

Microbes in Restoration of Polluted Ecosystems



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1 Introduction

1.1 *Why Polluted Ecosystems Are Considered a Threat?*

Global climate change and water scarcity are examples of problems humanity is facing as harmful after-effects of human actions over the environment ignoring sustainability principles (Santhakumari & Sagar, 2020). Pollution is an important consequence of these actions as it is a serious threat to the ecosystem, including the living beings present. Humans, for example, can be highly affected by pollutants in a negative manner not only at cellular level but also at the level of organs and systems (Fig. 1). A large array of organic and inorganic pollutants possesses the capacity to be persistent contaminants, accumulating in polluted areas for long periods of time and also entering the food chain (becoming a threat to food security) (Ojuederie & Babalola, 2017). This kind of pollution is especially difficult to deal with once the effects of legislative control inducing a reduction in new pollutant emissions take a long time to be noticed on the environment. The concentrations, for example, of these substances in freshwater predators still exceeded the limits considered safe for reproduction/survival decades after measures to reduce new emissions of persistent contaminants in the water (Kean et al., 2021).

Pesticides, for example, are ubiquitous environmental pollutants that present a risk to 64% of global agricultural land and a high risk to 31% of agricultural land worldwide. These organic persistent pollutants negatively impact biodiversity, water quality, and human health (Tang et al., 2021). In human organism this kind of

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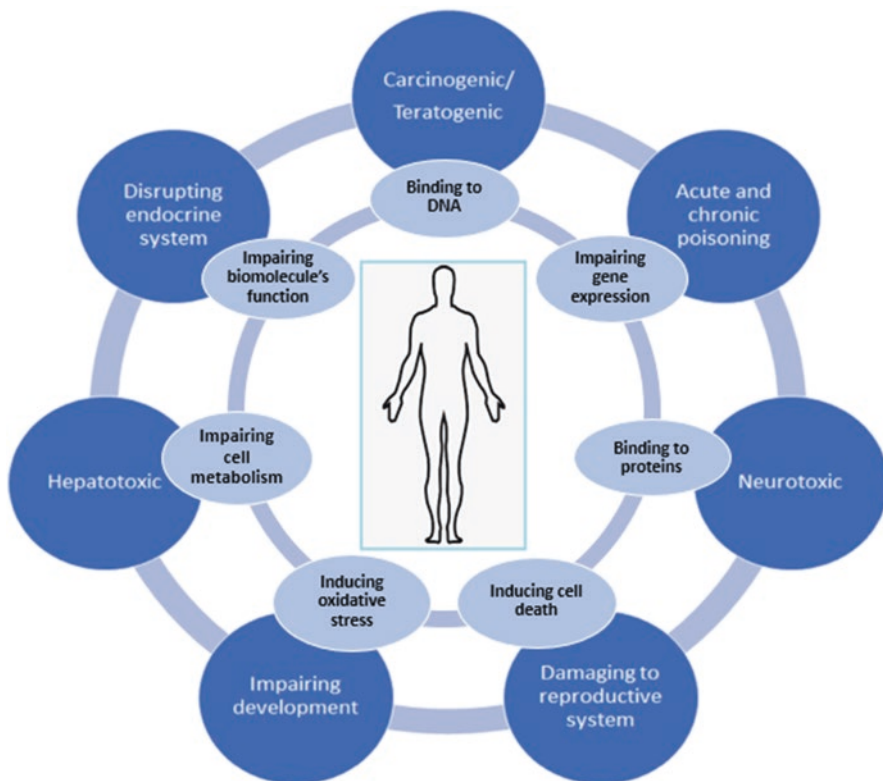


Fig. 1 Pollutants as threats to humans' health – main negative effects pollutants can cause on the human body at cellular level (inner circle) and also at the level of organs and systems (outer circle)

substance can impair the functioning of different organs and tissues through cytotoxicity and DNA damage, being neurotoxic, hepatotoxic, carcinogenic, disrupting endocrine system, and being also teratogenic (Kalyabina et al., 2021). Carbofuran [2,3-dihydro-2,2-dimethyl-7-benzofuranyl N-methyl carbamate] is widely used in agricultural practices and after inhalation, ingestion, or dermal absorption can do severe damage to different living beings causing also their deaths (Mishra et al., 2020).

When it comes to textile industry effluents, for example, the large spectrum of chemicals used during the process reflects on the polluted wastewater generated. Heavy metals and textile dyes present toxic effects on living organisms especially from the aquatic biota and to people who will drink the water even after treatment. It is common that treatments fail to remove metals or only disperse dyes (Methneni et al., 2021).

Heavy metals, for example, can enter human body through inhalation, gastrointestinal tract, or skin and cause not only membrane but also DNA damage. By binding to functional groups in proteins (such as thiol) they can disturb protein/enzyme

function (Witkowska et al., 2021). Mercury, for example, is a heavy metal that can accumulate in the human's body as same as in animals' bodies causing severe problems. Fish consumption can expose humans to the neurotoxicant methylmercury (Moriarity et al., 2020; Novo et al., 2021) poisoning them. This substance is also highly toxic to animals (Davis et al., 2021). Activities such as gold mining are also risky (Achatz et al., 2021) to allow mercury intoxication. These heavy metals also damage plants negatively impacting the photosynthesis rate and the metabolism as a whole (Hu et al., 2020).

1.2 Living Beings Performing Remediation: Bioremediation

Physicochemical methods of remediation, such as soil washing, soil flushing, electrokinetic remediation, solvent extraction, incineration, and chemical reduction in the gas phase, can be applied to deal with environmental contaminants (Ajiboye et al., 2020; Baldissarelli et al., 2019; Cameselle & Gouveia, 2019). However, commonly some disadvantages are faced especially when they are applied on a large scale. High cost and generation of additional pollution are examples. Bioremediation, however, can present interesting advantages regarding the costs and it is also a process eco-friendly (Gaur et al., 2018; Gong et al., 2018; Fernando et al., 2019).

Bioremediation can be performed by a large variety of living beings: bacteria, fungi, yeasts, microalgae, and plants that can degrade contaminants in a harmless state or provide mechanisms to reduce their concentration to levels considered safe (Estrada & Quijano, 2020; Ojhaa et al., 2021). These living systems present the ability to modify and/or decompose pollutants and this ability can be naturally found on the species or added through genetic engineering strategies (Zhu et al., 2012; Ye et al., 2017).

Strategies of bioremediation can also be applied together with physicochemical strategies. The inoculation of microbes such as bacteria can contribute to enhance the efficiency of pollutants removal and restoration of ecosystems by reestablishing water and/or soil biological function and also in the treatment of contaminated air (Chen et al., 2016). Bioactive coatings, for example, allow using microorganisms immobilized in bedding nanomaterials to improve air quality (Estrada & Quijano, 2020).

Restoration of contaminated areas through bioremediation can be performed *ex situ* (removing samples of the polluted environment, treating and returning it to its prior localization – more easily performed when the intention is to remediate soil) or *in situ* (treating the polluted area directly where it is) (Ortiz-Hernández et al., 2018; Parween et al., 2018).

The efficiency of bioremediation is influenced by various aspects related to the living being employed, environmental factors of the contaminated areas, number and amount of contaminants as same as their chemical nature, and also by the protocol of remediation applied (Azubuiké et al., 2016). It is common to have a redox process involved in remediation promoted by living organisms and consequently

addition of organic and inorganic amendments to regulate medium physicochemical properties can favor environmental decontamination/restoration (Beiyuan et al., 2017). For example, to remediate oily contaminated soil, protocols of bioremediation can have their efficiency improved by adding biosurfactants and lipases (Kreling et al., 2021). Biochar can be used to immobilize metals and organic pollutants enhancing the bioremediation success and this type of strategy has been reported by many researchers (Rizwan et al., 2016; Yuan et al., 2017).

2 Microbes Restoring Polluted Ecosystems

There are microbes that can naturally deal well with some types of environmental pollutants, metabolizing or sequestering them from contaminated areas (which is a process known as natural attenuation). However, it is generally a time-consuming strategy to be applied (Cui et al., 2020). In order to improve process' efficiency and speed it, microbes can be submitted to genetic engineering or receive stimulus: from substances added to the polluted spot (biostimulation), from aeration of the polluted area to increase biodegradation pollutants (bioventing) or from microbial taxa with useful biodegradation/detoxification capacity (bioaugmentation) (Gaur et al., 2018; Dell' Anno et al., 2021a).

Among microbes (bacteria, fungi, yeasts, microalgae, and protozoa) bacterium is the most applied on bioremediation protocols (Jain & Bajpai, 2012) since the 1980s (Delfino & Miles, 1985; Karns et al., 1986; van der Hoek et al., 1989) with a deserved highlight being directed to genera such as *Corynebacterium*, *Staphylococcus*, *Streptococcus*, *Shigella*, *Alcaligenes*, *Acinetobacter*, *Escherichia*, *Klebsiella*, *Enterobacter*, *Flavobacterium*, *Pseudomonas*, *Bacillus*, *Alcanivorax*, *Thalassolituus*, *Cycloclasticus*, *Oleispira*; *Vibrio*, *Pseudoalteromonas* and *Marinobacter* specially when it comes to organic pollutants (Haritash & Kaushik, 2009; Kafilzadeh et al., 2011; Dell' Anno et al., 2021a, b) (Table 1).

Species naturally able to remediate can use pollutants as nutrient source, surviving in contaminated areas such as *Pseudomonas stutzeri* OX1 dealing with tetrachlorethylene (Ryoo et al., 2000) and *Pseudomonas nitroreducens* and *Pseudomonas putida* metabolizing p-coumaric acid and p-hydroxybenzoic acid (Zhang et al., 2010).

Microalgae are also very useful to be used in bioremediation protocols, especially to remediate environs contaminated by polycyclic aromatic hydrocarbons, but also to deal with heavy metals as contaminants. The genera *Chlorella*, *Selenastrum*, and *Scenedemus* deserve a highlight. *Chlorella pyrenoidosa* could efficiently remediate heavy metals (Cr, Cu, Pb, Zn, Cd, Mn, and Ni) from wastewater collected from a common effluent treatment plant (Kothari et al., 2021). *Chlorella sorokiniana* could remediate wastewater contributing to the assimilation of Zn and Ni but also nitrogen and phosphorous (Lugo et al., 2020). The main mechanism involved in this process is related to reduction in bioavailability (and consequently toxicity) due to the exopolysaccharides that make it possible pollutants' immobilization and/or internalization (Dell' Anno et al., 2021a). However, some species can use

Table 1 Examples of bacteria that can be used to bioremediate environmental pollutants

Bacterium species	Pollutant	Reference
<i>Pseudomonas stutzeri</i> OX1	Tetrachlorethylene	Ryoo et al. (2000)
<i>Pseudomonas nitroreducens</i> , <i>Pseudomonas putida</i> , and <i>Rhodotorula glutinis</i>	p-coumaric acid and p-hydroxybenzoic acid	Zhang et al. (2010)
<i>Pseudomonas</i> sp. strain ADP	Atrazine and cyanuric acid	Neumann et al. (2004)
<i>Thalassolituus oleivorans</i>	Aliphatic hydrocarbons from C7 to C20 carbons	Yakimov et al. (2004)
<i>Flavobacterium</i> sp.	Organophosphate pesticides	Ortiz-Hernandez et al. (2004)
<i>Cycloclasticus</i> spp.	Polycyclic aromatic hydrocarbons	Niepceron et al. (2009)
<i>Achromobacter</i> sp. WM111, <i>Rhodococcus</i> TE1, <i>Pseudomonas</i> sp. 50,432, <i>Sphingomonas</i> sp. strain SB5, <i>Enterobacter</i> sp., <i>Burkholderia</i> sp. PLC3, <i>Bacillus</i> sp., and <i>Cupriavidus</i> sp. ISTL7	Carbofuran	Karns et al. (1986), Behki et al. (1994), Chaudhry et al. (2002), Kim et al. (2004); Park et al. (2006), Mohanta et al. (2012), Plangklang and Reungsang (2013), Onunga et al. (2015), Gupta et al. (2019)
<i>Staphylococcus succinus</i> HLJ-10	D-cyphenothrin	Huang et al. (2020)
<i>Novosphingobium</i> sp. PCY, <i>Microbacterium</i> sp. BPW, <i>Ralstonia</i> sp. BPH, <i>Alcaligenes</i> sp. SSK1B, and <i>Achromobacter</i> sp. SSK4, PCY	Polycyclic aromatic hydrocarbons (PAHs)	Wongwongsee et al. (2013)
Species from <i>Streptomyces</i> gender	Chlordane	Cuozzo et al. (2012)
<i>Alcaligenes faecalis</i>	Endosulfan	Kong et al. (2013)
<i>Sphingobium wenxiniae</i> strain JZ-1	3-phenoxybenzoate	Cheng et al. (2015)
<i>Corynebacterium variabilis</i> Sh42	2-hydroxybiphenyl (2-HBP), catechol, and benzoic acid	Younis et al. (2020)
<i>Staphylococcus aureus</i> V329	Uranium (VI)	Shukla et al. (2020)
<i>Bacillus cereus</i> WHX-1	Chromium (VI)	Chen et al. (2021)
<i>Escherichia coli</i> , <i>Streptococcus</i> <i>pyogenes</i> , and <i>Streptococcus</i> <i>pneumoniae</i>	Zoxamide	Ahmad et al. (2020)
<i>Shigella flexneri</i> FB5	Fomesafen	Yang et al. (2020)
Species from the genus <i>Alcaligenes</i>	Cu ²⁺ , Cd ²⁺ , Cr ⁶⁺ , Ni ²⁺ , and Zn ²⁺	Sodhi et al. (2020)
<i>Oleispira antarctica</i> RB-8	Hydrocarbons	Gregson et al. (2020)
<i>Acinetobacter</i> sp.	Fluoride	Shanker et al. (2020)
<i>Klebsiella variicola</i>	Chromium VI	Yu et al. (2021)
<i>Enterobacter</i> sp. MN17	Petroleum hydrocarbons	Ali et al. (2020)
<i>Alcanivorax borkumensis</i>	Oil hydrocarbons	Shaikhulova et al. (2021)
<i>Vibrio fluvialis</i>	Mercury	Saranya et al. (2017)
<i>Pseudoalteromonas</i> sp. SCSE709-6	Cadmium	Zhou et al. (2013)
<i>Marinobacter</i> sp.	Hydrocarbons	Al-Wahaib et al. (2016)

non-chlorinated hydrocarbons as carbon source degrading petroleum hydrocarbons (Chekroun et al., 2014). *Selenastrum capricornutum* and *Scenedesmus acutus* could efficiently promote the biodegradation of benzo(a)pyrene (de Llasera et al., 2016).

Fungi are capable of degrading environmental pollutants, especially organic ones (such as pesticides, dyes, and hydrocarbons) through mycodegradation, bioremediating environs (Bhattacharya et al., 2012). Important genera when it comes to this activity are *Aspergillus*, *Curvularia*, *Drechslera*, *Fusarium*, *Lasiodiplodia*, *Mucor*, *Penicillium*, *Rhizopus*, and *Trichoderma* (Dell' Anno et al., 2021a). *Aspergillus niger* could efficiently deal with environmental contaminant 2-chloro-N-(2,6-diethylphenyl)-N-(methoxymethyl)acetamide, as same as the bacterium *Xanthomonas axonopodis* (Ahmad et al., 2020). The dead *Aspergillus niger* O-5 biomass could also remediate Pt^{4+} from polluted samples (Lombana-Fraguela et al., 2020). *Aspergillus* sp. A31 and *Curvularia geniculata* P1 favored the growth and development of *Oryza sativa* L. under mercury stress by sequestering the heavy metal (de Siqueira et al., 2021). *Penicillium chrysogenum*, as same as *Alternaria alternata*, efficiently promoted polyaromatic hydrocarbons' degradation, bioremediating contaminated samples (Hamad et al., 2021). *Drechslera* sp. strain 678 proved to be an interesting option to the remediation of methyl tertiary-butyl ether, a common additive of gasoline (d'Errico et al., 2021). *Fusarium solani* exhibited high tolerance to Zn^{2+} ions and was capable of promoting their biotransformation (El Sayed, 2020); the capacity of this species to remediate metal-contaminated waste could be enhanced by the presence of the gram-negative bacterium *Comamonas aquatica* (Qurbani & Hamzah, 2020). *Lasiodiplodia theobromae* could remediate polluted samples containing benzo[a]pyrene by using enzymes such as lignin peroxidase and laccase (Cao et al., 2020). *Mucor irregularis* strain bp01 proved to be able to promote the biodegradation of fluorene (Bankole et al., 2020) and *Mucor hiemalis* could deal well with acetaminophen, especially after pH adjustment (Esterhuizen et al., 2021). *Rhizopus stolonifer* could remediate samples polluted with Cd in an efficient manner, and when associated with the bacterium *Bacillus megaterium* also proved to be highly efficient to deal with Pb pollution (Njoku et al., 2020). 2,4,6-trinitrotoluene could be degraded by *Trichoderma viride* eradicating the toxicity associated with the pollutant (Alothman et al., 2020).

Yeasts are particularly relevant when it comes to remediating pollution caused by heavy metals (Sun et al., 2020). For example, *Diutina rugosa* stood out among 213 strains by its capacity to remediate Zn pollution (García-Béjar et al., 2020). However, organic pollutants can also be metabolized by yeasts, such as aflatoxin B₁ by *Rhodotorula mucilaginosa* (García-Béjar et al., 2020) and azodyes by *Sterigmatomyces halophilus* SSA-1575 (Al-Tohamy et al., 2020).

The mechanisms applied by microbes to remediate (that depends on the pool of genes and consequently proteins/enzymes available) can influence the process' efficiency. However, accessibility and bioavailability of the contaminants as same as environment characteristics (salinity, temperature, pH, and redox potential) can also interfere on this capacity (Gaur et al., 2018; Fernando et al., 2019).

3 Microbes Assisting Bioremediation Promoted by Plants

Phytoremediation involves the use of plants to restore environs polluted by environmental contaminants. However, there are some researchers that also consider the microbes associated with plant roots as part of the process. That is due to the importance that plant growth-promoting rhizobacteria and plant endophytes possess in improving the remediations' efficiency making it easier for plants to deal with complex scenarios such as dealing with a large array of different types of contaminants in the same area (He et al., 2020).

Phytostabilization and rhizodegradation are examples of phytoremediation mechanisms in which the participation of microbes is crucial. For example, *Funneliformis mosseae* (a fungus species) could improve the capacity of the plant species *Robinia pseudoacacia* to remediate Pb contamination through phytostabilization. The microbe promoted Pb's immobilization, consequently reducing this heavy metal's toxicity to the vegetal species phytoremediating it (Huang et al., 2019). *Alcanivorax* and *Bacteroidetes* are microbes that can live well in stressful situation regarding salt level and also present the capacity to metabolize some organic contaminants. They proved to be important tools to favor remediation of petroleum hydrocarbons by plant species *Hylotelephium spectabile* (Cheng et al., 2019). In fact, a large array of petrochemical pollutants, and hydrocarbon in general, contaminating water and soil environment could be remediated by phytoremediation assisted by microbes through different mechanisms/strategies (Asemoloye et al., 2019; Singh et al., 2021).

It is well known, for example, that microbes can favor the removal of heavy metals and radionuclides (that generally come from industrial and municipal solid waste) performed by plants. And in situation in which removal is difficult, they can favor neutralization or conversion into less toxic substances by biotransforming, biosorbing, and biomineralizing (Thakare et al., 2021). *Enterobacter cloacae* ATCC 13047, an endophytic bacteria isolated from *Ficus septica*, for example, could remediate soil contaminated with Cr (VI) reducing the pollutant to Cr³⁺ and contributing to the survival of the vegetal species (Rohmah et al., 2020). *Streptomyces pactum* and *Bacillus* sp. co-application could improve *Brassica juncea*'s growth and also favored phytoextraction of Cd, Cu, Pb, and Zn promoted by the plant (Jeyasundar et al., 2021).

It is also interesting to mention that microbes associated with plants can also favor the vegetal's development besides improving remediation potential. For example, *Klebsiella pneumoniae* AWD5 not only enhanced the capacity of *Jatropha curcas* to deal with aromatic hydrocarbon's pollution, but also favored plant's growth in pyrene-contaminated soil (Rajkumari et al., 2018).

Not only plants are influenced by the microbes associated with them, but they can also stimulate the growth and development of microorganisms present in the rhizosphere through chemical substances such as growth factors (Dominguez et al., 2019).

It is also possible to genetically modify plants, using sequences of DNA originally present in microbes' DNA, or in other organisms, to improve the efficiency of phytoremediation (Ozyigit et al., 2021). *Arabidopsis thaliana* could have its capacity to promote mercury phytoextraction improved after genetically engineering the plant to express the bacterial mercury transporter MerC fused with SYP121 (a plant SNARE that favors protein transportation to cell membrane) under the control of a root epidermis-specific promoter. Mercury accumulation was enhanced in shoots and phytoremediation's efficiency was successfully improved (Uraguchi et al., 2019).

4 Engineered Microbes Restoring Polluted Ecosystems

The development of fields related to genomics, metagenomics, metabolomics, transcriptomics, proteomics, and genome editing technologies is crucial to the advancement of bioremediation techniques (Jaiswal et al., 2019; Marco & Abram, 2019). Synthetic biology, for example, presents strategies applicable for bioremediation that involve cell-mediated detection of pollutants and remediation by genetic circuit and microbial biosensor (Jaiswal & Shukla, 2020). Metabolic reconstruction, for example, can allow the generation of microorganisms with improved catabolic activities by genetic engineering, offering elegant strategies for the remediation of contaminated ecosystems (Janssen & Stucki, 2020).

Various examples of protocols to generate genetically modified bacteria (GMB) to perform bioremediation are available in the literature and new ones are still being proposed nowadays. Phytochelatin synthase from *Pyrus calleryan*, when overexpressed in *Escherichia coli*, allowed remediation of Cd, Cu, and Hg and also increased tolerance to the heavy metals' presence (Li et al., 2015). The expression of the azoreductase from *Enterococcus* sp. L2 (product of *azoA* gene) in *E. coli* DH5 α and *Pseudomonas fluorescens* PfO- allowed decolorization of recalcitrant azo dyes. This process has its efficiency enhanced by coexpression of *azoA* with *fdh* from *Mycobacterium vaccae* N10 (Rathod et al., 2017). *Deinococcus radiodurans* (a radiation-resistant bacterium) was recently engineered to overexpress the *smtA* gene from *Synechococcus elongatus* fused to sequences from the surface layer proteins Hpi and SlpA. The gene is responsible to encode the metal-binding metallothionein protein that is naturally located in the cell's cytoplasm but fusion proteins took it to cell surface. This strategy offered a extraction of cadmium 1.5–3 times higher when compared to the one performed by organisms expressing only the cytosolic version of the metal binding metallothionein protein and cell-free preparations presented a potential for uranium remediation (Misra et al., 2021). Recombinant *Rhodococcus erythropolis* expressing ammonia monooxygenase and hydroxylamine oxidase offered optimized results on the remediation of pollution associated with landfill leachate (Bai & Tian, 2021).

Fungi and yeast can also be genetically modified to offer optimizations in the results of bioremediation protocols. However, yeasts are more easily genetically modified than fungi, being more applied in remediation protocols. They can deal,

for example, with heavy metals' pollution promoting their accumulation, precipitation, and changing their redox state (Ayangbenro and Babalola, 2017). The gene *EpNramp* from *Exophiala pisciphila* encodes a metal transporter; yeasts expressing this protein could enhance their natural capacity to accumulate Cd^{2+} (Wei et al., 2016). The gene *lac I* that encodes a laccase from the fungus *Phlebia brevispora* BAFC 633 could be successfully expressed in *Pichia pastoris*, and the enzyme exhibited high tolerance to diverse solvents and NaCl, being also capable of degrading recalcitrant synthetic dyes (Fonseca et al., 2018). When the dye-decolorizing peroxidase from *Pleurotus ostreatus* (a white rot basidiomycete) was expressed in the filamentous fungus *Trichoderma atroviride* it allowed decolorization of mono-azo, di-azo, anthraquinone, and anthracenedione dyes (Cuamatzi-Flores et al., 2019).

Microalgae can also be modified to enhance bioremediation potential. Overexpression of *CrMTP4* gene in *Chlamydomonas reinhardtii* increased the potential of the organism to remediate Cd pollution. The gene encodes for a member of the Mn-CDF clade of the cation diffusion facilitator family of metal transporters (Ibuot et al., 2017). The potential to promote remediation of Cd^{2+} and Zn^{2+} ions could be enhanced in this species after recombinant expression of a protein from *Arabidopsis thaliana*: the AtHMA4 C-terminal domain protein (Ibuot et al., 2020). Cd^{2+} bioremediation could also be optimized through the expression by *C. reinhardtii* of a synthetic gene (*gshA*) encoding for a *gamma*-glutamylcysteine synthetase (Piña-Olavide et al., 2020).

5 Recovering Soil Microbial Community to Promote Ecosystems Restoration

It is also possible to apply microbes in other types of protocols aiming restoration of polluted ecosystems. For example, improving soil aggregation by influencing beneficial communities of microorganisms. The functional recovery of the soil microbial community (SMC) is essential for ecological restoration. Incorporating SMC measurement and monitoring into the study designs is a challenge, once there is still not available a metric that represents the diverse functional and compositional complexity inherent in the SMC. Focus must change from trying to compositionally recreate the “reference” SMC for the creation of functionally robust SMCs that provide ecosystem functioning and provide ongoing ecological resilience in restored ecosystems (Hart et al., 2020). Soil inoculation is a common form of microbial reforestation, which consists of moving soil from target sites to restoration sites (Wubs et al., 2016). This practice is known as “the whole community” rewilding, and although it is evident in soil inoculation studies, is very little researched outside of soil transplants and therefore rarely considered during restoration. The desired sites are on the practitioner’s premises criterion; so they can adapt the community built on the base of any site they choose. However, a summary of community-wide reforestation for restoration purposes highlights that nearby remaining sites are

chosen more often, which conforms to conventional restoration paradigms (Contos et al., 2021; McDonald et al., 2016).

6 Conclusions

Physicochemical methods to restore polluted areas may present some disadvantages (such as the high cost to be performed on large scale) that can be surpassed by bioremediation strategies. Among bioremediation strategies is a large array of protocols applying microbes as tools to remove environmental pollutants and contribute to the restoration of ecosystems. There are strategies that use only microbes on their wild form, protocols applying genetically modified versions of these organisms to optimize results, and strategies associating microbes and phytoremediation, among other types of protocols. Microorganisms proved to be efficient in performing remediation of contaminants from diverse chemical nature in different environments.

7 Future Perspectives

In order to enhance the opportunity of innovative protocols using microbes to promote remediation, it is essential that the metabolism of microbes be known in a deep way. So, advancements in the field of molecular biology and in omics platforms are highly relevant to the proposal of new rapid, eco-friendly, safe, and cost-effective technologies of bioremediation of polluted ecosystems by microbes. The improvements on the possibility of efficiently engineering the DNA of these organisms, for example, are directly dependent on these advancements. Biosafety related to the field use of microbes in bioremediation also needs to receive special attention considering also the impact of microbe-assisted bioremediation on the ecosystem as a whole.

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