Chapter

The Strategy and Future of Biotechnology in Protecting the Global Environment

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Abstract

Global warming is accelerating, and the average global temperature is projected to rise from 3.5 to 5.7°C by the end of this century. Therefore, there is a strong possibility that we will soon experience frequent global-scale abnormal weather events and severe water and food shortages. To avoid such crises, three issues must be urgently addressed: reduction of CO_2 emissions, securing of energy sources that can replace fossil fuels, and securing of groundwater and food supplies. In this introductory chapter, we first discuss the development of new biotechnology processes such as CO_2 sequestration by algae, biofuels, and biopolymers. Biofuels and biopolymers, in particular, will soon play an important role as alternatives to scarce fossil fuels. In addition, bioremediation technologies for widespread groundwater and soil contamination are discussed. Novel bioremediation technologies, such as gene editing and the use of artificial enzymes, have the potential to dramatically improve bioremediation throughput. This new biotechnological approach to the environment will be a decisive factor in ensuring food and beverage safety.

Keywords: CO_2 capture, biofuel, biopolymer, bioremediation, global warming

1. Introduction

The Earth's climate has been undergoing significant changes over the past centuries, and the impact of these alterations is becoming increasingly evident. According to the Fourth Assessment Report of the United Nations Intergovernmental Panel on Climate Change, the concentration of carbon dioxide (CO_2) in the atmosphere has surged by more than 40% since pre-industrial times (1750). It is estimated to surpass the level of 400 parts per million (ppm) by 2013. This rise in CO_2 and methane, the two primary greenhouse gases, has led to a 1.09°C increase in the average global temperature for over 260 years. Notably, the rate of temperature increase is accelerating rapidly, and it is projected that the average global temperature will further increase by 1.5°C by 2027 and 3.3–5.7°C by the end of this century [1].

With the intensification of global warming, the world is grappling with an increasing frequency of extreme weather events, such as storms and heavy rainfall, and the consequences are already taking a toll on the global economy. The expansion of seawater, induced by global warming, has led to a 16-cm rise in the world's average sea level within the last century. This phenomenon has raised concerns about the imminent inundation of the South Pacific Islands and the potential submergence of the entire landmasses in some regions. Moreover, the regions that rely on glaciers and melting snow as their water source, covering one-sixth of the world's population, will face water scarcity as these ice formations continue to vanish due to global warming. Mountainous areas, which serve as the origins of major rivers, will also experience similar challenges. The impact of global warming on land and marine ecosystems is immense, causing extensive damage to plants, animals, and marine life. This destruction leads to the disruption of ecosystems, exacerbating the problem further. Notably, phytoplankton, a crucial CO_2 sink responsible for absorbing 31% of CO_2 emissions, is severely affected, accelerating global warming. Considering the current trajectory, it is evident that by the end of this century, the average global temperature will rise by 3.3–5.7°C, pushing Earth into a critical situation [2].

In addition to the direct impacts of global warming, water shortages have emerged as a severe concern, exacerbated by various factors beyond climate change [3]. The extensive implementation of large-scale projects that utilize rivers for irrigation in agricultural lands, along with population growth and rapid global economic development, has significantly increased the demand for water for domestic and industrial purposes, resulting in severe shortages in many regions. For instance, Egypt suffers from severe water scarcity as countries upstream of the Nile River consume disproportionate amounts of water and contribute to pollution [4]. According to a report by the United Nations in 2007, approximately 660 million people in 30–40 countries face extreme water shortages, whereas severe water shortages affect about 1.1 billion people. Tragically, 1.8 million children succumb to diseases caused by drinking contaminated water every year. Additionally, the combination of water scarcity and pollution in these countries adversely impacts crop growth, leading to water shortages and food scarcity, potentially escalating into international conflicts over vital resources in the future.

Given these pressing concerns, it is highly probable that humanity may soon encounter a time when only specific regions on Earth can sustain life. To ensure a sustainable society, the world must mobilize science and technology to address critical issues, such as reducing CO_2 emissions, developing renewable energy sources as alternatives to fossil fuels, and remediating pollution to secure safe food and drinking water. This chapter outlines the crucial role that biotechnology can play in confronting these challenges and providing potential solutions (**Figure 1**).

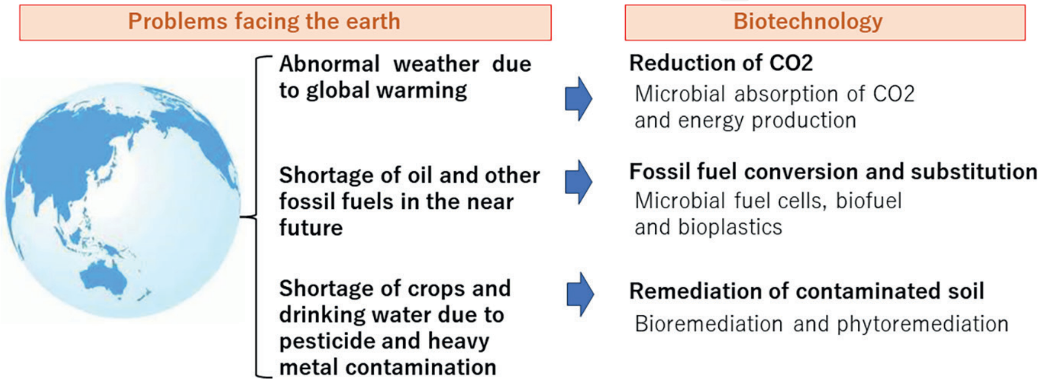

Figure 1.
Problems facing the earth and strategies for solving them using biotechnology.

2. Biotechnology strategies for global warming prevention and renewable energy development

2.1 CO_2 reduction

To address the urgent challenge of global warming, it is crucial to reduce atmospheric CO_2 levels. In this endeavor, countries worldwide have united under the "Paris Agreement," committed to mitigating CO_2 emissions. For instance, the Japanese government has taken a significant step by announcing the "2050 Carbon Neutral Declaration," setting the ambitious goal of achieving zero carbon emissions. To further prevent atmospheric CO_2 increase, global CO_2 emissions must be reduced to 50% by 2050 from 2006 levels, with the ultimate target of achieving a 100% reduction by 2075. However, despite the critical situation, major contributors to CO_2 emissions, such as China and the United States, are yet to embrace CO_2 capture technologies, emphasizing the need for innovative and effective technological advancements.

The most promising method of reducing atmospheric CO_2 is the "carbon capture and storage (CCS) process," in which atmospheric CO_2 is compressed and stored in liquid form at an underground site or the bottom of the deep ocean [5]. Although this method can significantly reduce the concentration of CO_2 in the atmosphere, its high cost is a major problem [6, 7]. Currently, converting CO_2 into value-added fuels instead of treating it as waste is considered economical, and efforts are being made to achieve this. For example, several studies have been conducted to convert CO_2 into fuels through electrochemical methods, and solar-electric lands have been developed to convert CO_2 into hydrocarbon fuels using only sunlight [8].

CO_2 capture using microorganisms is attracting attention as an environmentally friendly method [9]. Microalgae, in particular, are the best CO_2 capture sources compared with higher plants primarily due to their exceptional solar energy yield and year-round cultivability. Because the production cost of microalgae for CO_2 capture must be less than 500 t/ha/year, efforts are being made to reduce this cost. For example, there is a lower production cost in conjunction with the production of various useful substances. Using genetically modified microalgae, bioconversion processes from CO_2 to biofuels, polyhydroxybutyrate, fatty acid ethyl esters, and other useful substances have been considered [10, 11]. Various novel production processes have also been devised, including the direct use of exhaust gas from thermal power plants or wastewater treatment facilities as sources of carbon dioxide gas, the development of photobioreactors suitable for large-scale outdoor cultivation, and microbial mineral carbonization in combination with metal recovery [12], which are considered key to success.

2.2 Fossil fuel conservation and substitution

The rapid increase in energy consumption has raised concerns regarding the depletion of fossil fuels, such as oil and gas. According to the Hubbert Peak Theory, oil resources are expected to be severely depleted within the next 40 years [13]. It is also estimated that currently mined oil reserves will be exhausted in 42 years and natural gas reserves in 60 years. In reality, fossil fuel depletion is unlikely to occur because there are still unexploited fossil fuels on Earth, but the prices of newly mined oil and gas will be much higher than those of today. Furthermore, the consumption of fossil fuels is a major source of CO_2 emissions; thus, fossil fuel consumption must be reduced. Therefore, shifting from fossil fuels to sustainable renewable energy is important for securing energy and preventing global warming [14]. Here, we introduce renewable energy initiatives from a biotechnological perspective.

Biofuels are used as renewable alternative fuels [15, 16]. Ethanol and biodiesel, produced from food and animal feed such as corn, soybeans, rapeseed, or used cooking oil, have been developed as first-generation biofuels. They are already used in transportation as substitutes for conventional fuels, and their production continues to increase. However, producing these biofuels requires large tracts of farmland, which is undesirable because it pressurizes food production. Therefore, second-generation biofuels have been designed using inedible lignocellulosic feedstocks accumulated as agricultural waste. However, this method has not yet been put to practical use because of issues related to the stability of the feedstock supply and its preprocessing.

Third-generation biofuels are currently being developed via transesterification and hydrogenation of oil produced by microalgae [17]. As mentioned, microalgae are ideal for biofuel production and global warming mitigation because of their high CO_2 fixation capacity and, at the same time, high lipid content. Microalgae are also superior in that they can produce various biofuels, such as hydrogen and biodiesel by transesterification, bio-oil, and bioethanol, but their production costs are higher than those of fossil fuels and biodiesel. Currently, genetically modified algae and algae improved by gene editing using CRISPR/Cas9 have been developed to increase oil accumulation. Wastewater treatment and the use of photochemical and electric fuels are also being investigated [18, 19].

Cyanobacteria, considered as photosynthetic microorganisms, have been genetically modified to produce biofuels [20]. Cyanobacteria produce biofuels from the carbon produced by photosynthesis using CO_2 and water. For example, genetic modification has been reported to improve the production of bioethanol, isobutanol, isoprene, and fatty acids that are attracting attention as alternatives to gasoline [21], and strains that secrete biofuels extracellularly have also been created through genetic modification. If the use of genetically modified algae and cyanobacteria is accepted in biofuel production, they may be used as renewable alternative fuels.

To reduce CO_2 emissions and fossil fuel consumption, gasoline-powered vehicles are being replaced by electric vehicles (EVs). The European Union (EU) has effectively banned the sale of gasoline-powered vehicles, including hybrid vehicles, for 35 years and has decided to shift to EVs and fuel-cell vehicles. However, securing sufficient electricity from hydroelectric and wind power generation is difficult and replacing all cars with EVs will require generating vast amounts of new electricity. For example, Norway has become a leading EV country with revenues generated by its abundant oil and natural gas, but as a result, the fossil fuels that Norway exports emit CO_2, which has not led to a reduction in CO_2 emissions. In the future, it will be necessary to face an electricity shortage by generating electricity from solar and fuel cells in each household instead of supplying electricity from electric power companies, such as in the past.

Hydrogen fuel cell is one of the promising fuel cells [22, 23]. Hydrogen fuel cells generate electricity from chemical energy using hydrogen and oxygen and have many advantages, such as almost zero emission of CO_2 and environmental pollutants, low vibration and noise, easy installation inside buildings and in urban areas, and high energy efficiency using waste heat. Hydrogen fuel cells can also be used as fuel for cars and trucks. Hydrogen-fueled vehicles are being developed in China and Japan. Various fuels, such as natural gas and methanol, can be used to extract hydrogen, but the challenge is to produce hydrogen at a low cost. Although the production capacity is not yet sufficient, methods using photosynthetic microalgae and cyanobacteria have been investigated to produce hydrogen gas [24, 25]. Microalgae that absorb CO_2 while producing hydrogen are attracting particular attention because natural gas emits CO_2 during hydrogen production.

In contrast, the development of microbial fuel cells (MFCs) is also underway [26, 27] MFCs are bioreactors that convert organic compounds into electrical energy under anaerobic conditions through microbial catalysis and convert biomass into electricity, such as wood and food waste as well as wastewater. It is a desirable renewable energy source. Currently, MFCs have not been put to practical use because of their high cost, low expression rate, and lack of scaled-up capabilities. However, in recent years, with the use of molecular tools and genetic systems of microorganisms combined with genetic manipulation and electrochemistry, improvements in electric fields and a new style of MFC using wastewater [28, 29], the amount and stability of power generation have rapidly improved and future developments are expected.

2.3 Substitution of petroleum-based polymers

The CO_2 emitted by the production and incineration of plastics was about 850 million tons in 2019, whereas open-air combustion may emit 1 Gt [30]. In addition, newly initiated industrial projects continue to increase the production capacity of petrochemical plastics with a potential CO2 emission of 2.8 Gt by 2050 (10–15% of the global carbon budget). To reduce petroleum consumption and CO_2 emissions simultaneously, the consumption of petrochemical products, especially plastics, must be reduced; and the recycling and reuse of plastics must be implemented. Currently, PET bottles are being actively recycled, and plastic cups and other plastic products are being regulated in Japan. The substitution of paper products is progressing but not sufficiently. The dependence on paper as a substitute will lead to the deforestation of forests including mangroves, and the resulting sharp decline in the number of plants worldwide will spur an increase in atmospheric CO_2 emissions.

Bioplastics have attracted attention as alternatives to petroleum-based polymers [21, 31, 32]. Bioplastics are bio-based, biodegradable, or a combination of both. In addition to synthesizing new polymers, “drop-in polymers,” which are substitutes for various petroleum-based polymers, can also be produced from biomass-derived carboxylic acids, alcohols, vinyl monomers, and amides. For example, the aliphatic homopolymer PLA can be produced by the polycondensation of lactic acid from fermentation (or ring-opening polymerization of lactide), with an annual production capacity of over 250,000 tons [33]; PHA is a polymer that can be synthesized by bacteria and accumulates in high concentrations in the cells, and the production of polymers from biomass is a promising alternative to petroleum-based polymers. It is expected to be produced at 100,000 t/year within the next few years [34]. In addition, PET is produced by polycondensation of the bio-derived monomer monoethylene glycol with 2,5-furandicarboxylic acid, and bioPET is obtained synthetically or microbially from biomass. PET production is expected to increase dramatically in the future, owing to the progress of PET recycling [34]. Although biopolymers are used in significantly smaller quantities than petroleum-derived polymers, they are expected to play an increasingly important role in reducing fossil fuel consumption.

3. Biotechnology strategies for global environmental restoration

3.1 Soil contamination resulting in severe food and drinking water scarcity

The excessive utilization of persistent organic pollutants (POPs) has severely damaged soil and groundwater. In the 1980s, these pollutants were widely employed as pesticides. Because of their prolonged persistence in the environment, they contaminated groundwater and have accumulated in birds and fish. To address this issue, the 2001 Stockholm Convention prohibited the production and use of POPs. Following that, organophosphorus pesticides (OPPs), which exhibit a much shorter persistence in soil than POPs, were banned in the EU in the 2000s following the reports of contamination in drinking water. Triazine herbicides developed in the 1970s were found to have endocrine-disrupting effects on frogs. The EU established an upper limit for their field use, whereas the U.S. Environmental Protection Agency legally regulated the maximum amount (3 ppm) of atrazine in drinking water. Despite these efforts, the excessive use of pesticides to improve crop yields [35] continues, particularly in Asia, Africa, and South America, exacerbating food concerns. Additionally, some areas still employ the stocks of POPs produced before the implementation of the Stockholm Convention.

Soil contamination by toxic metals is also becoming increasingly serious. For example, arsenic (As), because of the widespread use of Ga-As and Se-As semiconductors in consumer electronics, is produced in the mining industry; and large amounts of crude ore, sediments, and wastewater containing As have contaminated soil. High concentrations of As have been reported in groundwater in Asian countries, such as Vietnam, Thailand, India, and China [36–38]. In the Indian state of West Bengal, approximately 8 million people are at risk of arsenic poisoning. Other poisoning cases include As and fluorine in low-quality coal fuel and in some areas of China's Sichuan and Guizhou provinces, where coal containing very high concentrations of fluorine (500 mg/kg) is used, leading to coal-fueled fluorine poisoning in approximately 20 million people.

Pollution by toxic heavy metals, such as lead, cadmium, mercury, and hexavalent chromium, has also caused health problems [39, 40]. For example, the production of lead-acid batteries has increased sharply owing to the increasing demand for automobiles; and the direct discharge of exhaust gases and wastewater containing high concentrations of lead from many metallurgical and mining operations with inadequate pretreatment, low-quality ore, or used sludge left on the soil has caused severe lead poisoning in areas surrounding these operations. Cadmium is mainly used in Li-Cd batteries in consumer electronics, but because the recycling rate of Li-Cd batteries is as low as 20%, cadmium poisoning has been reported in various areas and around mining sites.

In 2000, the EU promulgated the waste electrical and electronic equipment (WEEE) Directive, which mandated the recycling of end-of-life vehicles and strictly limited the use of hazardous metals [41]. The 2011 revised RoHS Directive (RoHS2) expanded the number of products subject to it to approximately 20,000 products. As a result of this strict directive, the recycling rate of lead-acid batteries exceeded 90%; and alternatives that did not contain toxic metals, such as lead-free solders, were developed. However, nearly 80% of WEEE is still disposed of, and technical solutions are required. The most effective measures to halt the progression of soil and groundwater contamination are to reduce the discharge of pollutants through strict legal regulations and treaties, but at the same time, the ever-widening contaminated soil and groundwater must be remediated as soon as possible. If this is not done, there is a certainty that food and drinking water shortages will occur in the future.

3.2 Soil remediation

Bioremediation is a promising technology that uses living organisms to remove harmful contaminants through degradation, adsorption, or absorption and has the advantages of being cost-effective and widely applicable compared with physicochemical methods [42]. For *in situ* methods, the bioaugmentation method that promotes decomposition by activating indigenous microorganisms with nutrients and oxygen has been primarily used rather than the biostimulation method that adds highly degradable microorganisms [43, 44]. This is because of concerns about the damage to the ecosystem caused by the introduction of nonindigenous microorganisms. However, biostimulation methods have limited restorative capabilities. Considering the wide variety of substances currently contaminating the soil and the wide range of contamination, bioaugmentation methods must be used for remediation in the future.

Phytoremediation, which involves the use of plants for restoration, is also expected. In the case of microorganisms, it is difficult to recover from their spread in the soil. In contrast, phytoremediation is a method in which contaminants near the soil surface are absorbed or adsorbed by plants; as contaminants can be removed from the soil together with the plants, phytoremediation is advantageous for the recovery of heavy metals that cannot be decomposed. Plants are susceptible to environmental influences, do not necessarily have a high remediation capacity, and are often not economically viable. To solve this problem, several plants that can effectively absorb and accumulate metal ions, called "hyperaccumulators," have been discovered [45]. For example, *Rinorea nicolifera*, found in the Philippines, can accumulate 18,000 ppm of nickel [46]. Furthermore, attempts have been made to reduce production costs by combining them with plants that produce useful substances, such as herb-producing plants.

Recently, enzyme-based bioremediation methods have attracted significant attention [47, 48]. Enzymes, such as oxidoreductase, laccase, hydrolase, and peroxidase, are used in bioremediation. Because they are proteins, they do not require the removal of accumulated biomass from the treated area; however, they are not suitable for the adsorption of heavy metals because they cannot be removed without immobilization. Therefore, various methods for enzyme immobilization have been investigated [48].

3.3 Biotechnological strategies for practical remediation

As mentioned earlier, wild strains of microorganisms and plants have not achieved economically viable remediation capabilities. For practical use, it is necessary to use strains with tens to hundreds of times higher decomposition capacities and excellent operability using the latest technologies, including genetic modifications. Another advantage of using recombinant microorganisms and plants is that they can provide the ability to decompose and absorb pollutants even under extreme climatic conditions. Widespread pollution occurs mainly in harsh climates, such as acidic, cold, and arid climates. However, microbial activity and plant growth are less resistant to extreme environments, which is a disadvantage of biological remediation processes. Therefore, it would be useful to utilize genetic technology to add the ability to maintain high activity in harsh climates. Many microorganisms that degrade and remove pollutants have been discovered [49]. For example, many microorganisms with improved degradation ability of triazine pesticides and organophosphorus herbicides and microorganisms with high absorption and adsorption ability for heavy metals have been discovered; their degradation pathways and absorption mechanisms have been clarified, and most degradation genes have been cloned.

Furthermore, new methods are being developed to identify and utilize enzymes from unknown organisms and artificial enzymes using new techniques. For example, metagenomics is a method used to identify the enzymes of unknown organisms. Ninety-nine percent of all microorganisms are unculturable, and their genetic information has been overlooked. Conversely, metagenomics makes it possible to extract the genetic information of unculturable microorganisms and dramatically increase the repertoire of enzymes. It is now possible to obtain genetic information on highly active enzymes and microorganisms in specific environments [50, 51]. For example, fewer than 20 nitrilases have been discovered using conventional screening, whereas more than 300 unknown nitrilases have been discovered in metagenomic libraries [52]. Using this method, metagenomic analysis is becoming essential for bioremediation, as new metallothionein families (environmental metallothioneins) have been found [53]; and metagenomic analysis is actively used for the removal of heavy metals, such as hexavalent chromium, which is becoming indispensable for bioremediation.

Molecular evolutionary engineering has facilitated the development of superior artificial enzymes [54]. Artificial enzymes with excellent activity and stability were produced after introducing mutation either by site-mutagenesis based on computational design or Et-PCR or DNA shuffling and repeating the selection process using DNA, RNA, or ribosomal display. For example, the thermostability of transglutaminase was improved 12-fold at 60°C by introducing saturating mutations [55]. In addition, computational design and site-mutagenesis methods have been used to create eight new enzymes that catalyze Kemp dephosphorylation reactions that are not found in nature [56]. The modification of enzymes for environmental remediation by molecular evolutionary engineering is still in its infancy, but it is expected to become an important area in the future.

CRISPR-Cas9 and other genome-editing technologies are also being actively used. CRISPR-Cas9 is a technology for silencing by cutting specific gene portions. However, further advances have led to the use of CRISPRa and CRISPRi to control gene expression, as well as CRISPR to rewrite gene sequences, such as BaseEditor or Target-AID. This novel technology is convenient for use in environmental restoration because (1) it can edit both genes, even if the chromosomes are diploid, making it excellent for gene editing in plants and other organisms; and (2) it is not part of a genetically modified organism, making it suitable for environmental restoration. Many genetically edited plants have been produced in recent years, and their decomposition and adsorption activities have increased. For example, gene editing has produced rice and corn with improved yields or plants that have been enhanced with carotene and other substances [57], and the application of plants for phytoremediation has rapidly expanded in recent years [58, 59].

In synthetic biology, attempts are being made to use these new technologies and databases to redesign and reconstruct microorganisms and create new biological systems (or reaction modules) with enhanced functions [60]. Optimized devices, or microorganisms carrying these devices, have been created by restructuring molecular tools and genetic frameworks to degrade pollutants. Wang *et al*. created a microorganism in which a phenol hydroxylase gene and seven catecholamine-degrading genes were incorporated into *Escherichia coli* [61]. This microorganism uses phenol as the carbon source in wastewater contaminated with crude oil. Dueber et al. increased the reaction rate nearly 89-fold by creating a synthetic protein scaffold (module) that closely aligned the binding domains with the enzymes involved in the pathway, mimicking the substrate tunneling effect [62].

Consortia are also important for improving bioremediation capacity. Plants and microorganisms, such as rhizobacteria, live in natural settings, helping each other; and there is excellent synergy, especially in the case of toxic metal ion adsorption [63, 64]. Although many microorganisms exhibit a high resistance and adsorption capacity for heavy metals, removing metal-adsorbed microorganisms from the site is complex. The removal of metal-adsorbed plants from soil is easy; however, the resistance of plants to heavy metals is lower than that of microorganisms, and they cannot adsorb contaminants below a depth of 1–2 m. Therefore, a combination of rhizobacteria and plants would enable effective remediation. Plants undergo apoptosis when the water content is insufficient. If microorganisms that secrete polymers, such as polyglutamic acid and chondroitin, are combined with plants, they can be used as soil humectants, and some plants can grow even in dry climates. Moreover, aloe can grow with little water, accumulate a large amount of water components in its leaves, and keep growing new leaves without withering [65], suggesting that aloe can be used as a host plant for genetic manipulation for more effective remediation.

Nanoremediation, a combination of nanotechnology and bioremediation, has attracted attention as a novel technology [66]. Nanoparticles (NPs) are useful tools for remediating contaminants because of their small size (1–100 nm), large specific surface area, and reactivity for adsorption, redox reactions, and precipitation. The combination of nanoparticles and microorganisms can promote purification efficiency. For example, in nano-phytoremediation, which combines NPs with plants, the NPs mitigate the toxicity of metal contaminants and promote plant growth. Consequently, the removal rate of toxic substances improved. However, the safety of NPs released into the ecosystem has not yet been well studied and warrants further research. However, nanoparticles are toxic for humans and for certain animals due to their small size (nano size). Especially in cases of inhalation, nanoparticles (NP) mixed with oxygen or air can cause serious life-threatening problems. Furthermore, during breathing, the lungs, brain, and cardiovascular neural system are exposed to serious life-threatening illnesses, such as pneumonia, chronic cough, inflammation of the trachea, lung diseases, heart diseases as well as mental disorders and disorders of the nervous system. Therefore, when working with NPs, it is necessary to observe the maximum safety and to be well informed about the toxicity of nanoparticles (NP) and the potential danger to the human body.

4. Conclusions

The global environment is deteriorating rapidly. To ensure a sustainable society, there is an urgent need to prevent global warming by reducing greenhouse gases, developing renewable energy sources to replace fossil fuels, and securing drinking water and food supplies by remediating soil and groundwater contamination. It is no exaggeration to say that if these measures are not accomplished, humanity will face a crisis by the end of this century. These issues must be addressed by combining various scientific technologies; biotechnology is also important. The production of biofuels, bioplastics, and microbial batteries using microorganisms can reduce CO_2 emissions and provide renewable energy. The remediation of soil and groundwater contamination using microorganisms and plants can increase the availability of drinking water. Biotechnology is expected to play a significant role in the future.

However, several problems to overcome in the future are still present. One is expensive running costs because of insufficient production and degradation capacities of microorganisms. To solve this problem, it is necessary to construct prominent microorganisms by the use of genetic modification, gene editing, and molecular evolutionary engineering and to build optimal processes. Another problem is the difficulty of using genetically modified organisms in the environment. The legal regulation prevents the widespread use of fuel production by recombinant algae and bioremediation with recombinant microorganisms or plants. Although, we must carefully consider the impact on ecosystems; I think that the active use of genetically modified bacteria and plants in the environment cannot be avoided because it may be a final option to realize a sustainable society in the future.

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