, **plant uptake may play a minor role** in their removal and other mechanisms, such as plant stabilization and/or rhizosphere bioremediation, might be involved in the remediation

Translocation: physiochemical properties of antibiotics, particularly ionization behavior and hydrophobicity, can also significantly influence the translocation in plants. Hydrophobic compounds tended to remain in the roots with limited in-plant redistribution, while hydrophilic compounds were susceptible to moving toward leaves in the direction of the transpiration stream. neutral compounds with $\log Kow$ values between -1 and 5 are considered mobile in the transpiration stream.



Role of macrophytes

Higher removal in presence of macrophytes

microbial and enzymatic activities through the secretion of exudates, thus enhancing the removal of antibiotics

Evidence that **antibiotics** caused a significant **increase in the ARGs in wetlands**, which were harbored in biofilms on plant rhizosphere and substrate, due to selection pressure on rhizospheric and microbes. In addition to causing the proliferation of their corresponding ARGs, antibiotics can cause the **spread of diverse ARGs**.



Role of microorganisms

Microorganisms play an essential role in biosorption and biodegradation of antibiotic in CWs.

Irrespective of the different removal mechanisms, antibiotics may undergo hydrolysis, oxidation, side chain breakdown, acetylation, hydroxylation, ring cleavage, demethylation, decarboxylation, and dihydroxylation.

Due to the complexity of microbial communities in CW systems and the differences in the chemical structure of antibiotics, **the biodegradation mechanism and functional microorganisms for specific antibiotics removal remain ambiguous.** Therefore, the contribution of different biodegradation pathways to the removal of specific antibiotics needs to be fully elucidated to optimize the design criteria of CW for removal enhancement.

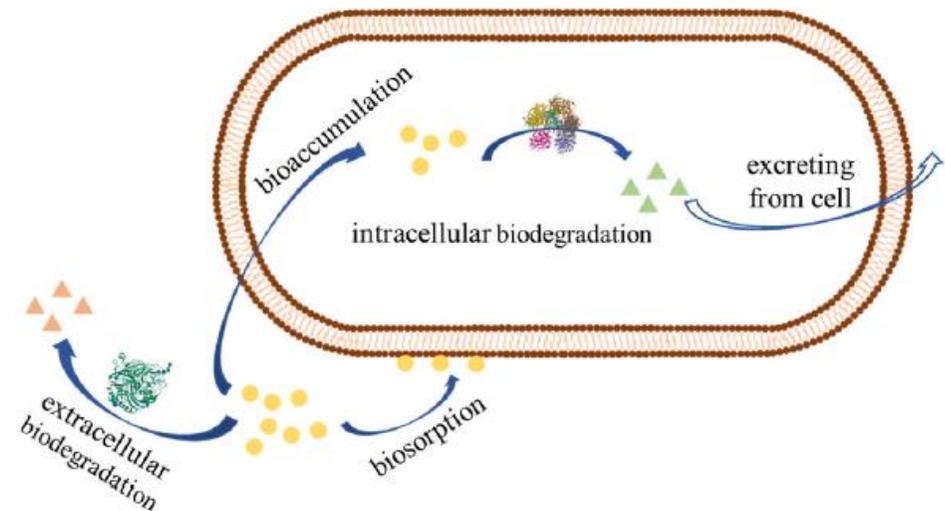


Fig. 6. Possible biological interactions between microorganisms and antibiotics.



Enhancement in the removal

Artificial aeration: the presence of oxygen is beneficial for most of the investigated compounds. Why don't increase the content of oxygen in the bed?

Tidal flow CW characterized by regular cycle of a “filled/wet” phase or a “drained/dry” phase promoting the aeration of the system: air is drawn into the soil pores and the bio-waterfilms is rapidly oxygenated.

Innovative filling materials

Bioaugmentation: this is an effective process for intensifying the degradation of organic pollutants and optimizing the optional conditions in CW systems (Tara et al., 2019). By introducing **bacterial strains** with the ability to degrade specific contaminants into CWs, the population, density, diversity and activity of functional microbes could be reinforced (Wang et al., 2018; Zhao et al., 2019), leading to the acceleration of the pollutant biodegradation.

Towards engineered CW... providing artificial electron acceptors...



Attempts to enhance the removal: *ARTIFICIAL AERATION*

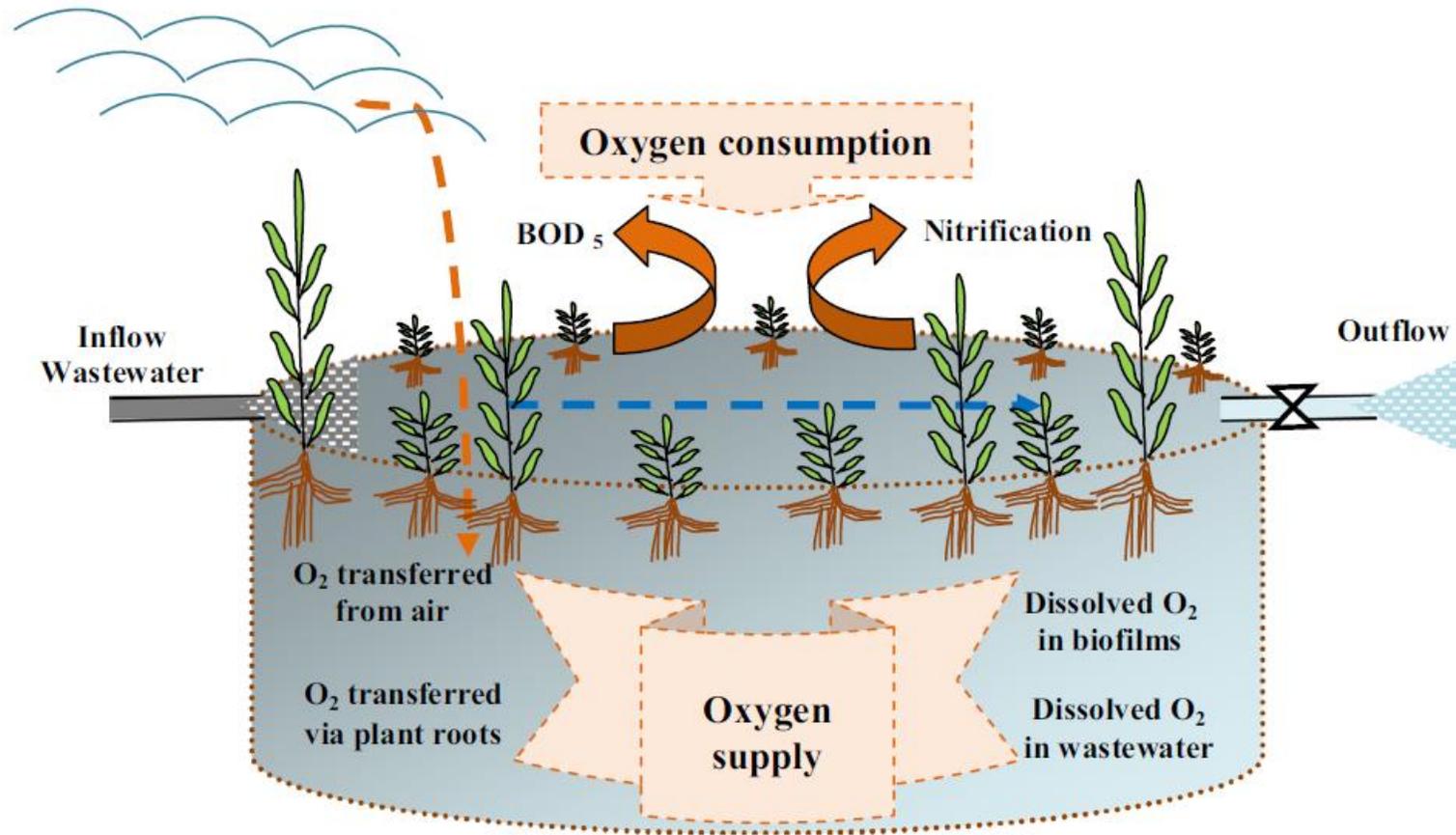
Advantages of **enhancing oxygen availability in CWs**

- (1) improved biodegradation;
- (2) reduced clogging;
- (3) enhanced removal of organic matter, nitrogen, and phosphorous; and
- (4) reduced land area requirement

Table 4 Removal efficiency (mean % and standard deviation) of PhCs in different types of aerated (AA) and non-aerated (NA) CWs

Parameter/ pharmaceutical	NA-FWSCW/AA- FWSCW	NA-HFCW/AA- HFCW	NA-VFCW/AA- VFCW	NA-HCW/AA- HCW	Major removal mechanism
DO (mg L ⁻¹)	6.0/na	1.6 ± 1.3/8.9 ± 0.7	4.5 ± 0.7/7.5 ± 1.1	2.3 ± 2.4/11	
Diclofenac	na	21 ± 12/48 ± 22	56 ± 7/68 ± 9	56 ± 32/99	Biodegradation (aerobic/anaerobic)
Ibuprofen	na	23 ± 8/99	96 ± 2/99 ± 1	na	Biodegradation (aerobic)
Naproxen	na	28 ± 6/99 ± 1	89 ± 2/94 ± 1	na	Biodegradation (aerobic/anaerobic)
Acetaminophen	99/97	na	na	na	Biodegradation (aerobic/anaerobic)
Tramadol	na	6.8/1.6	na	na/99.9	Biological transformation
Caffeine	82/94	89 ± 8/99.5 ± 0.7	97 ± 1/99	na	Biodegradation (aerobic/anaerobic)
Carbamazepine	na	12 ± 9/11 ± 11	- 8.5 ± 0.7/- 2.5 ± 2.1	27 ± 20/94	Adsorption/sorption
Atenolol	na	na/96 ± 1	na	73 ± 37/98	Biodegradation (aerobic/anaerobic)
Metoprolol	na	na	na	99/98	Biodegradation (aerobic)
Sotalol	na	24 ± 6/32 ± 13	na	82/99	Biodegradation (aerobic)

The oxygen supply routes and consumption processes in CWs



Ilyas and Van Hullebush (2020)



Another direction of the research for enhancing ECs removal: “engineered CW”

Major limitations of CWs = **slow reaction kinetics**, mainly due to the limited electron acceptor and **slow microbial metabolism** in presence of massive anaerobic conditions in the CW systems.

Incorporation of **bio-electrochemical systems (BESs)** into the CWs, such as

microbial fuel cells (MFCs)

and **microbial electrolysis cells (MECs)** (Ilyas and Hullebusch, 2020; Zhang et al., 2020b), has gained a considerably increasing attention.

CW-BESs can **provide artificial electron acceptors** in the anaerobic regions of CW, and electrical stimulation in CW-BESs may **enhance the variation in cell membrane structures and enzyme activities of some microorganisms**,

→ thus, promoting the utilization of mineral elements and carbon sources by microorganisms,

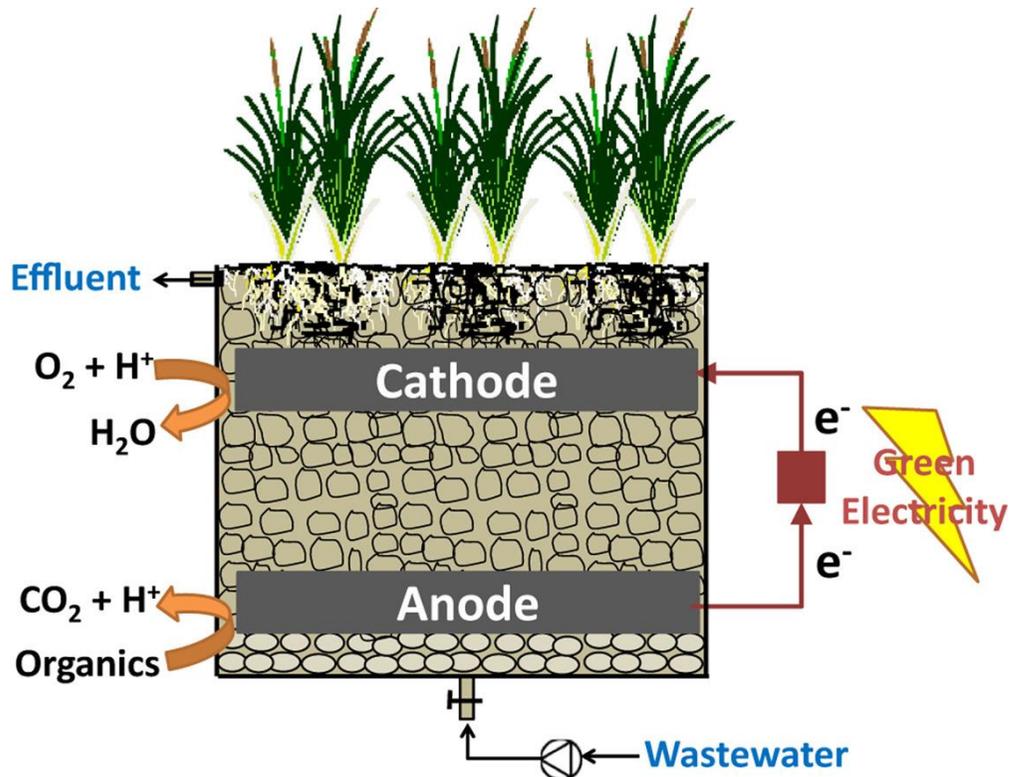
→ and thus, improving degradation performance to antibiotics.



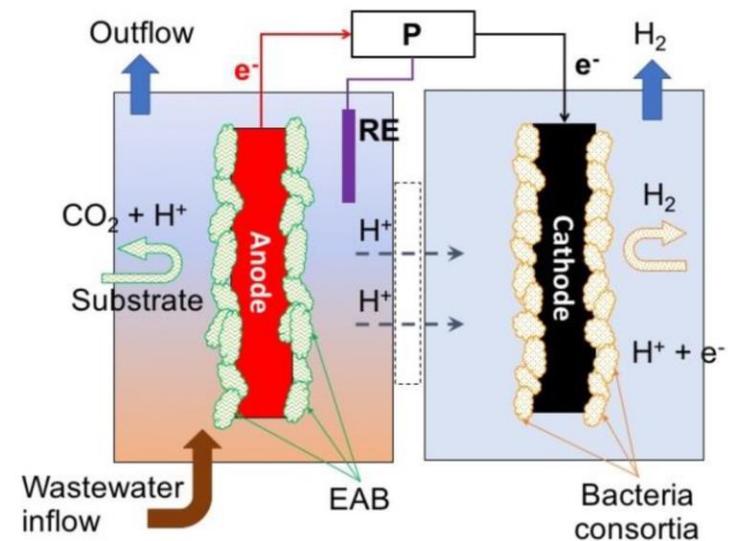
Enhancement in the removal: CW Microbial fuel cell

The application CW-MEC for contaminant removal is based on the same principle as CW MFCs, with a slight difference at configuration.

CW-MFC can transform chemical energy into electricity by oxidizing organic matter and produce bioelectricity



CW-MEC requires additional electron donors for the treatment (see the principle in the picture)



Zhao et al., 2013

Ramirez-Vargas et al. 2018

Considerations and hints for discussion - Conclusions

Different mechanisms affect the removal of these micropollutants presenting a variety of chemical and physical properties

Researches are dealing with:

- **Key metabolic and co-metabolic removal pathways** for the different compounds
- **Transformation products** (usually unknown compounds, and the relative research is currently viewed as a challenge)
- **Bioaugmentation** (some key issues remain unresolved in the bioaugmentation process, such as how the succession of microbial community affects the performance of bioaugmentation system and how the added strains interact with local microbial community). The integration of **microalgae into CWs** can enhance biodegradation performance by compensating for microbial activity at low temperature.
- **Artificial aeration**



- However, the **feasible applications** are still scarce and future research on the integrated systems is essential for improving a low-cost and environmentally sustainable alternative for micropollutants removal.
- Great efforts have been made to develop **intensified CW systems for enhancing the antibiotics removal ability in CWs**.
- However, most of the trials are limited to **lab-scale applications** without field validation. Consequently, the resilience and sustainability of these new approaches have to be thoroughly evaluated via pilot scale investigation to establish field-based data on accumulation and biodegradation of antibiotics in soil-plant system.
- It is clear that the research requires the synergies among different researchers: biologists, chemists, environmental engineers, chemical engineers, geologists, biochemists, biotechnologists, ... in order to match the different experiences and provide robust, reliable and “resilient” solutions to enhance the removal of target ECs and to reduce the environmental impact of their presence in the aquatic environment!



Thanks for your kind attention!

And now time for questions or discussion

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**Traitement des eaux usées et des
boues résiduelles par filtres plantés
et usage agricole durable**



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