

# Phytoremediation as a Tool to Remove Drivers of Antimicrobial Resistance in the Aquatic Environment

Kaniz F. Chowdhury<sup>1</sup> · Rebecca J. Hall<sup>2</sup> · Alan McNally<sup>2</sup> · Laura J. Carter<sup>1</sup>

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#### Abstract

Antimicrobials, heavy metals, and biocides are ubiquitous contaminants frequently detected in water bodies across the globe. These chemicals are known as drivers of antimicrobial resistance (AMR), as these chemicals can select for resistance. Tools and processes, are therefore, needed to remove these chemicals from the environment to tackle the environmental component of AMR. Aquatic phytoremediation is a nature-inspired green solution to remove contaminants from the environment. Phytoremediation utilises macrophytes' ability to sequester and degrade chemical pollutants in aquatic environments. In this review, we define the problem statement by highlighting the presence of AMR drivers in the aquatic environment. We also provide an in-depth review of phytoremediation to tackle chemical pollution by evaluating mechanisms for the removal and degradation of chemicals. This review identifies potential hyper-accumulators and understands how plant species and chemical composition can influence the potential for accumulation. Different pollutants accumulate to different extents in a range of aquatic macrophytes. Therefore, the combined use of floating, submerged and emergent plants would facilitate the optimum removal of AMR drivers considered in this review. A suggested configuration includes *Helianthus annus* around the edge of a contaminated site, followed by a belt of submerged plants (Myriophyllum aquaticum) and a bed of floating plants (e.g., Lemna species) together with the hyperaccumulator, Phragmites australis. Whilst phytoremediation offers a promising option to treat contaminated water, several critical knowledge gaps still exist. The effect of co-exposure to contaminants on the accumulation potential of plants and the fate of antibiotic-resistant genes and bacteria during the phytoremediation process are highlighted in this review. Based on this understanding, targeted areas for future research are proposed.

# Introduction: Pollution as a Driver of Antimicrobial Resistance

Antimicrobials, including antibiotics, antivirals, antifungals and antiparasitic, are prescribed to combat infections such as pneumonia, meningitis and sepsis (Amos et al. 2018).

Kaniz F. Chowdhury K.F.Chowdhury@leeds.ac.uk

> Rebecca J. Hall R.J.Hall@bham.ac.uk

Alan McNally a.mcnally.1@bham.ac.uk

Laura J. Carter l.j.carter@leeds.ac.uk

<sup>1</sup> School of Geography, University of Leeds, Leeds LS2 9JT, UK

<sup>2</sup> Institute of Microbiology and Infection, College of Medical and Dental Sciences, University of Birmingham, Edgbaston, Birmingham B15 2TT, UK

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When an antimicrobial treatment is effective, bacterial growth inhibition is achieved when the antimicrobial interacts with its target. However, when these chemicals are used or overused, the bacteria they are meant to kill can adapt and develop resistance, thus rendering these treatments often ineffective. Bacteria resistant or have acquired resistant traits can survive, multiply, and develop antimicrobial resistance (AMR) (Prestinaci et al. 2015). AMR is now recognised as an extreme global health concern in the twenty-first century, threatening the successful delivery of key UN Sustainable Development Goals (Samreen et al. 2021). There were an estimated 4.95 million (3.62-6.57) deaths associated with bacterial AMR in 2019 (Murray et al. 2022), with suggestions that AMR could kill 10 million people per year by 2050 (O'Neill 2016), which is more than deaths caused by cancer (Pires et al. 2017). It is also important to note that AMR is not only confined to a clinical setting; resistance also poses a threat to the effective use of antimicrobials in aquaculture, livestock and poultry production.

Antibiotic consumption in cattle and poultry has risen unprecedentedly across several continents (Nhung et al. 2017; Hedman et al. 2020) and is expected to increase by 67% by 2030 in rapidly developing countries (Van Boeckel et al. 2015). The environment has been suggested to play a role in the global spread of clinically relevant AMR (Larsson and Flach 2022; Murray et al. 2021). Wastewater and animal waste contain many antibiotic-resistant bacteria (ARB) and antibiotic-resistant genes (ARGs), which can be spread on our soils, sediments and water bodies following the discharge of urban wastewater, antibiotic manufacturing discharge, and organic waste (Kotwani et al. 2021; Larsson and Flach 2022) into the receiving environment (Murray et al. 2021; Stanton et al. 2022). Bacteria can also develop resistance following exposure to chemical contaminants commonly detected in our environment, known as AMR drivers. There are mainly three main classes of resistancedriving chemicals identified that this review will focus on, namely: (Singer et al. 2016).

- (a) antimicrobials with four subclasses, antibiotics, antifungals, antivirals, and antiparasitics;
- (b) heavy metals; and
- (c) biocides (i.e., disinfectants and surfactants).

It is important to note exposure to natural compounds (plant-derived) and xenobiotics (hexane, toluene and octanol) has also been reported to select for resistance genes (Fernandes et al. 2003; Friedman 2015; Samreen et al. 2021) but is beyond the scope of this review.

Bacteria can become resistant to antimicrobials through several mechanisms (Fig. 1) based on either modifying the target or reducing the concentration of the antimicrobial that can access the target. For example, antibiotic sequestration can block the antibiotics from reaching their target (Peterson and Kaur 2018), the bacterial membrane can be modified to protect the bacterium from an antibiotic insertion, or resistant genes can be transferred from other bacteria (Walsh 2000) Fig. 1; for a comprehensive review of resistance mechanisms see (Wanda 2018). However, the term 'ARG' is often misleading as antibiotics are not the only chemicals that select resistance genes (Singer et al. 2016). Contaminants such as heavy metals and biocides can also contribute to the dissemination of AMR by enriching resistance gene determinants via co-selection mechanisms (Thomas IV et al. 2020). Co-selection of resistance genes has been reported for hazardous chemicals such as solvents (Korshunova et al. 2016), biocides (Conficoni et al. 2016), heavy metals (Wales and Davies 2015) and antibiotics. Coselection can occur via (i) co-resistance, where the selection of one gene supports the selection of another gene that usually does not offer a selective advantage to the compound of interest (Pal et al. 2015b); and (ii) cross-resistance, where one resistance gene protects against a range of toxic chemicals (Hall et al. 2018; Samreen et al. 2021). The structural and functional characteristics of antibiotic resistance share

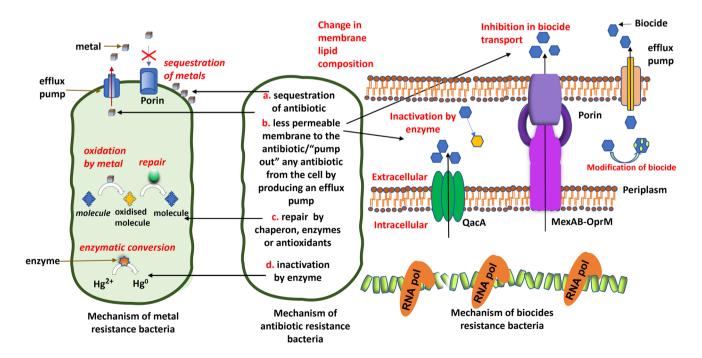


Fig. 1 Common resistance mechanisms of bacteria to metals (left), biocides (right) and antibiotics (centre). This figure's mechanisms are written in red, and biocides and metals are drawn as blue hexagons

and grey boxes, respectively. The mechanism of biocides resistance bacteria (right-hand side of the figure) was redrawn following Venter et al. (2017)

common themes with those of metal and biocides resistance, as illustrated in Fig. 1.

## **Resistance Driver: Antimicrobials**

Antimicrobials are medicines used to prevent and treat infections and play a significant role in human medicine, aquaculture, and livestock industries (Ahmed and Gareib 2016). China and the USA are the largest consumers of antimicrobials for animal use (Van Boeckel et al. 2014), and non-prescription antibiotics are also still common in many countries outside Europe and North America (Sohail et al. 2016), including for use as prophylactic agents and growth promoters (Landers et al. 2012). Their widespread use has resulted in the mass loading of bioactive antimicrobial compounds in the environment with concentrations typically in ug/L in wastewater to ng/L in surface waters (aus der Beek et al. 2016). In a recent review of antibiotic detections in the aquatic environment, fluoroquinolones were found in high concentrations in waters (<460 ng/L) and sea sediments (406 ng/g), with a frequency of 49% detection of all antibiotics. In rivers, sulfonamides were reported in the highest abundance (30%), with the highest concentration in lakes observed for fluoroquinolones (abundance of 34%) (Maghsodian et al. 2022). Poor removal methods are mainly responsible for the discharge of antibiotics into the environment following wastewater treatment, exposing ARGs to high-level antibiotic selection pressures (Phoon et al. 2020; Hou et al. 2019; Guo et al. 2018). For example, sulfonamides and trimethoprim are frequently detected in the aquatic environment, with WWTP removal efficiencies reported to vary from 20% to over 90% (Michael et al. 2013; Göbel et al. 2007; Ternes and Joss 2007). The fate and behaviour of antimicrobials in the receiving aquatic environment are also variable and are influenced by environmental parameters such as temperature and pH (Rosi-Marshall and Kelly 2015; Manzetti and Ghisi 2014; Cycoń et al. 2019; Kraemer et al. 2019). Selected antimicrobials are relatively persistent (Patel et al. 2019), such as fluoroquinolones (e.g., ciprofloxacin) and sulphonamides (e.g., sulfamethoxazole). Their residues are frequently detected in the environment, and their resistance is often reported (Ashbolt et al. 2013; Kümmerer 2009). In contrast,  $\beta$ -lactam antibiotics are readily degradable and not often detected in the environment but, interestingly, still contribute to developing resistance (Kümmerer 2009; Lundborg and Tamhankar 2017).

Theoretically, a chance interaction between a single molecule of an antibiotic and a bacterium can trigger natural selection for resistance or a mutation favouring resistance (Lundborg and Tamhankar 2017). Antibiotic concentrations found in the environment and released from anthropogenic sources are generally lower (ng/L–µg/L) than minimum inhibitory concentrations (MICs) defined to select for resistance (Finley et al. 2013; Hanna et al. 2023; Levy and Marshall 2004). Traditionally these concentrations have not been regarded as a risk in AMR selection. However, single species competition assays determined that the selection for resistance occurred at concentrations considerably lower than the MICs, where the resistant strain is enriched over the susceptible strain with the lowest selective concentration, termed the "minimal selective concentration" (MSC) (Gullberg et al. 2014, 2011). The MSCs have been determined for various antibiotics, e.g., 100 ng/L for ciprofloxacin to 3 mg/L for erythromycin and include concentrations commonly detected in the environment (Stanton et al. 2020). In addition to antibiotics, clear links have also been made between concentration levels of the antimicrobial triclosan in streams, and the proportion of cultivable benthic bacteria that were resistant to triclosan, demonstrating concentration of antimicrobials in the environment can affect native communities (Drury et al. 2013).

#### **Resistance Driver: Heavy Metals**

Although they are naturally occurring elements in the earth's crust, widespread heavy metal pollution is essentially a result of their multiple industrial, domestic, agricultural, medical and technological applications (Rahman and Singh 2019; Tchounwou et al. 2012). Studies have linked mining and smelting operations, particularly steel production, to releasing heavy metals into the environment, including lakes, rivers, and sediments. Rivers with the most significant pollution are typically near industries and mining areas (Scerbo et al. 2002; Di Cesare et al. 2016). Heavy metals such as copper (Cu), zinc (Zn), chromium (Cr), nickel (Ni), lead (Pb) and cadmium (Cd) are still used in some intensive dairy farming operations as feed additives. The land application of animal wastes can lead to metals being washed off into nearby water courses (Rahman and Singh 2019). Concentrations of heavy metals are typically reported up to low mg/L levels. For example, in Xikuangshan, the world's largest antimony mining region, river concentrations were 0.48 mg/L (0-4.34 mg/L), 2.58 mg/L (0-4.34 mg/L), 1.05 mg/L (0.0009–5.33 mg/L), 1.06 mg/L (<19.60 mg/L) and 0.00084 mg/L (< 0.0036 mg/L), for total nitrogen, total phosphate, antimony (Sb), arsenic (As) and mercury (Hg) respectively (Xie and Ren 2022).

Like antimicrobials, heavy metal release into the environment can also occur following wastewater treatment (Tytła 2019), with iron (Fe) recently observed to be the most abundant heavy metal in processed wastewater, followed by Zn (Rathi et al. 2021a). Comparatively, Cd has been reported as the lowest abundant metal in wastewater and sludge (Karvelas et al. 2003). Since metals are not biodegradable, they are persistent pollutants and remain present in the aquatic environment following the discharge of treated effluents. Metals including arsenic (As), Cd, Cr, Cu, Ni, Pb, and Zn are frequently reported in the aqueous phase as well as adsorbed onto microplastics, such as polyethene terephthalate and polyethylene (Sarkar et al. 2021).

The presence of heavy metals in aquatic systems has increased the selection of AMR genes in the environment (Singer et al. 2016; Bazzi et al. 2020; Yazdankhah et al. 2018). Cu resistance genes are among the most commonly detected in the environment in the BacMet database (Pal et al. 2013), with excessive use of Cu and Zn as feed additives in livestock production suggested to be responsible for this (Yazdankhah et al. 2014). Research has also shown the recovery of heavy metal-resistant bacteria from different environmental matrices, including water bodies (Eltahawy et al. 2022), with further studies demonstrating heavy metal-resistant mutants can exhibit multi-drug resistance. For example, Zn(II) evolved ciprofloxacin-resistant mutants are also resistant to chloramphenicol and tetracycline (Guo et al. 2021).

## **Resistance Driver: Biocides**

Biocidal products include various chemical compounds that exert microbiostatic or microbiocidal effects against various microorganisms. Disinfectants are commonly used in cosmetics, hospitals, household cleaning products, wipes, and industrial processes, including fouling management and souring of pipes (Maillard et al. 2018), with the most commonly used biocides including formaldehyde, chlorhexidine and quaternary ammonium compounds (QACs) (Jutkina et al. 2018). Biocide use is continually expanding with recent applications for use as antifouling agents in building materials. Biocide also can disseminate contaminated aerosols in cooling towers (e.g., Legionella spp.). not only that biocides are also using for development of antimicrobial surfaces (Adlhart et al. 2018; Jones and Joshi 2021; SCENIHR 2009). In contrast, biocides can be easily washed off outdoor materials following rain events and reach the aquatic environment via urban stormwater runoff (Hensen et al. 2017). Incomplete removal in WWTPs also presents a significant pathway by which biocides can end up in receiving aquatic environments (Paun et al. 2022). Between 2013 and 2020, pesticides were reported in 10,219 surface water samples from European countries (EEA 2022). Disinfectants (triclosan and triclocarban), preservatives (methylparaben and propylparaben) and the insect-repellent DEET are also commonly detected in surface waters (Jia et al. 2020). Maximum surface water concentrations have been reported up to 5160 ng/L for triclosan in India (Ramaswamy et al. 2011), 6800 ng/L for triclocarban in the USA (Halden et al. Halden and Paull 2005), 1060 ng/L for methylparaben, 2140 ng/L for propylparaben in China (Peng et al. 2008b) and 3700 ng/L for DEET in USA (Lee and Rasmussen 2006). Biocides and antimicrobials are similar chemicals that have comparable structures. The processes that determine the fate of antimicrobials in the environment are also crucial for biocides because of their chemical similarity (Singer et al. 2002; Thomas and Brooks 2010; Hensen et al. 2018). The presence of biocides in surface waters can enhance ARG development (Kampf 2018), with the MIC of biocides determined against multidrug-resistant pathogens reported to range from 0.40 to 1000 µg/mL (Samreen et al. 2021). Triclosan, an antimicrobial agent combined with biocides such as QACs and chlorhexidine, is suitable for antibiotic resistance in microbial pathogens (Buffet-Bataillon et al. 2012). Sub-lethal concentrations of biocides also facilitate the selection of mutations that confer antibiotic resistance, similar to the selection of ARGs at sub-lethal concentrations of antibiotics commonly detected in the environment (Bengtsson-Palme and Larsson 2016; Lu et al. 2018; Pal et al. 2015a).

Following the continuous discharge of AMR drivers into the environment, aquatic systems are now considered a source of resistance genes and a site of antibiotic-resistance evolution, thereby increasing the demand for practical remediation tools (Czekalski et al. 2014). AMR drivers also present a risk to non-target organisms that inhabit these matrices (Samreen et al. 2021; Singer et al. 2016); thus, there is a clear need to clean up and remove these contaminants from our aquatic environment. This review summarises our current understanding of phytoremediation as a tool to remove biocides, heavy metals and antimicrobials from aquatic systems to propose an ideal phytoremediation setup to maximise the removal of AMR drivers in a wastewater-effluent-dominated water body system. Potential areas of future research must also be identified to maximise the efficiency of aquatic macrophytes as a removal measure to and ultimately reduce the global spread of AMR.

# Phytoremediation and the Treatment of Contaminated Aquatic Systems

While the development of antibiotic resistance is a natural phenomenon, increasing exposure to AMR drivers increases the selection pressure. Thus, reducing exposure and removing these contaminants from our environmental systems is an important strategy to reduce selection pressure for AMR. A range of remediation techniques, including photolysis, UV-degradation, membrane and nanofiltration, reverse osmosis, ion exchange and adsorption, have been developed to remove antimicrobials, biocides and heavy metals from aqueous systems (Rahmanian et al. 2011; Zupanc et al. 2013; Lai et al. 2016; Kaur et al. 2019; Shen et al. 2019). However, these methods are commercially limited because of toxic sludge generation, partial chemicals removal, high-operating costs, and the need for skilled operating and maintenance personnel (Kaur et al. 2019). In contrast, phytoremediation is a cost-effective plant-based remediation approach that uses the ability of plants to accumulate and concentrate compounds from the environment with the potential to metabolise various molecules in their tissues (Delgado-González et al. 2021; Mustafa and Hayder 2021).

In phytoremediation, plants accumulate contaminants through their roots and translocate the chemicals to the shoots (Calamari et al. 2003; Lalumera et al. 2004). Phytoremediation can also be known as agro-remediation, green remediation, vegetative remediation, green technology and botany remediation (Hirsch et al. 1999; Pires et al. 2017; Sacher et al. 2001) and can take the form of in-situ and ex-situ remediation. Phytoremediation in-situ is more commonly adopted as it minimises the risk to the adjacent environment (Ashton et al. 2004). Multiple pollutants can be treated on-site by phytoremediation without needing additional disposal. Phytoremediation was introduced in the 1980s to remove heavy metals (Utsunamyia 1980). Certain plants, called 'hyper-accumulators', are good candidates for phytoremediation, particularly of heavy metals. Through repeated harvesting of the plant tissues, certain elements can be re-extracted and recycled for subsequent applications (Sarma 2011; van der Ent et al. 2013). Phytoremediation is now considered an incredibly versatile approach to removing various chemicals, including antimicrobials and biocides.

#### **Phytoremediation Mechanisms**

Aquatic macrophytes, defined here as emergent, floating or submerged plant species with distinct roots and shoots, have a significant capacity to uptake substances from their growth medium, thus lowering the pollution concentration of a target water body (Dhote and Dixit 2009; Fletcher et al. 2020). Phytoremediation comprises physical, chemical and biological processes (Garrison et al. 2000), and as outlined in Fig. 2, this approach utilises many mechanisms, including (1) accumulation (phytoextraction, rhizofiltration); (2) immobilization (phytostabilization); (3) degradation (rhizodegradation, phytodegradation; (4) dissipation (phytovolatilization) to remove, degrade or immobilise pollutants. The combination of specific mechanisms for pollutant removal and degradation by macrophytes depends primarily on the type of plants, properties of the pollutant and the location of the contaminant within the water body (i.e., water column, lake or streambed sediment) (Miretzky et al. 2004; Vymazal 2011; Xing et al. 2013; McAndrew et al. 2016). The phytoremediation potential of a plant can be evaluated by calculating a Bioconcentration Factor (BCF), which is the ratio of the pollutant concentration in the plant to that in the water body and is often reported in L/kg. Commonly observed phytoremediation mechanisms in aquatic plants are discussed below and summarised in Table 1.

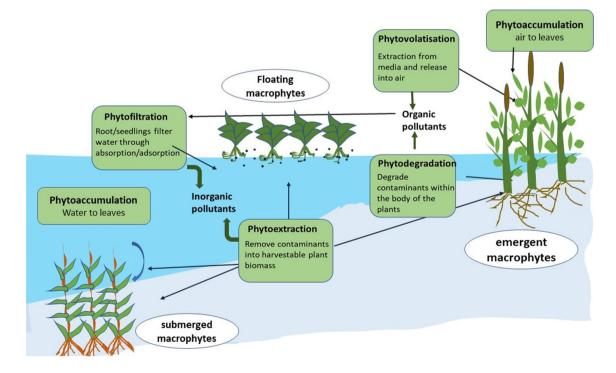


Fig. 2 Schematic diagram of specific parts (leaves, shoots and roots) of different kinds of aquatic plants (submerged, floating and emergent), outlining where phytoremediation processes are typically

observed. Following Fan et al. (2018); Fletcher et al. (2020) this figure was adopted, redrawn and modified

Example of plants

Site of action

Description

Phytoextraction/Phytoaccumulation Organic/Inorganic contaminants	Organic/Inorganic contaminants	Uptake by roots and translocation to upper parts. Uptake from water and air by leaves	Leaves	Juncus repens Water hyacinth
Rhizofiltration/phytofiltration	Organics/Inorganics including heavy metals	Organics/Inorganics including heavy Extraction from contaminated water Shoots/roots metals by adsorption/absorption	Shoots/roots	Lemna minor, Hydrocharis morsus, Eichhornia crassipes
Phytostabilisation/Phytoaccumula- tion/Phytosequestration	Heavy metals, Cd and Zn	Bioconcentration factors and translo- Roots cation factors are high	Roots	E. crassipes and P. stratiotes
Phytodegradation/Rhizodegradation Organics/Inorganics	Organics/Inorganics	Degradation Through microbial degradation or by metabolism within plant	Degradation in rhizosphere/pollutant Typha angustifolia, Typha minima, degraded in the plant to less harm- Phragmites australis, Myriophyll ful metabolite aquaticum (Parrotfeather)	Typha angustifolia, Typha minima, Phragmites australis, Myriophyllum aquaticum (Parrotfeather)
Phytovolatilization	Organic compounds	Conversation of containments to volatile form	Atmospheric release	Phragmites australis
Adopted from Dhir (2013), Rezania e	Adopted from Dhir (2013), Rezania et al. (2016), Sricoth et al. (2018), Fletcher et al. (2020) and Saleem et al. (2020)	her et al. (2020) and Saleem et al. (2020	6	
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## ccumulation

or organic chemicals, including biocides and antimicrobils, diffusion is widely accepted as the primary mechanism or in-plant accumulation, as plants do not have specific ransport systems in their cell membranes for these chemials (Patel et al. 2019). These chemicals are absorbed into he plant through passive uptake, primarily at the root surace (see rhizofiltration). Diffusion depends mainly upon he compound's hydrophobicity (i.e.,  $\log K_{ow}$  value). Comounds with  $\log K_{ow}$  values between 1 and 3.5 show modrate to high bioavailability to the roots of vascular plants ecause they have enough lipophilic character to move hrough cell membrane lipid bilayers and still have enough vater solubility to disperse through cell fluids once in the lant (Pilon-Smits 2005; Patel et al. 2019). The accumulaon of heavy metals can also occur via apoplastic pathways passive diffusion). However, heavy metals typically move cross the root membrane via active (pathway-dependent ependent processes, which are mediated, by metal ion cariers or a complexing agent (Yan et al. 2020a). Although ptake mechanisms are yet to be comprehensively defined, hree classes of membrane transporters have been detected nd implicated in transporting heavy metals across cell nembranes. It has been suggested these membrane transorters play a vital role in the phytoextraction and phytoacumulation of metals in plants. These membrane transportrs are mainly the heavy metal (CPx-type) ATPases (Solioz nd Vulpe 1996), the natural resistance-associated macophage protein (Nramp) family (Govoni and Gros 1998) nd members of the cation diffusion facilitator (CDF) family Williams et al. 2000; Paulsen and Saier 1997).

hytoextraction (or phytoaccumulation/phytoabsorption) the removal process of contaminants into harvestable lant tissues. Pollutants are taken up by plant roots, after which they are translocated to shoots, where they are deposted in the metabolically inactive parts of the plant tissue e.g., vacuole, cell wall) (Kafle et al. 2022). In the plant, netal cations form metal-phytochelatin complexes (M-PC) r metal-ligand complexes inside plant cells (Asgari ajayer et al. 2019), and these complex molecules can be eadily translocated to the plants' vacuole and stored (Yadav 2010). Genetic engineering offers a means of increasing the phytoextraction efficiency of plants by overexpressing genes whose protein products are involved in metal uptake, transport and sequestration (Cherian and Oliveira 2005).

**Rhizofiltration**, Known as phytofiltration, occurs in the root zone where contaminants are adsorbed/absorbed onto/into submerged plant organs (Dushenkov et al. 1995; Ansari et al. 2020; Olguín and Sánchez-Galván 2012). Given the significant role roots play in rhizofiltration, a suitable plant

Table 1 Common mechanisms involved in phytoremediation by aquatic plants

Contaminants

The mechanism in aquatic plants

for this is characterised by having a large rapid-growth root system (Mareddy 2017). The root environments or exudates create favourable biogeochemical conditions that can precipitated contaminants inside the aquatic plant's root in an insoluble form. For example, plants can filter Pb-contaminated water by the precipitation of Pb-phosphate in the root (Dushenkov et al. 1995). Bacteria that live inside the root or root surface have been shown to enhance the rhizofiltration process of heavy metals; for example, by reducing hexavalent chromium (Cr) (Cr-VI) into trivalent Cr (Cr-III). Bacteria can easily precipitate inside the plant root, thus maximising removal efficiency (Dimitroula et al. 2015). Following rhizofiltration, contaminants may remain in the root or be translocated to other plant organs. Rhizofiltration and phytoextraction are very similar processes in that they result in contaminant accumulation in the plant.

#### Immobilisation

**Phytostabilization**, or phytoimmobilization, occurs through the inactivation or immobilization of pollutants within the roots or the rhizosphere, reducing contaminant mobility (Ansari et al. 2020). This process has been widely documented for heavy metals, which can precipitate in the rhizosphere, be sequestered within root tissues or be adsorbed onto root cell walls (Yan et al. 2020a). The formation of bound residues in the roots or rhizosphere ensures that pollutants are not released from the matrix following accumulation and do not translocate to the shoots.

#### Degradation

Phytodegradation (or Phyto transformation) involves the transformation of pollutants within plant tissues. Plants can facilitate the complete removal of organic compounds such as antimicrobials or biocides (mineralization). Chemicals are transformed into inorganic products, such as carbon dioxide and water, by naturally occurring bacteria; for example, plants can break down organic chemicals into metabolites via processes such as phytodegradation (Ansari et al. 2020). The transformation of organic chemicals usually occurs in three stages and is driven by enzymatic processes (Macek et al. 2000; Geissen et al. 2015): (a) chemical modification (e.g., oxidation); (b) conjugation (e.g., with sugars or amino acids); and (c) sequestration or compartmentation where conjugates are deposited in plant vacuoles or bound to the cell wall and lignin (Zhang et al. 2014; Cherian and Oliveira 2005).

**Rhizodegradation** is the degradation of pollutants in the rhizosphere. Plant-associated microorganisms in the rhizosphere have been shown to degrade organic contaminants such as pesticides (Van Eerd et al. 2003). Emergent mac-

rophytes can supply oxygen to the root zone, thereby facilitating degradation processes in the rhizosphere. Hydrophobic compounds which do not typically translocate into the shoots can instead serve as a microbial carbon source and undergo degradation in the root zone (Fletcher et al. 2020). The degradation efficiency of plants has also been shown to be significantly improved using genetic engineering to develop transgenic plants capable of overexpressing bacterial enzymes, which can increase transformation efficiency (Cherian and Oliveira 2005).

#### Dissipation

**Phytovolatilization** involves the conversion of pollutants accumulated within a plant to a less toxic volatile form and subsequent release into the atmosphere by plant transpiration processes. Phytovolatilization primarily occurs following organic chemical accumulation, but it has also been shown to extract volatile elements such as selenium (Se) and mercury (Hg). However, releasing toxic metals into the atmosphere following phytovolatilisation raises questions regarding the suitability of this method for heavy metal remediation (Pang et al. 2023).

# **Phytoremediation of Common AMR Drivers**

## **Phytoremediation of Antimicrobials**

Phytoremediation offers a promising technique for removing pharmaceuticals, including antimicrobials, from water bodies (Mohebi and Nazari 2021; Mustafa and Hayder 2021; Maldonado et al. 2022b). A number of studies have documented the removal of a suite of antibiotics by aquatic plants (Table 2).

Different accumulation between plant species has also been observed in studies utilising similar exposure conditions to compare phytoremediation potential between species. For example, tetracycline antibiotics were very efficiently removed by the water lettuce, Pistia stratiotes, as almost all spiked oxytetracycline and tetracycline were removed in 6d. In comparison, it took parrot feather plants, Myriophyllum aquaticum 15d to reach a similar level of contaminant removal. A similar comparison of phytoremediation potential between P. stratiotes and M. aquaticum for commonly used antibiotics (norfloxacin, sulfamethazine, and tetracycline) was recently published (Park and Son 2022). Differences in accumulation potential between these plant species were also observed in this later study, with antibiotics only observed to accumulate in the plant roots of P. stratiotes.

In contrast, in the parrot feather plants (*M. aquaticum*), antibiotics were detected in both plant organs, with higher

Table 2	List of studied	plants according	to their remediated	antibiotics entities
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Antibiotics	Plants	References
Quinolones		
Ciprofloxacin	Eichhornia crassipes, Phragmites australis	(Hoang et al. 2013; Yan et al. 2020b; Carvalho et al. 2014; Liu et al. 2013b)
Norfloxacin	Lythrum salicaria, Acrostichum aureum, Pistia stratiotes	(Hoang et al. 2013; Park and Son 2022)
Difloxacin	Ceratophyllum	(Carvalho et al. 2014)
Enrofloxacin	Phragmites australis	(Carvalho et al. 2012)
Flumequine	Lemna minor, Lythrum salicaria	(Forni et al. 2001; Cascone et al. 2004; Migliore et al. 2000)
Tetracyclines		
Tetracycline	Spyrogia sp., Zannichella palustris, Pistia Stratiotes, Myriophyllum aquaticum, Phragmites australis, Azola Lemna. gibba and Azola. filiculoides)	(Garcia-Rodríguez et al. 2013; Gujarathi et al. 2005b; Carvalho et al. 2012; Park and Son 2022; Maldonado et al. 2022b)
Oxytetracycline	Myriophyllum aquaticum, Zannichellia palustris, Pistia stratiotes	(Garcia-Rodríguez et al. 2013; Gujarathi et al. 2005b)
Sulfonamides		
Sulfathiazole	Salvinia natans, Zannichellia palustris	(Garcia-Rodríguez et al. 2013)
Sulfamethoxazole	Scirpus validus, Zannichellia palustris, Lemna gibba	(Garcia-Rodríguez et al. 2013; Brain et al. 2008)
Sulfamethazine	Zannichellia palustris, Pistia stratiotes	(Garcia-Rodríguez et al. 2013; Park and Son 2022)
Sulfapyridine	Zannichellia palustris	(Garcia-Rodríguez et al. 2013)
Macrolides/ketolides		
Tylosin	Zannichellia palustris	(Garcia-Rodríguez et al. 2013)
Spiramycin	Pistia stratiotes	(Gujarathi et al. 2005b)
Josamycin	Landoltia punctate	(Carvalho et al. 2014)

amounts detected in the shoots than in the roots. However, interestingly, the water lettuce exhibited an overall higher uptake accumulation since BCF (bioconcentration factor) accumulated sulfamethazine (0.59–0.64) L/kg and tetracycline (0.72–0.78) L/kg compared to the parrot feather plant (Park and Son 2022). This is similar to where *P. stratiotes* were also observed to be much more efficient at tetracycline and oxytetracycline removal than *M. aquaticum* (Gujarathi et al. 2005b).

However, concerning antimicrobials, published studies have focused primarily on a select number of antibiotics, which neglects to consider other antimicrobials such as antivirals and antifungals. From the published data, the removal of antibiotics by aquatic plants (and possible degradation) depends on the compound, its bioavailability, and the plant. For example, studies have shown that the antibiotics flumequine and sulfadimethoxine can be removed/degraded by environmental factors. Still, higher removal was observed in the presence of plants (Lemna minor and Azolla filiculoides). Specifically, for sulfadimethoxine and flumequine, removal was < 73% and < 96% in the *L. minor* exposure and < 88% and < 96% in the A. *filiculoides* exposure, respectively. These studies also highlight the differences in accumulation between plant species with A. filiculoides responsible for overall antibiotic removal and the exposure medium concentration effect, with more significant chemical accumulation occurring at higher exposure concentrations (450 mg/L) (Forni et al. 2002; Cascone et al. 2004).

Similar exposure conditions were used in accumulation between different plant species in studies to compare phytoremediation potential between species. For example, tetracycline antibiotics were very efficiently removed by the water lettuce, Pistia stratiotes, as almost all spiked oxytetracycline and tetracycline were removed in 6d. It took the parrot feather plants, Myriophyllum aquaticum, 15 d to reach this result (Gujarathi et al. 2005b). With both species, the tetracycline removal followed first-order kinetics with a significant depletion in the first 24 h (Gujarathi et al. 2005b). A similar comparison between P. stratiotes and M. aquaticum was recently made where the phytoremediation of commonly used antibiotics in South Korea, norfloxacin, sulfamethazine, and tetracycline, was assessed (Park and Son 2022). In the water lettuce (P. stratiotes), antibiotics were detected only in the roots. In contrast, in the parrot feather plants (M. aquaticum), antibiotics were detected in both plant organs, with higher amounts detected in the shoots than in the roots. However, interestingly, the water lettuce exhibited an overall higher capacity to accumulate the BCF (Bioconcentration Factor) of antibiotics ranging from sulfamethazine (SMZ) (0.59–0.64) and tetracycline (TET) (0.72–0.78) compared to the parrot feather plant (Park and Son 2022). This is similar to the results where P. stratiotes were also observed to

be much more efficient at tetracycline and oxytetracycline removal than *M. aquaticum* (Gujarathi et al. 2005b).

The capacity comparison for erythromycin removal between free-floating (*Salvinia molesta* and *L. minor*) and submerged macrophyte species (*M. aquaticum* and *Rotala rotundufolia*) has also been studied. (Rocha et al. 2020). Erythromycin depletion was observed after 7 d exposure to erythromycin-spiked growth media (0 and 1.7  $\mu$ g/L) in the presence of the free-floating and submerged plants. When antibiotics were added to the water, more was removed in plants fully submerged (31–44%) compared to plants that floated on the surface (9–12%). This was because the submerged plants had a greater measured concentration of erythromycin (an antibiotic) than the floating plants (Rocha et al. 2020).

In addition to L. minor, one of the most widely studied aquatic macrophytes in understanding phytoremediation potential is Eichhornia crassipes, commonly known as water hyacinth. In a recent study, the uptake of antibiotics under hydroponic conditions was investigated in E. crassipes at both the seedling and mature stages (Yan et al. 2021). Ciprofloxacin measured in roots at the seedling and mature stages was  $< 2114.39 \ \mu g/g$  and  $< 3711.33 \ \mu g/g$ , respectively, indicating mature plant has a more significant potential to accumulate ciprofloxacin in the roots. The aerial parts of the plant also accumulated ciprofloxacin to a greater extent in the seedling stage, with concentrations of ciprofloxacin ranging between 16.4–24.2  $\mu$ g/g and 9.5–20.1  $\mu$ g/g in the seedling and mature stages respectively (Yan et al. 2021). This study highlights the importance of considering the age of the plant when evaluating its phytoremediation potential, as this appears to affect the location and extent of chemical accumulation in the plant. Interestingly, this study also demonstrated that E. crassipes could facilitate phytodegradation and the breakdown of the parent compound as eight and ten major metabolic products of ciprofloxacin were observed in the plant tissues at the seeding and mature stages, respectively (Yan et al. 2021).

As discussed, organic compounds such as antimicrobials can undergo a chemical transformation in the plant following the uptake and accumulation, where new transformation products are produced (Fu et al. 2019). Eight transformation products of ciprofloxacin have been identified following five transformation potentials possible transformation pathways: demethylation, dehydroxylation, oxidation, hydroxylation and cleavage processes of the piperazine and quinoline rings in another study using *E. crassipes* (Yan et al. 2020b). This study also revealed that the majority of ciprofloxacin accumulated in the root. The potential for a chemical transformation potential for a chemical is a necessary process to consider as it highlights the potential for plants to remove the parent compound. Still, the subsequent identification of metabolites with retained biological potency demands further evaluation as these metabolites could pose an additional environmental risk (Yan et al. 2020b, 2021).

## **Phytoremediation of Metals**

Aquatic plants remove heavy metals via absorption or surface adsorption, where they can accumulate within the plant in certain bounded forms (Rai et al. 1995; Sas-Nowosielska et al. 2008; Bhat et al. 2022). A wide array of aquatic plants like water hyacinths, Salvinia sp., water lettuce (P. stratiotes), giant duckweed (S. polyrhiza), and Azolla sp. have displayed significant ability for the phytoremediation of heavy metals (Soda et al. 2012; Rodríguez and Brisson 2015). Studies have primarily focused on the role of aquatic macrophytes in constructed wetlands to remove heavy metals from aqueous media; these examples are provided in Table 3. Given the widespread presence of heavy metals in municipal wastewater, laboratory mesocosms and constructed wetlands have been set up to explore the phytoremediation of heavy metals by aquatic plants in this specific scenario (Pedescoll et al. 2015; Sasmaz and Obek 2009). For example, Liao The and Chang (2004) reported water hyacinth as a promising candidate for phytoremediation of heavy metal-polluted wastewater with the concentration of Cu, Pb, Zn and Cd in the roots reported at 3-15 times higher than the shoots (Liao and Chang 2004). Southern cattail (Typha domingens) also showed maximum accumulation of Zn, Al, Fe, and Pb in the below-ground plant parts (roots/rhizomes) in comparison to the above-ground shoots (Hegazy et al. 2011) with the ability of the roots to accumulate heavy metals in the following order: Pb>Fe>Al>Zn. The same species, T. domingensis, has also been shown to be a useful tool for removing Cd, Ni, and Mn from municipal wastewater (in addition to Zn and Fe), with maximum accumulation occurring during the first 48 h (Mojiri 2012). This study also observed that the accumulation of heavy metals in roots was higher than in shoots, which is in line with the findings of Mojiri (2012).

In-situ field trials and laboratory mesocosms have also been used to evaluate the phytoremediation potential of aquatic plants to remove heavy metals from contaminated industrial wastewater, given the widespread use of heavy metals in smelting operations, mining and the textiles industry (Li et al. 2016; Pat-Espadas et al. 2018; Mugisa et al. 2015). Specifically, research has demonstrated that E. crassipes exhibited remarkable efficiency in phytoremediation, achieving a removal rate of over 94% for Cr, Zn, and Cu within a 96 h period. This was observed in industrial wastewater sourced from five distinct textile industries in the Lahore district of Pakistan (Mahmood et al. 2005). In addition to E. crassipes, aquatic plants such as Pistia stratiotes, Azollapinnata, S. polyrhiza, L. minor, and Salvinia molesta have also been found to show great potential for the removal of heavy metals from textile wastewater (Manjunath and

Aquatic Plant	Common names	Metals	References
Eichhornia crassipes	Water hyacinth	Pb, Hg, Cu, Cr, Ni, Zn	Molisani et al. (2006); Hu et al. (2007)
Pistia stratiotes	Water lettuce	Cr, Zn, Fe, Mn, Cu	Miretzky et al. (2004)
Salvinia minima	Water spangles	As Ni, Cr, Cd	Sooknah (2000)
Salvinia herzogii	Water fern	Cd, Cr	Suñe et al. (2007)
Lemna minor	Duckweed	Cr, As, Ni, Cu, Pb	Kara (2004)
Nasturtium officinale	Water cress	Cr, Ni, Zn, Cu	Kara (2004); Zurayk et al. (2001)
Myriophyllum spicatum	Parrot feathers	Pb, Cd, Fe, Cu	Ridvan Sivaci et al. (2004); Branković et al. (2012)
Ceratophyllum	Demersum hornwort	As, Cd, Cr, Pb	Bunluesin et al. (2004); El-Khatib et al. (2014)
Potamogeton	Crispus pondweed	Cu, Fe, Ni, Zn, and Mn	Borisova et al. (2014)
Potamogeton pectinatus	American pondweed	Cd, Pb, Cu, Zn	Singh et al. (2005); Peng et al. (2008a)
Typha latifolia	Common cattail	Zn, Mn, Ni, Fe, Pb, Cu	Hejna et al. (2020); Qian et al. (1999) Sasmaz et al. (2008)
Mentha aquatica	Water mint	Pb, Cd, Fe, Cu	Branković et al. (2012); Kamal et al. (2004)
spartina alterniflora	Cordgrass	Cu. Cr, Zn, Ni, Mn, Cd, Pb, As	Aksorn and Visoottiviseth (2004); Hempel et al. (2008)
Phragmites australis	Common reed	Fe, Cu, Cd, Pb, Zn	Ganjali et al. (2014); Ha and Anh (2017)
Scirpus	Bulrush	Cd, Fe, Al	Kutty and Al-Mahaqeri (2016)
Polygonum hydropiperoides	Smartweed	Cu, Pb, Zn	Rudin et al. (2017)

 Table 3
 Heavy metal accumulation potential of various aquatic plants

Kousar 2016; Rolli et al. 2007; Kumar et al. 2019; Sekomo et al. 2012). Aquatic plants S. polyrhiza, E. crassipes and L. minor were all observed to eliminate heavy metals from wastewater. Still, E. crassipes was the most efficient overall, a much higher percentage (71, 69, 77%) of Fe, Cr, and Cu, respectively (Mishra et al. 2008). During a 15d exposure, free-floating E. crassipes demonstrated remarkable effectiveness in removing approximately 99.5% of Cr (VI) from industrial mine effluents. Additional species, such as P. australis, P. karka, and T. dominguensis, are used to remove heavy metals from contaminated mine effluents specifically (Saha et al. 2017; Türker et al. 2013; Younger and Henderson 2014). Further research also supports the concentration of heavy metals in the roots of emergent plants followed by leaves and stems in a constructed wetland receiving refinery wastewater (Cyperus alternifolius and T. latifolia) (Mustapha et al. 2018).

In a large-scale evaluation of the phytoremediation potential of twelve aquatic plants in wastewater collected from the Swabi district, constructed wetlands were influential in the removal of heavy metals with removal efficiencies reported in the order of Cd > Cr > Fe > Pb > Cu > Ni and ranging between 74 and 92% for Cr, Fe and Cd specifically (Khan et al. 2009). However, *T. latifolia*, *P. stratiotes*, *P. australis*, *C. aquatilis* and *A. plantago-aquatica* were more efficient in removing heavy metals from the wastewater, and no relationships between plant species and removal efficiency were observed (Githuku et al. 2018). This highlights the variable phytoremediation potential of different plants, which has also been documented in several other studies. Aquatic macrophytes *Marsile aquadrifolia*, *Hydrilla verticillata* and *Ipomea* aquatic showed much better accumulation potential and translocation factor value for heavy metals (Zn, Al, Fe, Pb, Cr, As, Hg, Cd, Cu) as compared to the algal species (*Phormidium papyraceum, Spirulina platensis*) (Ahmad et al. 2011). Maine et al. also reported that *T. domingensis* showed much better survival and removal efficiency than *Salvinia herzogii* for Fe, Zn, Ni, and Cr released from industrial wastewater of a metallurgy plant.

Lack of proper landfill management can release heavy metals in landfill leachate, which risks the environment (Njoku et al. 2019). Chemical and physicochemical approaches to eradicate pollutants from leachate are generally expensive and complicated (Kamaruddin et al. 2015). However, water hyacinth's (E. crassipes) ability to remove five heavy metals (Cd, Cr, Cu, Ni, and Pb) is commonly found in landfill leachate. (El-Gendy et al. 2006). The experiment, conducted in batch reactors in a greenhouse, demonstrated that the living biomass of water hyacinth was a good accumulator for Cu, Cr, and Cd. However, Pb and Ni were poorly accumulated. Comparatively, the non-living biomass of water hyacinth (dry roots) could accumulate all metals except Cr (VI) in its anionic form. Total metal sorption by non-living dry water hyacinth roots was found to be pH specific, with maximum accumulation occurring at pH 6.4 (El-Gendy et al. 2006). In another study, (Abbas et al. 2019) investigated the phytoremediation potential of water hyacinth and water lettuce in landfill leachate for 15 d with an experimental setup where aquatic plants were fitted as a floating bed with the help of a thermopole sheet. Both plants

significantly reduced the concentrations of Zn, Pb, Fe, Cu and Ni from the landfill leachate and the physicochemical parameters (pH, BOD, COD). The removal rate gradually improved from day 3 to 15 of the experiment. The maximum removal of Zn (80–90%), Fe (83–87%) and Pb (76–84%) was observed (Abbas et al. 2019). Studies have also shown that *L. minor* can significantly reduce concentrations of heavy metals (Cu, Zn, Pb, Ni, and Fe) in landfill leachate from Pakistan (Daud et al. 2018).

#### **Phytoremediation of Biocides**

Biocides Directive (98/8/EC) covers 23 product types, including drinking water disinfectants, wood preservatives and insecticides to antifouling products. For this review, only the most commonly used biocides are discussed here in the context of phytoremediation, namely pesticides, paint as an anti-fouling agent and wood preservatives (Table 4). Pesticides are frequently detected in water bodies and include a range of substances to control pests, e.g., herbicides and insecticides (Olette et al. 2008). The uptake capacity of aquatic plants L. minor, Elodea Canadensis and Cabomba aquatica were observed for three pesticides: copper sulphate (fungicide), flazasulfuron (herbicide) and dimethomorph (fungicide), with L. minor, demonstrated to have the most efficient uptake capacity, followed by E. canadensis and then C. aquatica (Olette et al. 2008; Bhalla et al. 2022). The maximum removal rate of copper, flazasulfuron and dimethomorph was 30, 27 and 11  $\mu$ g/g fresh weight/ d, respectively (Olette et al. 2008). However, this study also observed pesticide toxicity, using chlorophyll fluorescence as a biomarker in the order of flazasulfuron > Cu > dimethomorph (Olette et al. 2008). Bouldin et al. (2006) also observed differences in accumulation between plant species and that some plants had a greater affinity for certain chemicals in their study, which evaluated the uptake of atrazine and lambda-cyhalothrin by two other plants (Juncus effusus and Ludwigia peploides) under hydroponic conditions. J. effuses showed higher atrazine uptake, whilst greater lambda-cyhalothrin uptake occurred in L. peploides. Atrazine was translocated to upper plant biomass in the macrophytes, while 98.2% of lambda-cyhalothrin was sequestered in the roots of *L. peploides* (Bouldin et al. 2006).

Similarly to Bouldin et al. (2006), Riaz et al. (2017) observed that some aquatic plants and algae were more effective than others in removing organochlorine and pyrethroid pesticides from the water. Among the plants tested, *E. crassipes, P. strateotes*, and certain types of algae (*C. sutoria, S. sticticum*, and *Zygnema* sp.) were found to be highly effective at removing both organochlorine and pyrethroid pesticides from water. The removal efficiency of these plants was higher for pyrethroids (68–76%) than for organochlorine pesticides (58–62%), and the difference was statistically significant (p < 0.01) (Riaz et al. 2017). This study also observed differences in pesticide distribution within the plant, with greater accumulation typically occurring in the roots with minimal translocation to the shoots (<76% root, <33% shoot). Turgut et al. (2005) observed differences in the accumulation of pesticides within the plant organs following a study to investigate the uptake of pesticides by parrot feathers (*M. aquaticum*) (Turgut 2005). Interestingly, root concentration factor (RCF) and submerged shoot concentration factor (SSCF) increased with increasing hydrophobicity ( $K_{ow}$ ) of the pesticide.

Turgut (2005) observed greater atrazine and cycloxidim accumulation by the roots than by shoots compared to other pesticides used in this study. Aquatic plants can also readily take up pesticide metabolites which are often weak electrolytes and formed following chemical or biological transformations in the environment (Turgut 2005). Similarly to antimicrobials, there is also the potential for pesticides to transform in the plant whereby in-plant processes can transform the parent compound. Ando (2020) evaluated the translocation and metabolism of a model compound 3-phenoxybenzoic acid in water milfoil (Myriophyllum elatinoides) with further investigation of the behaviour and metabolic pathways of the herbicide flumioxazin in two algae (Pseudokirchneriella subcapitata and Synechococcus sp.), duckweed (Lemna sp.), and the water milfoil. Results suggested that the pesticides underwent rapid decomposition in the water, and following uptake, significant constituents included the formation of glucose and GSH conjugates via phase II reactions (Ando 2020). Curly leaf pondweed (Potamageton crispus L.), common duckweed (L. minor), and their epiphytic microbes have been shown to contribute to the removal and degradation of pentachlorophenol from a stream in the range of 55% to 74% (Pignatello et al. 1985). However, following this early study, limited phytoremediation studies have demonstrated in-plant metabolism of pesticides following plant uptake (Ando 2020).

#### **Phytoremediation of Chemical Mixtures**

As reviewed by Rathi et al. (2021b), the aquatic environment contains a complex mixture of hazardous contaminants, including organic and inorganic chemicals. AMR drivers are, therefore, not present in isolation, and to assess realistic environmental exposure, an understanding of the phytoremediation potential of combinations of AMR drivers is needed. An assessment of the potential influence of mixtures of pollutants from different chemical classes on phytoremediation potential has previously been considered concerning soil (Zhu et al. 2022; Ma et al. 2016) and constructed wetland systems (Guo et al. 2020b). For example, the antibiotic ceftiofur has been shown to improve metal uptake by *P. australis* while not adversely impacting plant

lable 4 Studied aquatic plants for the phytoremediation of pesticides	oremediation of pesticides		
Pesticides	Plant species	Comments	References
Atrazine, cycloxidim, terbutryn, trifluralin Myriophyllum aquaticum	Myriophyllum aquaticum	The uptake of atrazine and cycloxidim was more significant than terbutryn and triffuralin	(Wilson et al. 2000; Anderson and Coats 1994; Anderson and Coats 1995)
Metolachlor, atrazine	Ceratophyllum demersum L. Elodea canadensis	Within the first 16 days, both plants demonstrated removal with $> 90\%$ of metolachlor and atrazine metabolised	(Rice et al. 1997)
Malathion, demeton-S-methyl, crufomate	Myriophyllum aquaticum, Spirodela oligorrhiza L., Elodea canadensis	58–83% of pesticides removed by <i>M. aquaticum.</i> A lag phase was observed for the first few days for <i>S. oligorrhiza</i> L. and <i>E. Canadensis,</i> followed by rapid extraction of the pesticides	(Gao et al. 2000)
Oryzalin	Canna × generalis (Canna), Pontaderia cordata L, Iris × germanica (Iris)	Growth reductions and photosynthetic parameters due (Fernandez et al. 1999) to oryzalin were minimal for all plants indicating these plants would be useful in phytoremediation systems where oryzalin is present	(Fernandez et al. 1999)
DDT	Myriophyllum aquaticum, Spirodela oligorrhiza, and Elodea canadensis	After 6-day incubation, almost all of the DDT was removed from the medium, and most of it accumu- lated in or was transformed by these plants	(Gao et al. 2000)
Simazine	Acorus gramenius (Sweet flag) and Pontederia cor- data (Pickerel weed)	Uptake in the transpiration stream was observed	(Wilson et al. 2000)
Simazine	Myriophyllum aquaticum and Canna x hybrida L	Uptake and conjugation to glutathione using glutathione-S-transferases were observed	(Knuteson et al. 2002)
Halogenated pesticides	Myriophyllum aquaticum and Elodea canadensis	Uptake and in-plant metabolism observed	(Nzengung and Jeffers 2001)
Organo-phosphate pesticides	Parrot feather (Myriophyllum aquaticum), Duckweed (Spirodela oligorrhiza), Elodea (Elodea canadensis)	The tolerant and enzymatic transformation was demonstrated	(Gao et al. 2000)

 Table 4
 Studied aguatic plants for the phytoremediation of pesticides

growth (Almeida et al. 2017a). However, we lack similar data for phytoremediation potential in co-contaminated aquatic systems. A mixed culture of tailored endophytic bacteria has been shown to enhance the phytoremediation of co-contamination of antibiotics (ciprofloxacin and sulfamethoxazole) and heavy metals (Zn, Ni, Cd) by the aquatic macrophyte J. acutus (Syranidou et al. 2016). However, as this study focused on the role of inoculated plants in aiding phytoremediation potential, the authors did not compare single chemical exposure and the accumulation resulting from the combined exposure to metals and antibiotics. In a more recent study, the aquatic plant Iris pseudacorus, was exposed to the pesticide atrazine together with Cd; however, this was carried out to understand the potential impacts on phytotoxicity and accumulation in the plant was not considered (Wang et al. 2022).

## **Microbial Assisted Phytoremediation**

Interactions between aquatic macrophytes and microbial biofilm communities around the aquatic plant largely depend upon the mutual supplies of nutrients. Microbes receive organic carbon and oxygen from the plant. Plants receive defensive immunity and essential minerals (Davey and O'Toole 2000). Microbial assemblage as a biofilm commonly occurs on the leaves of submerged plants, rhizosphere, especially on rhizoplane and the solid surfaces of sediments. In addition to the mutual benefits, plant-microbe interactions also influence the water quality, especially at the rhizosphere, by mitigating pollution from the water column (Srivastava et al. 2017). Microbe-assisted phytoremediation has gained attention in the past decade, with research aiming to establish microbes in treating contaminated water bodies to maximise phytoremediation efficiency. However, research has focussed mainly on the role of microbial-assisted phytoremediation in terrestrial plants, with microbes identified as bioremediation of various soil contaminants, including metals, pesticides, and hydrocarbons (Kumar et al. 2022). In aquatic systems, a majority of research has focused on using bacteria in constructed wetland systems to treat contaminated effluent; adding specific bacteria to various plant species has been shown to boost phytoremediation potential, for example (Riva et al. 2020).

In the aquatic environment, published research has primarily focussed on microbial-assisted phytoremediation's role in removing inorganic contaminants from polluted water bodies (Srivastava et al. 2017). The microbe Bacillus cereus has been shown to enhance Cr(VI) uptake by *Pistia stratiotes* (Chakraborty et al. 2013) and also enhance Mn uptake in *Eichhornia crassipes* roots (Abou-Shanab et al. 2007). Studies have also shown that *Nitrobacteria irancium* and *Ochrobactrum* anthropic microbes enhance Cr and Zn uptake in aerial parts of *E. crassipes* and enhance Cr and Mn uptake in E. crassipes roots (Abou-Shanab et al. 2007). The addition of specific bacteria to plant species can promote plant growth and improve the removal of heavy metals from the water body and therefore offers a promising strategy to boost phytoremediation efficiency (Prum et al. 2018). Microbial consortia, where microbes function synergistically, have also been observed to degrade heavy metals more efficiently than single bacterial strains due to the presence of exopolysaccharides, which help form biofilms. However, this has primarily been demonstrated in terrestrial systems. For example, a mixture of Viridibacillus arenosi B-21, Sporosarcina soli B-22, Enterobacter cloacae KJ-46 and E. cloacae KJ-47 was found to be more efficient in the bioremediation of Pb, Cd and Cu contaminated soil than an individual bacterial culture (Kang et al. 2016); further work is needed to explore the benefits of microbial consortia in aquatic systems.

As well as using microorganism-assisted phytoremediation to degrade heavy metals, the catabolic activity of microbes makes them ideal bioremediation with the ability to degrade virtually all classes of organic chemicals, including complex recalcitrant organic compounds such as surfactants (Mori et al. 2005). Specifically, with respect to chemicals which are also identified as AMR drivers, bacteria in combination with the aquatic plants, Acrostichum aureum and *Rhizophora apiculata* have been shown to play a pivotal role in the phytodegradation of antibiotic-contaminated sediments (Hoang et al. 2013). Studies that have demonstrated the role of plants in microbial-assisted phytoremediation of antimicrobial and biocide-contaminated waters are largely focused on constructed wetland set-ups (Chen et al. 2012; Christofilopoulos et al. 2019; Riva et al. 2020). One of the first studies to isolate and compare culturable endophytic bacteria, which were responsible for degrading pesticides such as chlorpyrifos and fenpropathria in different aquatic plants (Phragmites communis, Potamogeton crispus, Nymphaea tetragona and Najas marina) (Chen et al. 2012). More recent research has demonstrated that inoculating the wetland plant Juncus acutus with indigenous endophytic bacteria increases the removal of the antibiotic ciprofloxacin (Syranidou et al. 2016). At the end of the experiment, more than 79% removal of CIP was established by all inoculated plants compared to 73.5% removal by the non-inoculated plants (Syranidou et al. 2016).

## Phytotoxicity

Phytoremediation success depends on successful and fastgrowth plant species. This conserved biological potency of antimicrobials and biocides following plant uptake can result in a series of in-plant phytotoxic responses, posing a risk to phytoremediation efficiency (Table 5). For example, phytotoxicity occurred after exposure of the duckweed Spirodela polyrhiza to a range of environmentally relevant (0.0001-0.01 mg/L) and high (0.1 and 1 mg/L) concentrations of the antibiotic amoxicillin with reported impacts on growth, pigments, and antioxidative enzyme activity (catalase, CAT; superoxide dismutase, SOD; and ascorbate peroxidases, APX). Specifically, the high dose (1 mg/L) of amoxicillin caused a significant (p < 0.05) decrease in photopigments, protein, starch and lipid content and an increase in carotenoids/total Chl and Chl a/Chl b ratios in fronds of S. polyrhiza (Richter et al. 2016). Laboratory experiments have also demonstrated the effects of sulfadimethoxine and flumequine, widely used in intensive farming operations, on post-germinative development in various plant species, including Azolla, Lythrum and Lemna spp. Nevertheless, although toxic effects were observed, the three plants maintained the capacity to accumulate the antibiotics, with a high survival rate even at very high-exposure concentrations (Forni et al. 2001). Research so far suggests that whilst phytotoxic responses may be observed, this appears not to impact the long-term phytoremediation potential of macrophytes. For example, exposure to the antibiotic ciprofloxacin has been observed to change the soluble protein growth rate of E. crassipes leaves, with longer-term exposure resulting in increased soluble protein content in the leaves. However, results from this study also indicated that changes in activities of certain enzymes could maintain normal cellular metabolism of the plant under 'stressed' (i.e., contaminant exposure) conditions and therefore, overall growth of the plant was not inhibited, and phytoremediation efficiency was maintained (Yan et al. 2019b).

Limited studies are available which evaluate the phytotoxicity of biocides to understand how this impacts phytoremediation capacity. One study evaluated the capacity of four treatment wetland macrophytes, Phalaris arundinacea, T. angustifolia, and two subspecies of Phragmites australis, to treat leachate-containing wood preservatives (pentachlorophenol (PCP) and chromate copper arsenate (CCA)) whilst accounting for any potential toxicity (Demers et al. 2020). Following 70 d exposure across three concentrations, chlorinated phenols accumulated in belowground plant parts. The exposure did not significantly affect plant biomass for any species. Comparatively, more published reports are available on the phytotoxicity of heavy metals and subsequent impacts on the growth and development of aquatic macrophytes (Table 5). For example, Cu toxicity has been observed to lead to failure in photosynthesis, affecting plant growth and survival of the aquatic macrophytes Potamogeton pectinatus (Costa et al. 2018). Three aquatic macrophytes (Lemna minor, Elodea canadensis, and the moss Leptodictyum riparium) were considered good accumulators of heavy metals (Cd, Pb, Zn, Cu). However, at the ultrastructural level, accumulation of these contaminants resulted in induced cell plasmolysis and alterations of the chloroplast arrangement (Basile et al. 2012b). Research has also shown that metal hyper-accumulating plants typically have metal tolerance mechanisms (detoxification and exclusion) which help the plants cope with the toxic effects of metal ions at elevated concentrations (Lasat 2002). Therefore, whilst several studies have observed phytotoxic effects, the ability of the aquatic macrophytes to accumulate heavy metals remains relatively impaired (Drost et al. 2007; Basile et al. 2012a; Nguyen et al. 2021).

## Phytoremediation and AMR

As highlighted above, the sediment matrix and the plant roots provide a surface for developing microbial communities, which offer beneficial effects regarding nutrient exchange and the degradation of chemical pollutants. However, as also discussed, exposure to AMR drivers, including antibiotics, for example, can result in changes to microbial function and structure linked to the emergence and propagation of AMR. Therefore, there is the potential that chemicals, which aquatic macrophytes are trying to clean up and remove, can instead create a selection pressure on microbes and significantly increase the abundance of ARGs (Ohore et al. 2022). Following the acquisition of ARGs, dissemination across plant microbes is facilitated by mobile genetic elements such as integrons and plasmids (e.g., horizontal gene transfer events) (Berglund 2015), ultimately allowing for the spread of antibiotic resistance.

Regarding phytoremediation, several studies have investigated the role of constructed wetlands in the propagation of resistant genes. Evidence has shown that whilst constructed wetlands can remove high levels of antibiotics (e.g., <93% of ciprofloxacin), the treatment process is also responsible for fluctuations in the antibiotic resistance profile of bacteria and increased levels of resistance genes in the effluent (Christofilopoulos et al. 2019). Other AMR drivers, such as biocides and heavy metals, can also contribute to the enrichment of antibiotic-resistant genes and bacteria in constructed wetlands through selection or co-selection events (Hazra et al. 2022; Zhang et al. 2023). Conversely, constructed wetlands can also limit the diffusion of ARGs and ARB by removing AMR drivers from the wastewater via different mechanisms, such as biodegradation. For example, plant uptake in constructed wetlands can result in enhanced ARG removal compared to other treatment options, such as UV disinfection (Chen et al. 2016; Liu et al. 2013a). Aquatic macrophytes used to treat contaminated waters could therefore be considered effective bioremediation by decreasing the absolute abundance of ARGs, or on the other hand, regarded as "hotspots" or reservoirs of ARGs (Riva et al. 2020). Nevertheless, as an interface between plants and the environment, the aquatic plant microbiome has the potential to play an essential role in the dissemination dynamics of

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AMR driver	Plants	Phytotoxic effect	References
Amoxicillin	Spirodela polyrhiza	At 1 mg/L concentration, amoxicillin accumu- lation resulted in a significant reduction in photopigments	Singh et al. (2018)
Lincomycin	Medicago sativa	10 μg/L was observed to be toxic for root growth, 10,000 μg/L was observed to be toxic for root growth	Kalaji and Rastogi (2017)
Oxytetracycline	Phragmites Autralis	> 10 µg/L concentration resulted in a toxic effect on root and photosynthetic activities. Plant lethality was observed at 160 mg/L concentra- tion	Boelsterli (2003); Stirnimann et al. (2010); Kalaji and Rastogi (2017)
Sulfamethazine	Phragmites autralis Medicago sativa	> 10 µg/L concentration causes a toxic effect on root and photosynthetic activities, and 10,000 µg/L was toxic for root growth	Susarla et al. (2002); Teixeira et al. (2015); Kalaji and Rastogi (2017)
Sulfamethoxazole	Medicago sativa	10,000 μg/L observed to be toxic for root growth	Teixeira et al. (2015; Kalaji and Rastogi (2017)
Sulfactificthoxine	Amaranınus reiropexus	our mg/L was mgnly toxic during post-germina- tive development	teixeira et al. (2013); Kataji and Kastogi (2017)
Trimethoprim	Medicago sativa	Toxic for root growth at high (> 10 mg/L) con- centration	Teixeira et al. (2015); Kalaji and Rastogi (2017)
Tylosin	Medicago sativa	10 $\mu$ g/L was observed to be toxic for root growth	Teixeira et al. (2015); Kalaji and Rastogi (2017)
Cu, Cd	Lemna minor	Cd showed the highest toxicity to Lemna minor	Drost et al. (2007)
Fe, Mn, Cu, Cd, Zn, Cr, Pb	Ipomea sp., Eclipta sp., Marselia sp., Alternanthera sp., and Typha sp.	Significant reductions observed in total chlo- rophyll and soluble sugar with an increase in protein and proline content	Nayek et al. (2010)
Pb	Phragmites Autralis	Chlorophyll synthesis inhibition, chlorophyll reduction, loss of photosynthesis activity	Vesely et al. (2012)
ïN	Phragmites Autralis	Plant wilting, chlorosis in leaves, chlorophyll reduction, carotenoid reduction, water loss, browning of root tips, and root damage	Singh and Pandey, (2011)
Cd, Cu	Lenna minor	Pigment degradation and photosynthesis restric- tion	Miretzky et al. (2004)
Cd, Zn	Eichhornia crassipes	Growth reduction, retardation, new root growth inhibition, root function disruption, leaf chlo- rosis	Hasan et al. (2007)
Cr	Eichhornia crassipes	Yellowing of leaves, leaf chlorosis, and growth retardation	Mishra and Tripathi, (2009)
Biotin T and Preventol RI80	Evernia prunastri Brachythecium sp	Severe impairment of the photosynthetic appara- tus and the absence of recovery (up to 21 days)	Vannini et al. (2021)
2,4 dichlorophenoxyacetic acid (2,4-D), glyphosate, hexazinone, imazapyr, metsulfuron methyl, sulfometuron methyl	Myriophyllum sibiricum	Species sensitive to root growth (inhibitory con- centrations reported). 2,4-D, imazapyr, and sul- fonylureas were more toxic to the macrophyte	Roshon et al. (1999)

 Table 5
 Demonstrated phytotoxicity following exposure to antibiotics and metals

Wang et al. (2022)

The coexistence of Cd alleviated the individual phytotoxicities of atrazine, whereas combined

pollution of atrazine and Cd still induced the decline in the photosynthetic performance of

oseudacorus

References

Phytotoxic effect

Iris pseudacorus

Plants

AMR, and this is something which we know little about. Based on previously published research considering the fate of AMR drivers, ARGs and ARB in constructed wetlands, an evaluation of the role of gene exchange during phytoremediation in aquatic systems is needed to understand if these processes can promote the diffusion of ARGs and ARB, with the potential to enter on the broader environment.

# The Future of Phytoremediation to Remove AMR Drivers from Aquatic Systems

As demonstrated by this review, chemicals can accumulate to different extents in different plants (Table 6). Hence, it is vital to customize the phytoremediation approach according to the specific chemicals being targeted and the environmental conditions in the vicinity (Yan et al. 2020a; Sabreena et al. 2022). For example, free-floating plants would be ideal phytoremediation candidates in a shallow water body contaminated with heavy metals as they have been demonstrated to be more efficient in the uptake of heavy metals compared to submerged and emergent plants due to their specific morphology and higher growth rate (Table 6) (Rezania et al. 2016). However, as these plants are typically characterised by a shallow root system (e.g., Lemna minor), the demonstrated efficiency in pollutant removal must be considered alongside the limited potential to remove contaminants from deep-water bodies. The need for multiple species to maximise pollutant removal efficiency is further supported by the fact that chemicals may exist in a dissolved phase and the particulate phase suspended in the water or adsorbed sediments. Therefore a suite of different aquatic macrophytes may be needed to remove contaminants from aquatic systems (Perk 2006). The ideal configuration of plant species for optimum phytoremediation is also going to depend on where the treatment needs to take place, as additional factors such as pH, solar radiation, nutrient availability, and salinity greatly influence the plant's growth and, therefore, the phytoremediation potential (Cunningham and Ow 1996; Tewes et al. 2018).

Ideal candidates for phytoremediation are native plants with a quick growth rate, high biomass yield, and the capacity to accumulate contaminants and transport them to aboveground parts of the plant. As AMR drivers also have the potential to induce phytotoxicity, a mechanism to tolerate chemical toxicity is also important (Cunningham and Ow 1996; Ali et al. 2013). Figure 3 demonstrates an example phytoremediation configuration to maximise the removal of heavy metals, antibiotics and biocides from a polluted water body receiving industrial wastewater. The combined use of floating submerged and emergent plants is suggested to facilitate the optimum removal of AMR drivers considered in this review. Plants were selected based on having high-demonstrated accumulation capacity of the three AMR

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AMR driver Atrazine, Cd

	of frequently studied aquatic plants f	Table 6 Comparison of frequently studied aquatic plants for the phytoremediation of heavy metals, antimicrobials and biocides	ls and biocides	
<ul> <li>Azolla filiculoides • HM: Cd, Cu, Pb</li> <li>AB: sulphadimethox- ine, Tetracyclines and Chloramphenicol</li> <li>Bi: Cong Red Dye, diazinon and feni- trothion (organophos- phorus pesticides)</li> <li>TI <i>Sipes</i></li> <li>HM: Pb, Hg, Cu, Ni, sipes</li> <li>AB: ciprofloxacin, sulfadiazine</li> <li>Bi: organophosphorus</li> <li>Pesticides ethion</li> </ul>	of aquatic	Observations	Bio concentration factor (BCF) (where reported)	References
<ul> <li><i>Eichhornia cras-</i> • HM: Pb, Hg, Cu, Ni, sipes</li> <li>Zn, Cr</li> <li>AB: ciprofloxacin, sulfadiazine</li> <li>Bi: organophosphorus pesticides ethion</li> </ul>	•••	. p .		Tizro et al. (2022); Sundararaman et al. (2021); Al-Baldawi et al. (2022); Forni et al. (2002); Hassanzadeh et al. (2021)
accumulated at $21.5 \pm 0.1 \mu$ g/g in roots and $304.7 \pm 57.6 \mu$ g/g by 168 h in shoots. Accumulated ethion decreased by 55-91% in shoots and $74-81%$ in roots following phytodegradation/ phytovola- tisation		• • • • • • •		Yan et al. (2019a); Yan et al. (2006); Hammad Ma (2006); Molisani et al. (2006); Hammad (2011)

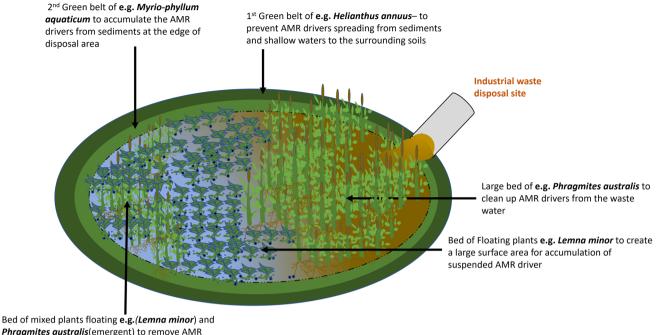
Clargory         Name of aganté huns         Ennostrated remonit: Attributés         Descritation (B)         Bio concentration factor (BCF)         References           Methan         Attributés         (B)         Attributés         (B)         Attributés         (B)           Methan         Attributés         (B)         Demonstrated Pound'In accumulation interaction         Attributés         Attributés         Attributés           Partin         Attributés         Attributés         Attributés         Attributés         Attributés           Partin         Attributés         Attributés         Demonstrated Pound'In accumulation water solution         Attributés         Attributés           Partin         Attributés         Attributés         Demonstrated Pound'In accumulation water solution         Attributés         Attributés           Partin         Attributés         Attributés         Demonstrated Pound'In accumulation water solution         Attributés         Attributés           Partin         Attributés         Attributés         Attributés         Attributés         Attributés           Partin         Attributés         Attributés         Attributés         Attributés         Attributés           Partin         Attributés         Partin         Partin acuributés         Attributés <t< th=""><th>Table 6 (continued)</th><th>(pər</th><th></th><th></th><th></th><th></th></t<>	Table 6 (continued)	(pər				
Ind         Heitanthus         HM: Ni, Cr Ph, Zh         Demonstrated Pb and Zn accumulation witch increased following EDTA treat- oxyteracycline           amuus         -AB: retrosycline oxyteracycline         -BB: retrosycline witch increased levels of Cr (VI) and NI (I) were found following expa- sing increasing the and oxyteracycline following update into the contrainated wate Suggested enzyme-assisted degrada- tion of treat-ycline and oxyteracycline following update into the contrainated wate Suggested enzyme-assisted degrada- tion of treat-ycline and oxyteracycline following update into the contrainated success of antibiotic oresult in rapid disappearance of antibiotic oresult in antibiotic oresult in and disappearance of antibiotic oresult in antibio	Category	Name of aquatic plants	Demonstrated removal: Heavy metal (HM) Antibiotics (AB) Bioc- ides (Bi)	Observations	Bio concentration factor (BCF) (where reported)	References
Lemna gibba• HM: Ph. Cd• Removal of Pb and Cd was pH (5, 7 and• BCF Pb 403–738 and BCF Cd• AB: tetracycline and chloramphenicol9) and concentration (2, 5 and 10 mg/L)445–616• AB: tetracycline and chloramphenicol9) and concentration (2, 5 and 10 mg/L)445–616• AB: tetracycline and chloramphenicol9) and concentration (2, 5 and 10 mg/L)445–616• AB: tetracycline and chloramphenicol98.1% (10 mg/L at 7 pH) and 41.6% (10 mg/L at 7 pH) and 41.6% (10 mg/L at 7 pH) and 41.6%• High demonstrated removal of tetracy- cline 84% (5–15 mg/L) and Chloram- phenicol 64% (10–20 mg/L)	Wetland plant	Helianthus annuus	<ul> <li>HM: Ni, Cr Pb, Zn</li> <li>AB: tetracycline and oxytetracycline</li> <li>Bi: persistent organic pollutants (POPs)</li> </ul>	<ul> <li>Demonstrated Pb and Zn accumulation which increased following EDTA treatment</li> <li>In all plant parts, statistically significant increased levels of Cr (VI) and Ni (II) were found following exposure &lt; 10,000 µg/L in contaminated water</li> <li>Suggested enzyme-assisted degradation of tetracycline and oxytetracycline following uptake into the root system to result in rapid disappearance of antibiotic</li> <li>Sixteen out of twenty-four persistent organic pollutants reached up to 87% remediation (including heptachlor, aldrin, heptachlor epoxide, trans-chlordane, chlordane, chlordane, dieldrin, DDE, DDT, methoxychlor, mirex and decachlorobiphenyl). Accumulation in sunflower roots ranged from 9.4% for 2,4,5,6-tetrachloro-m-xylene and 87.3% for 4,4'-DDT</li> </ul>		Almeida et al. (2017b); Gujarathi et al. (2005a); Garcia Pérez et al. (2014); Kaly- vas et al. (2022)
	Floating	Lemna gibba	<ul> <li>HM: Pb. Cd</li> <li>AB: tetracycline and chloramphenicol</li> </ul>	<ul> <li>Removal of Pb and Cd was pH (5, 7 and 9) and concentration (2, 5 and 10 mg/L) dependent, but pH 7 showed the optimum metal removal. Pb and Cd removal ranged between 60.1% (2 mg/L at 9 pH) and 98.1% (10 mg/L at pH 9) and 84.8% (2 mg/L at pH 7), respectively</li> <li>High demonstrated removal of tetracycline 84% (5–15 mg/L) and Chloram-phenicol 64% (10–20 mg/L)</li> </ul>		Maldonado et al. (2022a); Verma and Suthar, (2015)

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Table 6 (continued)	ued)				
Category	Name of aquatic plants	Demonstrated removal: Heavy metal (HM) Antibiotics (AB) Bioc- ides (Bi)	Observations	Bio concentration factor (BCF) (where reported)	References
Floating	Lemna minor	<ul> <li>HM: Cd, Cu, Pb, and Ni</li> <li>AB: cefadroxil metronidazole, trimethoprin, sulfamethoxa- zole</li> <li>Bi: copper sulphate (fungicide), flaza- sulfunon (herbicide), and dimethomorph (fungicide)</li> </ul>	<ul> <li>The removal efficiency was greater than 80% for Cd, Cu, Pb, and Ni metals and maximum removal was observed for nickel (99%) from sewage-mixed indus- trial effluent</li> <li>At 250 µg/L, 100% removal of cefadroxil in 14 d, followed by metronidazole (96%), sulfamethoxazole (73%) and trimethoprim (59%) over 24 d</li> <li>At 40 µg/L, moderate removal of Cu (50%) and Flazaulfuron 42%. Low removal of Dimethomorph: 27% at 400 µg/L</li> </ul>	• BCF < 1000 and maximum BCFs were found for copper (558) and lead (523.1), indicat- ing that the plant is a moderate accumulator of both metals	Olette et al. (2008); latrou et al. (2017); Bokhari et al. (2015)
Submerged to emergent	Myriophyllum aquaticum	<ul> <li>HM:Cd, Cr, Ni, Zn, Cu</li> <li>AB: norfloxacin, sulfamethazine tetra- cycline</li> </ul>	<ul> <li>The ratio of Cu/tetracycline greatly affected the removal and accumulation of tetracycline and plant growth. Low levels of Cu (II) (&lt;1000 µg/L) promoted tetracycline removal, but excessive Cu (II) (&gt;10,000 µg/L) impeded it</li> <li>Adsorption was the main mechanism for rapid tetracycline removal due to its large contact area and ion exchange, accounting for about 99% and 54% of the total tetracycline removed within 2 h and 5 d, respectively</li> </ul>		Gujarathi et al. (2005a); Guo et al. (2020a, 2019); Park and Son, (2022)
Floating	Pistia stratiotes	<ul> <li>HM: Cd, Cu, Fe, H</li> <li>AB: norfloxacin, ciprofloxacin sulfamethazine tetracycline</li> <li>Bi: clomazon</li> </ul>	<ul> <li>92% (5.3 mg/L) and 88% (7.5 mg/L) Cu removal with initial concentrations, over 14 days, with peak removal observed on day 3</li> <li>70% removal efficiency of ciprofloxacin in 10 ppm solution; steady growth of the plant was maintained at 0.1606 g/d</li> <li>Higher uptake of tetracycline in the <i>P. stratiotes</i> than in <i>M. aquaticum</i></li> <li>Demonstrated potential to eliminate &lt;90% of herbicide residues (clomazol)</li> </ul>	• BCF of norfloxacin was much lower, ranging from 0.24 to 0.38 than sulfamethazine (0.59–0.64) and TET (0.72–0.78)	Masiyambiri et al. (2023); Gujarathi et al. (2005a); Escoto et al. (2019); Park and Son, (2022); Novita et al. (2019)

Table 6 (continued)	(p:				
Category	Name of aquatic plants	Demonstrated removal: Heavy metal (HM) Antibiotics (AB) Bioc- ides (Bi)	Observations	Bio concentration factor (BCF) (where reported)	References
Emergent	Typha latifolia	<ul> <li>HM: Cd</li> <li>AB: ciprofloxacin and sulfamethoxazole</li> <li>Bi: atrazine</li> </ul>	<ul> <li>Initial concentration of Cd 5 and 10 mg/L resulted in 96% Cd removal</li> <li>34% removal for ciprofloxacin, 20% for sulfamethoxazole at 5 µg/L and 10 µg/L, respectively</li> <li>Significantly reduced overall atrazine (initial 20 µg /L) concentration loads by 45%</li> </ul>		Moore et al. (2013)
Floating with an extensive root system	Phragmites aus- tradis	<ul> <li>AB: norfloxacin, eerrofloxacin, tetracy- cline, ciprofloxacin, curacy- cline, ciprofloxacin, oxytetracycline and sulfamethazine</li> <li>HM: Fe, Mn, Ni, Zn, Pb, Cd, Co, Cu</li> <li>Bi: 13 Organophos- phate pesticides; tebu- conazole, imazalil</li> </ul>	<ul> <li>Observed removal of enrofloxacin (94%) and tetracycline (75%)</li> <li>The highest concentration of organo-phosphate pesticides was found in leaves (16.41–31.39 µg/kg dw), followed by roots (13.92–30.88 µg/kg dw),</li> <li>Demonstrated to remove of 96.1% of tebuconazole and 99.8% of imazalil</li> </ul>	• The highest BCF values were obtained for Ni (0.43), Ba (0.43), Mo (0.36), Cr (0.35), and Cd (0.31)	Carvalho et al. (2012); Milke et al. (2020); Liu et al. (2013b); Kovačević et al. (2019); Cicero-Fernández et al. (2016); Olisah et al. (2021)

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Phragmites australis (carried and e.g. (cerning minor) and Phragmites australis (cerergent) to remove AMR drivers from suspended sediments and create large surface area for uptake by roots

Fig. 3 Schematic diagram of suggested phytoremediation set-up at an industrial waste disposal site using a combination of submerged, floating and emergent plants

drivers of interest following our recent literature review, with supporting evidence provided in Table 6.

As emergent and floating macrophytes primarily take up contaminants (whether from the substrate or water column) through their roots, plants with large and deep root systems would therefore be preferred as contaminants can be filtered from the water and accumulate in the extensive root system (Shackira et al. 2021). A large plant with a deep root system and a demonstrated capacity to take up heavy metals, antimicrobials and biocides would include Phragmites australis, a common reed (Milke et al. 2020; Carvalho et al. 2012). P. australis is one of the world's most extensively distributed emergent plant species and has been used to remove chemicals from different types of polluted water bodies, including wastewater, since the 1970s (Rezania et al. 2019). P. australis is adaptive to a range of environmental conditions and, as demonstrated in Table 6, can remove micropollutants such as antibiotics whilst tolerating potential metal toxicity. Therefore, it is an ideal candidate species in any phytoremediation configuration.

A suitable plant for rhizofiltration is characterised by having an extensive root system with a rapid-growth rate (Mareddy 2017). Therefore a phytoremediation configuration including water hyacinth (*Eichhornia crassipes*) (Yadav et al. 2015) would be needed. Water hyacinth is widely used for phytoremediation as it can grow in highly polluted waters and bear significant variations in water quality parameters, e.g., nutrient levels and pH (Singh and Balomajumder 2021). In addition to root uptake, uptake via stem tissue of submerged macrophytes is an essential pathway for removing AMR drivers from the water column (Denny 1972; Dhote and Dixit 2009; Gabrielson et al. 2004; Fletcher et al. 2020). Species such as *Myriophyllum aquaticum* have demonstrated the efficient removal of heavy metals, antimicrobials and biocides from contaminated waters; and would therefore be a suggested candidate for a submerged species (Park and Son 2022; Gujarathi et al. 2005b; Guo et al. 2020a).

A green belt of Helianthus annus is suggested around the waste disposal site to minimise any pollutant migration into the surrounding soils. H. annus, is a well-known hyperaccumulator of metals (Kalyvas et al. 2022) with demonstrated capacity to accumulate other pollutants too (Table 6). As a moist-loving plant that can grow well in wet areas, it would be suitable at the edge of the contaminated water body. A second belt of submerged plants, e.g., M. aquaticum, at the edge of the disposal site would remove contaminants from the shallow water. The emergent perineal plant P. australis is suggested to cover the central section of the contaminated water body, given its extensive root system with a high surface area to volume ratio to maximise pollutant uptake and accumulation. After this bed, a bed of floating plants is suggested, e.g., Lemna minor or Lemna gibba. These floating plants have demonstrated efficiency in accumulating a wide range of chemicals such as Pb, Cd, Tetracycline and Chloramphenicol (Table 6) which fall within the classes of AMR drivers. Finally, a mixed bed of floating P. australis will maximise the phytoremediation process. Previous research has suggested that using only one species (P. australis) can result in up to 90% removal of heavy metals (Milke et al. 2020). However, given that other aquatic macrophytes are efficient in removing additional AMR drivers, such as antibiotics, it is suggested that a combination of different species should be used, where P. australis could potentially play a vital role. Given the greater affinity of some plants than others for antibiotics and biocides, using two or three species could result in a higher ability to reduce contaminant concentrations in treated waters. However, using multiple species would require consideration of interspecies competition between P. australis and other aquatic plants to ensure phytoremediation potential is not affected.

# **Knowledge Gaps**

Research has shown that floating and emergent aquatic plants can reduce pollutants in aquatic systems by solid filtration, assimilation, and microbial transformation. Phytoremediation, therefore, appears to be an environmentally friendly approach to remove drivers of AMR from contaminated waters. However, several knowledge gaps exist in the current literature and are discussed below:

- Across the suite of chemicals included in this review, most phytoremediation studies have focussed removal of metals, pesticides and antibiotics from contaminated waters. The available literature clearly shows that different chemicals accumulate to different extents in aquatic macrophytes, which is also a factor in the plant species used. Therefore, this leaves significant gaps in our understanding of phytoremediation potential for broader classes of antimicrobials (e.g., antifungals and antivirals) and biocides (e.g., wood preservatives and paint as an anti-fouling agent), all of which are routinely discharged into the aquatic environment.
- 2) An improved understanding of uptake and accumulation potential in the context of plant traits would enable the selection of the most efficient plant species for the phytoremediation of particular chemicals. Specifically, the mechanisms of migration and transformation of organic chemicals such as antimicrobials and biocides in plant tissues are still far from clear. Several studies have demonstrated that antibiotics can transform into simple compounds integrated with plant tissue and transform into metabolites with retained biological potency, thus altering the concentration of the parent compound. We need an improved understanding of the potential for this in a wider variety of aquatic macrophytes and across a

broader set of chemicals to maximise the potential for the completly removing of contaminants.

- 3) It is widely established that the plant species present affect the nature and functions of the bacterial communities, affecting microbial-driven removal processes (Ruiz-Rueda et al. 2009). Published research has shown that bacteria play a pivotal role in the phytodegradation of contaminants (Hoang et al. 2013). However, in most contaminated waters, the number of microorganisms is depressed, so there are not typically enough bacteria to either facilitate contaminant degradation or support plant growth (Huang et al. 2004). Microbe-assisted phytoremediation can play a key role in facilitating rhizosphere microbes to degrade organic contaminants. However, research into microbe-assisted phytoremediation in aquatic systems is lacking for most organic contaminants (e.g., biocides, antivirals, antifungals), with previous research primarily focussing on heavy metals and antibiotics. Mechanistic insights, such as information about the transformation pathways for a wider suite of chemicals, would enable the selection of the right plant species for treating contaminated water to maximise the uptake capacity and potentially facilitate its complete removal via degradation.
- Through phytoremediation, the processes of absorption, 4) transportation, and the transformation of contaminants have the potential to result in a toxic effect on plants following long-term exposure. However, limited phytotoxicity data exists in the literature for plants commonly used for phytoremediation (Table 5). This is partly because phytotoxicity data for aquatic plants have served a relatively minor role in regulatory decisions concerning the environmental hazard of potential contaminants (Lewis 1995). Where research has shown that chemical exposure results in plant toxicity, results also suggest that plants can still accumulate the contaminant (Nedjimi 2021). Nevertheless, we need a better understanding of how impacts on critical parameters responsible for plant growth and development may affect the long-term sustainability of phytoremediation.
- 5) Chemicals in the environment do not occur in isolation; however, studies have seldom evaluated phytoremediation potential in the aquatic environment concerning co-contaminants' presence (Almeida et al. 2017a). We need to build on existing knowledge to understand the phytoremediation potential of aquatic macrophytes in the presence of mixtures of heavy metals and organic contaminants such as antibiotics and biocides. This will reveal a plant's uptake and accumulation capacity when competing with other contaminants and a more realistic representation of pollution incidences.
- 6) Aside from the identified knowledge gaps concerning the fate and behaviour of biocides, metals and antimi-

crobials during phytoremediation, we know very little about how this dynamic process can impact the fate of ARGs or ARBs and the overall development of AMR in aquatic systems. By removing the AMR drivers from the aqueous media, are we just moving the problem to a different location? Research has recently been published, demonstrating that phytoremediation of heavy metals and antibiotics in soils using terrestrial plants (Lolium multiflorum and Brassica juncea) can impact the abundance of ARGs (Cui et al. 2021). The fate of ARGs and ARB during phytoremediation in aquatic systems remains undetermined however is a critical knowledge gap to address as rhizosphere/plant microbial communities provide opportunities for the exchange of genetic information (e.g., through horizontal gene transfer) conferring resistance. This must be explored further to understand the implications for AMR in the environment. In considering the wider context of AMR, the disposal of the plant following phytoremediation must also be considered. After phytoremediation, plants will likely transfer contaminants to the environment again, for example, through landfill disposal routes. There is a clear knowledge gap on adequately disposing of plants (Liu and Tran 2021) to prevent secondary contamination. This again comes back to the question if we are removing the problem to a different location.

# Conclusions and Recommendations for Future Studies

AMR drivers are routinely released into our environment. We have seen a notable increase in their detection in the past 2-3 decades, partly owing to improved analytical methodologies and an awareness of potential effects. This topic is an active research area, and there is a clear drive to find ways to extract these contaminants from the environment, thereby minimising any potential risk. Phytoremediation is a method which utilizes plants and the associated rhizosphere microorganisms to remove or transform toxic chemicals from the environment. Because of their abundance and limited mobility, aquatic plants have a great potential to function as in-situ and on-site accumulators and filters of aquatic pollutants. As highlighted in this review, plants can be beneficial for the remediation of chemical contaminants, specifically AMR drivers from aquatic environments.

Although phytoremediation is advantageous over other treatment methods, it is essential to account for existing knowledge gaps and potential limitations when considering its use as a remediation method. Despite many publications on this topic, the combined exposure from chemical mixtures in the environment is yet to be comprehensively evaluated. As highlighted in our review, some species have a greater affinity for certain compounds, and select plants can efficiently degrade some organic chemicals while unable to degrade others. This is not necessarily a limitation, but it is important to account for this to maximise the potential for effective phytoremediation. Potential solutions for the complex chemical cocktail of AMR drivers in the environment, for example, could include intercropping or the setup of constructing wetlands that take advantage of certain species' ability to accumulate select chemicals better than others.

Some plant species are sensitive to contaminants, and exposure to chemicals following phytoremediation can impact plant growth and development (Table 5). If phytotoxicity occurs and affects plant growth and development, there may be insufficient biomass for meaningful phytoremediation, and phytoremediation will become less effective. Our understanding of contaminant-induced effects on plant growth and development is inadequate to understand the implications of phytoremediation. In addition, there is a lack of information on biodegradation pathways and the transformation of organic chemicals, such as antivirals and biocides, during phytoremediation. Mineralisation offers the potential to remove the contaminant altogether. Therefore, we need to increase our understanding of plants' ability to facilitate these processes to allow us to capitalise on natural microbial processes to remove AMR drivers from the aqueous phase of the environment entirely, thereby significantly minimising any potential risk.

Ultimately, phytoremediation offers a cost-effective means of removing AMR drivers from aquatic systems. However, the broader implications of phytoremediation in AMR development remain undetermined. Specifically, an evaluation of the role of gene exchange during phytoremediation in aquatic systems is needed to understand if these processes can promote the diffusion of ARGs and ARB, with the potential to enter the wider environment. In this regard, we need to advance our understanding of phytoremediation before it may be considered an operational alternative to removing contaminants from polluted aquatic systems such as rivers, lakes and streams.

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**Data availability** Data sharing is not applicable to this article as no new data were created or analyzed in this study.

## Declarations

**Conflict of interest** The authors declare that they have no potential conflict of interest with respect to the research, authorship or publication of this article.

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