J. Aravind
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Strategies and Tools for Pollutant Mitigation

Research Trends in Developing Nations

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Part I

State of Art and Sustainable Remediation Approaches

Chapter 1
Utilizing Organic Wastes for Probiotic and Bioproduct Development: A Sustainable Approach for Management of Organic Waste

Raghuvandhanan Kumarasamy Sivasamy, Kumaresan Kuppamuthu, Lokesh Krishnasamy Nagaraj, Sakkthy Pradhieksha Manikandan, Raghul Kulandaivel, and Jenifer Gabriella Bastin

Abstract In recent years, organic waste, specifically food waste, has become a growing concern with the increased population. This surge is negatively impacting the environment. These food wastes have emphasized the importance of employing sustainable waste management strategies so that these wastes can be transformed into some value-added goods. Most of the food waste is rich in nutritive supplements that hold massive significance for bioconversion to value-added products and the growth of various microorganisms. One such type of microorganism is probiotics. Probiotics are not pathogenic microorganisms and have potential health benefits to the host when administered in modest amounts. It has proven benefits for humans and animals. This chapter widely focuses on and discusses the environmental impacts that are caused by different organic wastes, assess the existing methods for organic waste management and their limitations, probiotic strategies such as the utilization of organic waste as a supplement in the media for their growth, probiotic fermentation of organic waste. The microbial approaches discussed in this chapter offers a sustainable way for managing food waste by converting it into valuable bioproduct.

Keywords Organic waste · Bioconversion · Probiotics · Food waste · Organic waste management

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1.1 Introduction

Organic wastes are materials that originate from living organisms. These wastes include food waste, agricultural waste, and sewage waste from the treatment of wastewater. Among organic wastes, food waste is a significant source of organic waste (Raksasat et al. 2020). Food is an essential and indispensable part for the survival of any form of life. Organisms from different evolutionary levels take in food in various forms. Microorganisms take food in the form of macromolecules and higher eukaryotes take food in complex forms. Concerns about food develop when there is a considerable amount of waste produced from it which can be used for various purposes. Food and Agriculture Organization (FAO) describes food waste as the reduction in the quality or quantity of food resulting from actions and decisions by retailers, food service providers, and consumers. It is estimated that about 1.3 billion tons of food waste are spawned per year (FAO 2015), that is, one-third of the food produced for human consumption, either lost or wasted globally (FAO—Notícias: Food Wastage: Key Facts and Figures 2015).

Food waste generation is increasing and has much impact on the environment and the economy. It is estimated that food waste accumulates about 3.3 billion tonnes of CO_2 into the atmosphere per year (Paritosh et al. 2017). The negative impact has drawn attention to managing food waste. Food waste management has become a key to all the activities identified with reducing, avoiding, or recycling waste, throughout the production and consumption chain. Bioconversion and biotransformation of food waste by microbial methodologies using probiotics is one significant way of managing food waste. The key requisites for the use of food-based waste as a resource to develop value-added products are shown in Fig. 1.1.

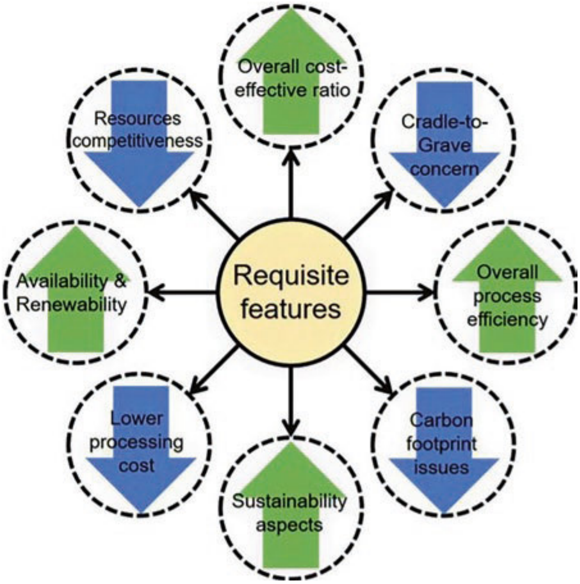

Fig. 1.1 Key requisite features that enable the use of food-based waste materials to develop value-added products of interest. (Reprinted from Bilal and Iqbal 2019 with permission from Elsevier)

In recent times, there has been a rapid rise in interest in probiotics and their applications. This chapter aims to present how organic waste, especially food waste, could be managed and utilized efficiently as a media for the growth of probiotics, bioconversion of the waste to bioactive compounds using probiotics, and the application of those bioactive compounds.

1.2 Organic Waste and Its Environmental Impacts

1.2.1 Fruit and Vegetable Waste

Fruit and vegetable waste (FVW) are commonly defined as waste intended for disposal from fruit and vegetable processing and production areas (Plazzotta et al. 2017). FVW is rich in energy, nutrition, and moisture content consisting of carbohydrates (glucose and fructose), polyphenols, fibers, minerals, and other bioactive compounds. Fruits and vegetables are considered waste only when the degree of acceptance from the consumer tends to reduce. This acceptance is reduced by many factors, including the degree of ripening, biochemical reaction, microbial attack, and discoloration.

About 59 million tonnes of fruit and vegetable waste is generated, which costs 2 trillion annually (Singh et al. 2007) and thus poses a threat to the environment due to high biodegradability, which will deplete valuable biomass and causes a financial burden to industries. Anaerobic digestion is one of the most commonly followed techniques to manage fruits and vegetable waste. Improper utilization of the digestate obtained through anaerobic digestion can pose serious environmental problems that include over-fertilization and pathogen contamination (Nkoa 2014). Most of the fruit wastes are decomposed in landfills and emit harmful greenhouse gases causing environmental burden (Gowman et al. 2019; Kumar 2012).

1.2.2 Agricultural Waste

Agriculture waste is an organic, biodegradable, and unwanted product produced by agricultural practices which include molasses, straw, spent grass, husk of rice, wheat and maize, shells of coconut, groundnut and walnut, the skin of avocado and banana, plant waste, livestock and poultry waste which is used as a manure. Nonedible waste from various sources is considered agro-industrial waste. For producing multiple high-valued commodities, a natural substitute can be agricultural wastes. Agricultural waste is rich in lignocellulose, which can be used to produce many microbial enzymes (Ravindran et al. 2018).

About 350 million tonnes of agricultural waste is produced, which imparts a negative impact on the environment. Runoff from the land has high amounts of nitrogen and phosphorus that can speed up the process of eutrophication in lakes and ponds (Atallah Abouelenien et al. 2014). Furthermore, the decomposition of agricultural waste like the organic matter in crops emits gases like hydrogen sulfide and methane which can cause air pollution by generating greenhouse gases (Qi et al. 2020).

1.2.3 Food Waste

Food waste (FW) is food products that do not get consumed; they are either nonedible parts or leftovers. FW mostly takes place in the consumption stage of the food supply chain (Parfitt et al. 2010). Food waste majorly consists of carbohydrates, lignin, lipids, and proteins. The carbohydrates can be broken down into oligosaccharides and monosaccharides; these are fermentable and can be used to develop bioproducts such as biopolymers, bioplastics, hydrogen, methane, and various enzymes (Uçkun Kiran et al. 2014).

About 67 million tonnes of food is wasted. There are several methods for managing food waste but, anaerobic digestion and composting are widely used methods. The gas emitted from this process is the main contributor to acidification, photochemical oxidation, and eutrophication (Al-Rumaihi et al. 2020).

1.2.4 Dairy Waste

Dairy products are the most cherishable products due to their complex organic constituents. Dairy products are diverse and include yogurt, cheese, butter, ice creams, and various milk products. Milk production has increased in the last few years as a result of industrialization. The processing of dairy products has also increased and is considered the primary source of industrial food wastewater (Slavov 2017).

About 5 million tonnes of dairy products are wasted. Dairy effluents that are sent out contain milk constituents like casein and inorganic salts, along with detergents and sanitizers. All of these components cause a rise in BOD and COD (Sinha et al. 2019). Dairy effluents have suspended solids and soluble organics. They promote the release of certain gases and eutrophication (Raghunath et al. 2016). Some chemicals like ammonia, nitrates, and nitrogen are present in raw milk, which are known to cause methemoglobinemia. When this is converted to nitrate, they pollute groundwater (Ahmad et al. 2019).

1.3 Management of Organic Waste

There are different types of managing organic waste based on the nature of the waste. The most common type of organic waste is food waste. The primary and the most followed way of managing food waste is through landfills (about 90%), then composting (about 1–6%), and anaerobic digestion (0.6%) (Thi et al. 2015).

1.3.1 Solid Waste Management

Anaerobic Digestion (AD) overcomes the traditional food waste management method, and it is comparatively cost-effective to other waste treatment options. In AD, anaerobic microorganisms convert various organic waste and biomass into biogas like carbon dioxide, methane, and small amounts of other gases like hydrogen (Xu et al. 2018). Complex organic polymers are converted into simple soluble biomolecules by hydrolysis and then into fermentation to form a volatile fatty acids mixture further converted into acetate (Kibler et al. 2018). The rate-limiting step of the AD is the hydrolysis process (Zhang et al. 2014). A successive co-digestion of food wastes with organic substrates by physical, thermochemical, and biological methods is performed to improve hydrolysis. The mesophilic and thermophilic conditions are suitable for the effective digestion of food wastes: higher buffer capacity and ammonia yield higher amounts of methane.

1.3.2 Incineration

Incineration is the combustion of food waste by supplying thermal energy source that occurs in a grate furnace. Incineration is not appreciated compared to other conventional food management systems because of the environmental and economic impact. The high capital requirement is due to the energy source, furnace designing, and minimizing the release of smoke into the atmosphere (Thi et al. 2015). The recovery of available nutrients and other valuable chemical compounds is hindered in this process, a notable economic impact. The time requirement is less in the incineration process and also a high volume of food waste is incinerated quickly. Potential health effects regarding inhalation of airborne pollutants resulting from incineration is possible (de Titto and Savino 2019). Burning food waste with moisture content leads to the release of dioxins and mercury compounds which causes several environmental problems and even acts as a carcinogen (Melikoglu et al. 2013).

1.3.3 Hydrothermal Carbonization

Hydrothermal Carbonization (HTC) is a wet process of thermal conversion technology that produces valuable energy-rich sources from food wastes under autogenous pressure and relatively low temperature (180–350 °C) (Lu et al. 2012). The reaction time of HTC is less than an hour, so it continuously degrades waste material daily, improves the food waste (FW) management, and high throughput of products. The carbonization process integrates more than 70% of carbon in the FW into carbon and results in hydro char with a higher density of energy source. It can be applied to the soil amendment process for plant growth, locks the moisture in it, and enhances the soil moisture content by increasing soil porosity (Li et al. 2013a). Studies on life cycle assessment of HTC treated water and emission of CO_2 can pose a significant footprint (Venna et al. 2021).

1.3.4 Landfills

Landfills are one of the traditional and most widely used methods for managing food waste and solid waste. The waste generated is dumped in a particular area separated from the living areas and requires many resources like land and money (Kim and Kim 2010; Xu et al. 2018). Landfilling method is known to have a more considerable effect on climate change; it is ten times as much as anaerobic digestion, composting, and incineration combined (Gao et al. 2017). Methane is usually emitted from landfills because of the degradation of organic waste (Kibler et al. 2018). Methane, when released into the atmosphere, causes global warming at a higher rate than carbon dioxide. Leachate, a potent toxic liquid, is also leaked into the soil and groundwater because of landfills (Melikoglu et al. 2013).

1.3.5 Composting

Composting is a process of the biological decomposition of organic waste under aerobic conditions. The end products of this process are fertilizer, fuel, or biofilter material. Unlike landfills, composting does not threaten underground water because the chemical pollutants are low comparatively (Ayilara et al. 2020). During composting, some factors can be considered, such as the addition of moisture during the process (Kibler et al. 2018). There may be some chemical changes and metabolic changes of the microorganism. Compared with other waste, food waste has particular physical and chemical properties like loose physical structure, high nitrogen content, low C/N ratio. So composting became an essential method for managing organic waste (Li et al. 2013b).

1.3.6 Animal Feed

Organic waste like food waste is generated during the consumption phase of the supply chain is rich in nutrients (Bakshi et al. 2016), which can be used for livestock feed, and modern treatment technologies can be used to convert food waste that is high in moisture content and susceptible to deterioration into feed that is safe for animal feed (Dou et al. 2018). Waste is treated to become free of contamination; it is sterilized and dehydrated by hot air at 390 °C. It should be heat treated at greater than 80 °C for 30 min. The animal feed ingredients are dependent on the feed consumption of the pigs for pork production (Salemdeeb et al. 2017).

1.3.7 Biovalourization

Food waste biovalorization converts food waste or its byproducts into higher-value products that contribute to the food supply chain. It is one of the recycling pathways for food waste that will help to close the food waste loop. This technique is aimed at the generation of biofuels and biomaterials. The production of biofuels from organic waste has become necessary because of the depletion of fossil fuels (Nayak and Bhushan 2019). Organic waste can be used to produce biogas with a yield of 150 Nm^3/ton of waste. It is used to produce electricity (Cristóbal et al. 2016).

1.3.8 Dairy Waste Management

The milk industry has a more significant environmental impact because it consumes large amounts of water and produces effluents. Different types of dairy waste management methods include mechanical treatment, physiochemical treatment, and biological treatment.

1.3.8.1 Mechanical Treatment

Mechanical treatment involves removing suspended solids from dairy wastewater. These are removed by using screens. The removed material is collected in the bottom and is known as sludge (Slavov 2017).

1.3.8.2 Physiochemical Treatment

The physiochemical treatment removes colloidal particles and reduces milk fat. Electrocoagulation and adsorption are two widely used methods for removing dissolved organic waste (Birwal et al. 2017; Slavov 2017).

1.3.8.3 Biological Treatment

The preferred way of managing dairy waste is biological treatment. This method is mainly used for removing organic material. Biological treatment is classified into aerobic process and anaerobic process based on the oxygen requirements.

In aerobic process, the microorganisms used in this process grow in an environment rich in oxygen. The microorganism breaks down the organic compound into the water, carbon dioxide, and cellular material. The aerobic process is not as efficient as the anaerobic process because of acidification and filamentous growth (Birwal et al. 2017; Slavov 2017).

In anaerobic process, The microorganisms used are grown in the absence of oxygen. Organic matters are converted to biogas and are cost-effective than the aerobic process. So, the anaerobic process is preferred to the aerobic process (Birwal et al. 2017; Slavov 2017).

1.4 Sources of Probiotics

Probiotics that are helpful for human beings' well-being can be isolated from various sources such as dairy products, wastes like vegetable and fruit waste, kitchen waste, plant material, animal material, human guts, human feces, and human milk (Sornplang and Piyadeatsoontorn 2016) (Fig. 1.2).

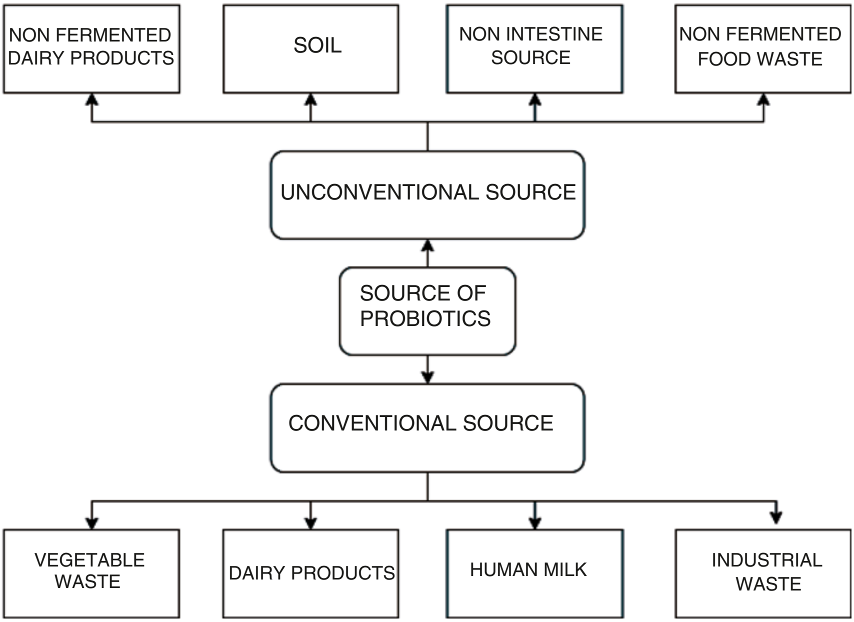

Fig. 1.2 Different conventional and unconventional sources of probiotics

1.4.1 Fruit Waste

During the processing of fruits and vegetables, thousands of tonnes of solid and liquid waste are produced. Solid waste is generated in the form of skins, pips, and stalks. Fruit waste and vegetable waste may contain many valuable sources for the growth of bacteria. Fruit and vegetable waste are rich in many nutrients like iron, magnesium, and carbohydrates which are the primary source of the growth of probiotic bacteria. Some strains of Lactobacillus were isolated from byproducts of fruits. *L. fermentum 139* and *L. fermentum 141* were isolated from the byproducts of *Mangifera indica*. *Lactobacillus plantarum 60*, *Lactobacillus fermentum 56*, and *L. fermentum 53* were isolated from the byproducts of *Malpighia glabra* (Barbados cherry). *Lactobacillus paracasei 106* was isolated from the byproducts of *Annona muricata* (soursop). *L. fermentum 250* and *L. fermentum 263* were isolated from the byproducts of *Ananas comosus* (pineapple). *L. fermentum 296* was isolated from the byproducts of *Fragaria vesca* (strawberry). These strains were identified using the 16S rRNA sequence (De Albuquerque et al. 2018). The bacteriocin-producing bacteria such as *Lysinibacillus JX416856* was isolated from the fruit and vegetable waste and was identified phenotypically and molecularly (Ahmad et al. 2014). The probiotic strain *Lactobacillus rhamnosus AW3* was isolated from the date processing wastewater. Wastewater was collected from a date fruits processing center, the bacterial strain was isolated from date effluent, and complete 16S rRNA was done to identify the strain at the molecular level (Al-Dhabi et al. 2020). The probiotic strains such as *Pichia kudriavzevii* and *Issatchenkia terricola* were isolated from pomegranate and grape seed, respectively. These were identified by the 18S rDNA sequence using ITS1 and ITS4 method (Prabina et al. 2019).

Fermented vegetables and fruits are some of the potential sources of probiotics because they nurture various lactic acid bacteria. Some of which include *Lactobacillus pentosus*, *Lactobacillus plantarum*, *Lactobacillus fermentum*, *Lactobacillus acidophilus*, *Lactobacillus mesenteroides*, and *Lactobacillus brevis* (Swain et al. 2014). Fermented vegetables predominantly contain *Lactobacillus plantarum* and *Lactobacillus brevis* because of their ability to break down phenolic acids present in food (Viridiana et al. 2018).

1.4.2 Dairy Products

Dairy products are one of the essential sources of probiotic microorganisms. In the Asian market, fermented milk and yogurt is an essential probiotic product. *Streptococcus thermophilus*, *Streptococcus cremoris*, *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus rhamnosus*, *Lactobacillus bulgaricus*, *Lactobacillus kefir*, and *Streptococcus lactis* were isolated from a variety of dairy products like yogurt, cultured buttermilk, acidophilus milk, lassi, kefir, and leben (Oh 2015). Certain probiotic species like *Enterococcus* and *Bacillus* were isolated from raw milk and identified using the 16S rRNA method (Panda et al. 2017).

1.4.3 Human Sources

The human gut microbiome harbors many microorganisms like probiotics. Potential probiotic bacterias like *L. rhamnosus*, *L. fermentum*, *L. plantarum*, and *L. paracasei* were isolated from the feces of infants less than 24 months. These bacteria were examined for probiotic characteristics like acid pH resistance, bile tolerance, adhesion assay, and inhibition of enteric pathogens (Jomehzadeh et al. 2020). In healthy women, vaginal microbiota is rich in probiotics. This microbiota is dominated by *Lactobacillus* species and certain species such as *L. gasseri*, *L. salivarius*, *L. crispatus*, *L. helveticus*, *L. fermentum*, *L. rhamnosus*, *L. paracasei*, and *L. plantarum* (Er et al. 2019; Pino et al. 2019). Breast milk is recognized as one of the primary sources of potential probiotic bacteria. There is vast biodiversity of bacterial species in human milk. Certain probiotics like *Lactobacillus casei* and *Lactobacillus rhamnosus* were isolated from human milk and identified using the 16S rRNA sequencing method (Riaz Rajoka et al. 2017). Seven strains of Lactic acid bacteria were isolated from human milk (Kavitha and Devasena 2013). Some major probiotic species isolated from human milk include *Streptococci*, *Staphylococci*, *Lactic acid bacteria*, *Bifidobacteria*, and *Corynebacteria* (Martín et al. 2003, 2009, 2012).

1.4.4 Fish Intestine

The fish intestine is a rich source of probiotics. Lactic acid bacteria (LAB) were isolated from the kitchen waste and fish intestine. About five strains such as KT1T, KT2W, KT1B, KA2, and FS was identified as *Lactobacillus casei*, and KT1 strain was identified as *Lactobacillus delbrueckii* (Rauta et al. 2013). Five strains of the *Lactococcus lactis* subsp. *lactis*, one *Lactobacillus plantarum*, two *Enterococcus* spp., and one *Leuconostoc mesenteroides* were isolated from the guts of 12 marine species (Alonso et al. 2019).

1.4.5 Soil

Probiotics that are usually found in soil are called soil-based probiotics. *Bacillus amyloliquefaciens* is a probiotic bacteria and it has been isolated from North East Himalayan Soil (Hairul Islam et al. 2011). Soil (rhizospheres) samples from Taiwan and Japan had possible probiotic bacterias like *Lactococcus lactis*, *Enterococcus faecium*, *Enterococcus mundtii*, *Lactobacillus plantarum*, and *Sporolactobacillus inulinus*. These strains were identified using the 16S rDNA sequence method (Chen et al. 2005) (Table 1.1).

Table 1.1 Probiotic organisms isolated from different sources, the media used to cultivate them, and the identification techniques

Source	Probiotic strain	Identification techniques	Medium	Reference
Human milk	*Lactobacillus casei*, *Lactobacillus rhamnosus*, seven strains of lactic acid bacteria	16S rRNA	MRS agar, TPY agar, MRS cysteine agar	Riaz Rajoka et al. (2017)
Mango pulp	*Bacillus JHT3*, *DET6*	16S rRNA	Nutrient agar	Patel et al. (2009)
Dairy waste	*Siderophoregenic Bacillus DET9*	Partial 16S rRNA, biochemical characterization	MRS agar	Patel et al. (2010)
Fermented vegetable, silages, grass	*Lactococcus lactis* subsp. *lactis*	PCR	MRS agar	Kimoto et al. (2004)
Fish intestine	*Lactic acid bacteria KT1T*, *KT2W*, *KT1B*, *KA2*, *FS*, *Lactobacillus delbrueckii*	Gram's staining and biochemical tests	MRS agar plate	Rauta et al. (2013)
Fruit and vegetable waste	*Lysinibacillus JX416856*	Phenotypical and molecular method	MRS agar	Ahmad et al. (2014)
Fish intestine	Five strains of the *Lactococcus lactis* subsp. *lactics*, one *lactobacillus plantarum*, two *Enterococcus* spp., and one *Leuconostic mesenteroides*	PCR	MRS agar	Alonso et al. (2019)
Mango, barbodos cherry, soursop, pineapple, strawberry	*L. fermentum 139*, *L. fermentum 141*, *Lactobacillus plantarum 53*, *Lactobacillus fermentum 56*, *L. fermentum 60*, *Lactobacillus paracasei 106*, *L. fermentum 250*, *L. fermentum 263*, *L. fermentum 296*	Complete 16S rRNA	MRS agar	de Albuquerque et al. (2018)
The seed of pomegranate and grape	*Pichia kudriavzevii* and *Issatchenkia terricola*	18S rRNA	Yeast extract peptone dextrose (YEPD) agar supplemented with chloramphenicol	Prabina et al. (2019)
Traditional fermented dairy products	*Lactobacillus plantarum P-8*	16S rRNA, PCR	MRS agar	Wang et al. (2015)

(continued)

Table 1.1 (continued)

Source	Probiotic strain	Identification techniques	Medium	Reference
Laying hens	*Propionibacterium acidipropionici LET 105*	16S rRNA	Lactate agar	Argañaraz-Martínez et al. (2013)
Young calves	*Lactobacillus johnsonii*, *L. salivarius*, *L. murinus*, *L. mucosae*, *L. amylovorus*, *L. mucosae*	PCR, 16S rRNA	MRS and LAPT medium	Maldonado et al. (2012)
Chickens	*Lactobacillus salivarius 15K*	PCR,16S–23S rRNA	MRS agar	Bujnakova et al. (2014)
Date processing wastewater	*Lactobacillus rhamnous AW3*	Complete 16S rRNA	MRS agar	Al-Dhabi et al. (2020)
Soil	Bacillus strains 12, 17 S10, S3, 14, 13, 8	16S rRNA	Tryptic soy agar (TSA)	Mohkam et al. (2016)
Indigenous and broiler chickens	*Streptomyces* sp. *JD9 (KF878075)*	16S rRNA, PCR	MRS agar	Latha et al. (2016)
Indigenous poultry	*Lactobacillus plantarum TN8*	PCR, 16S rRNA	MRS medium	Ben Salah et al. (2012)
Broiler chickens	*Lactobacillus salivarius DSPV 001P*	PCR, 16S rRNA	MRS agar	Blajman et al. (2015)
Cows, pigs, chickens, and ducks.	*L. plantarum (strain P6)*, *L. paraplantarum (strain P25)*, *L. reuteri (strain P30)*	PCR,16S rRNA	MRS agar with 0.1% $CaCO_3$	Pringsulaka et al. (2015)
Weaned pig	*Bacillus subtilis KN-42*	PCR, gel electrophoresis, 16S rRNA	MRS agar	Hu et al. (2014)

1.5 Organic Waste for the Growth of Probiotics

The interest in probiotics has increased recently, but the cultivation of probiotics is expensive, especially the media used for growing it. The cost of the media has a negative effect on the economic aspect of growing probiotics. Technologies for the production of active probiotic strains require low-cost media for their growth. MRS media is a suitable media for the growth of probiotics, but it is costly; specific food waste can be used as a substitute for MRS media.

1.5.1 Agricultural Waste as a Probiotic Growth Media

1.5.1.1 Banana Peel Waste as a Probiotic Growth Media

India accounts for 29% of the total banana production globally (Panigrahi et al. 2021) and food industries produce a vast amount of banana peel waste. The banana peel waste can be used as an alternative medium for the MRS medium. The banana peel is chopped into small pieces and made into a paste. Twenty grams of this paste is mixed with 100 mL of distilled water. This mixture is autoclaved, and probiotic organisms like *L. sporogene* and *L. acidophilus* are inoculated. Submerged fermentation of the Banana peel medium is carried out. The maximum growth of these probiotic Lactobacilli strains was observed at pH 6.0 and 37 °C. On comparing the growth of these strains with the traditional MRS medium and the banana peel medium, the study indicated that there was no significant difference ($p > 0.05$) (Farees et al. n.d.).

1.5.1.2 Barley Spent Grain (BSG) as a Media for Probiotics

During Beer production, byproducts are generated and 85% of it is barley spent grain (Aliyu and Bala 2011). BGS consists of lignocellulosic biomass, mainly consisting of fiber (30–70%) and proteins (20–30%). *Lactobacillus* and *Bifidobacterium* sp. can use these byproducts for their growth. Hence, this can be a component of the growth media (Song et al. 2012). *Bifidobacterium adolescentis 94 BIM* and *Lactobacillus* sp. Firstly, the BGS is separated into coarse polysaccharide fraction (FF) and fine protein fraction (PF). Two grams of these fractions are added to 100 mL of distilled water. This mixture is autoclaved and the pH is adjusted to 7.2. The growth, acetic acid production, and morphology of *Bifidobacteria* and LAB were assessed by inoculating them in 11 different media compositions, including BHB (Brain heart broth) medium, media with FF and PF supplemented with additives like lactose, ascorbic acid, yeast extract, and mineral salts. The results indicated that the proteins and polysaccharide fractions of BSG supplemented with the right amount of additives can be used to cultivate probiotics (Novik et al. 2007).

1.5.2 Cheese Whey and Molasses: Media for Probiotic Bacteria to Produce Biosurfactant

Cheese whey is a byproduct of the cheese industry. Because of its high organic load, it is considered one of the most polluting byproducts of the food industry (Addai et al. 2020). However, it also contains high amounts of protein, lactose, organic acids, and vitamins, making it a suitable substrate for biosurfactant production. Molasses is an agricultural waste and byproduct of the sugarcane industry. It is composed of vitamins, organic compounds, and minerals, which are considered valuable for fermentation. The cheese whey is heated to denature proteins, and the precipitate is removed by centrifugation. The supernatant consists of 50 g/L lactose supplemented with peptone and yeast extract used as a culture media. Molasses is diluted such that the sucrose concentration is 20 g/L and supplemented with yeast extract and peptone used as a culture media. In order to check the sugar consumption, biomass yield, and biosurfactant production, 12 different media compositions were prepared with MRS broth being the control for *Lactococcus lactis 53* and M17 broth being control for *Streptococcus thermophilus A*. The best results were obtained with media supplemented with molasses. A 1.2–1.5 times increase in the mass of the produced biosurfactant per gram cell dry weight was also observed. This reduced 60–80% of the expense in preparation of media (Rodrigues et al. 2006).

1.5.3 Kitchen Waste as a Media for Probiotic Production

Kitchen waste is composed of organic fragments like carbohydrates, lipids, proteins, and fat. The conversion of this waste is challenging because of its low calorific value and high moisture content. It is also a valuable source that can be used as a medium for producing probiotic bacteria. Studies have been done in this arena. In one such study, five strains of microorganisms (*Lactobacillus*, *Bacillus licheniformis*, *Bacillus subtilis*, Yeast isolated from broiler chicken gut and another yeast isolated from an inoculum used to produce alcoholic food) were mixed in an equal ratio. Kitchen waste was added to a rotary drum bioreactor and the pH was adjusted to 7.2 using Na_2CO_3; this waste mixture was heated at 110 °C for 30 min, this is then cooled to 37 °C and 5% inoculum was added to it. The growth of Lactobacillus in kitchen waste was higher than the growth of Lactobacillus in pure culture media, which implies that kitchen waste can be used for probiotic production (Yin et al. 2013).

1.6 Fermentation of Organic Waste and Production of Value-Added Products

Value-added products or compounds are generally produced by fermenting fruit and vegetable wastes. Generally, hydrogen and alcohol are produced by fermenting soluble sugars which are a result of the hydrolysis of food and vegetable waste. Acidogenic fermentation produces lactic acid and solid-state fermentation of wastes is hydrolyzed by making use of mixtures of crude enzymes for the production of succinic acid. Food waste can produce several high-end products through the fermentation process.

1.6.1 Probiotic Drinks

They are produced by fermenting a substrate with probiotic microbes. These drinks can be produced at significantly lower costs and with more valuable properties by using wastes as a substrate. Such manufactured drinks possess high antioxidant properties, good taste, and aid in improving health as they contain probiotics.

1.6.1.1 Production of Probiotic Drinks from Fruit and Vegetable Waste

The food processing industries, namely fruit and vegetable industries produce waste that can be valorized due to their bioactive potential. Around 15–30% of the raw material is wasted (Calinoiub et al. 2019). Beetroot is rich in betalains which can be utilized for making functional beverages using probiotics. Nondairy probiotic drink was developed using beetroot, rich in *Lactobacillus plantarum*, *Lactobacillus rhamnnosus*, and *Lactobacillus delbrueckii* sp. This drink can be used as an alternative to dairy drinks for people with Lactose intolerance (Panghal et al. 2017).

An inexpensive drink rich in antioxidant and probiotic properties is produced using pomegranate peel extract (POPE) and pasteurized cow milk. *Lactobacillus Plantarum* and *Bifidobacterium longum* are probiotic strains. *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus* were the starter culture strains. Both were grown in sterile skim milk and inoculated into pasteurized cow milk before use. Antioxidant activity of fermented milk beverages supplemented with pomegranate peel (FMPO) has been identified as better than regular milk. Phenol contents were high in POPE and decreased after fermentation (Al-Hindi and Abd El Ghani 2020).

Kefir grains were cultured in milk (Lactic acid bacteria) and the biomass obtained was used as a pre-inoculum for fermentation. Mango peels were freeze-dried and ground into powder. This powder mixed milk acts as culture media to which kefir grains were added for fermentation under static submerged conditions. Tests conclude that mango peel milk fermented with kefir grains have high antioxidant properties because of the release of phenolic compounds from the peel. Also, the bacterial count and growth in medium containing mango peel have shown significant results. Hence, a potential probiotic drink can be produced inexpensively using mango peels (Vicenssuto and de Castro 2020).

1.6.1.2 Production of Probiotic Drinks from Dairy Waste

Cheese whey, a byproduct of the cheese industry, has many nutrients that can harbor probiotics and can be used for producing probiotic drinks. Whey was directly used and sometimes with supplements like buttermilk powder or skim milk powder with different ratios to develop a probiotic beverage. The whey was fermented with common probiotic strain *Lactobacillus acidophilus La-5* and *Bifidobacterium animalis Bb-12*. It was stored for 21 days and throughout this period the viable cells of La-5 and Bb-12 were above 8 log CFU/mL. The whey and buttermilk powder formulation had better sensory scores than the other formulations (Skryplonek and Jasińska 2015).

1.6.2 Polyhydroxybutyrate

Polyhydroxyalkanoates are carbon and energy storage compounds present in gram-negative bacteria. As they resemble properties of synthetic plastic, PHAs especially polyhydroxybutyrate (PHB) can be used as biological plastic of lower cost and good biodegradability. PHB can be extracted from strains capable of producing it fermented in a food waste medium (Tsang et al. 2019).

1.6.2.1 Polyhydroxybutyrate Production from Dairy Waste Using Probiotics

A possible probiotic strain *SRKP-3* capable of producing polyhydroxyalkanoates (PHA) similar to *Bacillus megaterium* was isolated from brackish water. The fed-batch process is carried out and dairy waste was fed at the 12th and 24th h of fermentation. Dairy waste was given as a substrate, and PHAs were isolated from dried cells. Production of PHB was maximum at 36th h of fermentation with a yield of 11.32 g/L. Hence, using a cheap medium highly useful polymer, PHB, was produced (RamKumar Pandian et al. 2010).

1.6.2.2 Polyhydroxybutyrate Production from Agricultural Waste Using Probiotics

Probiotics such as *Bacillus megaterium* and *Pseudomonas aeruginosa* were grown in mineral salt medium (MSM) with different carbon sources such as sucrose, fructose, cane molasses, orange peel powder, and also with amino acids and vitamins supplementation. PHB analysis was done. PHB yield (1.73 g/L) and the samples inoculated in a medium containing cane molasses showed better results than the rest of the carbon sources. PHB yield was enhanced due to amino acid and vitamin supplementation and obtained polymer can be used in food packaging applications (Tripathi et al. 2019).

1.6.3 Production of Biosurfactant from Food and Agricultural Waste Using Probiotics

Biosurfactants are surface-active agents that are produced extracellularly or as a part of the cell membrane. The biochemical and the 16S rRNA analysis identified the most efficient surfactant product by *Azorhizobium* strain (Pendse et al. 2018). Non-septic production of biosurfactant from molasses by a mixed culture was investigated in the stirred-Batch reactor (Ghurye et al. 1994). The production was directly correlated with biomass production and was improved by pH control on the addition of yeast (Bakshi et al. 2016).

Nutritional requirements of the microorganism producing biosurfactant play a vital role in developing a suitable growth media. Food waste like maize powder, potato peel powder, and sugarcane bagasse can be used as a carbon source. Rhamnolipid is a type of biosurfactant produced by *Pseudomonas aeruginosa*. Paneer whey is another byproduct of the dairy industry and is used to produce rhamnolipid using *Pseudomonas aeruginosa*. Oil waste can be used for the production of rhamnolipid (Nitschke et al. 2005). The carbon source in the feed produced a low dry cell weight concentration (g/L), but the rhamnolipid concentration was very high. So, optimizing carbon and nitrogen sources available for utilization is vital for higher productivity of biosurfactants. Four different Pseudomonas species were taken. *Pseudomonas cepacia*, *Pseudomonas pickettii*, *Pseudomonas fluorescens*, and *Pseudomonas acidovorans* in low-cost substrates, which were different. *Pseudomonas cepacia* showed the best result (Lee et al. 2004). The results show that low-cost agricultural waste can be used as a renewable source for producing biosurfactants. Peanut oil cake is a byproduct of oil industries and it is a suitable substrate for lipopeptide production by *Bacillus cereus SNAU01* (Nalini et al. 2016). Fish waste has also been shown to produce lipopeptide using *Bacillus subtilis N3-1P* (Zhu et al. 2020).

1.6.4 Production of Cosmetics

Bacterial fermentation, especially probiotic fermentation, is one of the emerging fields in the cosmetic industry. Fermented probiotic products reduce the cosmetic resources' toxicities and improve absorption into the skin by altering the molecular structures and improving certain pharmacological activities. The fermented products used in beauty products are rich in antioxidants, nutrients, omega-3 fatty acids, and enzymes. Conventional skincare products incorporate probiotics in them. The tropical probiotic products are now rising to trend wellness in the beauty industry (Tkachenko et al. 2017).

S. thermophilus is a probiotic bacteria that has many benefits to the skin. *S. thermophilus YIT 2001* and *S. thermophilus YIT 2084* has a skin hydration effect and seems to show antioxidative effects (Yamada 1982). The latter one also can produce hyaluronic acid, which is a conventional cosmetic ingredient (Izawa and Sone 2014).

1.6.5 Production of Biofuels

Biofuels is a renewable source produced by the transesterification and fermentation of vegetable oils or animal fat with alcohol (methanol or ethanol) which has recently sustained interest due to its contribution to petroleum-based diesel global dependence production. Three probiotic strains of Lactobacillus, such as *Lactobacillus acidophilus*, *Lactobacillus delbrueckii*, and *Lactobacillus plantarum* was tested to check their capability to acquire and metabolize glycerol. Biodiesel-derived glycerol is used as a major carbon and energy source in microaerobic growth. These strains were able to acquire glycerol, consuming between 38% and 48% in approximately 24 h. *L. acidophilus* and *L. delbrueckii* showed similar growth, higher than

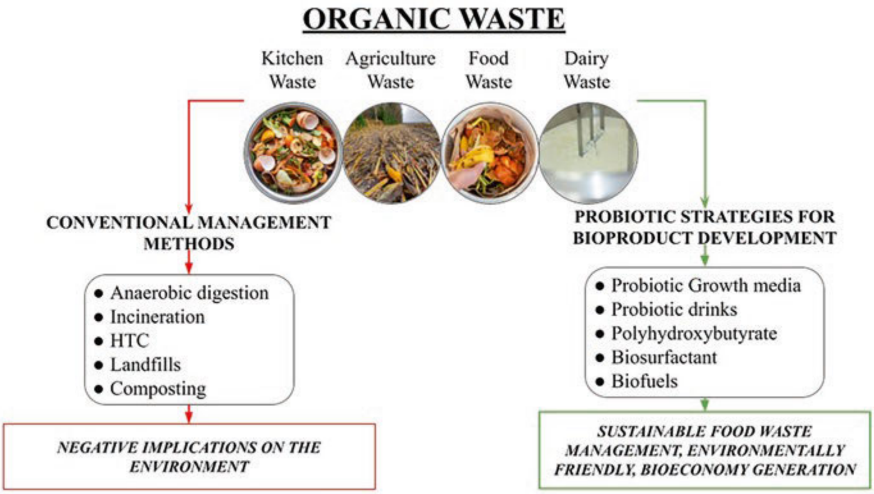

Fig. 1.3 Probiotic strategies as an alternative and its positive implications over conventional organic waste management

Table 1.2 Production of various value-added products from organic waste using probiotics

Nature of waste	Probiotic organism	Final product	Reference
Cheese Whey	*Lactobacillus acidophilus La-5* and *Bifidobacterium animalis Bb-12*	Probiotic whey drink	Skryplonek and Jasińska (2015)
Pomegranate Peel	*Lactobacillus plantarum*	Probiotic antioxidant milk beverage	Al-Hindi and Abd El Ghani (2020)
Beetroot	*Lactobacillus plantarum*, *Lactobacillus rhamnnosus* and *Lactobacillus delbrueckii* sp.	Non-dairy probiotic drinks	Panghal et al. (2017)
Mango peel	Lactic acid bacteria	Probiotic milk	Vicenssuto and de Castro (2020)
Dairy waste	*Bacillus megaterium SRKP-3*	Polyhydroxybutyrate (PHB)	RamKumar Pandian et al. 2010
Cane molasses	*Bacillus megaterium* and *Pseudomonas aeruginosa*	Polyhydroxybutyrate (PHB)	Tripathi et al. (2019)
Soy molasses	*Pseudomonas aeruginosa ATCC 10145*	Glycolipid biosurfactant	Rodrigues et al. (2017)
Kitchen waste oil	*Pseudomonas aeruginosa*	Biosurfactant	Chen et al. (2018)

(continued)

Table 1.2 (continued)

Nature of waste	Probiotic organism	Final product	Reference
Mill waste (olive oil)	*Pseudomonas aeruginosa* and *Bacillus subtilis*	Biosurfactant	Moya Ramírez et al. (2015)
Dairy effluent (cheese whey)	*Pseudomonas aeruginosa SR17*	Biosurfactant	Patowary et al. (2016)
Noodle waste	*Saccharomyces cerevisiae K35*	Biodiesel and bioethanol	Yang et al. (2014)
Olive oil (cooked)	*Penicillium expansum*	Biodiesel	Papanikolaou et al. (2011)
Banana Peel, potato peel, household waste	Mix of α and β amylase, and glucoamylase; *Saccharomyces cerevisiae H058*	Bioethanol	Karmee (2016)
Cane molasses and starch-rich food waste	*Clostridium acetobutylicum*, *Clostridium beijerinckii P260*	Biobutanol	Ujor et al. (2014)
Mixed food waste	*Exoelectrogenic bacteria*	Methane	Park et al. (2018)

L. plantarum. All strains catabolize glycerol mainly through glycerol kinase (EC 2.7.1.30) (Rivaldi et al. 2013) (Fig. 1.3 and Table 1.2).

1.7 Conclusion

Large amounts of organic waste are generated annually as a result of the growing population globally. As the waste generated is high, it has prompted sustainable approaches for managing and reusing food wastes. Currently, the methods used for food waste management have environmental impacts and uses much energy. Microbial strategies for food waste management, particularly utilizing probiotics, can reduce the impact on the environment. This chapter concludes that bioconversion and utilization of organic waste by probiotics for its growth and substrate for producing various value-added bioproducts is a promising and feasible method for cost-effective and environmentally friendly food waste management.

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Chapter 2
Bioremediation as an Alternative and Sustainable Strategy Against Environmental Pollutants

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Abstract Pollution has been the most discussed subject in the past decade due to uninterrupted intervention of humankind in the nature's environmental balance in the form of release of toxic substances into air, soil, and water. Overexploitation of the natural resources in the postindustrial era has led to complete destruction of the environment. Researchers and government agencies have employed various strategies to mitigate the impact of environmental damage but to a larger extent all the efforts employing physical and chemical treatment methods have been unsuccessful due to the unsustainability during treatments. The use of bioremediation for treating pollution at various levels has been contemplated in the past as well but a concerted effort from the various fields is required for the method to gain practical acceptance. Bioremediation involves the use of living organisms in neutralizing the harmful effects of pollutants. There has been exploration on the materials that aid in the degradation either by augmenting the speed of degradation or by providing a suitable environment. Hence, a concerted effort has been employed in the development of advanced techniques that have been used in the past decade for the degradation of pollutants like hydrocarbons, fertilizers, and pesticides which exist as residues in all biotic environments. Since bioremediation is one of the cheapest alternatives that is available for treating the pollutants in the environment, a review of the past studies and assessment of the possible future methods is very much important for protecting the environment and as well as repairing the already damaged environment.

Keywords Bioremediation · Degradation · Phytoremediation · Electricigens · Sustainability

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2.1 Introduction

Modern agricultural practices postindustrialization involving extensive use of toxic chemicals for better yield have impacted the environment at large. This phenomenon has not slowed down in the past few decades even after the impact of pollutants being felt in all spheres of life. With the health of humans at stake due to various pollutants that are carcinogenic, mutagenic, and having ability to cause chronic effects, it becomes pertinent to remove the contaminants from the natural environment. Treatment strategies are devised based on the type of pollutants and it is broadly classified into physical, chemical, and biological pollutants. In spite of these strategies that have been followed previously in mitigating the effects of the pollutants from soil, water, and air, the results have not been satisfactory. This situation necessitated the need for novel methods in treating environmental pollutants through bioremediation strategies (Azubuike et al. 2016).

Bioremediation is a process of rehabilitating the natural resources to its native state by performing bioprocesses involving microorganisms and plants. Since the application and handling of microorganism is less cumbersome and cost-effective, the use of microorganisms has gained significance over other methods. The bioremediation process is classified into two categories based on the site of application of the microorganisms, namely ex situ and in situ methods (Kumar et al. 2018). The choice of application of these methods will be dependent on the type of pollutant, soil composition, and cost incurred on removal of the pollutant from the site. These parameters enabled the researchers in identifying new methods and also in modifying the existing strategies based on the requirement.

With bioremediation process by microorganism gaining prominence over other remediation methods, there has been an attempt to employ bacteria, fungi, algae, and yeast for the removal of contaminants. These organisms in combination with other biotic and abiotic factors augment the process of remediation in the environment. The microorganisms have been broadly classified into aerobic and anaerobic microbes. The efficacy of the remediation process is also dependent on the concentration of the pollutants, accessibility, and other environmental factors including pH, temperature, and nutrients. Anaerobic microorganisms process toxic hydrocarbons into less toxic forms. The utilization of anaerobic forms of microorganism for degradation of the toxic aromatics and chemicals has gained prominence in the recent past (Rabus et al. 2016).

In this scenario, identification and extensive study of the various other modifications of the bioremediation processes gain importance to keep abreast with the emerging pollutants of the world's industrial atmosphere. Thus, in this chapter, we have tried to enlist and describe the important research activities in the past decade pertaining to the advancements in the bioremediation processes and their efficiency in dealing with the pollutants. This chapter will delve into the various developments in the "omics" of the microorganisms and thereby providing information on the molecular and genetic level of organisms and their effect on pollutants, plant-based strategies involved in degradation, use of the microbial electrochemical systems

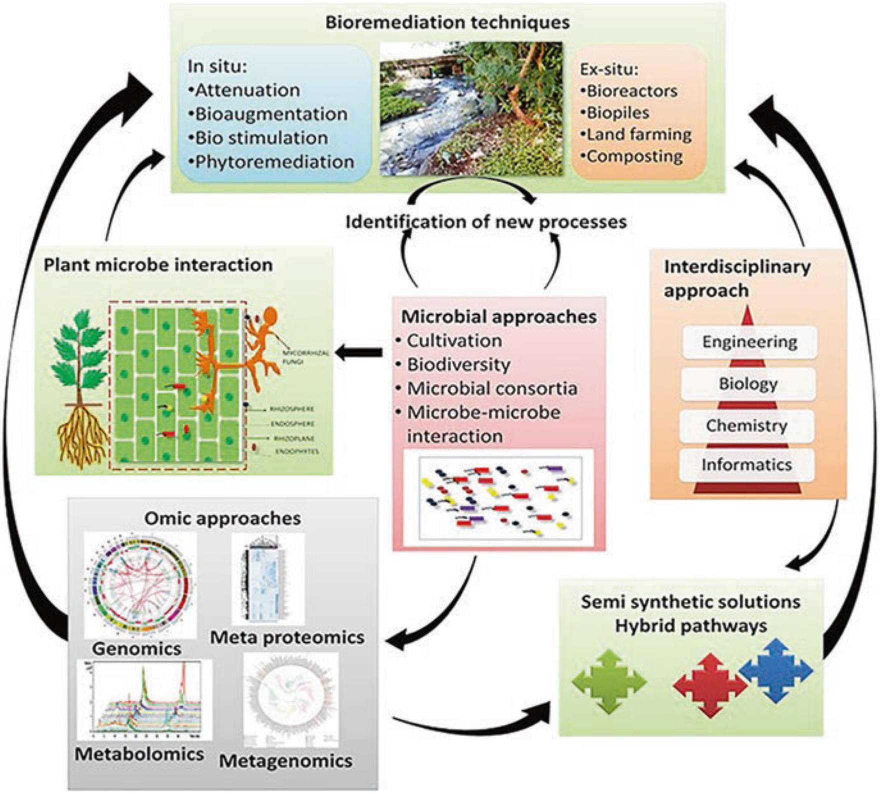

Fig. 2.1 Representation of various bioremediative procedures employed in removal of environmental contaminants. (Source: Adopted from Malla et al. 2018)

during the ex situ degradation process and finally on the use of nanoparticles and their advantages in augmenting the speed and efficiency of the remediation process (Fig. 2.1). Thus, the primary objective of this chapter has been to consolidate the various advancement in the past decade in the field of bioremediation.

2.2 Microbial Bioremediation

Microorganisms are present all over the environment, given their metabolic potential is noteworthy, and they could undoubtedly fill in a broad scope of natural conditions. It has been noticed that the presence of contaminants for a more extended period creates havoc on the environment, resulting in various side effects to the ecology. So, it is mandatory to treat harmful and toxic contaminants with nontoxic eco-friendly products. Different physical and chemical techniques are involved in the bioremediation process, even though it has more limitations and create secondary environmental contamination (Yoshikawa et al. 2017). So, there is a need to manage the levels of hazards by improving the techniques and processes involved in bioremediation. Researchers are very keen to discover advanced technologies to overcome the limitations and get the best result in the bioremediation process.

Biology has immense potential as a tool for developing microbial and plant-based solutions for environmental remediation and restoration. Among various biological treatments, microbe-based treatments are versatile because of the simple, chief, and eco-friendly clean-up method. The effectiveness of microorganisms in remediation is based on their incredible adaption and metabolic diversity. Initially, the biodegradation technique focused on the isolation and identification of cultivable microorganisms for use in bioremediation processes. Due to less explored cultivable and uncultivable organisms and lack of knowledge on factor influence on the growth and metabolism of microbial population limits the treatment process. This limitation extended time-consuming, less removal of pollutants (Bharagava et al. 2019) and environmental disturbances like foul smell formation, etc. Hence, advanced techniques need to overcome the above limitations and achieve more efficient remediation strategies.

The recent advancement in molecular techniques helps to evaluate the uncultivable microorganisms in the natural environment. Primarily the microorganisms use metabolic pathways and catalysts for enzyme production that take part in biodegradation. Moreover, understanding metabolic pathways is necessary to study microbial remediation (Plewniak et al. 2018). Notably, *Archaeoglobus fulgidus* and *Syntrophoarchaeum butanivorans* degrade organic pollutants by releasing novel alkyl succinate synthase and alkyl-coenzyme M, respectively (Park and Park 2018). Moreover, the enzyme alkali-stable carbonic anhydrases (CAs) used for biomineralization of CO_2 is anticipated as an inexpensive and best method for mitigating global warming (Bose and Satyanarayana 2017).

The use of single microbes in the bioremediation process can metabolize only a limited range of environmental pollutants where mixed populations have a high capacity to survive stress and release of different metabolic capabilities. The mixed microbial culture plays a significant role in improving its removal abilities in bioremediation process. The mixed culture of *Leptospirillum ferriphilum* CS13, *Acidithiobacillus caldus* S2, and *Sulfobacillus acidophilus* S5 strains shows a good removal rate (~99%) of heavy metals by the bioleaching process (Hu et al. 2020). Moreover, studies have shown that sulfur reducing bacteria utilizes the minerals cadmium and zinc for their metabolic processes and degrades into nontoxic metabolites (Nordstrom et al. 2015). Single cultures were used for specific contaminant removal, but their efficacy is low compared to mixed cultures. Furthermore, endophytic bacteria are proposed as an alternative to mitigate heavy metal pollution. The endophytic bacteria associate with plants and remove lead and zinc by bioaccumulation and phytoextraction (Fan et al. 2018). The plant *Robinia pseudoacacia*, in association with root nodule bacteria *Mesorhizobium loti* HZ76 and *Agrobacterium radiobacter* HZ6, effectively degrade Pb/Zn in a very short time (Fan et al. 2018). In addition, more potential microbes reported for remediation of heavy metals are tabulated in Table 2.1.

Table 2.1 List of selected potential microorganisms used in remediation of heavy metals

Organisms	Genus/species	Pollutants	Process	Source
Bacteria	*Sporosarcina saromensis* M52	Hexavalent chromium	Bioreduction	Zhao et al. (2016)
	Bacillus sp. and *Aneurinibacillus aneurinilyticus*	Arsenic	–	Dey et al. (2016)
	Rhodobacter sphaeroides	Lead	Accumulation	Li et al. (2016)
	Bacillus cereeus	Cr		Nayak et al. (2018)
	Cellulosimicrobium sp. (KX710177)	Pb	Bioabsorption	Bharagava and Mishra (2018)
Archaebacteria	*Filo crenarchaeota*	Cd, Cu, Ni, and Zn	–	Sandaa et al. (1999)
Fungi	*Sporosarcina saromensis* M52	Hexavalent chromium	Bioreduction	Zhao et al. (2016)
	Aspergillus tamarii	Chromium complex	In batch and continuous bioreactors	Ghosh et al. (2017)
	Saccharomyces cerevisiae	Cr	–	Benazir et al. (2010)
	Candidapara psilosis	Hg	–	Muneer et al. (2013)
	Penicillium, *Aspergillus*, and *Rhizopus*	Mercury, arsenic, lead	Accumulation	Dixit et al. (2015)
	Geotrichum sp.	Hexavalent chromium	Bioleaching system	Qu et al. (2018)
Algae	*Anabaena inaequalis*	Cr	–	Kannan et al. (2012)
	Nostoc sp.	Hg, Pb, Cd	Adsorption	Girish and Mohammad (2013)
	Spirulina spp.	Pb and Cd	–	Chen and Pan (2005)

The recent studies on microbial glycoconjugates could be a notable advancement in the bioremediation process. The glycoconjugates make a link between microbes and organic pollutants and uptake desired contaminants from the external source (Bhatt et al. 2021). The rhamnolipid is one of the extensively used glycoconjugate in the bioremediation process, which is involved in the degradation of chlorinated phenols, oil pollution contaminants β-Cypermethrin, and many other organic pollutants. It has been observed that glycoconjugates like surfactin and sophorolipid are also commercially used for the enhanced bioremediation process. Moreover, these glycoconjugates also play a significant role in biofilm formation that accelerates the biofilm-based degradation of the organic pollutants. In addition, there are many other cost-effective advanced techniques for bioremediation such as microfiltration, electrodialysis, precipitation, or flocculation used to remove dissolved solids and heavy metals very efficiently. These techniques can work even at high temperatures to remove the contaminants effectively using cellular biomass.

The focus has been on microbial-based remediation among the various other bioremediation techniques because of their very effective degradation ability. These microbes degrade various types of pollutants by just taking up, transporting, engulfing, and then detoxifying them by using their metabolic processes. Many bacterial species synthesize many metabolites with an excellent binding capacity to attract contaminants but their degradation potential is found too insufficient. The microbes contain many potential genes and proteins for the degradation process, specific for a specific pollutant. Still, they may slow in the bioremediation process (Malik et al. 2021). Therefore, it is the need for time to fasten up the bioremediation process to save time and brings new technologies to overcome the side effects of the process.

The studies of microbial genes and their mechanisms are becoming a boon to the degradation processes in upcoming years (Malik et al. 2021). Ayangbenro and Babalola (2020) reported that the genome of *Bacillus cereus* NWUAB01 contains putative genes for transport-specific effective metal ion degradation. The recombinant photosynthetic bacterium *Rhodopseudomonas palustris* would take up harmful mercury ions and metallothionein from the wastewater contamination. This engineered strain displayed multiple times enhanced the degradation process compared to wild strain (Deng and Jia 2011). Likewise, the recombinant *Escherichia coli* engineered by the addition of metallothionein (MT) degrading gene and two more genes Nix A from *Helicobacter pylori* and Nis A from *Staphylococcus aureus*. These genes are specific for nickel-metal binding and bioaccumulation of nickel ions from waste pollutants (Deng et al. 2013). Also, the genetically engineered strains of *Rhodococcus* sp. IN306 and *Mycolicobacterium frederiksbergense* IN53 with unique genes that encodes specific enzymes for removal of hydrocarbons in waste petroleum contaminants (Steliga et al. 2020). Similarly, the strain *Comamonas* sp. CD-2 and *Pseudomonas* sp. CB-3 is engineered in such a way that the genes for degrading polychlorinated biphenyls (PCBs) have increased their expression multiple times than the original strain (Xing et al. 2020). In addition, the genetically engineered *Bacillus cereus* and *Pseudomonas putida* strain have enhanced their efficacy by adding genes that help them degrade more complex hazardous compounds simultaneously (Filonov et al. 2020).

Synthetic biology (Synbio) plays a crucial role in environmental remediation and restoration. The Synbio approach could grasp an organism's metabolic and catabolic complexity to evaluate the potentiality of the microbial community synthetically. The mining genes from the database confer the basic information to develop a synthetic microbial model for bioremediation (Fajardo et al. 2019). Since applying synthetic biology in remediation would advance the bioremediation process. In recent times, bioinformatics tools are used to explore at genetic level which could be used to identify potential microbes for bioremediation. This computer-assisted technology is very helpful to find the solutions to some limitations in the bioremediation processes in a very short period. The UM-BBD (University of Minnesota Biocatalysis/Biodegradation database) is the most important and frequently used database which is freely available and consists of all information related to degradative microbes. Apart from this, BioCyc, MetaCyc, Bionemo, etc., databases are also used in this field to get information about potential microbes.

In addition to the contribution of bacteria towards the bioremediation process, algae-mediated bioremediation of contaminants in the form of heavy metals, hydrocarbons from oil spills, and organic sewage from urban establishments has also been undertaken in recent years. It has been observed in a study that as early as 2000 itself, microalgae *Tetradesmus obliquus* has showed extensive nitrogen and phosphorus removal abilities from the urban wastewater (Martinez et al. 2000). Hydrocarbons like phenol have been treated with algal species such as Chlorella spp. including *T. obliquus* and *Limnospira maxima* (Scragg 2006). Similar studies on the removal of heavy metals using microalgae have also been conducted in the early part of this century (Sreekumar et al. 2020). There have been efforts in the removal of pesticides from the agricultural runoffs which contain a significant amount of nitrogen, phosphorus, and potassium. These efforts have started showing results in the use of microalgae as a substitute for bacteria and also works complementing the bacteria during degradation.

2.3 Phytoremediation

Phytoremediation is a plant-based technology that uses genetically engineered or naturally occurring plants to treat the contaminated site. It is identified that various types of vegetation, such as aquatic plants, trees, and grass have also been used to decontaminate soil, surface water systems, and as well as the groundwater systems. The ability of the plants to behave as hyper accumulators augurs well in accumulating the contaminants and subsequently breaking down the pollutants (Sridhar et al. 2020). Besides, phytoremediation has varied methods of dealing with contaminants namely, Phytoextraction, Phytodegradation, Rhizofiltration, Phytostabilization, Rhizodegradation, Phytovolatilization, and hydraulic control. Though enough studies have been performed in the phytoremediation process, there has been a recent development in the form of modifications in the techniques, type of pollutants, and the identification of new plants species for countering the ever-emerging pollutant species.

Phytoextraction is a process of absorption and translocation of the contaminants in the soil by the plants which can be accumulated as biomass and thus can be harvested. Some plants have the potential to accumulate heavy metals like Ag, Se, Hg, Cd, Cr, Co, and Pb which are essential for their growth (Lu et al. 2015). The selection of plant species for successful phytoextraction is dependent on the factors such as high biomass accumulation, extended root system, fast-growing, high metal tolerance in plant tissues, adaptability to contaminated sites, high translocation factor, and easy agricultural management (Kuppens et al. 2015). Hyperaccumulators like *Thlaspi caerulescens* and *Alyssum bertolonii* have been identified for their ability to accumulate high concentrations of metal. For improving the uptake of metal ion, recent studies have used synthetic or natural chelating agents along with improved agronomic practices like crop management, and genetic engineering to increase the capability of the plant species in the accumulation of metal ions (Sheoran et al. 2016). In addition to the phytoextraction method, further improvements have been introduced in the phytoremediation concept, by the use of plant–microbe relationship. The accumulation of *Pb* in the roots can be done by *Acacia mangium* with the addition of organic fertilizer (Meeinkuirt et al. 2012).

The relationship of plants with the microbes present in the rhizosphere enhances the degradation process. The plant root exudates help in enriching the degrading microbes generally referred to as rhizosphere effect. This has been mainly utilized in the degradation of PAH. Generally, root exudates improve the plant defense mechanism and thus increase the number of microbes in the rhizosphere (Sivaram et al. 2020). The petroleum oily sludge (POS) is a toxic and mutagenic material that has a serious implication on the environment. The degradation of the material is greatly reduced by the toxicity of the compounds towards the microbes during bioremediation. In this scenario, plants along with microbes have shown potential to overcome the toxicity of POS. Recently, a leguminous plant *Cajanus cajan* was found to be efficiently involved in phyto-remediating of petroleum oily sludge-spiked soil (Allamin et al. 2020).

Rhizofiltration is the eco-friendly, cost-effective method to adsorb, concentrate, and precipitate the contaminants onto plant roots (Haldar and Ghosh 2020). The sources of pollutants in the groundwater are Pesticides, fertilizers, improper sewage management, Industrial effluent, Landfill leachate leakage, Mining, petroleum by-products, and heavy metals. The secondary metabolites which are released by the plants from the roots may adsorb within the root and translocate to the phyllosphere (Sharma and Juwarkar 2015). In general, plants adopt the following methods to tolerate the phytotoxicity namely symbiotic association with mycorrhiza for heavy metal uptake, utilization of plasma membrane to reduce influx, storage of heavy metals in epidermal tissues and vacuoles, utilization of antioxidants and enzymes to neutralize ROS, and detoxification of heavy metals by producing metallothioneins and phytochelatins (Kristanti et al. 2021).

Plants that can interact with many organic compounds and inorganic compounds can be used for volatilization of these compounds. Previous studies on various other contaminants of groundwater contaminants like trichloroethylene (TCE), tetrachloroethylene (PCE), methyl tert-butyl ether (MTBE), and 1,1,1-trichloroethane (TCA). The organic compounds which are volatilized from the leaves or stem are called direct Phytovolatilization and the compounds from the soil due to root activities are called indirect Phytovolatilization. Similarly, few studies have shown that heavy metals such as mercury, selenium, and arsenic compounds can also be volatilized from the soil (Limmer and Burken 2016).

Studies on degradation of Trinitrotoluene (TNT) and Nitroglycerine (GTN) using plants like *Avena sativa* and *Beta vulgaris* has paved way for similar approaches by various other researchers. The concept of phytodegradation of the chemical compounds has helped in identifying plants that have immense potential in performing the degradation in soil and aquatic environment. In the past decade with the utilization of dyes in industries becoming more rampant, a plant belonging to the clade tracheophytes *Salvinia molesta* was identified to have the ability to degrade azo dye (Chandanshive et al. 2016).

A novel method such as hydraulic control has also been identified for controlling the spread of contaminants from the site of contamination to farther sites. This technique is performed by the presence of phreatophytic trees and plants to transpire water in large volumes and in turn reduce the migration of contaminants. The plant species like Eucalyptus, Birch, Willow, and poplar can be used for this approach (Fortin et al. 2021). Transgenic plants have also played a major role in the detoxification of heavy metal contaminants present in the soil and water sources. Oilseeds such as transgenic canola consisting of a rice transcription factor *OsMyb*4 have also been used for bioremediation of copper and zinc salts (Raldugina et al. 2018). The plants like transgenic *A. thaliana* have been used for enhanced lignin biosynthesis and accumulation of Cd (Xia et al. 2018). Similarly, Engineered *P. trichocarpa* and *A. thaliana* (L.) have been found to accumulate Hg (Sun et al. 2018) and nettle plant *Urtica dioica* has attained the ability to accumulate Polychlorinated biphenyls after modifications using CaMV 35s promoter (Viktorova et al. 2017).

These various methods that have been developed on the basis of using plants in the remediation process have yielded good results till now (Table 2.2). Also, phytoremediation is an eco-friendly and cost-effective technique to degrade harmful pollutants without leaving any toxic by-products. Besides, the knowledge on the utilization of plants along with the microbes has shown immense potential in decontaminating the pollutants in a site-specific manner. Further, the introduction of transgenic plants in the removal of contamination from natural sources has only

Table 2.2 Selected plants used for phytoremediation of metal contaminants

Name of the contaminants	Method of remediation	Plant type used	Reference
Cd, Zn, and Pb	Phytoaccumulation	*Scots pine (Pinus sylvestris L.)* and *Norway spruce (Piceaabies L.)*	Placek et al. (2016)
Al and Fe	Phytoaccumulation	*Ipomoea aquatica* and *Centella asiatica*	Hanafiah et al. (2020)
Pb, Cd, Cu, Zn, and Ni	Phytoextraction	*Amaranthus* spp.	Ziarati and Alaedini (2014)
Cd, Cr, Pb, Zn, Ni, and Cu	Phytoaccumulation	*Jatrophacurcas*	Chang et al. (2014)
Mercury (Hg)	Phytoaccumulation	*J. curcas*, *P. marginathum*, *C. annuum*, and *S. bifidus*	Marrugo-Negrete et al. (2015, 2016)
Chromium	Phytostabilization	Weed plant (*Ipomea carnea* and *Jatrophagos sypiifolia*)	Nirmalkumar and Kavitha (2020)
	Phytoaccumulation	*Suaedavera*, *Vetiveria zizanioides*, *Tageteserecta*	Nayak et al. (2018), Din et al. (2020)
Arsenic (Ar)	Phytoaccumulation	*P. vitatta*, *P. cretica*	Anning and Akoto (2018)
Cadmium (Cd)	Phytoaccumulation	*Paspalum conjugatum*, *Pseudotsuga menziesii*	Zhang et al. (2020), Astier et al. (2014)

increased the confidence on phytoremediation in dealing with novel and emerging contaminants.

2.4 Biomolecular Engineering of Microbes

The OMICs advancements, specifically metagenomics, meta-transcriptomics, meta-proteomics, and metabolomics are being utilized as a bioremediation strategy in recent times. This approach promotes exploring a biomolecule such as RNA, DNA, proteins, and metabolites from individuals and the entire community simultaneously (Gutierrez et al. 2018). The OMICs approaches from genomics to metabolomics, provide complete knowledge of microbial action and better understanding in field applications of bioremediation. There is no doubt that the OMICs approaches provide a better knowledge on molecular mechanism involved in microbial remediation and thereby booming the efficiency of microbial degradation and assisting in designing more robust approach to restore contaminated sites (Ma and Zhai 2012). The newly seeded OMIC approaches such as metagenomics, meta-transcriptomics, meta-proteomics, metabolomics, fluxomics, and interactomics provide more insight into the microbial communities inhabiting specific ecological niches and their dynamic alterations within a time span. Moreover, postgenomic technology offers a better understanding and monitoring of the bioremediation process.

Microbial remediation strategies pose effective restoration of contaminated environments without/minimum damage to the environment. A large portion of the microorganisms in the environment are uncultivable in laboratory conditions. This "OMICs" technique could help to screen such uncultivable microorganisms growing in a diverse environment. The uncultivable microorganisms might possess novel metabolic pathways that could be more effective removal of contaminants. The search of new potential genes in both cultivable and noncultivable microorganisms is being achieved by modern genomic and metagenomic sequencing technology (Bharagava et al. 2019). This metagenomic approach provides to identify several functional genes in various microorganisms that code enzymes for bioremediation (Rodriguez et al. 2020).

The metagenomic screening of contaminated environments can accurately detect microbial community interactions. Most recent studies proposed that metagenomic applications are widely accepted and used to screening and remove environmental contaminants. Moreover, advanced bioinformatic tools have been integrated with metagenomics to investigate phylogenetic and functional aspects of microbial communities for metagenomic bioremediation. It is believed that the advancement in metagenomics provides the best degradation rate in microbial remediation. The genomic study of oil-degrading bacteria *Franconibacter pulveris* possesses genes that also degrade diverse petroleum hydrocarbons, chemotaxis, metal resistance and transport, biosurfactant synthesis (Pal et al. 2017).

Transcriptomics is one of the extensively studied OMICs methods which deals with the study of a whole set of RNA from protein-coding to noncoding in an organism. Metagenomic analysis has certain limitations, usually combined with transcriptomics technique to recognize gene expression. The gene expression of microbial remediation in the contaminated site is an important one to achieve effective remediation. Understanding gene expression like different genes has expressed differentially under different conditions could be facilitated by transcriptome analysis. It imparts knowledge on the up- and down-regulation of the gene in microorganisms under varying environmental conditions (Chandran et al. 2020). The techniques like microarray and sequencing, etc., are generally used for transcriptome analysis while DNA microarray assists in evaluating and examining mRNA expression in gene organisms. Transcriptome analysis of *P. aeruginosa* reveals the importance of distinctively expressed genes related to oil degradation (Das et al. 2020). The transcriptomic study of the pesticide reduction would permit the physiological optimization of the strains that are used in bioremediation and construct novel pathways to remove contaminants (Rodriguez et al. 2020).

The proteomics approach provides insight into the abundance of protein and type of critical protein involved in the response by microorganisms. Since the bioremediation process is a metabolic activity conducted by the intrinsic mechanisms of the microorganisms over the pollutants, the environmental factors playing the role of the external stimulus also determines the type of proteins and its levels expressed during the remediation process (Mattarozzi et al. 2017). The study on the protein profile of the microorganism provides information on the type of pollutant that the organism can metabolize in the form of pollutants. It also provides insight into the type of ecosystem and microbial community structure. Besides the analysis of the proteomics of the microorganism, the data helps in analyzing the post translations modifications of the proteins and its mechanism of action during degradation. In addition to the proteomics approach, recent studies have revealed that metabolomics also play a major role in the degradation process of pollutants, as the downstream processing of the pollutants into nontoxic metabolites is significant in the bioremediation process. The metabolomics approach deals with screening and quantification of the metabolites released by organisms at a specific point in time. The information on metabolomics facilitates in establishing the sequence of microbial activities, allows us to determine the potency of microorganism in bioremediation, and also provides data on the ecosystem of the remediation site.

2.5 Nonliving Biomass

The microbial organisms can be used for the treatment of organic and inorganic contaminants; the usage of nonliving biomass is becoming more attractive than living organisms because of its benefits. The usage of living microorganisms can also be used for the treatment of the contaminants but due to some drawbacks like after reaching the threshold level the microbes may die due to toxicity and also there is a possibility of the amplification of toxicity in the food chain. Bacteria, fungi, and algae biomass can be used for accumulating heavy metals through biosorption which includes precipitation, ion exchange, Complexation, physical adsorption, and transport across the cell membrane (Javanbakht et al. 2014). The cell wall and microbial extracellular substances play an important role in the biosorption process. Microbes such as *Bacillus* sp., *Aspergillus niger*, and *Penicillium* sp. were used for the biosorption of heavy metals like the Pb, Cr, and Cd (Abioye et al. 2018). Similarly, *Desulfovibrio desulfuricans* has been shown useful for the removal of Cr, Cu, and Ni (Tarekegn et al. 2020). Studies have shown that the biomass of *Scenedesmus* spp., *Chlorella* spp., and *Tetraselmis* spp. can be utilized for the uptake of Cd (Sbihi et al. 2012). Also, the dried biomass of *Spirulina* sp. can be used for the removal of heavy metals like Pb and Zn (Cheng et al. 2017).

2.6 Microbial Electrochemical System

Bioremediation is a technique that employs a sustainable approach in removal of contaminants from a natural source without harmful secondary contaminants, and thus restoring the site of pollution to its native state. Since bioremediation is a process involving cumbersome activity in treating the polluted environment, it involves large areas of land, time, and utilization of other resources for treatment. Also, it has been observed that in situ remediation procedures have limitations in the aspects of lesser oxygen availability and lack of indigenous microbial species in the site of treatment (Wang et al. 2015). These limitations during the remediation procedures have led to researchers searching for other more effective or modified forms of remediation techniques based on Bioelectrochemical system (BES).

The alternative to this is a more compact, speedy, and chemical retrieval process, which uses an external power to oxidize metabolic wastes and further reduce the oxidized contaminants (Wang and Ren 2013). A microbial electrochemical system (MES) has been one of the ever-evolving technologies with its utilization still in the nascent stages. MES primarily produces chemical energy from bioorganic substances to generate electrical energy. This system is an integration of the microbes, electrochemistry, and the environment, as the fundamental objective of this latest technology has been to bioremediate the polluted biotic components and, in the process, generate electricity through Extracellular/external electron transfer (EET) (Kumar et al. 2017).

In BES, the interaction of live microorganisms with the electrodes thus produce electricity and remove the pollutants from the natural sources. Natural sources such as soil and water can be treated, with slight modification in these technologies itself like microbial desalination cells (MDC), photomicrobial fuel cells (PhotoMFC), microbial electrosynthesis (MES). The anodes in the system are involved in oxidation reaction whereas the cathode does the reduction of the molecules. Since the microorganisms are involved in these redox reactions, these electrodes are referred to as bioanodes and biocathodes (Rabaey and Rozendal 2010). These redox reactions along with the electrical energy result in the production of chemicals such as H_2 and methane on the cathode and thereby referred to as MEC (Microbial electrolysis cell) (Kadier et al. 2015).

The fundamental mechanism of BEC is based on the interaction between the microbial cell and the electrodes, which can be classified into capacitive interactions and faraday type interactions. In capacitive interactions, the interaction between the microbial cell and the electrode leads to displacement of the water and ions between the bilayer and the electrodes, whereas in faraday-like interactions biofilms and cells that form it are charged/uncharged through redox reactions. The current generated during the oxidation of substrates can either be used as electricity through the MFCs or produce value-added chemicals (Fornero et al. 2010).

The basic functionary in a BES is the microorganisms and their respective abilities in transferring the charges to the electrodes while degrading the pollutants in a closed environment. The ability of degrading the pollutants and thereby transferring the charges to the electrodes varies among the microbial species involved in the process. These microorganisms that have the ability to transfer the generated electrons to the anodes of the MFCs are referred to as electricigens (Cao et al. 2019). This transfer of electrons happens through two methods: direct transfer and indirect transfer.

During the direct transfer of electrons, there is a physical contact established between the microorganism and the outer membrane of the cell through the formation of the biofilms by the electricigens or by involving the conducting apparatus such as cilia and flagella. Unlike during direct transfer, indirect transfer of electrons occurs with the aid of a soluble mediator, which can be a transporter protein present in the outer membrane of the electricigens (He et al. 2017). Also, it has been identified that there are limitations in these methods with regards to the access of active sites on electron transport proteins in case of direct transfer and the potential difference between the mediators and the redox proteins in case of indirect transfer (Evelyn et al. 2014).

In the past decade, many self-mediators like pyocyanin (Dantas et al. 2013) and most recently another molecule of importance phenazine (Peng et al. 2018) were identified, which has enhanced the ability to transfer the electrons from the electricigens to the bioanodes. The utilization of self-mediators for electron transfer will avert the necessity of the external mediators in the MECs thus making the system more efficient and environment-friendly.

The efficiency also depends on whether a pure culture, enriched culture, or a mixed culture is utilized in the BES (Saratale et al. 2017). Of these many pure culture microorganisms or electricigens have been identified predominantly belonging to Proteobacteria and Firmicutes. Studies by Abrevaya et al. (2011) have revealed that two species of halophiles *Haloferax volcanii* and *Natrialba magadii* had an ability to produce electricity when supplemented with exogenous mediators. Likewise, studies on acidophiles as early as 2014 by Mark et al. (2014) showed the ability of these microorganisms to withstand a very high load of metal pollutants. Similarly, studies are underway to find potential microorganisms under the categories of alkaliphiles, psychrophiles, and various other extremophiles that could provide enhancement in the MEC technologies.

2.7 Current Trends in Bioremediation

In recent times, the use of various bioremediation strategies for the detoxification or removal of toxic emerging contaminants has been on the rise. The use of enzymes for the bioremediation process seems to have gained attention recently owing to its efficiency and biocompatibility. The process of identifying and isolating novel enzymes with specific properties from microbes and using them in contaminated sites would offer numerous benefits. However, the activity and mechanism of action of these enzymes have to be explored so that they can be efficiently used for the bioremediation process (Mousavi et al. 2021). Moreover, Dutta et al. (2021) have suggested the use of synthetic biology and machine learning process for engineering enzymes that could be used in the bioremediation process. The enzymatic properties and bioremediation pathways could be redesigned or optimized using data-assisted synthetic biology approaches thereby enhancing the process of enzyme-mediated bioremediation (Dutta et al. 2021).

Emerging trends in bioremediation involve the use of in silico techniques for the mitigation of environmental pollutants. These include the prediction of pathways involved in biodegradation using QSAR (Quantitative Structure-Activity Relationship) and QSBR (Quantitative structure-biodegradation relationship) model system (Singh et al. 2021). Similarly, glycobiotechnology is another area that has been explored upon in bioremediation strategies. Glycoconjugates which consist of glycoproteins and glycolipids produced by microbes play an important role in biofilm formation. These biofilms create an interface and facilitate the microbial interaction with contaminants thereby accelerating the degradation process (Bhatt et al. 2021). Leong and Chang (2020) have emphasized on the use of microalgae-based strategies for the bioremediation of heavy metals. Microalgae has bioaccumulation capability and could be used for the biosorption of heavy metals thereby offering a promising avenue in the remediation of emerging contaminants.

Recent studies have explored the possibility of using nanomaterials for the remediation process. The current improvements in the bioremediation process have been through the combination of bacteria and nanomaterials for the removal of contaminants from the environment. High efficiency has been achieved by modulating the interactions between these nanoparticles and living organisms (Cecchin et al. 2017). Various factors such as size, shape of nanoparticles, chemical nature of nanoparticles, method of synthesis, pH, and temperature determine the level of interactions between them (Patra and Baek 2014). The bacterial and nanomaterial interaction in the form of chemical or photocatalytic processes helps in effectively removing the contaminants (Vazquez-Nunez et al. 2020). Recent advancement in the field of nano-bioremediation has provided scope of continuous monitoring of the site of contamination using the nano-sensors as well, and thus providing us live data on the

Table 2.3 Nanomaterials used for bioremediation process

S. no.	Nanomaterials used	Degraded pollutants	References
1.	Carbon nanotubes	Cationic dyes Copper Nickel	Shabaan et al. (2020) Popuri et al. (2014) Adolph et al. (2012)
2.	Cyclodextrins (CD)	Metals and organic pollutants	Barbosa et al. (2019)
3.	Dendrimer-nanoparticle composite	Wastewater treatment	Guo et al. (2012)
4.	Iron oxide nanomaterials	Heavy metals	Dave and Chopda (2014)
5.	Nanocellulose composite	Diuron	Liu et al. (2018)
6.	Nanocrystalline zinc sulfide	Arsenic and Lead	Piquette et al. (2012)
7.	Nanoscale zero-valent iron (NZVI)	Arsenic Chlorinated hydrocarbons Chromium Lead	Zhu et al. (2020) Pavelkova et al. (2020) Yin et al. (2020) Moazeni et al. (2017)

status of degradation at the site (Mohamed 2017). The use of nanomaterials for the remediation process has gathered momentum in recent times (Table 2.3).

The recent trends in bioremediation have been widely used in Asian countries and especially in China, since pollution due to excessive industrialization and urbanization has been observed in the past two decades. Heavy metal contamination, petroleum hydrocarbons, chlorinated organics, and accumulation of plastics are the major pollutants (Pratush et al. 2018). Different bioremediation strategies like hyperaccumulators including *Elsholtzia splendens* for copper, *Pteris vittata* for Arsenic, *Cardamine violifolia* for Selenium, *Sedum plumbizicola* and *Sedum alfredii* for Zinc and Cadmium, respectively (Li et al. 2018). Microbial electrochemical system (MES) is another methodology used to treat organic and inorganic pollutants; In pilot scale 82.1–89.7% Petroleum hydrocarbons can be treated using this technique (Wu et al. 2018). Genetically Modified organisms are also used for bioremediation; bacteria like *E. coli*, *Staphylococcus aureus* RN4220, *Achromobacter* sp. AO22, *Methylococcus capulants*, *B. subtilis* BR151, *P. fluorescens* 4F39 were used to treat Arsenic, Lead, Mercury, Chromium, Cadmium, Nickel, respectively (Pratush et al. 2018). Microbes like bacteria, fungi, and enzymes associated with that can be used for the degradation of synthetic plastics; some of the examples include organisms like *Rhodococcus ruber* C208, *Xanthomonas* sp., *Pseudomonas stutzeri*, *Aspergillus niger*, *Chaetomium globosum*, *Thermobifida fusca* for biodegrading the synthetic plastics polyethylene, polystyrene, polypropylene, polyvinyl chloride, polyurethane, polyethylene terephthalate, respectively (Ru et al. 2020).

2.8 Conclusion

This chapter on bioremediation provided insights into different strategies that have been employed currently for biodegradation of the pollutants generated from various sources. The study on the recent advancements has revealed that the techniques such as genetically engineered organisms, nanomaterials, and microbial electrochemical systems have actually contributed in the recent past for cleaning up the polluted environment in a faster and efficient manner. A concerted approach in developing new techniques in this regard will help in mitigating the effect of environmental pollution on a large scale.

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Chapter 3
Role of Nanomaterials in Environmental Remediation: Recent Advances—A Review

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Abstract Production of consumer products, pharmacological compounds, and rapid growth in automobiles profoundly releases waste materials which are being pollutant causing threat to the environment. Emerging pollutants such as engineered nanoparticles/heavy metal ions, phenolic compounds, water-soluble pollutants, e-wastes, and other toxic gases significantly affect the nature and stability of air, soil, and water environment. Polluted environment in turn directly and indirectly affect the lives of all the living organisms either via food chains or water. Environmental pollution is a world-threat problem as it is closely related to climate change, global economy, disease outbreak, etc. Several physical, chemical, and biological remediation methods are currently exploited at different levels to eradicate polluted environment. Nanomaterials are synthesized and extensively utilized in diversified fields starting from energy to environment applications. Exotic physicochemical and biological properties of nanomaterials will give a wide range of environmental applications such as waste management, detoxification, conserving resources, and bioremediation of heavy metal ions. Carbon nanomaterials, metallic/metal oxide nanoparticles, polymeric nanocomposites, magnetic materials, quantum dots are the prominent nanomaterials widely exploited for different environmental applications due to their high photo/chemo-catalytic, mechanical, magnetic, porous nature, etc. Ample studies have been demonstrated on the role of different nanomaterials in soil, water, and air pollution management and many of those technologies are executed into real-time practices. This chapter will unfold the ongoing investigation on exploring nanotechnology in remediation and the challenges in implementing the novel technologies.

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3.1 Introduction

Environmental pollution is undeniably one of the major problems that pose a huge threat to the world. The enormous amount of pollutants from various industries has led to the contamination of soil, water, and air. The constant increase in population along with a rapid unplanned industrialization has caused a huge impact on the quality of air, soil, and water. Various pollutants have found their way into the environment due to both anthropogenic activities and natural causes, and, thus, leading to environmental pollution. These pollutants from different sources are harmful to both life and the environment. Heavy metals, pesticides, synthetic fertilizers, insecticides, industrial wastewater effluents, plastics, toxic gases, sewage, pharmaceutical wastes, radioactive wastes, hazardous wastes, etc., are few of the major environmental pollutants released from different industries (Ferdous et al. 2016; Ayangbenro and Babalola 2017; Khulbe and Matsuura 2018; Gaviria-Arroyave et al. 2020; Danish et al. 2021).

Recently, environmental cleaning up of the contaminated sites due to various industrial activities is gaining importance for a better tomorrow. Remediation of the contaminated sites is being done to redevelop them or return them to their natural state. Environmental remediation is the elimination of pollutants or contaminants from air, water, and soil, which may be done in situ or ex situ. Conventional physical remediation techniques such as pollutant substitution, separation, vitrification, and electrokinetic method and chemical treatment such as using chemicals for immobilizing the pollutant, liquid wash, vapor extraction are very expensive, time-consuming (Khalid et al. 2017; Sharma et al. 2021). Bioremediation strategy involves specific microbial consortia to detoxify or remediate several kinds of emerging pollutants in an inexpensive way (Bhuvaneswari et al. 2021). However, it has its own growth limitations on specific bioremediative applications. Researchers are constantly exploring the possibility of using different types of engineered materials for environmental remediation. Due to the complex nature of the contaminated sites with a mixture of different compounds, high volatility, and low reactivity; the exploit of nanomaterials for ecological application is in recent times gaining much importance.

Nanomaterials are those having size between 0 and 100 nm, synthesized by physical, chemical, and biological methods and it has different dimensions as zero-dimensional (0D), one-dimensional (1D), two-dimensional (2D), and three-dimensional (3D) nanostructures. Nanomaterials have unique physicochemical properties which are suitable for environmental applications in the past decades (Manawi et al. 2018; Gaviria-Arroyave et al. 2020). The physical properties of the

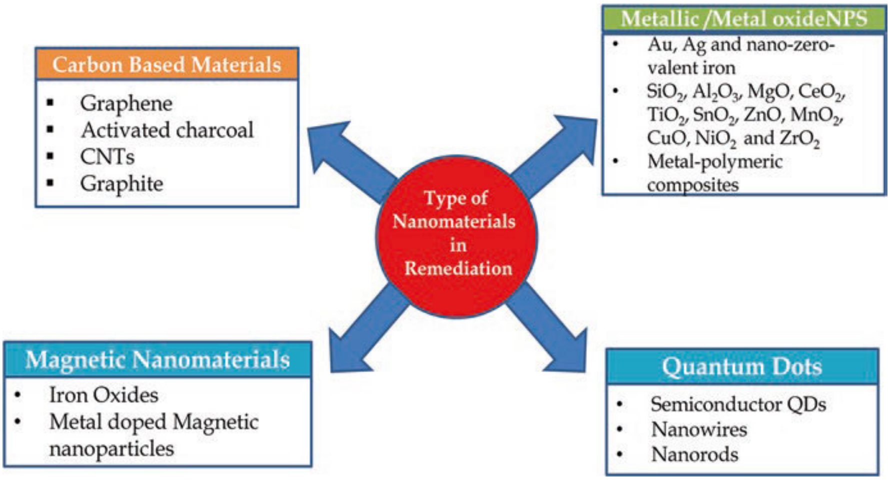


Fig. 3.1 Applications of different nanomaterials in environmental remediation processes

nanomaterials like crystallinity, texture, size, etc., higher surface-to-volume ratio and surface modification with desired functional groups that can target defined pollutants make nanomaterials efficient in environmental remediation (Guerra et al. 2017). Recently, plenty of reports are published related to the investigation on the possible contribution of nanomaterials in soil, water, and air pollution management and their technology transfer approaches for commercial applications (Danish et al. 2021; Mensah et al. 2021; Sharma et al. 2021). This report will unveil the immense collection of recent investigations that have been made on the role of different nanomaterials in the remediation process for pollution-free environment. Figure 3.1 shows the recent application of different nanomaterials in environmental remediation processes. Four classes of nanomaterials have been recently studied based on their innate properties and the mechanism of interaction with the pollutants. They are (a) Metal/Metal oxide nanoparticles, (b) Carbon Materials, (c) Quantum dot, and (d) Magnetic nanoparticles.

3.2 Type of Remediation Process by the Nanomaterials

3.2.1 Pollution Monitoring

Commonly, physical and chemical properties of the pollutants are differing and their occurrence in different environment also varies. Hence, identifying the presence of pollutants such as pesticides in food, heavy metals in soil and consumer products is essential either by quantitative and qualitative methods. Nanobiosensors are one of the upcoming technologies used in monitoring the emerging as well as existing pollutants. At present, optical, electrochemical, biological, and fluorescent

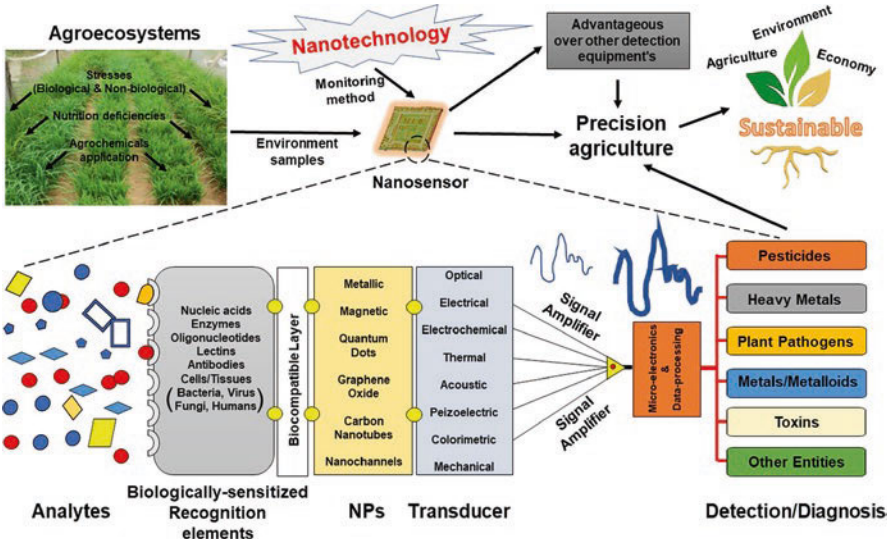


Fig. 3.2 Multiple functions of nanosensors in detecting the soil pollutants

biosensors are used to detect chemical/metal contaminants at very low regimes in soil (Fig. 3.2) as well as in other environments (Gaviria-Arroyave et al. 2020; Sharma et al. 2021; Guan et al. 2021). Among nanomaterials, metallic nanoparticles, carbon nanotubes, carbon dots, graphene, quantum dots (QD), transition metal chalcogens, and metal-organic frames (MOFs) are mainly used in biosensors (Gaviria-Arroyave et al. 2020; Wang et al. 2018). Table 3.1 shows the specific nanomaterials that are used to detect and monitor environmental pollutants (Gaviria-Arroyave et al. 2020). The detection limit is found to be very low when the nanoscale materials are used to detect the pollutants (Pesticides and Heavy metals).

In fact, graphene and metallic nanoparticles (MNPs) such as Au, Ag, or zinc are preferred as effective quenching agents in fluorescent biosensors due to their better surface characteristics. In this, AuNPs and carbon dots are used to detect organophosphates (Yan et al. 2018; Cheng et al. 2018) which are major chemical pollutants present in pesticides causing long-term disorders to consumers. Contaminants like heavy metals, nanobiosensors based on Forster resonance energy transfer phenomenon (FRET) are advantageous to detect multiple metal ions (Yun et al. 2017) over other conventional sophisticated spectroscopic techniques such as atomic absorption spectroscopy, UV–visible spectroscopy, and other chromatographic techniques as they are time- and energy-consuming processes. Medical wastes such as antibiotics (Kanamycin, Glutothion, Sulfadimethoxine, Thaimphenicol) are detected using carbon dots (CDs) and SiO_2–MnO_2 nanocomposites (Wang et al. 2020; Peng et al. 2020). Figure 3.2 represents the role of nanosensors for agriculture applications (Sharma et al. 2021) to be also applied for pollutant detection in soil.

In addition to the above applications, metal oxide nanoparticles (NiO, ZnO, TiO_2, SnO_2, and WO_3) based gas sensors are served as "e-noses" (Danish et al.

Table 3.1 Exploitation of advanced nanomaterials for high sensitive detection of pesticides, heavy metals, and phenol pollutants

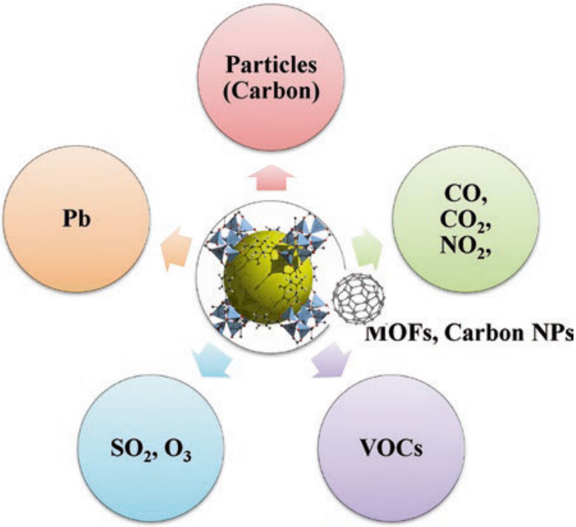
S. no.	Nanomaterial	Pollutant detected	Detection limit	Reference
1.	Carbon dots (CDs) conjugated with acetylcholinesterase	Organophosphates	1 ng/l	Gaviria-Arroyave et al. (2020), Li et al. (2018)
2.	Boron nitride quantum dots conjugated with acetylcholinesterase	Paraoxon	33.3 ng/l	Zhan et al. (2019)
3.	CdTe QDs aerogel with ache, stabilized with L-glutathione (GSH)	Paraoxon, Parathion, Dichlorvos, and Deltamethrin	1.2, 0.94, 11.7, and 0.38 pm, respectively	Hu et al. (2019)
4.	Acth—1,2-bis[4-(3-sulfonatopropoxyl)phenyl]-1,2-diphenylethene (bspotpe)-SiO_2 NPs conglomerate with MnO_2 nanosheets	Paraoxon	0.7 mm	Wu et al. (2019)
5.	Gold-doped carbon dots and DNAzyme	Pb(II)	0.25 nm	Li et al. (2020)
6.	Unlabeled aptamer, syber with AuNPs	Bisphenol a	9 pg/ml	Rajabnejad et al. (2020)
7.	CdSe/S-functionalized MoS_2 with poly (diallyldimethylammonium chloride)—QD labeled DNA probe	Hg(II)	0.1 pm	He et al. (2020)
8.	Green fluorescent copper nanocluster	Hg and sulfide ions	1.7 and 1.02 nm, respectively	Maruthupandi et al. (2020)
9.	Silica—quantum dot graphene, nanomaterials	Cr ions, Cu^{2+}, and Hg^{2+}	Cr—0.708 ppb, Cu, and Hg ions—<10 nm	Gaviria-Arroyave et al. (2020)

2021) in detecting the foul smell and odors that arise in food packaging and quality assessment which prevent further contamination. Generation of poisonous gases like ammonia from industries can be detected using the above metal oxides is known as e-nose technology.

3.2.2 *Toxic Gas Absorption*

Industrial emissions and burning of fossil fuels produce toxic gases such as CO, H_2S, NO_2, and SO_2 that are considered for air pollution (Singh and Ahuja 2020; Peng et al. 2021). It leads to many respiratory diseases and neurological disorders if

Fig. 3.3 Action of MOFs and carbon nanomaterials used for the absorption of toxic pollutants



it is exposed at long-term period. Metal organic frameworks (MOFs) possess unique characteristics like adsorption capacity and affinity to gases and self-cleaning upon light irradiation (Meicheng Wen et al. 2019). It contributes in the elimination of primary and secondary pollution. Figure 3.3 depicts the action of MOFs and Carbon nanomaterials used for the absorption of toxic pollutants in industries. The adsorbed gas pollutants can also be used as renewable energy resources.

Different carbon nanomaterials such as fullerenes, multiwalled nanotubes, nanofibers, and graphene are being widely used in wastewater treatment as adsorbents and also to remove air pollutants due to their high surface area, biocompatibility, inexpensive, effectiveness, and eco-friendliness (Scida et al. 2011; Anjum et al. 2016). Graphene-based materials are employed for the adsorption of gaseous contaminants like greenhouses (carbon dioxide (CO_2), hydrogen (H_2), methane (CH_4), and nitrogen (N_2) gases) (Ghosh et al. 2008; Zhao et al. 2012).

3.2.3 Agricultural Waste Management

Fullerenes' physicochemical features make them ideal candidates for extracting various substances from aquatic environments. They are especially used to remove the Cu^{2+} pollutant which is relatively higher in the first and the observed equilibrium isotherm of Cu^{2+} which is adsorbed on the fullerene fits the Langmuir model (Zhang et al. 2013). Despite fullerenes possess exotic properties on water adsorption, the production cost of the materials hinders the application at a large scale. However, they are hardly used to improve the adsorption efficiency of carbon materials such as activated carbon, lignin, and zeolites. Fullerene production increases the hydrophobic character of materials, making it more suitable for adsorption and facilitating recycling (Samonin et al. 2014). 4-Nitrophenol and its derivatives in agrochemicals like herbicide, insecticide, pesticide, etc., are efficiently degraded by metallic nanomaterials (PdNPs, CuNPs, AuNPs, and AgNPs) due to their exotic catalytic potential on the reactive groups of 4-nitrophenol derivatives (Singh et al. 2018).

3.2.4 Heavy Metal Leaching

Heavy metal contaminants in soil and water cause a significant threat to living organisms in the terrestrial environment. Use of nanometal frameworks as nano adsorbents is one of the efficient strategies to remove heavy metals from the contaminated sites in which high surface area nanoparticles (e.g., silica and graphene) sequester the heavy metals (Mensah et al. 2021). Guo et al. used an in situ coprecipitation approach to amalgamate a nanocomposite of partly reduced graphene oxide to remove Pb^{2+} ions from water. The prepared nanocomposites reveal a good adsorption capacity of Pb^{2+} ions (373.14 mg/g) from aqueous solution. In the same way, Zhang et al. (2018) used functionalized reduced graphene oxide (rGOs) to remove different heavy metal ions (Pb^{2+}, Cu^{2+}, Ni^{2+}, Cd^{2+}, and Cr^{3+}) from aqueous solution. It is found that the nanocomposites showed maximum adsorption capacities of 689, 59, 66, 267, and 191 mg/g, respectively, against the above heavy metals treated aqueous solution.

Khulbe and Matsuura (2018) have reported that multiwalled CNTs (MWCNTs) can be well dispersed and be easily separated from water. Hence, they were exploited to effectively remove Pb(II) and Mn(II) from wastewater. CNTs are further oxidized with potent oxidizing agents with Potassium permanganate, Hydrogen peroxide, and Nitric acid and showed high adsorption efficiency of heavy metals (Uranium, Cadmium, and Lead) (Anna and Krystyna 2007). MWCNTs and SiO_2 nanocomposite has been observed to be a potential adsorbent in removing Pb(II) from the contaminated aqueous environment (Saleh 2015). Similarly, MWCNTs-Fe_2O_3 for the removal of Cu^{2+} (Tang et al. 2012), MWCNTs-ZrO_2 for the removal of As^{3+} (Ntim and Mitra 2012), MWCNTs-Fe_3O_4 for the removal of Ni^{2+} (Yang et al. 2009), MWCNTs-MnO_2-Fe_2O_3 for the removal of Cr^{6+} (Luo et al. 2013) also observed. Zhao et al. (2010) have reported that plasma-oxidized nanotubes with TiO_2 and MnO_2 can be utilized for the removal of lead ions from water. Porous graphene nanomaterial and reduced graphene oxide fabricated with Fe_3O_4 have been applied and shown the removal of As^{3+} (Tabish et al. 2018) and Pb^{2+} ions (Guo et al. 2018) from aqueous solution.

3.2.5 *Dye Degradation*

The zinc oxide NPs under ultraviolet (UV) radiation are capable to reduce the toxic dye Methylene Blue as reported by Li et al. (2019). Zinc oxide NPs fabricated with TiO_2 or other nanomaterials act as a valuable dye adsorbent from the solution by enhancing the number of adsorption sites (Jain and Vaya 2017). ZnO NPs coupled with chitosan have been used as a nanocomposite in the removal of Direct Blue 78 and Acid Black 26 (Salehi et al. 2010) and CuO-ZnO composite nanofibers have been utilized for the adsorption of Congo red (Malwal and Gopinath 2017). TiO_2 nanoparticles are very much resistant to photochemical corrosion, alkali and acid treatment, and nonhazardous in nature. Hence, they are widely used as a photocatalyst for degrading many dyes under UV. Nano-TiO_2 can be activated under light leading to the production of free radicals with high catalytic activity causing effective photodecomposition of numerous organic and inorganic substances. Laishram et al. (2018) have reported that the dyes solo-chrome black (SB), thymol blue (TB), cresol red (CR), methyl blue (MB), and methyl orange (MO) have been photocatalytically degraded by catalysts such as titanium dioxide (TiO_2), hafnium oxide (HfO_2)/TiO_2, and hydrogenated HfO_2 doped TiO_2 (H-HfO_2/TiO_2). RGOs fabricated TiO_2 was successfully applied for the photodegradation of alizarin red S (Rommozzi et al. 2018). Exploring the use of inexpensive semiconductors like Tungsten oxide and its nanocomposites with GO/Dopants is gaining attention in dye degradation from industrial effluents.

3.2.6 *Disinfection*

By combining zinc oxide with tea polyphenol and reducing graphene oxide, Zheng et al. created nanocomposites (TPG-ZnO). Heavy metal ions were removed using a specially designed polymer that also has antibacterial properties. They used this material to remove Pb^{2+} ions from an aqueous solution with 98.9% adsorption efficiency, and the adsorbent was discovered to have antibacterial activities against *Streptococcus mutans* with a 99% eradication rate (Zhang et al. 2018).

3.2.7 *Wastewater Treatment*

A range of carbon nanostructures, each with its own set of functions, are being researched and developed and used in the remediation of contaminated water, industrial effluents, and textile wastes (Thines et al. 2017). In the last few decades, the researchers are focusing on the synthesis and development of magnetic nanomaterials (MNMs) because of their unique properties like high magnetic susceptibility, exhibition of superparamagnetism, and lower coercivity and as compared to their bulk counterparts (Ali et al. 2016; Mensah et al. 2021). Superparamagnetic iron oxide nanoparticles particularly the magnetite (Fe_3O_4) and maghemite (Fe_2O_3) NPs are the most exploited and effective magnetic nanomaterial because of exceptional properties such as ease of synthesis, supraparamagnetic, biocompatibility, and non-hazardous nature. Due to their strong magnetic properties and oleophilicity, magnetite and maghemite NPs are extensively used in wastewater treatment (Li et al. 2011), heavy metal removal (Dave and Chopda 2014), remediation of oils, organic pollutants, and dyes from water (Kumar et al. 2015; Doshi et al. 2018). TiO_2 and zerovalent iron nanoparticles have been reported as effective and potent adsorbents for the removal of surfactants from the contaminated water (Dasgupta et al. 2018; Abd El-Lateef et al. 2018).

3.2.7.1 Photocatalytic Action

Hydrophilic fullerenes (C60) are shown potent photocatalysts to kill pathogenic microbes in water as they are readily transformed to C–C bonds and then to C–H bonds. This is more suitable in developing hydrogen storage units using such uncontaminated green nanomaterials (Brunet et al. 2009). Water-soluble fullerene compounds are used as a sensitizer to produce reactive oxygen species (ROS) in water which have the ability to photodegrade organic pollutants in water, and it can be used as anti-oxidants. It is important to mention that one can easily recover the fullerenes from treated water after photodegradation function (Pickering 2005). CNTs improve the process of photocatalysis which are widely being used for the wastewater treatment due to their excellent surface properties (Mauter and Elimelech 2008). Activated carbon, lignin, and zeolites fabricated with fullerene have improved the adsorption efficiency.

Metal oxide nanoparticles such as Titania, Zinc Oxide, Tungsten Oxide, and their composites are reported as promising photocatalytic nanomaterials due to their high tunability in their structures, low-cost synthesis, photocatalytic performance, heterogeneous action, etc. (Danish et al. 2021). Along with the above metal oxides, the dopants and carbon nanohybrids enhances the photocatalytic performance on a wide range of organic pollutants.

3.2.7.2 Removal of Oil

Fullerenes have been reported for the efficient removal of Cu^{2+} ions (Samonin et al. 2014; Alekseeva et al. 2016). Recent research reports demonstrated the application of fabricated carbon nanotubes especially single-walled (SWCNTs) as a photocatalyst in combination with Titania NPs for photodegradation and purification of oil-contaminated water (Gupta et al. 2017). Yang et al. (2017) have also proved that functionalized Multiwalled CNTs with 3-aminopropyltriethoxysilane (APTES) anchored on the polyvinylidene fluoride (PVDF) membrane purifies oil spills in aquatic environment. In oil under simulated seawater conditions, Magnetic nanomaterials (MNMs) functionalized with polyvinyl-pyrrolidone (PVP) magnetically remove entire lower hydrocarbons (C9–C21) and partially higher hydrocarbons (C22–C25) (Mirshahghassemi and Lead 2015). In addition, chitosan grafted MNMs on functionalized silica resulted in better degradation of diesel in water (Lü et al. 2017). MNMs coated with many surfactants like oleic acid (OA), sodium dodecyl sulfate (SDS), sodium dodecyl benzene sulfonate (SDBS), Tween 80, and Triton X-100 also confirmed significant results when they tested under simulated seawater added with diesel (López et al. 2019).

3.2.8 Renewable Energy Resources: Generation

Degradation of inorganic and organic pollutants in industrial wastes (batteries), botanical wastes, and aquatic pollutants serve as a precursor material for nanoparticles' synthesis (Abdelbasir et al. 2020). Nano metal oxides such as Silica and ZnO are feasibly synthesized using agri wastes rice husk ash and plant leaves, respectively, for electronic, biomedical, and cosmetic applications (Suriyaprabha and Rajendran 2018; Suriyaprabha et al. 2019). Because mesoporous silica nanoparticles are of highly demanded resource for Si conversion which is majorly employed in energy fields such as batteries solar cells and semiconductor device fabrication. Similarly, ZnO nanoparticles are nontoxic in nature and have equal potential to TiO_2 which is widely used for photocatalytic activity, and thereby generating renewable energy resources in terms of water splitting, hydrogen gas generation, organic degradation, heavy metal sequestration, etc. (Danish et al. 2021; Mensah et al. 2021). Many heavy metals, rare earth metals, and magnetic materials are recovered from the effluents and sledges (Samaddar et al. 2018). Nanoadsorbants used in heavy metal sequestration are also used in nanoparticles amalgamation process in which they are used as reducing agents and, in turn, avoid the use of hazardous chemicals like sodium borohydrides. Certain organic wastes are also used as the source for renewable energy production like biofuels. This approach is considered as lucrative and effective remediation process in eliminating pollutants as well as regenerating the novel materials of industrial importance.

3.2.9 Mediwaste Management

Disposal of medi-wastes from hospitals, pharmaceutical industries, and surface contamination are considered as one of the major organic pollutants which cause serious threat to the life of humans (Danish et al. 2021). Metal oxide/graphite-based carbonization of organic pollutants and infected medical accessories such as cotton fibers can be the immediate solution of efficient waste management. Husein et al. (2019) have reported that the green-synthesized copper nanoparticles have been utilized effectively for the removal of drugs like Diclofenac (Dic), Naproxen (Nab), and Ibuprofen (Ibu) from wastewater. Similarly, nano-Fe_3O_4 synthesized by heterogeneous electro-Fenton process is also used to remove the drug Metronidazole from solutions (Rahmatinia and Rahmatinia 2018). In fact, the development of resorbable polymeric nanomaterials and their metal oxide nanocomposites prevent the disposal and bioremediation issues in the biomedical field (Prabhu et al. 2014) as those materials degrade and excreted themselves in the hosts. Metal oxide nanocomposites also prevent microbial infection in the body and in vitro environment by biomedical materials due to their antimicrobial properties.

3.3 Nanobioremediation (NBR)

Environmental remediation will be very effective and more practical when the pollutant treatment is combined with biological sources along with nanomaterials. Especially, heavy metals and agricultural pollutants are effectively remediated by the action of microorganisms which chelate the detoxification of metal ions and it

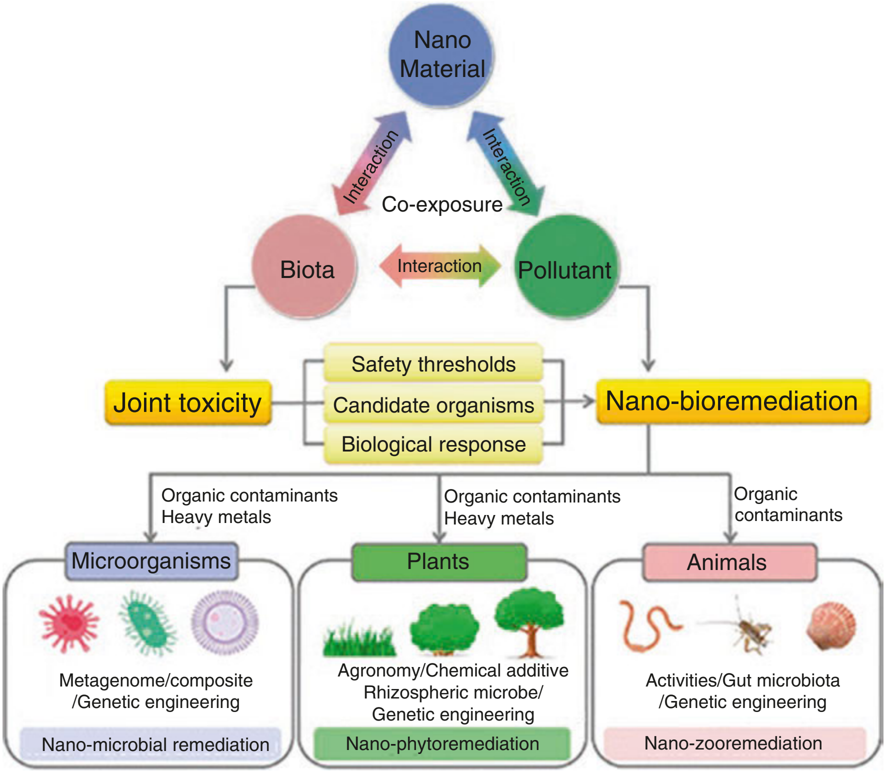


Fig. 3.4 Schematic representation of nanobioremediation process. (Reprinted from Hou et al. 2019)

can either produce nanoparticles or sequester heavy metal resources (Bhuvaneswari et al. 2021). On the other hand, nanoparticles are immobilized with microbes as a career for absorbing emerging contaminants both in situ and ex situ conditions. Interaction of nanomaterials with pollutant and living organisms are schematically well represented by Hou et al. (2019). NBR process mainly depends on the kind of pollutants (Heavy metals/organic compounds), nanoadsorbents, and Biota (Fig. 3.4). Hence, nanobioremediation significantly speeds up the volume and duration of the pollutant treatment process.

3.4 Challenges

Nanoscale materials are very effective as they physically and chemically interact with environmental pollutants and change their toxicity into nontoxic as well as eco-friendly end products. Figure 3.5 conveys the possible interaction of various nanomaterials with the pollutants and their modification. The illustrated image clearly demonstrates the action of nanomaterials on the pollutants.

Transition metal oxides, magnetic nanoparticles often react with environmental pollutants and readily transform into nontoxic forms and separate them. Hence, nanomaterials-driven remediation is widely considered for real-time applications especially in wastewater treatment, heavy metal detection, detoxification, etc.

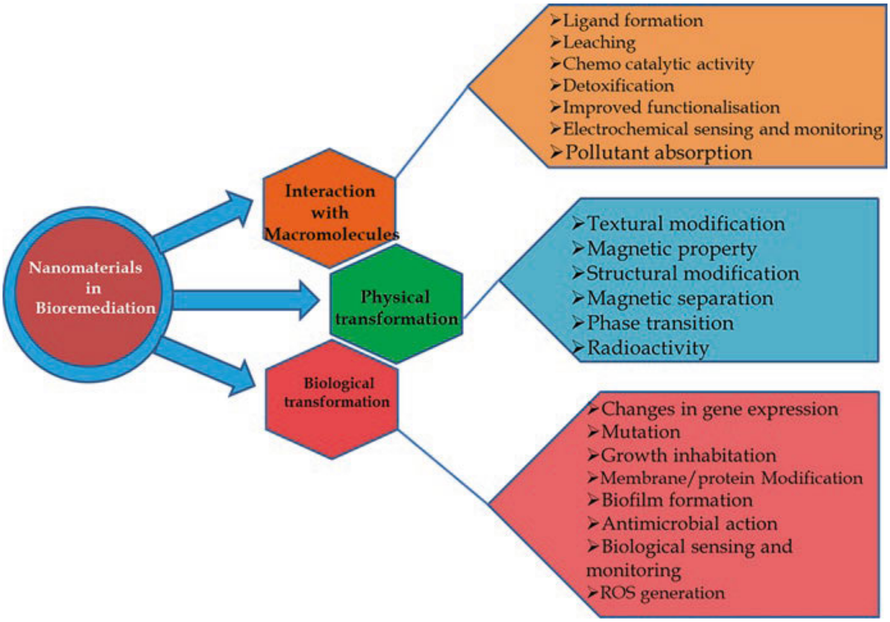


Fig. 3.5 Possible interactions of various nanomaterials with the pollutants and their modification

However, commercialization of engineered nanomaterials for environmental applications is still facing hurdles in terms of the following parameters

(a) Identifying the precise interaction of nanomaterials with inorganic and organic parts of the pollutants.
(b) Optimization of nanoparticles' use and safety upon release in environment.
(c) Production cost of quantum dots, MOFs, sensors, etc.
(d) Stability of nanomaterials as frameworks and nanocomposites.
(e) Vivid demonstration of degradation pathways for specific pollutants.
(f) Diversity in their environment makes the nanomaterials changes in their reactivity and stability.

Overcoming the above hurdles many research groups come out with the outcomes. Exploiting nanomaterials in remediation of pollutants into real-time practices are gradually increasing in many countries by companies and research institutes. Table 3.2 summarizes few technology transferred nanomaterials for environmental application developed in different countries.

Table 3.2 Technology transferred nanomaterials for environmental applications

S. no.	Nanomaterials	Application	Organization	References
1.	Graphene nanomaterials	Heavy metal removal	Sparc Technologies Limited, South Australasia	Cotton (2021)
2.	Phosphate elimination and recovery lightweight (pearl) membrane	Phosphate sorption	North-Western University, Evanston, IL, USA	Ribet et al. (2021)
3.	Green chemistry solutions	Industrial-born pollution	U Spinoff Claros Technologies, University of Minnesota	https://www.startribune.com/minnesota-s-claros-commercializes-green-chemistry-pollution-fixes/600036941/
4.	Electrochemical nanosensors	Pesticide detection	Institute of Functional Materials and Agricultural Applied Chemistry, China	Xie et al. (2018)
5.	Aptamer-based paper sensor	Pathogen detection	Chungbuk National University, South Korea	Shin et al. (2018)
6.	Picolyl-functionalized rhodamine sensors	Glyphosate detection	Northwest A&F University, China	Guan et al. (2021)

3.5 Conclusion

Nanoscale materials such as nanometal oxides, magnetic nanoparticles, carbon-based nanomaterials, and polymeric/aptameric nanocomposites attract much attention on detecting and remediating the environmental pollutants due to their excellent interaction with organic and inorganic waste materials, physicochemical modifications, and stable integration with biological molecules. Even though the production cost and implementation hamper the commercialization of such nanomaterials, it is essential to implement this strategy to protect environment-related illness. In fact, lucrative synthesis of highly porous nanomaterials is rapidly progressing in the present scenario, and hence they serve as better alternatives to conventional chemical treatment methods in near future. By reviewing the recent investigations of the environmental remediation process in detail, it is noted that metal oxide nanoparticles are predominant and widely applicable to most of the pollutant treatment methods followed by carbon-based nanomaterials. Notable outcomes are observed by the action of potent nanomaterials on organic and inorganic pollutants. Moreover, precise validation and understanding of the health risks, cost estimation and threshold level of nanoparticles to be used for pollutant degradation pathways are yet to be essentially carried out. By overcoming the hurdles, many researches are emerged from proof-of-concept level to scale-up level and finally technology transfer in the field of environmental treatment processes.

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Chapter 4

Production Techniques, Mechanism, and Application of Biochar in Remediating Soil Contaminated with Heavy Metals: A Review

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Abstract The ultimate goal of heavy metal contaminated soil remediation is to increase crop yields on the premise of ensuring food production safety. Soil contaminated by heavy metals threatens the quality of agricultural products and human health. Hence, it is necessary to choose appropriate economic and effective remediation techniques to control the deterioration and revive the land quality. Among the methods available, biochar application for adsorption and remediation of heavy metal contaminated soil is emerging to be a sustainable approach. Biochar introduction to the soil provides organic matter and essential macro and micronutrients like C, N, P, K, Ca, Mg, etc., which enhances soil enzyme and microbial activities. Additionally, the plant root environment, soil water retention, and saturated hydrau-

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lic conductivity can be improved in the presence of biochar. This chapter is intended to present an overview of the production techniques of biochar, its properties, and characteristics required for effective heavy metal removal and the corresponding process conditions, mechanisms involved in the interaction of biochar with heavy metals, and the benefits as well as bottlenecks of biochar application in soil.

Keywords Biochar · Heavy metals · Remediation · Soil contamination · Mechanism

4.1 Introduction

Soil contamination with heavy metals is a matter of global concern due to the impending damage of the natural ecosystem and human health hazards through bioaccumulation in the food chain. Some of the soil contaminants include heavy metals (cadmium (Cd), zinc (Zn), copper (Cu), nickel (Ni), lead (Pb), arsenic (As), etc.), pesticides, and polycyclic aromatic hydrocarbons (PAHs). Though minuscules of heavy metals are naturally present in the soil, the concentration rises beyond admissible limits (10–1000 times) due to anthropogenic activities such as mining, solid waste disposal, and wastewater irrigation (Gao et al. 2020). Scientific research articles reported the average concentration of heavy metals in soil (worldwide data) to be 20 mg/kg Cu, 0.06 mg/kg Cd, 20–200 mg/kg Cr, 10–150 mg/kg Pb, 40 mg/kg Ni, and 10–300 mg/kg Zn. National soil surveys revealed that out of the 19.4% contaminated arable land, more than 80% was polluted with inorganic compounds. The potential risk of heavy metal leaching to surface or ground waters should also be taken into consideration. It is claimed that millions of people from India, China, and Bangladesh are facing health-related threats due to the negative impacts of metal contamination (Sonone et al. 2020).

Globally, all developed and developing nations require new technology to renew nature and negative emissions technologies for greenhouse gases. Soil amendments are being developed to carry out in situ metal stabilization and avoid the risk of soil and groundwater ecosystems. Of the many strategies employed to remediate heavy metal contaminated soil like phytoremediation, soil dressing, soil washing, etc., chemical immobilization proved to be the most efficient, in terms of its economic feasibility and simplicity. Chemical immobilization includes the addition of amendments to the soil to help retain contaminants. Tons of crop residues are produced worldwide which are neither effectively used nor recycled (Wang et al. 2018; Maji et al. 2020; Pathy et al. 2020b). The raw materials for adsorbent production include waste products of humans, animals, and plants. So, this technology also helps to improve the waste management of the nations and give more potential economy of the nation. China is one of the most populated countries that produce 2.5–3 billion tons of animal wastes, 1.2 billion tons of vegetable and fruit wastes, 2 billion tons of municipal waste (Chen et al. 2020a), 0.5–0.7 billion tons of domestic garbage wastes, and 0.17–0.3 billion tons of domestic meat wastes per year. These are fundamental raw materials of the large-scale production of biochar (Wang et al. 2013).

Biochar production technology is proving to be the best method to treat soil contamination by heavy metals. Biochar production is a currently developing technology to enhance the soil nutrients retention capacity, water holding capacity, reduction of greenhouse gas emission, and stabilizes the carbon (Kamali et al. 2021). Biochar is a product of the thermochemical conversion of biomass to carbon-rich materials, under limited oxygen conditions, which is used for environmental management. This carbon-negative or carbon-neutral material can be used as a soil ameliorant, adsorbent, and in several climate mitigation approaches. The process greatly reduces the volume of the residues while eliminating the pathogens and improving the nutrient utilization efficiency. Their specific activities include high cation exchange capacity (CEC), high pH, moisture content, and total nitrogen and phosphorus ions of soil, promoting root development and decreasing soil erosion (Abhijeet et al. 2020; Mandal et al. 2021). Due to the aromatic nature of biochar, it can effectively adsorb both organic and inorganic contaminants. Application of biochar to soil is reported to effectively immobilize heavy metals due to their high surface area, pore volume, and sufficient adsorption sites. The high porous structure of biochar can be attributed to the presence of tubular arrangement of plant cells. Being a slow-release organic material, biochar releases the adsorbed metal ions at a slower rate, fulfilling the requirements of the plants (Wang et al. 2018; Pathy et al. 2021). Table 4.1 shows various sources of biochar that have been used for removing heavy metals in soil and Table 4.2 includes information on remediation of other contaminants using biochar.

Table 4.1 Sources of biochar, their production conditions, and heavy metals remediated from soil

Source of biochar	Pyrolytic temperature (°C)	Residence time (min)	Removal of pollutants	References
Bamboo	750	180	Cd, Cu, Pb, and Zn	Lu et al. (2014)
Rice straw	500	30	Cd, Cu, Pb, and Zn	Lu et al. (2014)
Diary manure	350	240	Pb, Cd, and Zn	Liang et al. (2014)
Sugarcane straw	700	60	Cd, Pb, and Zn	Puga et al. (2015)
Corn straw	600	120	As	Yu et al. (2015)
Olive mill waste	400–450	30	Zn, Pb, and Cd	Hmid et al. (2015)
Bamboo	750	180	Pb and Cd	Xu et al. (2016)
Sludge	400	120	Cr and As	Tsang et al. (2016)
Soybean stover	300 and 700	180	Pb and As	Ahmad et al. (2016)
Pine needle	300 and 700	180	Pb and As	Ahmad et al. (2016)

Table 4.1 (continued)

Source of biochar	Pyrolytic temperature (°C)	Residence time (min)	Removal of pollutants	References
Bamboo	750	180	Cd, Cu, Pb, and Zn	Lu et al. (2017)
Rice straw	500	30	Cd, Cu, Pb, and Zn	Lu et al. (2017)
Pine cone	200 and 500	120	Pb	Igalavithana et al. (2017)
Vegetable waste	200 and 500	120	Pb	Igalavithana et al. (2017)
Switchgrass	400	30	Pb, Ni, and Co	Mohamed et al. (2017)
Wheat straw	450	120	Cd	Liu et al. (2018)
Sugarcane bagasse	450	240	Cd, Cu and Pb	Nie et al. (2018)
Rice husk	550	120	Hg	O'Connor et al. (2018)
Bamboo hardwood	550	300	Cd	Wu et al. (2019)
Rice straw	500	180	As, Cd, Cu, and Zn	Tang et al. (2020)
Sewage sludge	500	120	Zn	Penido et al. (2019)
Silver grass	500–600	60	As and Pb	El-Naggar et al. (2020)
Rice straw	500–600	60	As and Pb	El-Naggar et al. (2020)
Umbrella tree wood	500–600	60	As and Pb	El-Naggar et al. (2020)
Rice straw	500	300	Cd and Pb	Fan et al. (2020)
Maize straw	400	480	Cd and Cu	Tu et al. (2020)
Wood	550	120	Cu, Cd, and As	Zhang et al. (2020)
Bamboo	550	120	Cu, Cd, and As	Zhang et al. (2020)
Cornstalk	550	120	Cu, Cd, and As	Zhang et al. (2020)
Rice husk	550	120	Cu, Cd, and As	Zhang et al. (2020)
Corn straw	350, 500, and 700	120	Zn	Song et al. (2020)
Seweage sludge	300 and 500	30	Cu, Mn, Pb, and Zn	de Figueiredo et al. (2019)
Rice hull	450	120	Cd and Cu	Wang et al. (2021b)
Oriental plane	650	120	As, Cd and Pb	Wen et al. (2021)
Pig carcass	650	120	As and Pb	Pan et al. (2021)
Green waste	650	120	As and Pb	Pan et al. (2021)
Wheat straw	600–900	120	Pb, Zn, and Cd	Li et al. (2022)
Switchgrass	300	30 min	U(VI)	Kumar et al. (2011)
Sugar beet tailing	300	~2 h	Cr(VI)	Dong et al. (2011)

(continued)

Table 4.1 (continued)

Source of biochar	Pyrolytic temperature (°C)	Residence time (min)	Removal of pollutants	References
Spartina alterniflora	400	2 h	Cu(II)	Li et al. (2013)
Soybean straw	400	3.75 h	Cu(II)	Tong et al. (2011)
Sludge	400	2 h	Cr(VI) and Pb(II)	Zhang et al. (2013a)
Sludge	550	2 h	Pb(II)	Lu et al. (2012)
Rice straw	700, 400, and 100	6 h	Al	Qian and Chen (2013)
Rice husk and pinewood	300	20 min	Pb(II)	Liu and Zhang (2009)
Pine needles	200	16 h	U(VI)	Zhang et al. (2013b)
Miscanthus sacchariflorus	300, 400, 500, and 600	1 h	Cd(II)	Kim et al. (2013)
Hardwood	450	<5 s	Zn(II) and Cu(II)	Chen et al. (2011)
Corn straw	600	2 h	Zn(II) and Cu(II)	Chen et al. (2011)
Cattle manure	100, 400, and 700	6 h	Al	Qian and Chen (2013)
Canola straw	400	3.75 h	Cu(II)	Tong et al. (2011)
Spartina alterniflora	400	2 h	Cu(II)	Li et al. (2013)
Sludge	400–700	2 h	Fluoride	Oh et al. (2012)
Rice straw	100–700	6 h	Aluminum	Qian and Chen (2013)
Rice husk	350	4 h	Pb, Cu, Zn, and Cd	Xu et al. (2013)
Rice husk and pinewood	300	20 min	Lead	Liu and Zhang (2009)
Orange peel	400–700	2 h	Fluoride	Oh et al. (2012)
Miscanthus sacchariflorus	300–600	1 h	Cadmium	Kim et al. (2013)

This chapter investigates the application of biochar for remediating soil contaminated with heavy metals. It elucidates various techniques available for biochar production and the optimum process parameters required for synthesizing biochar with characteristics suitable for removing heavy metals efficiently. The mechanisms involved during the interaction of biochar with heavy metals are discussed. In addition, the advantages and disadvantages of biochar application in soil and the future perspectives in this research area.

Table 4.2 Sources of biochar, their production conditions, and other contaminants remediated from soil

Source of biochar	Pyrolytic temperature (°C)	Residence time (min)	Removal of pollutants	References
Swine manure	400	1 h	Herbicide paraquat	Tsai and Chen (2013)
Wood	200–600	1 h	Fluorinated herbicides	Sun et al. (2011)
Sugarcane bagasse	300 and 700	6 h	Hydrophobic organic compounds (HOCs)	Chen et al. (2012a)
Sugarcane bagasse	450 and 600	–	Sulfamethoxazole	Yao et al. (2012)
Soybean stover	300 and 700	3 h	Trichloroethylene	Ahmad et al. (2012)
Pinewood shavings	150–700	6 h	Naphthalene	Chen et al. (2012b)
Pine needle litters	100–700	6 h	Naphthalene (NAPH), nitrobenzene (NB), and m-dinitrobenzene (m-DNB)	Chen et al. (2008)
Pine needle	300 and 700	6 h	Hydrophobic organic compounds (HOCs)	Chen et al. (2012c)
Pine needle	300–700	3 h	Trichloroethylene	Ahmad et al. (2013)
Peanut shells	300 and 700	3 h	Trichloroethylene	Ahmad et al. (2012)
Palm bark	400	30 min	Methylene blue dye	Sun et al. (2013)
Orange peel	150–700	6 h	Naphthalene and 1-naphthol	Chen and Chen (2009)
Orange peel	300 and 700	6 h	Hydrophobic organic compounds (HOCs)	Chen et al. (2012c)

4.2 Techniques for Production of Biochar

The application of biochar in wastewater treatment and water purification is due to their inheriting characters including porous structure, high specific surface area, large pore volume, acid and alkali corrosion resistance, and rich functional groups. The preparatory conditions of biochar are crucial and should be chosen wisely depending on the type of biomass and intended properties and application. The characteristics of biochar and byproducts formed during the production depend on the process conditions such as temperature, heating rate, residence time, etc. These features influence the surface properties and porous structure of biochar (Wang et al. 2017).

In general, biochar production involved three processes including simultaneous carbonization, magnetization, and activation. Biochar has carbon-rich molecules depending on the organic content of the biomass and some specific characters including cation exchange capacity, large specific surface area, stable structure, and a large number of carbon sources. More than 200 scientific research articles were published about the production and application of biochar in the last decade (Wang and Wang 2019). There are several techniques for biochar production including pyrolysis, torrefaction, hydrothermal carbonization (HTC), gasification, and microwave carbonization. Among these methods, pyrolysis, HTC, and microwave carbonization are reported to be better due to ease of process controls, no requirement for drying steps, no hysteresis, rapid heating, and energy efficiency.

Pyrolysis is the most common method for the production of biochar. In this method, the organic materials are burned under high temperatures in inert atmospheric conditions and oxygen-free conditions (Selvam and Paramasivan 2021b; Selvam et al. 2021; Pathy et al. 2020a). This thermochemical process can be used to convert biomass into biochar, bio-oil, and syngas, whose composition depends on the range of operational conditions (Swagathnath et al. 2019b). Pyrolysis can be categorized as slow and fast pyrolysis corresponding to the heating rate. In slow pyrolysis (1–20 °C/min), the biochar is produced with high fixed carbon content and low minerals, making them suitable for carbon sequestration in soil. In addition, the composition of biochar produced in slow pyrolysis (35%) is much higher compared to fast pyrolysis (10%), where the bio-oil composition is considerably large (70%). The slow pyrolysis biochars are very stable in soil and contribute more to carbon sequestration. Decomposition of biomass during pyrolysis generally takes place between 200 and 500 °C and the carbon content increases with an increase in temperature. However, elements other than carbon like sulfur, nitrogen, oxygen, and hydrogen diminish at high temperatures (Vithanage et al. 2017).

Hydrothermal carbonization is another method of biochar production that uses low temperature to produce the biochar than pyrolysis. In this method, waste materials are converted into biochar under temperatures ranging from 150 to 375 °C with a residence time of 30 min. The operating conditions and product compositions are shown in Table 4.3. Mostly, hemicellulose and cellulose degrade at temperatures below 250 °C temperature (confirmed through FT-IR and ^{13}C NMR) and lignin

Table 4.3 Various techniques for biochar production and its process conditions (Tahir et al. 2020; Vithanage et al. 2017; Wang et al. 2020)

	Process conditions				Product composition (%)		
Production technique	Temperature (°C)	Oxygen	Heating rate	Residence time	Biochar	Bio-oil	Syngas
Fast pyrolysis	300–1000	Absent	High	Seconds	10	70	20
Slow pyrolysis	Low—350–550 High—600–900	Absent	Low	Seconds to hours	35	30	35
Hydrothermal carbonization	150–375	Presence	Low	Minutes to hours	60	30	10
Torrefaction	200–300	Absent	Low	Minutes to hours	85	10	5
Gasification	700–1200	Present	Moderate/high	Hours	10	5	85

degrades at high temperatures during hydrothermal carbonization (Ornaghi et al. 2020). Agricultural wastes are mostly used in hydrothermal carbonization to produce solid fuel (brown coal) and achieve maximum fuel (16.3 L CH_4/kg FM) (Oliveira et al. 2013). The biochar produced through HTC have high O-containing groups and render efficient cation exchange capacity. Though this method utilizes unconventional wet biomass, the energy consumption is very high as the carbonization depends on the moisture content of the biomass (Vithanage et al. 2017).

Microwave carbonization is also known as low-temperature pyrolysis that has a temperature ranging from 245 to 390 °C to burn waste materials (Selvam and Paramasivan 2021a) is an emerging technology where the carbonization efficiency is increased by combining with microwaves. Torrefaction is a form of slow pyrolysis technique, where the biomass is decomposed at relatively low temperatures (200–300 °C) for a high residence time. Biochar is obtained as a by-product in the first step of torrefaction, where the hemicellulose is decomposed into an unsaturated solid product. Gasification involves the production of high composition of syngas comprising of hydrogen, nitrogen, carbon dioxide, carbon monoxide, etc., at the expense of biochar and bio-oil. This technique is adopted mostly in energy sectors (Vithanage et al. 2017).

4.3 Biochar Properties for Potential Soil Amendment

Biochar possesses a large specific surface area, microporous structure, water, and nutrient retention capacity, high pH, and oxygen-containing surface functional groups that benefit the stabilization of heavy metals. Their properties solely regulate the mechanism and hence should be thoroughly analyzed before identifying the research needs. It is reported in the literature that biochar produced at higher pyrolytic temperatures has shown better adsorption capacities for Cd and Zn (Chatterjee et al. 2020). However, at very high temperatures, there is a loss of oxygenated functional groups which can reduce the cation exchange capacity of the biochar. Though its agronomic value is determined by the nutrient concentration of biomass, temperature >750 °C has a significant undesirable effect on the adsorption capacity of biochar (Domingues et al. 2017).

Ultimate and proximate analyses assist in evaluating the agronomical potential of biochar. The high ash content of biochar can be used to predict the presence of alkaline compounds such as $CaCO_3$ and $KHCO_3$ which can act as liming agents for improving the soil condition (especially acidic soils) and improve nutrient availability, irrespective of its pH. This characteristic has to be considered when correction of soil acidity is attempted. High ash content can be positively correlated with the chemical and nutrient composition of biomass. In addition, such biochars exhibit the high cation exchange capacity necessary for enhancing the nutrient and water retention capacity in soil (Domingues et al. 2017; Pathy et al. 2020a).

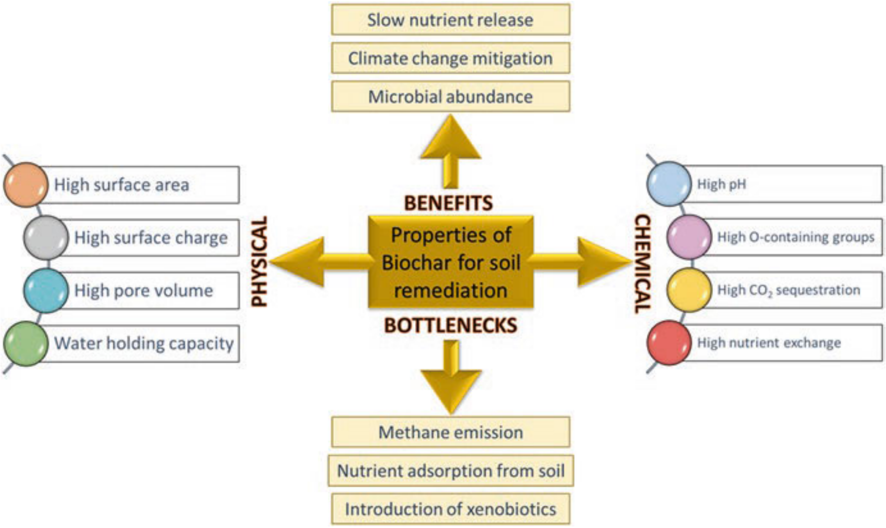

Fig. 4.1 Physical and chemical properties of biochar for potential soil amendment

Biochar produced at higher temperatures gets mineralized at a higher rate when applied to the soil. The carbon fraction of the biochar stimulates the decomposition of organic matter due to the presence of volatile materials. The magnitude of these materials helps in the evaluation of nitrogen cycling and carbon bioavailability in biochar. Conversely, biochar synthesized at low temperatures (300–450 °C) contains high aliphatic character making them susceptible to degradation by microbes in the soil. As a result of this, the duration of metal immobility decreases (Li et al. 2019; Rodriguez et al. 2020). Some of the inherent physical and chemical properties of biochar for efficient heavy metal remediation are depicted in Fig. 4.1.

4.4 Interaction of Biochar and Heavy Metals

Biochar is an organic substance that can uptake heavy metals and reduce their harmful effect in the contaminated soil environment. The functional groups of biochar responsible for this effect depend on the type of feedstock used and influence the surface charge to determine the adsorption of transition and non-transition metals. Biochar can interact with heavy metals and transform them from natural species to stable species in soil. The mechanism of action involves adsorption, ion exchange, redox reactions, volatilization, methylation/demethylation, precipitation, and complexation to abate the mobility of contaminants and ensure the bioavailability of metals. If the pH of biochar is greater than that of the soil, then there are higher chances of metal immobilization. This condition is more suitable for acidic soils where the solubility is comparatively higher (Gao et al. 2020; Yu et al. 2020).

In general, heavy metal remediation by biochar occurs through two different mechanisms: direct mechanism and indirect mechanism. In direct mechanism, biochar immobilizes the heavy metal components through chemisorption, physisorption, electrostatic attraction, complex formation, and precipitation. In indirect mechanisms, the application of biochar contributes to the soil environment by increasing soil pH, microbial biomass, organic carbon, water holding capacity, and nutrient use efficiency (Beesley et al. 2015).

4.4.1 Direct Mechanism

Biochar removes heavy metals from the contaminated soil environment through several types of mechanisms. In physisorption, the heavy metals are absorbed through the large surface area, porosity, and diffusion movements of biochar. Biochar has a large surface area with well-distributed pores such as micropores (<2 nm), mesopores (2–50 nm), and macropores (>50 nm) to adsorb the heavy metals. On the other hand, biochar can also adsorb via chemisorption (electrostatic attraction, ion exchange reaction, complex formation, and precipitation). Biochar has a negative surface charge due to the presence of a functional group. The electronegativity of biochar helps in attracting positive heavy metal ions due to the electrostatic attraction. The surface charge becomes more negative at higher pH and could facilitate more interaction between biochar and heavy metals (He et al. 2019). Biochar can release cations, such as Ca(II) and Mg (II), which got to exchange with the positive metal ions, and in this manner, metal got adsorbed onto biochar surface. The cation exchange capacity (CEC) determines the amount of cation being exchanged (the corresponding amount of HMs will get adsorbed), and hence biochar having a higher CEC value is desirable for maximum heavy metal adsorption. It was observed that biochar derived from animal biomass have a higher amount of Ca and hence they have a higher CEC value as compared to plant biomasses (Lei et al. 2019). Complexation is one of the important mechanisms that form the multiatom structures (complexes) with specific metal–ligand interactions. Biochar has a diverse function group on them which can immobilize the heavy metals by forming metal complexes. Some of the groups that drive the complex formation on biochar surface are –OH, –COOH, –C=O, and –C=N. Moreover, higher content of Fe(II), Mn(II), and carbonate in biochar helps in an increased amount of complex formations. Similarly, inorganic components such as Si, S, and Cl also play an important role in forming complexes with metal ions (Tan et al. 2017). Biochar has mineral elements in them, these elements interact with biochar and got precipitated on the surface. This is another mechanism via which the biochar immobilizes the heavy metals onto them (Xiao et al. 2020).

4.4.2 *Indirect Mechanism*

Instead of interacting directly with the heavy metals biochar can impact the soil's chemical properties which consequently enhance the heavy metal immobilization. When biochar is amended in the soil it increases the soil's pH. This leads to several reactions such as hydrolysis of HM, increased HM complex formation, and oxidation of residual fraction of HMs (Duan et al. 2017). These reactions help in metal immobilization. Similarly, biochar application in the soil increases its CEC, and that consequently enhances its metal adsorption capacity. It was also reported that minerals present in the biochar get transferred to soil, these minerals interact with metal ions and form complexes, and reduce their bioavailability (Rees et al. 2014). Biochar application in the soil also improves soil organic carbon content, this results in complex formation between metals and an oxygen-containing group of biochar. In this way, the heavy metals get converted to a less mobile fraction (organically bound fraction) in the soil and become unavailable for plant uptake (Abdelhafez et al. 2014). The most prominent mechanisms by which biochar removes heavy metals are shown in Fig. 4.2.

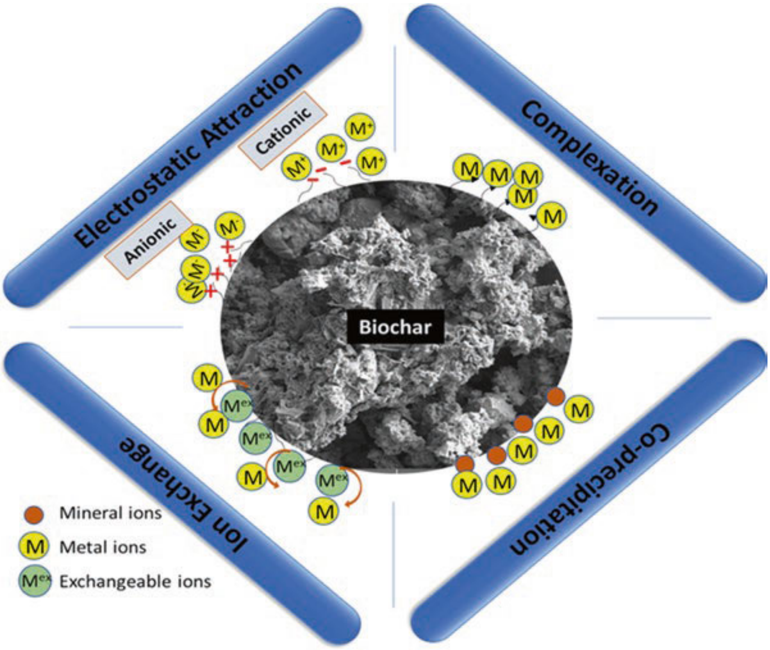

Fig. 4.2 Mechanisms of action behind heavy metal remediation by biochar

4.5 Modifications of Biochar for Enhanced Heavy Metal Removal

Biochar produced from different feedstocks exhibits different physicochemical properties, which also intend to change with respect to operational conditions such as the method of biochar production, temperature, heating rate, and time. The functional groups present on the surface of biochar that are solely responsible for adsorption and remediation of heavy metals from contaminated soils are highly influenced by the temperature at which the biochar is produced. For example, C–O–C, –OH, C=O, and $–CH_2$ were prevalent at moderate temperatures, whereas, only C=O and C=C were preserved at higher temperatures (Wang et al. 2021a). Also, at higher temperatures, the presence of oxygen-containing groups on the surface decreases corresponding to a decrease in adsorption of heavy metals ions (Ambaye et al. 2020). Biochar produced at lower temperatures has lower pH and high cation exchange capacity, making them suitable for soil with high pH and poor fertility (Tahir et al. 2020; Domingues et al. 2017). There are several other factors that determine the adsorption capacity of heavy metals on biochar, leading to decreased efficiency of remediation. In adverse cases, the biochar produced might not be functional enough to remove heavy metals from soil (Wang et al. 2021a). These scenarios call for the development of pretreatment modifications to improve the surface properties of biochar for enhanced remediation.

The modifications rendered on biochar can be physical (choice of feedstock, temperature, time, etc.) or chemical (acid/base treatments, steam, magnetization, impregnation of minerals, etc.). It can also be pre- or post-preparation of biochar. Magnetic biochar produced by Wang et al. (2021) with iron-based modification was found to be effective for simultaneous removal of As, Pb, and Cd. Magnetization was also induced by coprecipitation of Fe^{2+}/Fe^{3+} on orange peel powder prior to pyrolyzing the biomass (Tang et al. 2013). It was also confirmed that sulfur-based modifications can offer long-term and stable remediation of Hg in soil. The sulfur medium can be thiols, sulfur dioxide, or carbon disulfide and the immobilization takes place formation of hydrogen sulfide on the surface. However, this condition holds only in aerobic conditions, other than which the hydrogen sulfide molecules will be assimilated by sulfate-reducing bacteria (O'Connor et al. 2018). Oxidization of biochar with agents like sulfuric acid or hydrochloric acid rendered more carboxylic groups on the surface demonstrating higher entrapment of Cu, Pb, and Zn (Tang et al. 2013). Almost 100% remediation of copper and cadmium was achieved when biochar produced from switchgrass by HTC was treated with alkali. In advanced cases, a combined modification strategy was applied to enhance the surface area of biochar, followed by heavy metal immobilization. Treatment with sodium hydroxide has been reported to improve the surface area of produced biochar, after which the latter was modified with hematite for incorporating good adsorption capacity of heavy metals (Pan et al. 2021).

4.6 Applications of Biochar

Biochar is one of the best materials that are derived from woody biomass and crops residues for the removal of pollutants from the contaminated environment (Cheng et al. 2020). They have some unique features including large specific surface area, porous structure, and surface functional groups to remove the pollutants from the contaminated environment (Swagathnath et al. 2019a). Scientific research literatures on biochar focused on the removal of pollutants from the contaminated environment such as heavy metal removal (46%), removal of organic pollutants (39%), removal of nitrogen and phosphorous (13%), and other pollutants (2%) (Tan et al. 2015). The biochar production was mainly used to improve soil properties, crop production, and remediation enhancement of polluted environment (including remediation of heavy metals, remediation of organic pollutants, pesticide removal from soil environment, and remediation of other pollutants) (Chen et al. 2020b; Lebrun et al. 2021). The biochars have been widely used in various fields of soil environment including soil physical health alternation, soil acidity management, crop yield and production enhancement, soil micronutrients mineralization, soil quality and fertility restoration, nutrient retention and sorption, sequester soil carbon, soil chemical properties modifications, influence plant physiological parameters, and increased water availability. It can enhance the properties of the soil including soil microbial biomass carbon, phosphorus, nitrogen, carbon mineralization, and various enzymatic activities (Das et al. 2020). The biochar technology has been used in wastewater management to remove the pollutants from water due to its benefits including cost-effectiveness, high specific surface area, and surface reactive groups (Wei et al. 2018).

4.7 Advantages and Disadvantages of Biochar

Biochar production has a lot of advantages including recovery of components, water, and energy and efficiency enhancement strategies for resource recovery and removal of pollutants from the contaminated environment (Ye et al. 2020). Biochar production is a carbon-negative process that has additional benefits including nutrient retention, high stability against decay, the high adsorption capacity of carbon-oxygen complexes, and high capacity to adsorb cation per unit of carbon. At the same time, some of the issues were found in biochar technology including limiting nutrients and amount of C in soil, soil acidity, microbial activity, and low NPK compared to commercial fertilizers. The biochar production from wastes was beneficial for a few fields including clean water and sanitation, industry innovation and infrastructure, responsible consumption and production, climate action, and land on life. Biochars are also effective in removing organic contaminants (phenol, pesticides, and dyes) and inorganic contaminants (As, Pb, and Cd) from contaminated environments.

4.8 Toxic Compounds Present in Biochar

However, there exist certain risks associated with biochar amendments in the soil. For instance, the thermal degradation of biomass could lead to the generation of harmful compounds such as perfluorochemicals, polycyclic aromatic hydrocarbons (PAHs), polychlorinated dibenzo-*p*-dioxins and furans (PCDD/F), and volatile organic compounds (VOCs) (Godlewska et al. 2021). Although the presence of these chemicals in biochar is not common and primarily depends on the feedstock selections and pyrolytic conditions, the potentially harmful effect cannot be avoided completely. When the biochar is amended into the soil, these compounds can leach into the soil from biochar, subsequently entering into the food chain. The harmful effect on humans includes disruption of the endocrine system, which results in oxidative stress, apoptosis, and many of them are carcinogenic in nature (Weidemann et al. 2018). Hence, it becomes highly crucial to check the presence of these harmful chemical compounds in biochar before applying them in the field. Hence, it becomes highly crucial to understand the reason behind the toxicity of the biochar to choose biochar that will not contaminate the land on its application.

For PAHs, both the choice of biomass and the experimental conditions determine its concentration in biochar. For instance gasification results in more PAHs than any other thermos chemical technique. Moreover, in pyrolysis itself, a faster heating rate (Fast pyrolysis) produces more PAHs than that of a slower heating rate (slow pyrolysis). It was also observed that when N2 is used as a carrier gas instead of CO_2, it results in a higher amount of PAHs in the biochar. Biochar having a lower surface area could have higher bioavailable PAHs, as it will be limited adsorption sites that bind with the generated PAHs (Visioli et al. 2016). Similarly, it has been also observed that biochar produced from green garden waste contains more PAHs as compared to other biomasses. PCDD/F in biochar presents in an insignificant quantity; however, their presence becomes significant in the biomasses having higher chlorine content (i.e., food wastes). Hydrochar was observed to have higher PCDD/F content as compared to biochar because the former was produced at a lower temperature, which was not sufficient to degrade the generated PCDD/F (Hilber et al. 2017). On a similar note to reduce the amount of VOCs biomass needs to be highly carbonized and a slightly aerobic condition will reduce the concentration of VOCs in the biochar. And lastly, the presence of certain heavy metals (such as Pb, Cd, Hg) could also posses a challenge. It was reported that biochar produced from sewage sludge has a higher heavy metal concentration. Moreover, biomass collected from heavy metal contaminated sites could also result in a higher concentration of heavy metal in the biochar (Godlewska et al. 2021). It is also important to note that most of the toxic pollutants present in the biochar are not immediately bioavailable; however, a cautious approach needs to be adopted to evaluate biochar before applying it in the field.

4.9 Conclusion and Future Prospects

The use of biochar as a soil amendment is emerging considering its beneficial characteristics like surface area, pore volume, high pH, nutrient, and water retention capacities. This chapter provides an overview of the production methodologies, properties, and mechanisms involved in the adsorption of heavy metals. Soil remediation with biomass is a fast-growing field with the introduction of novel techniques to improve removal efficiency in a sustainable way. Some of the further perspectives of this research are given below:

- Co-pyrolyzed biochar from biomass and orthophosphate has been shown to be very effective against the adsorption of heavy metals such as Pb, Cu, and Cd. The biochar was able to complex, immobilize, and precipitate the metals due to the presence of phosphate and hydroxyl groups on their surface. Further research is needed to evaluate the role of surface carboxyl groups on the enhanced complexation of heavy metals.
- Biochar is being used as microbial immobilized carriers for abating PAHs using immobilized microorganism technology (IMT). This technology can couple bioremediation and bioaugmentation for complexing and degrading such high molecular weight compounds. Hence, attention should be paid to this emerging technology.
- Once biochar is applied to soil, they interact with heavy metals, organic and inorganic compounds in soil and hence it is important to understand the mechanism and changes in biochar properties with time. This would enable the utilization of biochar in crop productivity for a prolonged duration. The properties of biochar play a chief role in defining the environmental effect and delineating the direction for future use.
- Based on the selection of biomass and the experimental conditions biochar's properties can vary widely, and hence it becomes crucial in determining biochar's interaction with heavy metals. For instance, pH, CEC, and the presence of diverse functional groups affect the mechanism and potential of biochar in immobilizing the heavy metals from soil.
- Biochar's interaction with heavy metals can be broadly classified into two mechanisms; direct and indirect. In direct mechanism, biochar immobilizes the pollutants via electrostatic force, ion exchange reactions, complex formation, and precipitations. Whereas, in the case of indirect mechanism the biochar impacts the soil properties such as pH, soil CEC, mineralization of heavy metals, and soil organic carbon content thus affecting biochar's heavy metals uptake potential.
- To achieve the desired physicochemical characteristics necessary to remove heavy metals, biochar is being modified/engineered through various methods. Biochar can be modified chemically by treating it with various chemicals (based upon the requirement). A large number of current research is being focused on developing novel treatment methods for improving biochar metal removing properties. However, in certain cases, the modification techniques become either

chemical-intensive or economically costlier. Hence, future studies should address this challenge while developing various modification techniques.

- Most of the studies have been carried out on the laboratory scale and a limited number of studies have been undertaken in the field. Hence, more studies need to be done at the field level to understand the difficulties faced in scaling up the process. Moreover, attention should be given to carrying out the techno-economic assessments for the overall process, and thus it could be acceptable at an industrial level.

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Chapter 5
Vermitechnology: An Eco-Friendly Approach for Organic Solid Waste Management and Soil Fertility Improvement—A Review

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Abstract The variation in agricultural patterns and climate change makes the farming community refine the conventional procedures to maintain soil fertility and crop productivity. The sustainable method of farming is a current focus for long-term productivity to supplement nutrition for the growing population. The utilization of traditional agricultural practices helps to maintain soil quality that in turn supports productivity. The organic waste generated from various anthropogenic activities can be utilized to produce manure. The vermicomposting uses the earthworm and converts the voluminous waste into manure. During the process, bioremediation also occurs due to the activity of earthworms that help to reduce any toxicity present in the wastes. During this bioconversion, earthworms, along with microorganisms in different stages of composting, help to convert the wastes. The enzymes secreted by the microbes during the degradation of organic materials help to transform various organic and inorganic components present in them. The agricultural products obtained from organic farming can be more beneficial. This chapter discusses the possibilities of utilizing different organic wastes for manure production using earthworm and their applicability in organic farming and environmental pollution mitigation. The organic manure thus produced using earthworms can be applied for reclamation of salt-affected soil and soil that have reduced organic matter in place of chemical fertilizers.

Keywords Crop improvement · Solid organic waste · Waste management · Soil fertility · Vermiremediation

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5.1 Introduction

Globally, agrochemicals have impacted the health of farmers and the adverse effect on the population that consumes chemically grown food is now more evident. Organic waste positively influences soil properties and enhances agricultural productivity. In developing countries across the globe, there is a search for a cost-effective and sustainable alternative to chemical fertilizers. Small cultivation groups opt for compost production as a technique for maintaining soil fertility and crop yields. In India, composting is a significantly older technique, and farmers mainly favor the conventional technique of pit composting method. Earlier literature shows that the inclusion of microorganisms in compost enhances the plant nutrient concentration (Awasthi et al. 2015, 2016; Kumari et al. 2016). In India, annually, approximately 320 million tonnes of cultivated waste are produced. The decomposition process releases a higher amount of greenhouse gases when it is dumped with other municipal solid wastes. These productions are primarily because of a result of the landfill and further life cycle actions. After reaching landfills, the organic wastes from the vegetable market cause much nuisance because of their high biodegradability nature. Many reports suggest that the burning of these wastes generally produces many poisoning gases and noxious substances that remain in the land (Manisalidis et al. 2020).

An enormous quantity of solid and liquid waste material is produced from agricultural activities, food processing units, pulp industries, paper industries, or cellulose-based industries. It is a severe problem for the whole world to dispose of and manage this generated industrial waste through an environment-friendly method. Because of this in recent years, focus on changing these nutritious organic wastes into valuable products for justifiable land-use applications through low-inputted efficient technologies. Vermicomposting is defined as the fragmentation of multiplex organic waste into an odorous humus-like material by earthworm activities. In other words, vermicomposting is the equalization of organic waste by the coaction of microbes and earthworms. In the whole process of vermicomposting, the microbes are mainly answerable for the biochemical decomposition of organic waste, while earthworms are the leading operator of the whole operation, acclimatizing the materials and altering the activities of microorganisms (Pathma and Sakthivel 2012).

The efficiency of organic waste degradation by a composting process involving various kinds of microbes mainly depends on the substrate-based hydrolytic enzymes secreted by these microorganisms to hasten the degradation procedure. These enzymes can be extracellular enzymes or intracellular enzymes. Intracellular enzymes catalyze biochemical reactions within the cellular system, and extracellular enzymes are liberated into the external composting system and decompose complex organic materials into simpler units. Released extracellular enzymes convert the structure of the polymer into monomers. Intracellular enzymes act on soluble components in water, and after release, dissolve in water and are secreted inside the microbial cell (Gajalakshmi and Abbasi 2008). The importance of organic farming is that the food produced through this method for society is free of chemicals and naturally protective. The vegetables and fruits produced through organic farming are highly nutritious and contain antioxidants in contrast to fruit and vegetables grown chemically. Mie et al. (2017) reported that organically grown foods contain a huge considerable amount of “organic acids” and “poly-phenolic compounds” with a potential to improve health by acting as an antioxidant.

Vermicomposting technology has become the most popular solid waste management technique throughout the world. Vermicomposting is defined as “the biotransformation of organic wastes into a biofertilizer through the activities of earthworm” (Kaur 2020; Pandit et al. 2020). In the process, the earthworms feed on the organic wastes and the earthworm’s gut performs as a bioreactor, where vermicast is produced (Maharjan and Regmi 2020). When earthworms excrete the organic wastes, it is rich in nitrogen, phosphorus, and potassium content, additionally, rich in many trace elements based on the type of feedstock used. The vermicomposting process is mesophilic and operating conditions like temperature, pH, electrical conductivity, and moisture content levels must be optimized. Generally, vermicomposting happens in vermi-reactors, including plastic, mud pots, and worm bins of wood (Manyuchi et al. 2013). Besides improving soil fertility and crop production, earthworms are also responsible for the removal of contaminants from the soil and the process is called vermiremediation. The previous studies of vermiremediation focused mainly on organic wastes, with comparatively less attention paid to inorganic contaminants (Dada et al. 2021).

Solid organic wastes generated by various activities, including agriculture, industrial, and domestic, can be converted to nutrient-rich organic manure by vermitechnology. This helps reduce the accumulation of wastes at landfills that generate various gases and leachate that pollute groundwater. Solid wastes pollute soil, water, and air while dumped in any place. This article documented research and review articles explaining the possibilities of utilizing various organic wastes for manure production using earthworm and their applicability in organic farming and environmental pollution mitigation. This could be a measure to reduce pollution and produce an eco-friendly manure for improving soil fertility and crop production.

5.2 Organic Waste Generation

More than a quarter of the urban solid waste in India comes from fruits, vegetables, and animals (Dey 2018) and the organic fraction of the municipal solid waste markup 40–85% of the total garbage produced. Because of its high decomposable nature, the vegetable waste can easily and effectively be composted for their reutilization as fertilizers and conditioners of soil. Compared to landfilling and incineration, the composting of plant waste may cut down the natural effect of changing climate. When composting, the organic material is biologically decomposed through a large group of microbes providing a final product full of alleviated carbon, nitrogen, and other nutrients. There is about 500 million MT of biomass available in

India, waste to the energy sector may project to a market size of 14 billion ~USD by 2025 with an annual increase of around 7% (Bhatia et al. 2020). It is further projected that approximately 686 metric tonnes (MT) of gross agro biomass residues per annum are produced through 26 separate crops from which 234 MT (34%) are additional agro residues. Composting technology generated a huge amount of farmed biomass residues. Composting technology generated a huge amount of farmed biomass residues, which are valued suppliers of carbon that can be used to improve soil organic carbon. Production and treatment of compost have been having huge attention in India to limit the improved utilization of chemical fertilizers that are mainly responsible for the deterioration of soil health, release of greenhouse gases because compost is a good source of humus and nutrients which is required for rising the soil fertility (Gabhane et al. 2012). To restore soil productivity and proper management of organic wastes, the agricultural application of organic waste manure generated by vermicomposting is one of the main possible approaches (Bhat et al. 2018a).

5.3 Vermicomposting Process

The vermiculture technology is almost 50 years old as it began in Holand, during 1970 when the first serious experiment on vermiculture was conducted and simultaneously in Canada and England also. The word “vermi” in the word “vermicomposting” is taken from the Latin word “vermis” which means a worm. However, vermicomposting is defined as a composting process performed by the epigeic, endogeic, and anecic species of earthworms that have a natural capability to colonize and decompose organic waste materials (Bhat et al. 2018a). Earthworms are coming under the class Oligochaeta and phylum Annelida having a dark brown color of body and clitellum for cocoon production and are hermaphrodites in nature with bilaterally symmetrical, segmented invertebrates. They crush the complex organic materials into simpler or smaller sizes with the help of a gizzard. There are many microorganisms (bacteria fungi, actinomycetes, and protozoa) that are the reason for the decomposition of organic matter are live symbiotically in the gut of earthworms (Medina-Sauza et al. 2019).

Processing of vermicomposting involves collecting degradable wastes and subjecting them to predigestion. The variation in temperature during predigestion changes indicated the activity occurring in the decomposition process. There were changes in pH and moisture content also during these periods (Arshad and Hiranmai 2018). There was considerable variation in observed parameters during the predigestion period which makes the larger particles smaller than earthworms can easily ingest. The vermicomposting is carried out in controlled conditions with maintained temperature and moisture. Once earthworms are introduced, they convert the waste by consumption and ejection as vermicast. The vermicast contains various microbes and the product is rich in macro and micronutrients, microbial population, growth hormones for plants, and enzymes.

Through the decomposition process microorganisms convert complex organic matter into simpler forms of organic matter. The microbial community is affected by the pH and raw material composition, as suggested by the functional profiling of microbiota in the cast of *Eisenia fetida* during the vermicomposting process (Budroni et al. 2020). The low molecular weight organic acids like citric acid, formic acid, oxalic acid, and acetic acids are produced at the time of the vermicomposting process, especially in the cast-associated processes (Busato et al. 2012).

In vermicomposting, the transformation of organic matter to simpler forms of carbon and nitrogen by microorganisms and their enzymes takes place. The augmented environmental conditions like temperature, moisture content, light, and season are essential for the better survival of microbes and it varies according to the composting practices. Several articles on the isolation and microbial diversity in compost (Varma and Kalamdhad 2014). In particular, the vermicompost is rich in a variety of microbes, as earthworms are propagated by useful microorganisms based on organic substances present in the soil (Jack 2010). At the time of composting process, almost 50% of included organic material has been fully mineralized because of the conversion of simply decomposable materials like proteins, cellulose, and hemicelluloses by microbes. Upon completion, the last residual composting organic material consisted of type humic substances that are not biodegradable and the more stable mature compost fraction. In the composting process of organic matter, decomposition is accomplished with many types of microbes, such as mesophilic bacteria, spore-producing bacteria, fungi, actinomycetes, and converting them into steady humic components (Bhatia et al. 2012). However, it is believed that during composting, the nature of degradation and humification changes are based on the raw organic materials utilized for composting process.

For sustainable soil management, the application of compost in agricultural fields as a soil conditioner is one of the most important methods to recycle organic waste. Decomposition of biological materials usually causes complete or partial mineralization of organic compounds to form carbon dioxide, water, ammonia or nitrate, sulfate, and carbonates of calcium, magnesium, and potassium, oxides and phosphates of iron and manganese. Few of these compounds are lost from mineralized composting biomass in the form of gaseous compounds, such as CO_2, H_2O, and NH_3, and some are dissolved with wastewater and some remain in the form of precipitated or adsorbed compounds in the last product compost (Ayilara et al. 2020). The formation of fulvic and humic substances is a small metabolic side pathway of the entire composting process. Even under highly oxidizing conditions, it leads to the decomposition of biological masses. They are intended for mummification of decaying organic tissues or for strong deposition in the form of humates on the surface of clay particles (flour formation). These are comparatively stable or even nonreactive by-products in both cases and create the dark blackish-grey color of all compounds.

Through the complex mechanical and biochemical interactions with oils biotic and abiotic components, earthworms promote plant growth and productivity. Due to the burrowing nature, earthworms ingest soil and resulting in the mechanical breaking of soil particles and increased surface areas for biotic actions. Burrows of earthworms act as a pathway for water movement, particle movement, nutrient flow, and aeration. There are millions of enzymes and microbes present in the earthworm's gut, which helps in rapid bioconversion and mineralization of soil organic matter thus increasing soils' nutrient status. These all processes along with other factors helps in increasing plant growth and crop yield (Sinha et al. 2008).

5.4 Potential of Different Earthworms for Waste Management

The name earthworm is also known as "farmers friend" or "nature's plowman." Worldwide more than 4000 species of earthworms are reported and in India, about 420 valid species/subspecies of earthworms are documented which belongs to about 70 known genera. Out of the total worldwide earthworm diversity, India alone contributes about 10.5% (Kale and Karmegam 2010). In the production of vermicompost through the decomposition of organic materials, it is much important to select an appropriate species of earthworms. The selected species of earthworms for the decomposition of organic matter should have the adaptability to waste, a fast growth rate, high reproductive potential, and minimal gut transit time. Generally, a few earthworm species have been successfully cultured for scientific or commercial purposes. Vermiculture has often concentrated on culturing a restricted number of litter-dwelling species (including *Eisenia fetida* (Savigny), *Dendrodrilus rubidus* (Savigny), *Dendrobaena veneta* (Rosa), *Lumbricus rubellus* (Hoffmeister), *Eudrilus eugeniae* (Kinberg), *Perionyx excavatus* (Perrier), and *Pheretima elongata* (Perrier) that have commercial applications like processing organic residues into a potentially saleable product). Litter-dwelling species are ideally suited to large-scale breeding programs due to their high growth, maturation, and reproductive rates. Based on their properties and nature, earthworms are classified under three main types as shown in Table 5.1.

The species used for organic waste decomposition globally is *Eisenia foetida* while in tropical and sub-tropical countries, the popular species of earthworms are *Eudrilus eugeniae*. At many times the mixed culture of different species of earthworms is also used for the degradation of organic matter. *Eisenia fetida* came into the subcontinents of India from the European subcontinents. As compared to the other species of earthworms like *Eudrilus eugeniae* and *Perionyx excavates*, the reproduction rate of *Eisenia fedida* is very high. It is well known that earthworms are producing vermicompost during the vermicomposting process by the intake of complex organic materials and secreting simpler forms of organic materials (Van Groenigen et al. 2019). The *Eisenia fetida* has the magnificent capability to transform organic materials such as garden waste, food waste, agricultural wastes, and municipal solid organic wastes into valuable compost (Yuvaraj et al. 2020). It was reported that due to the metal stress the skin tissues of earthworms releases an extracellular polymeric substance that binds heavy metals on their skin and reducing

Table 5.1 Classification of potential earthworms based on their properties

S. no.	Properties	Epegeic species	Endogeic species	Anecic species
1.	Habitat	Surface dwellers, 3–10 cm	Upper layer soil, 10–30 cm	Deep burrowing, 30–90 cm
2.	Color	Coloration of the body is uniform	Weak pigmentation	Pigmentation only at the anterior and posterior end
3.	Body size	Small in size	Moderate in size	Large in size
4.	Life cycle	Short	Medium	Long
5.	Live in	Near the surface litter or dung	Below the surface	Deep soil
6.	Temperature tolerance	Wide range of temperature tolerant	Poor in temperature tolerance	Poor in temperature tolerance
7.	Feeding habitat	Undecomposed litter, plant litter, or mammalian dung	Subsurface soil materials, organic-rich soil	Surface litter, decomposed litter
8.	Reproduction rate	High	Low	Moderate
9.	Main role	Good for vermicomposting because of efficient biodegrading nature	Aeration process and soil mixing	Decomposition and distribution of organic matter in the soil
10.	Vermicomposting potential	Good	Poor	Poor
11.	Examples	*Eisenia fetida*, *E. andrei*, *Eudrilus eugenie*, *Lumbricus rubellus*, *L. festivus, L. castaneus*, *Bimastus minusculus*, *B. eiseni*, *Dendrodrilus rubidus*, *Drawida modesta*, *Dendrobaena veneta*, *Perionyx excavatus*	*Octochaetona thurstoni*, *Aporrectodea caligenosa*, *Allolobophora rosea*, *A. caliginosa*, *Metaphire posthuma*, *Pontoscolex corethrurus*, *Drawida barwelli, Amynthas species*	*Lumbricus terrestris*, *L. polyphemus*, *Lampito mauritii*, *Aporrectodea trapezoids, A. longakc*

their mobility and removing them from the substrate (Khan et al. 2019). These extracellular polymeric substances also play a role as an enzyme for the bacterial decomposition of waste material and improve their population. In addition, it was also reported that such extracellular polymeric substances are also responsible for the improvement of compost quality and improvement of the nutritional value of compost as it consists of protein and carbohydrates as its primary substrates (Guhra et al. 2020).

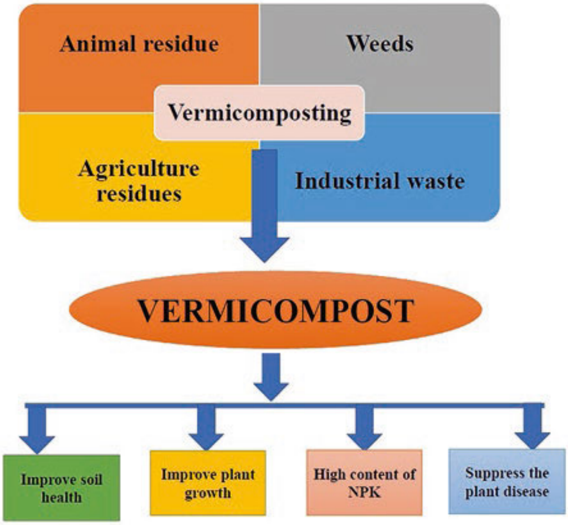
Earthworms are mainly responsible for the enhancement of composting process directly because they breakdown the complex organic materials by utilizing their gizzards mechanically; because of this, the substrate surface area is increased and consequently altering the microbial activities, into the process that is collectively known as gut-associated process (Gómez-Brandón et al. 2012). Consequently, besides the mechanical action of the earthworms, the microbes which are present in the gut of earthworms and the compost are responsible for the production of many bio-compounds, such as enzymes and organic acids, which are the main reason for the acceleration of the biodegradation and nutrient mineralization within vermicompost (Medina-Sauza et al. 2019).

5.5 Vermicompost for Soil Quality Improvement and Plant Growth

Vermicomposting mainly involves the bio decomposition of organic wastes like agricultural waste, industrial waste, animal residues, weeds, etc., which results in the production of a nutrient-rich product called vermicompost, which is very helpful in enhancing soil health and fertility, increasing the nutrient content of soil and promotes plant growth, and suppresses plant diseases (Fig. 5.1). Arshad and Hiranmai (2017) showed that the manure prepared from the vermicomposting technology was rich in nutrient contents suitable for maintaining soil fertility and the growth of plants.

Vermicomposting is very efficient as compared to any other composting method. The most important measure to enhance soil fertility and increase crop yields is the constant use of fertilizers and improvers. Thus, innovative organic fertilizer represented bioorganic perspective, allowing alternatively carry out as an option of conventional fertilizers, additionally a few numbers of pollutants of different nature in their structure. The natural biological process of nitrogen fixation within the soil is

Fig. 5.1 Vermicompost for soil fertility and plant growth



suppressed by excessive use of nitrogen fertilizer (60 kg active ingredient per hectare) which causes the accumulation of nitrate and nitrite in the plant (Anas et al. 2020).

Organic fertilizers, such as manure or decomposed form, such as compost, perform a very important part in maintaining and improving soil fertility (Yadav et al. 2011; Amossé et al. 2013). This type of fertilizer is good for the nutrition of plants, soil properties, and acts as an important reserve for the replacement of soil organic matter and increases crop yields. In soil, compost or compost forms a valuable agronomic structure enhancing its capability to retain water and air. Due to the presence on their surface of ions of soluble salts, they provide a balance with the ion exchange solution and the soil conditions affect plant nutrition (Tejada and Benítez 2011).

The worms act on waste and their excretory stuff like vermicast induces excellent development of plants. The growth parameters such as seed germination improvement, seedling growth rate enhancement, flower and fruit of main crops like paddy, wheat, sugarcane, corn, potato, tomato, okra, brinjal, grape, spinach, and strawberry, including flowering plants such as marigolds, petunias, chrysanthemums, sunflowers, and poinsettias (Singh et al. 2020). In the vermicomposting process of waste materials, the activities of the earthworms are physical as well as biochemical. Substrate aeration, mixing, and actual grinding are the main physical activities of the earthworms, while the biochemical activities are influenced by the substrate microbial decomposition within the intestine of earthworms which results in the formation of chelating and phytohormonal elements by the vermicomposting of organic matter which has a large content of microbial matter and stabilized humic substances. The chemical analysis of prepared vermicompost exhibits potassium (K), available phosphorous (P), Available nitrogen (N), and available magnesium (Mg) as compared to surrounding soils (Mahmud et al. 2018). Papafilippaki et al. (2015) also documented that mature compost prepared from municipal solid organic waste increased the bioavailability of trace elements such as Fe, Mn, Zn, Cu, Pb, Ni, Cr, and Cd in soils and invigorated spiny chicory yield.

Due to its ability to improve the properties of soil and the activity of microbes that are linked to the fertility of the soil, municipal solid waste (MSW) is utilized primarily as a nitrogen and organic matter source. Organic waste like biological waste and food waste modifies pH, nitrogen content, water-holding capacity, the soil cation exchange capacity, and microbial biomass of soil. The sewage sludge contains a huge quantity of organic substances and many nutrients for the plants. To enhance the quality of the soil by the addition of nutrients and increasing microbial and enzymatic activity in the soil is a very common practice. According to Hossain et al. (2017), organic residues have a large positive effect on the soil's physicochemical and biological properties, additionally, stimulate plant growth and crop yields. The use of compost and vermicompost increases soil organic matter, that is, the carbon in the soil, to a more sustainable level. Adding compost to clay soils improves the structure, quality, and fertility of the soil. Soil organic matter acts as a "glue" as it binds soil particles into aggregates and thus improving the structure of soil, infiltration, air and water porosity, and nutrient retention. Soil erosion and compression increase when there is less organic matter in the soil. Since carbon is constantly removed from the soil on farms because of harvesting grain, hay, and cutting of stubble, fed to cattle, as well as by the oxidation of a greenhouse gas named “carbon dioxide.” The carbon content of soil on farms is not replaced naturally. The application of compost reloads the organic matter of soil and adds the lost soil carbon and helps maintain the quality and fertility of the soil and improves production time (Sinha et al. 2011). As soil organic matter degrades over time, it produces a more stable carbon compound called humus. Humus increases cleavage minerals as well as improves the availability of nutrients for the plants. Stable, highly mature compost such as vermicompost contains a long-term carbon form known as humic and fulvic acids or humates, which are much essential for soil fertility and health (Compost Australia 2011). Porosity, aeration, drainage, and water-holding capacity of vermicompost are very high. To ensure retention of nutrients and strong absorption, vermicompost has a huge surface area. They can retain nutrients longer. There are many studies that have shown that the soil treated with vermicompost has a significantly higher bulk density of the sun, becomes porous and light, and never compacted. In addition, the constant application of vermicompost over the years has led to a complete enhancement in the soil quality and agricultural land, even degraded and sodic soils, since vermicompost functioned as soil conditioners (Nelson and Rangarajan 2010).

Organic substances present in the soil, are also a good source of food for beneficial soil microorganisms and help improve the population and the diversity of microbes. The transformation, release, and cycling of nutrients and essential elements take place by these microorganisms. Microbes are also responsible for the transformation of a variety of nutrients in their available form for the plants, as well as for facilitating the absorption of nutrients by plants. These soil microorganisms also generate a “glue” because of which different soil particles stick together and form soil crumbs and pore space that decrease soil hardness and make good soil structure (Jacoby et al. 2017). It is well known that in the processes like decomposition of organic materials, formation of soil organic matters, and nutrient cycling, the microbes play a significant role and these processes decide the quality and fertility of the soil. That is why the utilization of organic fertilizers is suggested management practice as they stimulate growth and microbial activity, which leads to a physically and chemically favorable soil environment for plant growth. Microbes perform these processes with the help of extracellular enzymes which they secret from the body. These extracellular enzymes could remain in the soil for a longer duration and become more and more with the continuous use of organic fertilizers (Song et al. 2017). They have beneficial microbes that produce extracellular enzymes for the release of nutrients associated with organic compounds. Since the organic fertilizer has compounds that act as a substrate for these enzymes, they also stimulate local microbes to complete these processes. Thus, the activity assay may be used to assess the influence of organic additives on the microbial soil condition. On the other side, the enzymatic action of the soil is used as soil quality indicators because of their sensitivity to any changes that may happen in the soil. The influence of various factors, including organic additives on the activity of soil enzymes, has been broadly studied by many scientists over the past four decades. It is well known that each enzyme is specific to the substrate, so a separate measurement of the enzyme activity in the soil is insufficient for this reason. The activities of various enzymes like dehydrogenase, urease, β-glucosidase, and alkaline phosphatase are analyzed in much research (Adetunji et al. 2017).

5.6 Vermitechnology in Environmental Pollution Mitigation

At present almost all the scientists of the world are searching for a technology which should be "cost-effective," "environment friendly," and "socially acceptable." Vermiculture technology, which depends on the utilization of earthworms, fulfills all these requirements. Earthworms are performing their role as "environment engineers" for more than 600 million years. Throughout the world vermiculture scientists understand the earthworm's importance as "waste and soil engineers" and "plant growth promoters" from ancient times. Besides this there are a few new discoveries of its application in the "treatment of wastewater" and "remediation of polluted soil" and the presence of a number of important "biologically active compounds" for the manufacturing of "lifesaving modern medicines" and "raw materials for the few consumer industries" are a revolution in the studies of vermiculture technology (Sinha et al. 2009).

For the appropriate management of most of the organic wastes, treatment of wastewater, cleaning of chemically contaminated soils, improving soil quality and fertility, and the production of food crops, vermiculture is the most sustainable technology (Fig. 5.2). Utilization of earthworms in the manufacturing of "life-saving modern medicines" and "raw materials for many industries" are some "new discoveries." It is successfully studied in vermicomposting of "municipal solid waste," vermifiltration of "industrial and municipal wastewater," vermiremediation of "chemically contaminated soils," and production of "cereals and vegetable crops" with outstanding results. Wastes are decomposed by more than 75% faster,

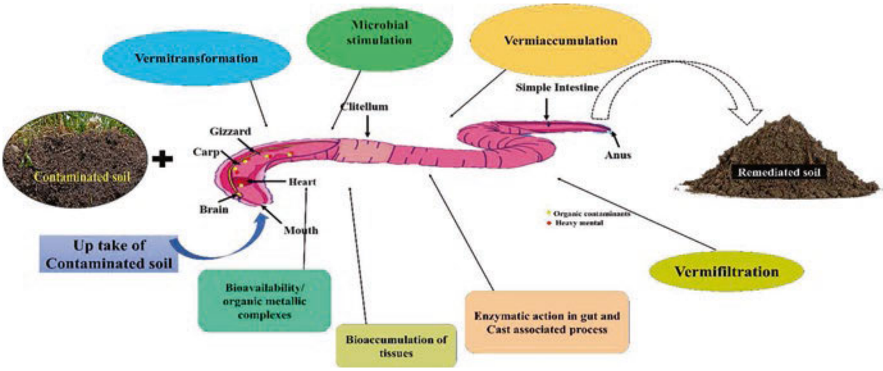


Fig. 5.2 Vermitechnology for remediation of organic and inorganic contaminants

biological oxygen demand and total dissolved, suspended solids of wastewater is decreased by more than 95%, and growth of crop plants are improved by 30–40% higher as compared to chemical fertilizers, by worms and vermicast application (Sinha et al. 2010a).

It is suggested that earthworms also have the potential to solve many environmental problems. The science of utilizing earthworms to improve food production and solve environmental and other human problems is called vermitechnology (Dada et al. 2015). Vermiremediation is a process that describes the process through which earthworms clean up soil contaminants and it is the least attention received important aspect of vermitechnology. Earthworms' biotic and abiotic interactions, life cycle, burrowing and feeding behavior of earthworms to transform, degrade, or remove contaminants from the soil environment are the key points of vermiremediation technology (Shi et al. 2020).

Moreno et al. (2020) utilized a biobed mixture using a combination of soil, peat, and straw and another with soil, vermicompost of wet olive cake, and olive tree pruning for remediating pesticides. Despite the limited available information about vermiremediation, for the removal of organic pollutants vermicomposting has proven to be an effective process, even in some complex substrates like sewage sludge (Rorat et al. 2017) or heavy metals in coal fly ashes (Usmani et al. 2017). Besides, the utilization of vermicompost as soil amendment decreases the availability and mobility of potentially toxic metals (Liu et al. 2019a) and pesticides (Fernández-Bayo et al. 2015), which favors their removal from the soil (Castillo-Diaz et al. 2016).

The inoculation of polluted soils with different earthworm species, including *Eisenia fetida,* directly or in combination with organic matter, enhanced the degradation and removal of a vast range of pollutants from the substrate (Shi et al. 2020). There are some limitations of vermiremediation also because of a high concentration of contaminants and residual fractions of very toxic compounds in soil and organic wastes may negatively affect the survival of used earthworms (Rodriguez-Campos et al. 2014).

Environmental functions, such as soil remediation and management of lands, a revolution is started in vermiculture technology (earthworm's rearing) also known as "unheralded soldiers of mankind" as per Charles Darwin which are working day and night within the soil surface. Earthworms can be utilized for recovery and convalescence of suboptimal soils like soils having poor minerals, open cast mining sites, closed landfill sites and cutover peats, polder soils, and used for land recovery. A sphere influenced by the earthworms is known as "drilosphere system" within the soil environment. This includes the burrow system, surface and below ground earthworm casts, the earthworm surface in contact with the soil, internal earthworm gut and process, and linked biological, chemical, and physical interactions, in addition to the soil microbes. Earthworms are also helpful in the removal of heavy metals, pesticides, and lipophilic organic micropollutants such as polycyclic aromatic hydrocarbons (PAHs) from the soil. Throughout the world, for cleaning up the chemically polluted/contaminated sites/lands, vermiremediation is practiced, in which earthworm species that are chemical tolerant are used as an emerging, cost-effective, and convenient technology (Sinha et al. 2008).

It is predicted that bioremediation assisted by the earthworm (vermiremediation) approaches might include

1. Direct utilization of earthworms to the polluted soils (Schaefer and Filser 2007).
2. Utilization of earthworms with the other organic materials like compost as a coapplication to the contaminated sites/soils (Ceccanti et al. 2006).
3. Utilization of polluted media (soils) to the earthworms as a portion of feeding material (Getliff et al. 2002).
4. Indirect application of earthworms by its digested (composted) matters (vermicompost).

The vermicast of the worm is high in catabolic activities because it is high in degrader microorganisms. It was reported that in vermicast the bacterial counts/gram of vermicast is about 32 million, which is far better than bacterial counts/gram of surrounding soils which is about 6–9 million/g of soil. Vermicomposting is simple in construction, operation, and maintenance. It is a technology that is promoted itself, regulated itself, improved itself, and enhanced itself, and has low or no energy-requiring zero-waste technology. Because of this, it utilizes organic materials that otherwise cannot be utilized by any other processes, vermiremediation is an excellent technology compared to any other bioconversion technology. It is also excellent in "biotreatment" among all the technologies because it attains greater application as compared to the rate of destruction attained by any other technology. Compared to any other biological technology, it concerns about 100–1000 times higher value addition (Alvarez-Bernal et al. 2006).

5.6.1 Remediation of Organic and Inorganic Contaminants

There are mainly two types of pollution mitigation processes of vermicomposting. Detoxification is mainly helpful in the removal of hazardous chemicals from the contaminated raw materials and bioremediation is mainly applicable in the remediation of the different pollutants from the contaminated soils. Raw material which is used to produce vermicompost contains many hazardous materials, including heavy metals and different organic pollutants like polycyclic aromatic hydrocarbons, polychlorinated and polybrominated biphenyls, pesticides, pharmaceuticals, and personal care products. The application of vermicompost to enhance soil fertility needs that this microbiologically active and nutrient-rich organic amendment fulfill the quality standards. Thus, vermicompost must be free from these contaminants. The heavy metal concentration in vermicompost is affected by chemical speciation. Due to the change in chemical speciation, the bioaccumulation of heavy metals through earthworms is affected and it helps in the sorption of heavy metals to the organic legends in the vermicompost. There are many studies that state the bioavailability of many heavy metals (As, Cu, Cd, Cr, Ni, Pb, and Zn) and metalloids is reduced by the vermicomposting significantly with respect to earthworm species and type of raw material used (He et al. 2016; Lv et al. 2016; Goswami et al. 2016; Sahariah et al. 2015; Singh and Kalamdhad 2013). Different kinds of organic pollutants are also detoxified and reduced in concentration by the earthworms through the vermicomposting process. It also reduced the toxic effect of organic pollutants on the earthworms, microorganisms, and extracellular enzyme activities, which are the main factor of organic matter decomposition.

Apart from the detoxification of hazardous materials, vermicomposting is also responsible for the bioremediation of contaminants from the organic waste and from the soil because of the excellent sorbent property of vermicompost, thereby reducing their bioavailability and toxicity in soil. Zhu et al. (2017) have also documented that vermicompost can efficiently remediate toxic metals from the soil solution. Vermicompost can act as a metal sorbent. Hoehne et al. (2016), using metal contaminated soil in a laboratory experiment, examined the capacity of vermicompost to immobilize Cd, Cr, and Pb. High organic matter content, microbial abundance and diversity, and the existence of pollutant-detoxifying exoenzymes are the three main properties of vermicompost, which are responsible for the bioremediation of organic pollutants from the soil.

There are separate contaminant remediation mechanisms of earthworms for the organic and inorganic contaminants. The soil is facing anthropogenic pollution and contamination from industrial, farming, and other activities throughout the globe. Soil contaminants mainly include chemicals, organic wastes, inorganic compounds, or elements, especially metals (Dada et al. 2015). The traditional physicochemical remediation methods are very costly and not environment friendly just because of this the attention is shifted from them to biological in situ alternatives. Dada et al. (2021) reported the effectiveness of the application of earthworms to remediate organic and inorganic (metals) soil contaminants.

There are several interlinked mechanisms or processes through which an earthworm can remediate organic contaminants categorized under direct and indirect activity. Direct activities may be physical or physiological in nature. The burrowing activities of earthworms are the direct physical action of the earthworms. Earthworms make burrows for the proper movement of water, particles, and for aeration. Because of this burrowing effect, soil and substrate particles are broken down mechanically and thus increase surface area for microbial activities. Besides, earthworms intake and digest a huge amount of organic matter or contaminated soil at the time of burrowing activities. There is a significant reduction in the size of soil or contaminant particles due to the digestion process. This is also responsible for the enhancement of surface area for microbial composting and enzyme action (Sinha et al. 2010b) (Table 5.2).

The availability of nutrients and transformation and some heavy metals were evaluated in the integrated composting–vermicomposting of both wastes activated sewage sludge and primary sewage sludge utilizing aged vermicompost as a native bulking material and applying *E. fetida* as the earthworm species (Hait and Tare 2012). Vermicomposting process has been shown to remove about 90% of the heavy metals from spent mushroom compost and sewage sludge combination (Azizi et al.

Table 5.2 Vermiremediation contaminated soil

S. no.	Vermiremediation	Effectiveness of remediation	References
1.	*Eisenia fetida and Lumbricus terrestris*	*L. terestris* reduced petroleum hydrocarbons (PH) by 28%, *E. fetida* reduced by 33%, and both jointly reduced PH by 35%	Almutairi (2019)
2.	*E. fetida* with and without activation biopreparation	*E. fetida* aided petroleum oil degradation (99%)	Chachina et al. (2016)
3.	Earthworm species not clearly indicated	Earthworms aided degradation of up to 64.3–66.5%	Ahmed et al. (2020)
4.	*E. andrei*	High removal of PAHs. Earthworms accumulated PAHs	Rorat et al. (2017)
5.	*Dendrobena veneta*	Hydrocarbons decreased by 95% in the presence of *D. veneta*	Chachina et al. (2018)
6.	*Alma milsoni, Eudrilus eugeniae*, and *Libyodrilus violaceus*	Presence of earthworms decreased glyphosate residues in the contaminated soil	Owagboriaye et al. (2019)
7.	*E. fetida*, *E. andrei*, and *D. veneta*	Soil engine lubricant oil decreased by up to 99.9%	Chachina et al. (2015)
8.	Earthworm species not specified	87–100% reduction in soil metals. Increasing soil metals decreased remediation efficiency	Shameema and Chinnamma (2018)
9.	*L. rubellus*	50% reduction in soil amended with mushroom compost	Cheng-Kim et al. (2016)
10.	*Libyodrilus violaceus*	Cd, Zn, and Pb were reduced	Dada et al. (2016)

2013). Vermiremediation of dyeing sludge present in a textile mill has also been demonstrated with the assistance of the exotic earthworm species *E. fetida* (Bhat et al. 2018b). The influence of metal-rich tea factory coal ash on composting, reproduction and the metal deposition capability of *E. fetida* and *Lampito mauritii* has been observed by Goswami et al. (2014). Suleiman et al. (2017) observed that *E. fetida*, *E. andrei*, and *D. veneta* accumulated various heavy metals; among those species, *E. fetida* has shown the highest ability to accumulate heavy metals. Inoculation of local species for vermicomposting is a viable option and is recommended to the farming community for recycling sugar industry wastes (Shah et al. 2015). The capability of inoculating cow dung–paper waste mixture (fly ash) plus a specialized microbial concoction termed as an effective microorganism in vermicomposting using *E. fetida* earthworms.

Promotion of microbes and enzymes in their gut and in the contaminant containing soil substrate are the indirect biotic activities of the earthworm for the remediation of soil organic contaminants. Earthworms have millions of microorganisms in their gut which are biodegrader in nature and they also release them into the soil as vermicast. Many studies revealed that the microorganisms present in earthworms' gut are mainly related to the species and strain of genera Bacillus, Azotobacter, Pseudomonas, Enterobacter, Klebsiella Streptococcus, and Proteus (Bamidele et al.

2014). Several studies also documented a number of enzymes interconnected with the remediation activities of earthworms such as cellulases, lipases, proteases, and amylases (Ravindran et al. 2015). These enzymes and microbes released on the surface of substrates are very helpful in biodegradation, biotransformation, and mineralization of contaminants and organic matter as they pass through the earthworm's aliment canal.

As the inorganic contaminants (metals) are generally nonbiodegradable, just because of this, dermal absorption and intake through the intestine, and accumulation on their bodies are the main known mechanisms by which an earthworm can remediate inorganic contaminants from the soil environment. There are many metals that are toxic in nature. To overcome these problems, many plants and animals that have metal accumulators must possess a suitable mechanism. Earthworms are taking metals inside their body and release some of these metals by the calciferous glands and some of these metals are accumulated in the body of earthworms which are not excreted by the earthworms. The induction of metallothionein and subsequent sequestration and storage of the metallothionein-bound metals in structures like waste nodules and chloragogen are the main mechanisms by which earthworms are accumulated and deal with the high concentration of metals (Dada et al. 2015). Metallothionein induction is not only responsible for the remediation of contaminants (Metals) from the soil, but it also played a significant role in the survival of earthworms in contaminated environments.

5.6.2 Saline Soil Reclamation Using Vermicompost

Agriculture in Indo Gangetic plains is affected by salinity caused by irrigating water and fertilizer application. Irrigated land in arid and semi-arid regions are affected by increasing soil salinity and alkalinity. Climate change leads to a rise in temperature, leading to drought, causing land degradation and deterioration (AghaKouchak et al. 2014). Saline-sodic soils have poor aeration and hydraulic conductivity (HC) due to the migration of fine dispersed clay particles through the conducting pores (Matosic et al. 2018), which also form a crust on the soil surface (Nachshon et al. 2018). These soils harm the growth and yield of crops because of their low fertility (Matosic et al. 2018) and need effective, low-cost, and environmentally acceptable management practices (Kheir et al. 2019). Providing Ca^{2+} to remove excess sodium from the cation exchange complex is common to ameliorate sodic soils (Singh et al. 2016). The organic amendments could be used instead of chemical amendments to reclaim saline-sodic soils for a crop which alleviate saline soils by modifying bacterial community and soil aggregates (Liu et al. 2019b) enzyme (urease, acid phosphatase, acid invertase, and catalase) activity of saline soil (Deng et al. 2017). Vermicomposting, eco-friendly technology for crop residue management through non-thermophilic biodegradation of organic materials using earthworms and microorganisms, can provide the soil with macronutrients (Nurhidayati et al. 2018). In vermiconversion, earthworm gut phosphatases release the available P from organic

Fig. 5.3 Use of biowaste for vermicomposting and their application in saline soil remediation

wastes that be in available form to plants (Goswami et al. 2013). Vermicompost also provides plant growth hormones (gibberellins, auxins, and cytokinins) and improves plant nutrition, growth, photosynthesis, and chlorophyll content of leaves (Ravindran et al. 2015). Figure 5.3 depicts the saline soil remediation using vermicompost prepared from biowastes and their impact on soil characteristics.

5.7 Conclusion

The global scenario of population growth also leads to the use and disposal of various solid wastes in everyday life. They get accumulated in large quantities that need to be recycled by efficient technology to manage the pollution problems. Accumulation of solid wastes affects the atmosphere, aquatic and terrestrial ecosystems. Vermitechnology is a traditional method that can be utilized for converting organic solid wastes into manure. This vermicompost can be applied to soil to improve nutrient content and organic matter, thereby balancing biogeochemical cycling. Vermicompost is also helpful for the remediation of toxic soil. Vermitechnology is an eco-friendly technique applied for remediating problematic soil, detoxification and as a method of bioremediation. This could be implemented at different levels to achieve the goal of reducing, recycle, and reuse.

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Chapter 6
Application of Biochar from Waste for Carbon Dioxide Sequestration and Sustainable Agriculture

S. Sri Shalini, K. Palanivelu, and A. Ramachandran

Abstract Biochar is a stable carbon-rich and porous material produced by the thermochemical conversion of organic waste in low-oxygen conditions. Recently, biochar is shown to have enormous applications in the different fields of environment, climate change, and agriculture. Significantly, biochar production from waste is promising as it minimizes the waste quantity, produces energy, and supports climate-related aspects. This chapter critically explores the various solid waste feedstock possibilities for biochar production, an in-depth analysis of its material properties, and applications for carbon dioxide sequestration and sustainable agriculture. Biochar has the potential to reduce carbon dioxide concentrations and store carbon for several decades. For large-scale applications, it requires an immense study on the biochar-soil mechanism and climate impact studies. Various strategies involved in biochar for carbon capture and storage, waste utilization, soil amendment benefits, physicochemical properties, biotic and/or abiotic factors impact on the soil, and other climate aspects are detailed in this chapter. Scientific technicalities of biochar for the carbon-negative and carbon trading markets addressed. Overall, this review will find the insights of the biochar for waste utilization, global climate change mitigation, and sustainable agriculture.

Keywords Biochar · Waste · Carbon dioxide sequestration · Sustainable agriculture · Carbon storage and properties

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6.1 Introduction

Climate change is obvious and unavoidable in today's scenario of greenhouse gas (GHG) emissions in the atmosphere causing global warming. Major GHG contributing to global warming are anthropogenic carbon dioxide (CO_2) emissions. Its emission is increasing more than 3% every year for the past two decades due to the various anthropogenic activities that cause drastic changes in the climate that is irreversible and severe impacts on the environment (Solomon et al. 2009). According to the Intergovernmental panel on Climate Change (IPCC) sixth assessment report (AR6), it showed that the global warming is affecting almost all the sectors and the global average air temperature may exceed 1.5 °C (IPCC 2021). Nearly, in two decades we may have to face climate impacts of rising sea levels, erratic rainfall, and hotter temperatures. An international treaties "Kyoto protocol" aims to reduce the GHGs and "Paris agreement" that addresses the global warming is aiming to limit the global warming with below the 2 °C temperature preferably to 1.5 °C, compared to preindustrial levels (UNFCCC 2021). Many countries are opting for different climate mitigation options for addressing the climate change with their nationally determined contributions (NDCs). Recently, the new trend on low-carbon solutions, carbon neutrality, and zero-carbon solutions are emphasized.

"Biochar" a carbonaceous substance produced by thermochemical conversion of organic materials is mainly determined for the carbon sequestration and soil amendment. Biochar can mitigate climate change effectively as their production and application in soil have shown to reduce 12% of the present anthropogenic carbon dioxide emissions (Woolf et al. 2010). The thermochemical process called pyrolysis is widely applied for converting the materials into biochar a porous with large specific surface areas and stable carbon compounds under the oxygen-limited conditions (Fan and Zhang 2019). The biochar can be prepared from various feedstocks such as agricultural waste (crop residues, straws, stalks, tailings, hulls, cobs, etc.) (Novak et al. 2009; Zheng et al. 2010; Yao et al. 2012; Bian et al. 2014), forest waste (wood waste, bark, etc.) (Yamato et al. 2006; Reed et al. 2017; Tomczyk et al. 2019), municipal solid waste (sewage sludge, anaerobic sludge, poultry litter, etc.) (Mierzwa-Hersztek et al. 2016), industrial wastes, etc.

The pyrolysis processes for biochar production varied with their operating conditions classified as slow pyrolysis with lower heating rate and temperature, intermediate pyrolysis with low heating and moderate vapor residence time, flash pyrolysis with higher heating rate, and fast pyrolysis with high heating rate and short vapor residence (Kumar et al. 2020; Sri Shalini et al. 2021). The various pyrolysis reactors operated are fixed-bed, fluidized bed, tubular, rotary kiln, etc. (Zaman et al. 2017; Benavente et al. 2018; Veses et al. 2020). Based on the feedstock and pyrolysis operating conditions the biochar properties of surface area, pore structure, particle size, elemental composition, heavy metal content, etc., are varied (Tomczyk et al. 2020; Kumar et al. 2020). According to their biochar properties, they are applied in various fields. Biochar is widely applied for climate change mitigation, waste management, soil improvement, building materials, energy production, environmental remediation, etc. (Sri Shalini et al. 2021).

Many studies have been carried out for the biochar in soil applications (Reed et al. 2017; Tomczyk et al. 2020). It revealed to enhance soil fertility, increase plant growth, decrease leaching, and reduce carbon dioxide, nitrous oxide, and methane emissions. Soil carbon sequestration studies have been carried out by many researchers (Ramachandran et al. 2007; Lal 2008). It is a process where the carbon dioxide in the atmosphere is removed and stored in the soil carbon pool, which stores carbon many times higher than that present in the atmosphere and plants. The soil studies with biochar showed that it can sequester carbon and store it in a stable form for greater than 1000 years (Singh et al. 2012; Kuzyakov et al. 2014). It is a carbon entrapment route. Therefore, the reduction of carbon dioxide occurs rather than its emission during degradation of biomass and its mineralization rates are very low than biomass (Spokas et al. 2010). This feature of sequestering the carbon in the soil during conversion to biochar is a boon to decrease the climate change effects on agriculture (Matovic 2011; Wang et al. 2013). The benefits of the biochar in soil amendment are decided upon the soil properties and climate parameters.

Hence, this chapter in-depth explores the biochar feedstock and soil properties for carbon dioxide sequestration and sustainable agriculture applications. It exhibits the biochar-soil mechanism and climate impact processes with critical review on the various soil physical, chemical, and biological properties and biotic/abiotic factors that govern the soil amendment. The scientific approach towards carbon-negative, carbon trading, climate change mitigation, and carbon storage is highlighted. Overall, this chapter will find the insights of the biochar for waste utilization, global climate change mitigation, and sustainable agriculture.

6.2 Biochar in Climate Change Mitigation

The biochar production process is a carbon-negative system as it eliminates high quantities of carbon dioxide than it produces (Lehmann 2007a; IBI 2008). These negative emission systems have the ability for permanent carbon sequestration and have many benefits with respect to energy, water, and land-use (Smith 2016). Various climate benefits associated with biochar are described below.

6.2.1 Biochar Production and Their Climate Benefits

The pyrolysis process is more advantageous than other processes for biochar production. A study by Roberts et al. (2010) evaluated the projected biochar costs for various feedstocks in pyrolysis process scenarios revealed possible revenue generation. The revenue generated was mainly from their offsets which were approximately two dollars per ton and from their energy production. Similarly, Lehmann et al. (2006) and Woolf et al. (2010) showed offset potential from biochar was two petagrams of carbon per year (i.e., 2 billion tonnes) and can go up to five and a half petagrams of carbon per year (or 5.5 billion tonnes), when all the biofuels are generated from pyrolysis system in the year 2100. During biochar production, heat and gases are produced which can be captured to generate energy fuels for electricity, hydrogen, and bio-oil.

The biochar generation followed by soil application has a major part in mitigating climate change (Woolf et al. 2010). In addition to biochar reducing CO_2 emissions, it is also shown to decrease the other highly potent GHGs of methane and nitrous oxide demonstrating mitigation options for climate change (Zhang et al. 2010; Rehrah et al. 2016). The climate mitigation potential is evaluated by various researchers through different modeling scenarios and experiments with the considerations of feedstock quantity, yield efficiency, offset, and carbon sequestration potential (Fowles 2007; Woolf et al. 2010; Lefebvre et al. 2020). A recent study showed the carbon sequestration potential using sugarcane biochar (*Saccharum officinarum* L.) in Brazil by RothC model. The biochar application rates were around 4.2 t/ha/yr giving an increase in the soil carbon pool of 2.35 ± 0.4 tC/ha/yr. The model outcome scale to State gave 50 Mt of CO_2 equivalent/yr which is 31% of carbon dioxide equivalent emissions in 2016 (Lefebvre et al. 2020).

6.2.2 Biochar for Carbon Sequestration

The biochar carbon sequestration is basically varied from other bio-sequestration methods, way fundamentally different from other forms of bio-sequestration. The role of biochar in the soil carbon sequestration process and climate change mitigation is depicted in Fig. 6.1. The carbonized biomass, i.e., biochar is highly stable than its original biomass (Lehmann et al. 2006). This stable nature of biochar along with its aromaticity supports permanent carbon sequestration in soil (Lehmann 2007b). Biochar is stable for several years due to their recalcitrant property and much resistant to microbial decomposition and mineralization (Zheng et al. 2010). Despite the fact that it can stay stable for several years in soil, its stability is mainly based on the biochar properties and environmental parameters (Bai et al. 2014). The biochar with atomic ratios of O/C_{org} and H/C_{org} with 0.2 and 0.4, respectively, have relatively higher carbon sequestration potential (Spokas 2010; Enders et al. 2012). The turnover rate and mass of carbon have been used for evaluating the carbon sequestration potential (Gaunt and Cowie 2009).

Based on the properties of 76 biochars, it was exhibited that those biochars generated at higher temperatures possibly have larger carbon sequestration potential and biochars with lesser nitrogen content can have nitrous oxide mitigation potential (Brassard et al. 2016). A meta-analysis by Wang et al. (2016) showed that the majority of carbon in biochar is involved in the stability in soil and only 3% of biochar carbon is bioavailable. The biochar amendment has agronomic and soil benefits. The biochar addition in soil improves the soil fertility and intensifies the

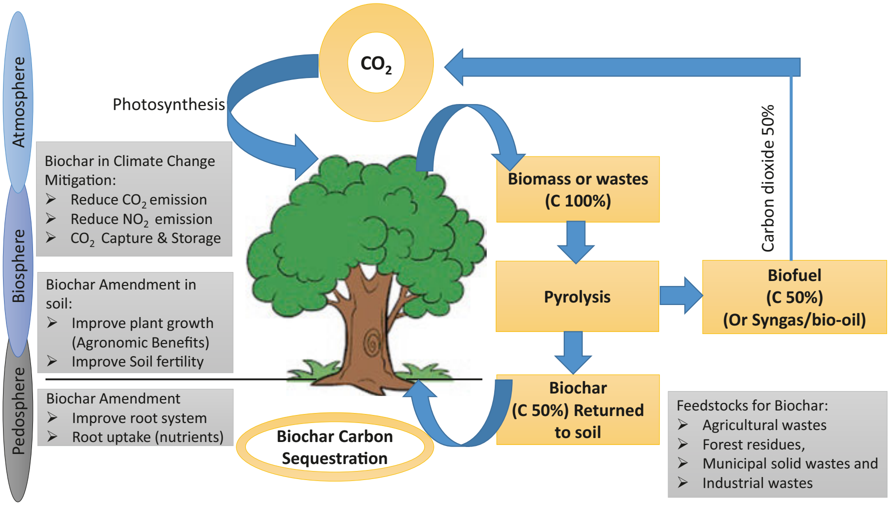

Fig. 6.1 Biochar for climate change and carbon sequestration

carbon sequestration as the decomposition rate is reduced and overall improves the soil physical, chemical, and biological properties (McHenry 2009; Hao et al. 2010; Khare and Goyal 2013; Malghani et al. 2013). The benefits of biochar amendment and its impacts on the soil properties are explored in detail as below.

6.3 Biochar Soil Amendment Studies

The soil-biochar mechanism mainly depends upon the type of soil and biochar properties. The different soil types with their biochar application rates are tabulated in Table 6.1. The biochar application rates varied from 0% to 2% or 10–40 Mg/ha by various studies. The yields differ based on the biochar application rates and the output results sometimes increase or decrease. Sometimes higher biochar addition rates in soil may adsorb contaminants and reduce soil fertility. Some toxicant pollutants that might be present in the original feedstock may release. The process of soil dilution losses soil fertility. So, careful and cautious biochar application rates must be considered along with the addition of supplementary nutrients such as nitrogen (Beesley et al. 2011). The biochar application has many environmental and agricultural benefits as depicted in Fig. 6.2. The biochar addition has improved the soil fertility, agronomic benefits, reduced soil nutrient leaching thereby accumulating moisture and nutrients that substantially, decrease the usage of fertilizers and water demand (Sohi et al. 2009). During the soil amendment, the biochar increases the soil pH, water retention, microbial activity, cation exchange capacity, and nutrient retention resulting in an enhancement in soil fertility and quality (Lehmann et al. 2006) and increases the crop yield.

Table 6.1 Biochar application rates in various soils

S. no.	Soil type	Biochar feedstock	Pyrolysis (°C)	Biochar application rate	References
1.	Acidic soil	Bark of Acacia mangium	260–360	37 Mg/ha	Yamato et al. (2006)
2.	Loamy sand	Peanut hulls Pecan shells Poultry litter Switchgrass	250–700	2.0% or 40–44 t/ha	Novak et al. (2009)
3.	Silt loam	Corn cobs Wood chips	450	40 t/ha	Zheng et al. (2010)
4.	Alfisol Vertisol	Poultry manure Wood	400; 550	10 t/ha	Singh et al. (2010)
5.	Pasture soil	Pinus radiata D	–	30 t/ha	Taghizadeh-Toosi et al. (2011)
6.	Sandy soil	Sugarcane bagasse Peanut hull Bamboo	300 450 600	2.0%	Yao et al. (2012)
7.	Sandy Loam soil	Willow wood Swine manure	350 700	10.0 Mg/ha	Ameloot et al. (2013)
8.	Hydroagric Stagnic Anthrosol	Wheat straw	350–550	40 Mg/ha	Bian et al. (2014)
9.	Alkaline	Sawdust	300–350	20 Mg/ha	Mete et al. (2015)
10.	Eutric Cambisol	Poultry litter	300	2.3 and 5.0 t/ha	Mierzwa-Hersztek et al. (2016)
11.	Sandy clay loam	Harwood	450	10 t/ha	Reed et al. (2017)
12.	Haplic Luvisol Haplic Podzol	Wood waste	650	0%, 0.01%, 0.05%, and 0.1%	Tomczyk et al. (2019)

6.3.1 *Biochar Effect on Soil Chemical Properties*

The main chemical property of the soil that has a significant effect is on soil organic carbon (SOC) and nutrients. The priming effects due to biochar addition have been studied by many researchers (Wang et al. 2016; Lefebvre et al. 2020). The impact on the present SOC might increase the SOC mineralization called “positive priming” and decreasing the SOC mineralization is called “negative priming.” Different studies showed varying priming effects on biochar addition as negative, positive, or no effect, usually positive priming for a year and then negative priming occurs. A meta-analysis study showed different priming effects on biochar and soil impacts, higher priming impacts in the sandy soils with lower fertility which are of sugarcane areas (Wang et al. 2016). The fine nature soil with biochar is shown to be better for carbon storage in soil microaggregates and resulting to increase in the soil organic matter in the soil (Wang et al. 2017). Another meta-analysis of biochar was carried out by Jeffery et al. (2011) with both pot and field trials. The results revealed that

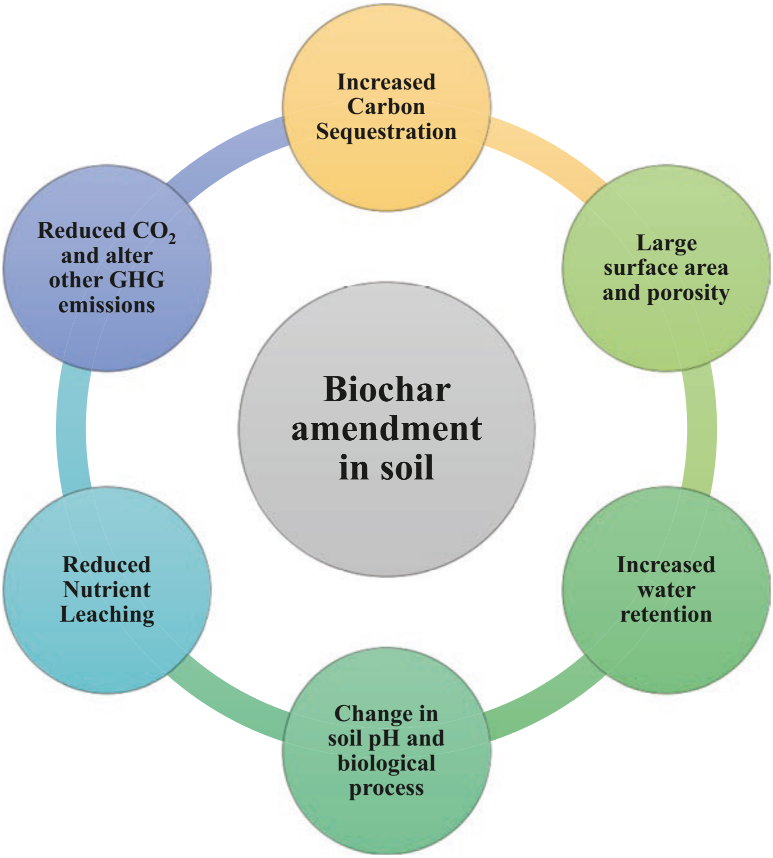

Fig. 6.2 Advantages of biochar amendment in soil

the biochar compared to control, the crop productivity increased and positive effects for acidic soils, neutral pH soils, coarse, medium texture, and hundred tonnes per hectare.

The effect of biochar addition on the nutrient retention capacity of a soil depends upon both the biochar property and soil nutrient property. In a study by Dari et al. (2016), it was indicated that the phosphorus accumulation in non-calcareous soil was due to the soil property and not based on the feedstock property. The biochar addition from the same feedstock application in different soils results in different effects based on the soil properties. The varying biochar properties also has a different impact on the soil. The specific biochar properties that are advantageous to the soil properties are listed in Table 6.2.

6.3.2 Biochar Effect on Soil Physical Properties

Adding biochar as finely grounded particles has a beneficial effect on enhancing the surface area, density, and ease of transport. Further, adding it as slurry decreases the losses due to the wind (Beesley et al. 2011). The advantage of biochar addition to soil is a liming effect, i.e., increased pH, and water holding capacity with improved nutrient availability (Jeffery et al. 2011). The texture of the soil also plays a role in the soil-biochar mechanism. Wang et al. (2017) showed that biochar addition on a fine nature soil had better aggregate stability than the coarse-nature soil with no effect. The biochar effect on water retention and bulk density have been reported by many authors (Novak et al. 2009; Laghari et al. 2016; Anwari et al. 2020). The biochar in the soil increases water retention, whereas reduces the soil bulk density. As shown in Table 6.2, the higher biochar surface and porosity contributed to the rise in

Table 6.2 The biochar property and its impact on soil

S. no.	Biochar property	Effect on soil	Reference
1.	Surface area and porosity	Improve soil structure Water and nutrient retention Immobilization organics and heavy metals Increased the porosity Increased cation exchange capacity Decreased nitrous oxide emissions Decreased nutrient leaching	Atkinson et al. (2010) Singh et al. (2010) Biederman and Harpole (2012) Zhang et al. (2012) Herath et al. (2013) Jien and Wang (2013) Anwari et al. (2020)
2.	Surface functional group	Improve soil moisture Enhance binding between biochar and soil Reduced soil nutrient leaching	Anwari et al. (2020)
3.	Organic C, N	Improve soil carbon respiration loss Increased organic matter Improve soil fertility Increased carbon sequestration	Kameyama et al. (2010) Matovic (2011) Zhang et al. (2012) Anwari et al. (2020)
4.	Exchangeable cations	Remove heavy metals Improve soil pH and CEC Nutrient bioavailability to plant	Anwari et al. (2020)
5.	Alkaline nature	Increased pH	Zhang et al. (2012) Jien and Wang (2013)
6.	pH	Increased crop yield Decreased methane emissions	Feng et al. (2012) Zhang et al. (2012)
7.	Bulk density, cation exchange capacity	Increased crop yield	Zhang et al. (2012)
8.	Colour, P and K cycling	Increased plant productivity	Biederman and Harpole (2012)

water holding capacity leading to an increase in plant growth (Novak et al. 2009; Laghari et al. 2016). The sandy soil such as desert soil with lesser surface area and particle size might be enhanced for plant growth by biochar amendment (Laghari et al. 2016).

6.3.3 Biochar Effect on Biological Properties

The biochar addition on soils has a pronounced effect on the soil microbial activities, microbial population, microbial enzyme, mineralization, and respiration rates (Thies et al. 2015). It also has an effect on the bacterial population to fungal population and soil-borne diseases. The soil biological properties vary based on the biochar and soil properties and also vary with respect to the climatic conditions. It was shown that the biochar addition improved the soil fertility, plant growth, and reduced GHG emissions (Woolf et al. 2010; Zhang et al. 2010; Brassard et al. 2016; Rehrah et al. 2016; Lefebvre et al. 2020). Hence, the biochar for soil amendment is a favorable option for sustainable agriculture.

6.4 Biochar for Sustainable Agriculture

The agricultural landmass is reducing due to the increasing population and its rising demand for food production causing more environmental pollution from the increased usage of fertilizers, pesticides, and herbicides. The inorganic compounds of nitrogen leaching pollute the water bodies and reduce soil fertility. The GHGs emissions from the soil add to climate change. The biochar addition in soils is a promising option to address the above issue as the biochar amendment in soil demonstrated better carbon sequestration, climate change mitigation potential, improved plant growth, and soil fertility (Novak et al. 2009; Woolf et al. 2010; Singh et al. 2010; Zhang et al. 2010, 2012; Brassard et al. 2016; Rehrah et al. 2016; Anwari et al. 2020; Lefebvre et al. 2020). Biochar can act as fertilizer, the addition of synthetic fertilizer that harm the environment is not required. Local feedstock can be used to produce biochar, and hence fossil fuel usage can be reduced.

The low carbon availability in the agricultural and forest fields necessitates the need for carbon addition to have better crop yield and plant productivity (Ramachandran et al. 2007). The biochar addition can increase the soil organic carbon, moisture retention, and nutrient retention thereby increasing the plant growth (Novak et al. 2009; Laghari et al. 2016), that eventually leads to agricultural sustainability. As shown in Table 6.2, the biochar properties has a strong positive impact on the soil properties to improve the soil structure, surface area, porosity, cation exchange capacity, fertility, organic carbon, organic matter, water retention, nutrient retention, carbon sequestration, crop yield, reduced GHG emissions, and removed soil pollutants. Especially, the biochar properties of porosity, alkalinity, and nutrient had a significant effect on soil fertility, pollution control, and disease management leading to sustainable agriculture. A meta-analysis study by Biederman and Harpole (2012) showed the biochar impact on plant growth and nutrient cycling. It demonstrated that the biochar had a positive impact on the aboveground productivity with enhanced crop yield, soil biological properties, rhizobial nodulation, and potassium content. Also, the nitrogen, phosphorus, potassium, and pH was higher than the control ones. The meta-analysis is powerful tool but its outputs are mainly based on the input data (Jeffery et al. 2013). Hence, the biochar addition to soil develops a sustainable agriculture approach. The biochar in market availability and carbon trading has to be explored.

6.5 Carbon Trading

The biochar can be a marketable commodity as revenue can be generated from its production and usage (Roberts et al. 2010). Recently, biochar is sold as a soil amendment for garden purposes at a cost around 1000 dollars per ton. It can offset approximately 5.5 billion tonnes of carbon in a year by the near future (Woolf et al. 2010) showing the potential in the carbon trading market.

As of now, no carbon markets are available that give carbon credits or financial compensation for biochar addition in soil for carbon sequestration or reducing soil emissions. Some of the chief regulatory carbon market systems mainly are the Clean Development Mechanism (CDM) of the Kyoto Protocol, the European Union Emissions Trading Scheme (EU ETS), California Cap and Trade market, etc., and voluntary markets such as Verified Carbon Standard, the American Carbon Registry (ACR), the Climate Action Reserve (CAR), etc. The biochar projects are not considered under the carbon offset protocol. However, biochar projects should be considered for carbon offset sooner for combating climate change.

As the biochar market grows, it is important to understand the merits and demerits before implementing them in a large scale. The environmental and public health risks evaluation and cost analysis are necessary for the biochar projects. The biochar projects are promising as revenue generation, positive outlook in carbon trading, increase the job opportunities and alternative for synthetic fertilizers makes it potentially marketable product however risk and cost evaluations are needed.

6.6 Conclusion

This chapter explored various application of biochar for carbon sequestration and sustainable agriculture. The biochar production from various feedstocks along with their climate benefits was highlighted. Biochar showed a positive carbon sequestration potential, reduction in greenhouse gases emissions, and possible revenue generation from their production and usage. The various biochar properties having a significant impact on the soil physical, chemical, and biological properties were exhibited. Especially, the biochar properties of surface area, porosity, nutrient content, and alkalinity have a significant impact on the soil for improved fertility and plant productivity. The opportunities for marketability and carbon trading of biochar are significant. Overall, biochar is a promising solution for carbon sequestration and sustainable agriculture.

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Part II
Waste Management for Cleaner Environment

Chapter 7
Propelling the Future Biofuel Research: Plant Breeding, Genomics and Genetic Engineering Strategies for a Cleaner Environment

Hemalatha Palanivel, Shipra Shah, M. Kamaraj, and Alazar Yeshitla

Abstract Substitution of fossil fuel with biofuel produced from crop biomass has the prospect to hugely contribute to meeting rising energy demands and mitigating air pollution. Current research interventions for biofuel production are focused on transforming plant biomass into alternate nonrenewable liquid fuels. The foremost constraints for biofuel production include lack of domestication of biofuel crops, limited oil yield from crop plants, and recalcitrance of lignocellulose breakdown by a chemical and enzymatic process. Scientists are exploiting the genetic and genomic resources available for crop improvement, elucidating the gene cascades for lipid metabolism to manipulate fatty acid metabolic pathways and explicate plant cell walls synthesis and assemblage. None of the present and probable crops have been domesticated or bred for higher polysaccharides or oil recovery for biofuel production. Due to this reason, biofuel research is targeted towards understanding the genetic architecture of plant biomass traits that need to be improved to optimize crops for efficient biofuel production.

Keywords Biofuel · Plant breeding · Genomics · Genetic engineering · Lignocellulose

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7.1 Introduction

Renewable energy has reached the highest interest and funding in recent times in the wake of high crude oil prices, incertitude in fossil fuel reserve and supply, international policies, and environmental concerns interconnected with nonrenewable fuel sources (Masjuki et al. 2013). The direct and apparent effects of global warming include a rise in temperatures around the world and environmental inequality, besides universal risk to natural ecosystems, posing a significant threat to the food chain and human race (Barneche et al. 2021). Moreover, the exhaustion of conventional resources and increasing greenhouse gases emission such as carbon dioxide released from the burning of fossil fuels drives countries to find alternative solutions. In pursuit of solutions to the petroleum crisis, the principles of environmentally friendly and renewable bioenergy gained popularity. These features have made biodiesel usage more compliant and irresistible to the current energy scenario, boosting sustainable pathways to greater energy security, environmental amelioration, and improved rural development by shifting power from hydrocarbon molecules to bio-based industries, simultaneously.

As the bio-based economy develops, issues of production and processing will be upgraded, and the demand for new products will also be expanding. The new products need innovative raw materials. In a bio-based economy or industry, the basic raw materials are genes and enzymes (Sticklen 2008). Thus, as we shift from a geology-based economy to one based on biology, identification of alternate fuel sources such as biofuel (ethanol, methanol, and biodiesel) and genetic improvement through plant breeding and biotechnological approaches have gained momentum (Davidson 2008). An important approach to address the depletion and the negative impacts of fossil fuels on the environment is the use of biofuels derived from renewable sources for transportation (Fig. 7.1). Biofuels derived from plant sources are among the most cost effective, clean and renewable source of energy (Awogbemi et al., 2021). They allow mass production of ethanol and butanol (gasoline additives) as well as long-chain hydrocarbons (diesel supplements or jet fuels) from organic materials (biomass).

7.2 Biofuels and Climate Change Mitigation

The rising popularity of biofuels as an alternative to fossil fuels is attributed to several factors. One of the most important factor is the scale down of GHG emissions from the transport sector. The biofuel industries in the United States (US), the European Union (EU), Brazil and progressively in Southeast Asia are presently supported by technologies that utilise food or feed crops as feedstocks (Searchinger et al., 2008). The rationales for promoting biofuel production are disparate and entail energy security, rural development and transition to a low carbon economy in addition to climate change mitigation. The degree to which biofuels can mitigate

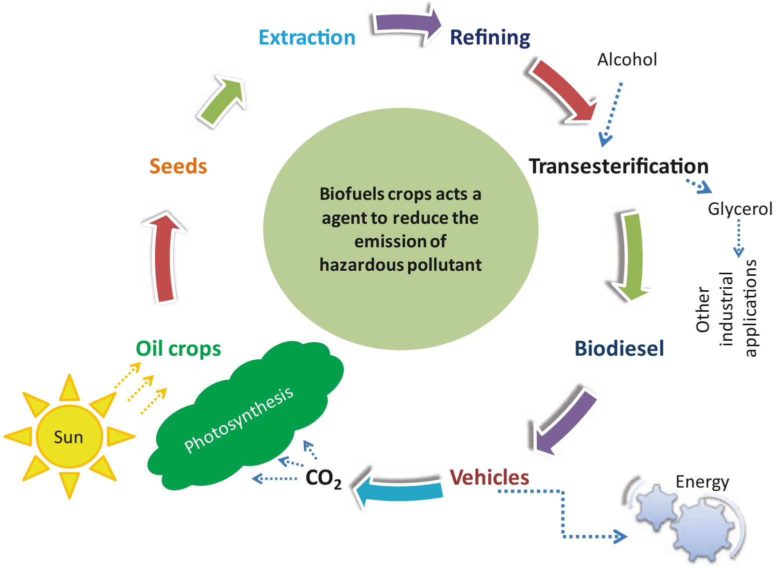

Fig. 7.1 Fuel crop's role in fuel circulation

climate change is determined by their GHG intensity in comparison with the liquid fossil fuels they substitute. Farming and land-use change already recorded for approximately 15% and 13% of global greenhouse gas emissions, respectively (Houghton 2008). Agricultural residues such as stalk are a promising source of feedstock for biofuel production since they have lesser climate impact and do not need excess land.

The most common renewable fuel is ethanol, which is produced from direct fermentation of sugars (e.g., from sucrose of sugarcane or sugar beet) or polysaccharides (e.g., starch from corn and wheat grains). Cellulosic biomass also has great potential to contribute to the demand for liquid fuel (Himmel and Bayer 2009). There has also been recent interest in the conversion of gaseous feedstocks such as CO_2 and H_2 (as off-gases, as syngas, or as producer gas from biomass gasification) to products such as ethanol, butyrate, and acetate. Overall, biomass residues may be grouped as follows: primary ones from agricultural and tree crops (crop residues) or forest (logging residues), secondary ones from agriculture (from food processing, animal manure) and forest (mill and manufacture residues), and tertiary ones including all type of final biomass waste.

There are several advantages and returns derived from the deployment of biofuels as a type of renewable fuel that can be broadly categorized into economic, ecological, social, and energetic (Gheewala et al. 2013; Awogbemi et al. 2021; Pryshliak et al. 2021):

Economic: Biofuels are inexhaustible compared to fossil fuel reserves. The development of the biofuels sector generates additional revenues to the national budget and reduces dependence on imported fossil fuels thereby conserving foreign exchange reserves. Biofuels can also increase income for rural communities through increased prices of feedstock, land, and labor. Another economic advantage of biofuels is reduced costs for combating the negative impacts of fossil fuel emissions on the environment.

Ecological: Biofuels play a critical role in climate change mitigation. They are carbon—and CO_2/GHG—neutral, and therefore green, sustainable, and environmentally friendly. Additionally, lower GHG emissions are produced from biofuels compared to fossil fuels. This also improves the health and well-being of populations, particularly in urban environments.

Social: The cultivation of biofuel feedstock may benefit rural communities including smallholder farmers by enhanced access to energy, farm income, and rural employment opportunities. Moreover, the production and utilization of biofuels increase home-grown agricultural development and investment. This can potentially contribute to poverty reduction and rural development.

Energetic: Energy security remains the overarching policy driver of biofuel expansion. Biofuels enhance national energy security by fostering independence from imports of conventional fossil fuels.

7.3 Classification of Biofuels

Biofuels are classified as the first-, second-, third-, and fourth-generation biofuels based on the origin and production technology. The choice of feedstock affects the development and utilization of biofuels as a potential substitute for fossil fuels. Feedstocks may be selected on the basis of price, hydrocarbon content, and biodegradability (Awogbemi et al. 2021). The first-generation biofuels are produced from crop plants as energy-comprising molecules like sugars, oils, and cellulose. They however generate inadequate biofuel yields and have a negative effect on food security since they utilize edible biomass. Research is now needed to speed up the generation of advanced biofuels by characterizing and engineering potential nonfood feedstocks, upgrading the performance of conversion technologies and the attributes of biofuels for various transport sectors as well as reducing the costs.

Second-generation biofuels tap the nonedible biomass but there are still some disadvantages associated with their cost-effectiveness (Naik et al. 2010). These biofuels utilize lignocellulosic material as feedstock such as straw, bagasse, forest residues, and purpose-grown energy crops on marginal lands. Projects are needed to maximize the amount of renewable carbon and hydrogen that can be converted to fuels from “second-generation” biomass. The third-generation biofuels are based on algal biomass production. They are presently under extensive research in order to improve the metabolic production of fuels and the separation processes in bio-oil generation for removing nonfuel components and lowering the overall costs. Fourth-generation biofuels direct attention towards genetically modifying microorganisms to accomplish preferable hydrogen to carbon (HC) turnover along with the formation of an artificial carbon sink. The third- and fourth-generation biofuels are in the preliminary stages of development (Li-Beisson and Peltier 2013).

7.4 Potential Plants for Biofuel Production

Biofuel production from food crops such as sugarcane, sunflower, soybean, sugarbeet, rapeseed, and maize is susceptible to a food security crisis. Biofuel extraction from fuel crops is widely influenced by several factors including the ecological nature, farming procedures, and industrial activities (Fig. 7.2). Whether or not biofuel production has displaced a considerable proportion of food production is still profoundly debated. Nonetheless, this does throw up an important question: which of the two should be given greater importance when making a choice between energy and food crops? The answer to this ambiguity lies in the exploitation of crop residues and the many prospective exclusive nonfood biofuel crops, including perennial grasses, such as switchgrass (*Panicum virgatum* L.) and *Miscanthus* spp., rapidly growing trees including poplar (*Populus* spp.) and willow (*Salix* spp.), fiber crops such as kenaf (*Hibiscus cannabinus* L.) and oil-rich nonedible crops.

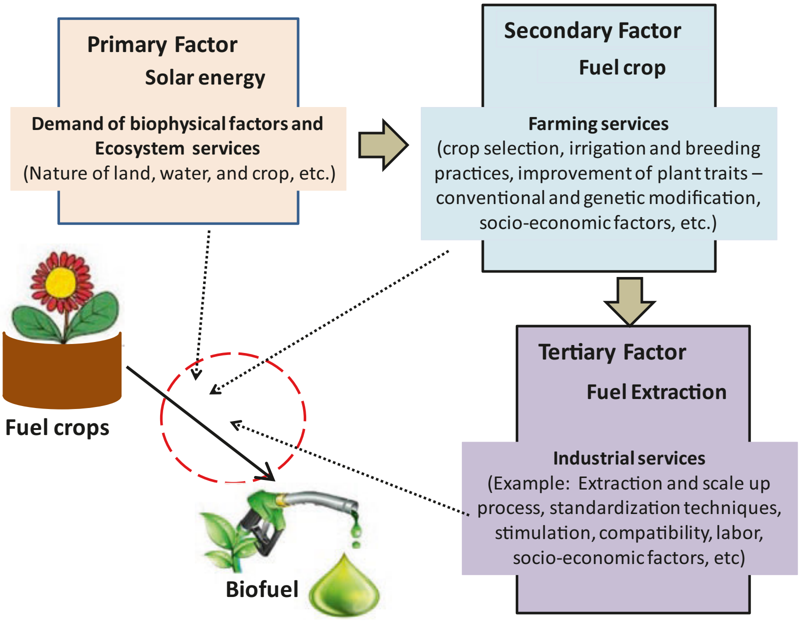

Fig. 7.2 Factors that influence the biofuel development from fuel crops

7.5 Tree Borne Oilseed Crops

Several underutilized crops can be prioritized for biodiesel production. These species are adapted to environments not suitable to grow food crops, providing a viable economic alternative that helps reduce poverty and rural migration. Tree-borne oilseeds (TBOs) are multipurpose tree species in agriculture systems. TBOs can be cultivated in diverse agro-climatic conditions in the forest and non-forest areas as well as in wastelands. TBOs include *Jatropha curcas* L., *Moringa oleifera* Lam., *Pongamia pinnata* (L.) Pierre, *Calophyllum inophyllum* L., *Hevea brasiliensis* (Willd. ex A. Juss.) Müll. Arg., *Azadirachta indica* A. Juss. which have been discussed in the following sections.

7.5.1 Jatropha

Jatropha curcas is a small tree that belongs to the family Euphorbiaceae. Although native to tropical America, it now has a pantropical distribution and is found in both Africa and Asia. The rapid rate of growth, easy propagation, drought tolerance, unpalatability to animals, small gestation period, and ability to grow in a wide range of environmental conditions are some characteristics that highlight the potential of *Jatropha* as a biofuel crop. The oil has a calorific value of 38.20 MJ/kg; higher than anthracite coal and comparable to crude oil (Moniruzzaman et al. 2017). The oil content of seeds is also high, varying between 30% and 35%. Contrary to popular belief pest damage has been observed in monoculture plantations of *J. curcas* in Africa and India. Some of the major pests of *J. curcas* are blister leaf miner *Stomphastis thraustica*, scutellera bug *Scutellera nobilis*, leaf webber *Pempelia morosalis*, bark-eating borer *Indarbela quadrinotata*, semi-looper *Achaea janata*, and flower beetle *Oxycetonia versicolor* (Brittaine and Lutaladio 2010). Low seed yield is often considered a major drawback in realizing the full potential of the *Jatropha* biodiesel industry (Moniruzzaman et al. 2017). Seed yields are economically unviable varying between 1 and 1.6 t/ha in sub-Saharan Africa and South Asia (Brittaine and Lutaladio 2010). At a plantation density of 2500 plants/ha, the average seed yield is around 2.50 t/ha and the oil yield is around 2 t/ha (Azam et al. 2005). The tree can grow on marginal land eliminating any direct competition with food crop cultivation. However, seed yield in such areas remains low, highlighting the importance of soil fertility, irrigation, and management practices (Mogaka et al. 2010). The unavailability of high-yielding commercial varieties is a key limitation. The high viscosity of the oil is another major limitation concerning its utilization as biodiesel particularly in cooler climates since it reduces the efficiency of fuel injectors (Moniruzzaman et al. 2017).

7.5.2 Moringa

M. oliefera commonly called drumstick is native to the foothills of the Himalayas in northwestern India. Extensively cultivated in the tropics and subtropics, the tree is fast-growing, drought-tolerant, and can grow in poor soil conditions under a wide rainfall range between 250 and 2000 mm (Rashid et al. 2008). It is a multipurpose tree with a diversity of uses including food, medicine, fuel, fodder, green manure, and water purification. The potential of *Moringa* seed oil as a promising feedstock for biodiesel production has been reported by da Silva et al. (2010). The seeds contain around 35–45% oil which has a high oleic acid concentration (>70%), remarkable oxidative stability (da Silva et al. 2010), and a calorific value of around 38.05 MJ/kg (Mofijur et al. 2015). Biswas et al. (2013) reported a seed yield of 3.03 t/ha in dryland and 6.06 t/ha in irrigated land in Australia. At a plantation density of 2500 plants/ha, the average seed yield is around *M. oliefera* is however susceptible to attack by several pests. Notable pests include the leaf caterpillar *Noorda blitealis*, hairy caterpillar *Eupterote mollifera*, itch caterpillar *Euproctis pasteopa*, leaf miner cum webber *Protrigonia zizanialis*, budworm *Noorda moringae*, stem borer *Coptops aedificator*, stem and root borer *Plocaederus ferrugineus*, and long horn beetle *Batocera rubus* (Rashid et al. 2008; Joshi et al. 2016). Among the major diseases affecting *Moringa* are twig canker *Fusarium pallidoroseum*, root rot *Diplodia* sp., and fruit rot *Cochliobolus hawaiiensis* (Mridha and Barakah 2017).

7.5.3 Pongamia

Pongamia pinnata commonly called Indian beech, Pongam and Honge tree is a member of the Leguminosae family. The tree is native to India, Northern Australia, and South East Asia (Karmee and Chadha 2005). It is a medium-sized evergreen, multipurpose, fast-growing, nitrogen-fixing, salinity, and drought-tolerant tree. *P. pinnata* can grow on marginal land and has an adaptability to a wide range of agro-climatic conditions (Kesari et al. 2010). The tree has been traditionally utilized as folk medicine, fuel, fodder, green manure, and fish poison (Islam et al. 2021). The seed oil of *P. pinnata* has several characteristics that highlight its potential utilization as biodiesel. The seed oil content varies between 35% and 45%, which can then be converted to biodiesel by transesterification with methanol (Kazakoff et al. 2011). Similarly, the oil yields of *P. pinnata* are higher than *Jatropha*. At a plantation density of 1111 trees/ha, seed yield is around 5.50 t/ha while oil yield is around 4.40 t/ha (Azam et al. 2005). The oil of *P. pinnata* has a calorific value of 40.76 MJ/kg; higher than other seed oils and slightly lower than kerosene (Halder et al. 2014).

However, the oil has a high viscosity and poor combustion characteristics. This can cause fuel injector blockage, poor fuel atomization, and engine oil contamination (Karmee and Chadha 2005). The establishment of commercial plantations is important to meet the demands of the biodiesel industry. However, *P. pinnata* has not yet received attention as a plantation crop. Lack of improved planting stock is a major bottleneck impeding any efforts for its large-scale domestication. Tree improvement programs should focus on the identification of elite genotypes. Selection traits include high seed yield, high oil content, and desirable fatty acid composition (Kesari et al. 2010).

7.5.4 Alexandrian-Laurel

Calophyllum inophyllum commonly called *Alexandrian-laurel*, *Indian-laurel*, *beach-touringa*, and *tamanu* is an evergreen tree belonging to the family Clusiaceae. It has multiple origins and is native to East Africa, India, South East Asia, Australia, and the South Pacific (Atabani and César 2014). Commonly found along with coastal areas, the tree has a high degree of tolerance to strong winds, salt sprays, strong tidal waves, and brackish water and is therefore recommended for sand dune stabilization. Traditionally the oil has been used as medicine, cosmetics, lamp oil, dye, and varnish (Dweck and Meadows 2002). Flowering occurs almost throughout the year with two distinct peaks in late spring and autumn. Fruits can therefore be collected twice a year for oil extraction. The tree has a high seed and oil yield per unit area than most other potential biodiesel sources. At a density of 400 trees/ha, the seed yield is around 4.68 t/ha and oil yield is around 3.74 t/ha (Azam et al. 2005). The seeds have a high oil content ranging between 65% and 75% (Ashok et al. 2018), with an unsaturated fatty acid content of about 71% (Kurniati et al. 2019). *C. inophyllum* oil has a calorific value of 39.25 MJ/kg (Atabani and César 2014). However, a major limitation is the high concentration of free fatty acids, up to 30%, which prevents the effective conversion of the oil to biodiesel. Transesterification is recommended to lower the free fatty acid concentration (Ashok et al. 2018).

7.5.5 Rubber

Hevea brasiliensis a natural source of rubber is an evergreen tree in the family Euphorbiaceae. It is native to the tropical rainforests of the Amazon basin in South America. It was introduced to South and South East Asia which account for 81% of the world's rubber plantations and 95% of the world's latex production (Pizzi et al. 2020). While latex from the rubber trees is utilized for rubber production, the seeds are currently discarded. Rubber seed oil is a viable biodiesel option considering the current extent of rubber plantations in the world, around 11 million ha out of which 9.2 million ha is concentrated in South East Asia (Azizan et al. 2021). Seed oil content is also high around 40–50% (Takase et al. 2015), with 17–20% saturated fatty acids and 77–82% unsaturated fatty acids (Ikwuagwu et al. 2000). However, the low seed yield of *Hevea brasiliensis* (2 t/ha) (Pizzi et al. 2020) is a key limitation for meeting the raw material requirements of the biodiesel industry. Furthermore, since the oil has high free fatty acid content, the alkaline catalyst forms soap during transesterification decreasing the ester yield (Ramadhas et al. 2005).

7.5.6 *Neem*

Azadirachta indica is an evergreen tree of the family Meliaceae native to India and Burma. It was later introduced in Africa, the Middle East, America, and Australia (Awolu and Layokun 2013). All parts of the tree: leaves, flowers, fruits, seeds, bark, roots, oil, and gum are traditionally used as medicine. Neem trees have a long lifespan of around 150–200 years and begin producing fruits at age 3–5 years. The seed oil content ranges between 35% and 40% (Madai et al. 2020). At a plantation density of 400 trees/ha, the average seed yield of neem is around 2.67 t/ha and the average oil yield is around 2.14 t/ha (Azam et al. 2005). The calorific value of neem oil is around 39.11 MJ/kg (Agarwal and Agarwal 2009). The oil also has a high concentration of free fatty acids 2.73% which results in a low yield of methyl esters (Bhandare and Naik 2015). The proportion of unsaturated fatty acid is 63% while saturated fatty acid is 37% (Aransiola et al. 2012). High viscosity (45.89 cSt) (Bhandare and Naik 2015) is the main limitation of neem oil affecting its utilization as biodiesel (Agarwal and Agarwal 2009). Table 7.1 summarizes the selected biofuel crop and the properties related to their fuel content.

7.6 Biofuels and the Contribution of Plant Breeding

When the significance of plants in liquid fuel generation was realized, the initial focus was given to food crops such as sugarcane (sugar), maize (starch), or soybean (oil) for biofuel production. Biofuel production from food crops has been criticized for triggering a food crisis in recent years. However, the production of biofuels from food crop residues or from unique nonfood lignocellulosic crops employs the whole plant thus gaining more energy per unit of land area. Furthermore, TBOs are increasingly being recognized for their potential as bioenergy sources.

Liquid biofuel genesis from plant cell wall material is nearly a half-century-old practice (Himmel and Bayer 2009), but its potential is yet to be realized. One of the major impediments in producing liquid fuels from plant biomass is that transformation of cellulosic matter into fermentable sugars is much more challenging than the conversion of starch. Biofuels produced from plant lignocellulosic biomass are also recognized as second-generation biofuels superior to first-generation biofuels from plant starches, sugar, and oil with respect to net energy and CO_2 balance and, more significantly, they do not affect food industries.

Table 7.1 Selected biofuel crop and its fuel properties

Tree	Seed yield per hectare (t)	Oil yield per hectare (t)	Oil content (%)	Density (g/cm^3)	Kinematic viscosity (cSt)	Flash point (°C)	Calorific value (MJ/kg)	References
Jatropha	2.50	2.0	30–35	0.932	52.76[a]	210	38.20	Azam et al. (2005), Brittaine and Lutaladio (2010), Moniruzzaman et al. (2017)
Moringa	–	–	35–45	0.898	43.33[b]	268.5	38.05	da Silva et al. (2010), Halder et al. (2014), Mofijur et al. (2015)
Pongamia	5.50	4.40	35–45	0.912	29.65[a]	241	40.76	Azam et al. (2005), Kazakoff et al. (2011), Halder et al. (2014), Fu et al. (2021)
Alexandrian-laurel	4.68	3.74	65–75	0.951	55.68[b]	236.5	39.25	Azam et al. (2005), Ashok et al. (2018), Mofijur et al. (2015)
Rubber	2.00		40–50	0.922	41.24[a]	294	37.50	Atabani and César (2014), Ikwuagwu et al. (2000), Takase et al. (2015)
Neem	2.67	2.14	35–40	0.910	45.89[a]	218	39.11	Agarwal and Agarwal (2009), Azam et al. (2005), Bhandare and Naik (2015), Madai et al. (2020)

[a] Kinematic viscosity at 30 °C
[b] Kinematic viscosity at 40 °C

7.6.1 Genetic Improvement of Dedicated Biofuel Tree Crops

TBOs are cultivated under different agroclimatic conditions in the forest and non-forest land. However, genetic improvement is limited due to long breeding cycles, late flowering, variable juvenile-maturity associations, insects, pests and diseases, climate, and market changes (Grattapaglia et al. 2018). Large-scale utilization of TBOs for biofuel production involves an increase in the frequency of beneficial genes for several traits at the same time in a synchronized manner in the target population. Genetically improved and quality planting material can be produced through multiple cycles of recurrent selection. This involves increasing the genetic gain per unit time while reducing associated costs. Attempts have been made for changes in oil composition, improved oil yield, enhanced plant growth, early and biannual flowering, and insect, pest, and disease resistance.

Jatropha is a promising biofuel crop; however, it is not domesticated, includes several toxic accessions, and has low productivity (Van Eijck et al. 2014). *J. curcas* is a semi-wild plant species that require at least 15 years through conventional breeding to reach a level of domestication (Maghuly and Laimer 2013). This period could be shortened through plant tissue culture and molecular breeding tools. However, the success of breeding programs depends on high levels of genetic variation. It is therefore important to identify the genetic diversity of *Jatropha* resources around the world for the breeding of better commercial cultivars. Moreover, quantitative trait mapping and association genetics have been ineffective in driving operational marker-assisted selection (MAS) due to the complex multifactorial inheritance of several traits. Grattapaglia et al. (2018) suggest the convergence of quantitative genomics and genetics in contemporary tree breeding programs. The identification of transcription factors and key genes associated with lipid metabolic pathways in *Jatropha* is possible through RNAseq data (Costa et al. 2010). Deconstructing metabolic networks allows the development of natural products and novel molecules with desirable traits. Synthetic Genomics Inc. (SGI) and Asiatic Centre for Genome Technology (ACGT) completed the *Jatropha* genome project which revealed that similar to the rice genome, *Jatropha* genome is about 400 million base pairs in size. Genome annotation facilitates the identification of genetic variation through marker-assisted breeding. It also provides information on alleles regulating oil synthesis, yield, biotic and abiotic stress tolerance, and low-curcin variants (Divakara et al. 2010).

Pongamia is an important potential feedstock in countries such as Australia where it is expected to meet a significant part of the domestic diesel demand. *Pongamia* is predominantly outcrossing resulting in broad genetic diversity evident in several physiological characteristics of the tree including seed size, seed shape, and oil composition. DNA markers have been utilized to assess the genetic diversity of *Pongamia* (Kesari et al. 2010). This is essential for the selection and maintenance of elite accessions for enhanced biomass, oil yield, and stress tolerance (Thudi et al. 2010). Biswas et al. (2013) applied genome sequencing techniques to develop two datasets of *Pongamia* genome consisting of short paired-end "reads" of 36 and 75 bp. Three complete fatty acid biosynthesis genes were identified and sequenced from the leaf tissues of the tree. These reactions involve multiple enzymes which are well described in several plant species (Kazakoff et al. 2011).

Sreeharsha et al. (2016) used the Illumina NextSeq platform for whole transcriptome analysis of *Pongamia* generating 2.8 GB of paired-end sequence reads, and data mined circadian clock and lipid biosynthetic genes. RAPD, AFLP, and inter-simple sequence repeat (ISSR) markers have been used to assess the genetic diversity and variability of natural populations and clonally propagated *Pongamia* germplasm (Kesari et al. 2010). Induced mutagenesis, M2 and M3 selection, RNA interference (RNAi), and virus-induced gene silencing (VIGS) are some techniques for genetic improvement of *Pongamia* germplasm. The fatty acid profile can be modified through up- or downregulating gene expression governing fatty acids biosynthesis. Breeding strategies in TOBs such as *Jatropha*, *Moringa*, and *Alexandrian-laurel* are summarized in Table 7.2.

7.6.2 First-Generation Biofuel Crops

7.6.2.1 Genetic Improvement of Sugarcane Biomass for Biofuels

To date, developing bioethanol from sugarcane has been one of the world's greatest saleable and successful biofuel production systems, with the prospective delivery of second-generation fuels from bagasse and trash resulting in an appreciable positive energy balance at a reasonably low production cost. Sugarcane is one of the most competent crops in the world together with other C4 grasses such as switchgrass (*Panicum virgatum*), Miscanthus species (*Miscanthus* × *giganteus*), and *Erianthus* species (*Erianthus arundinaceus* Retz.) due to high rates of conversion of solar energy into chemical energy and biomass (Hoang et al. 2015). The sucrose concentration ranges between 14% and 42% of the dry weight in sugarcane culm (Whittaker and Botha 1997). However, most of the carbohydrate in sugarcane is the cell wall lignocellulose (cellulose, hemicellulose lignin, and ash) (Pereira et al. 2015). Breeding and biotechnological interventions can improve sugarcane biomass for biofuel production. Among these interventions are: investigating the beneficial traits of sugarcane, exploring existing genetic resources and germplasm, cell wall modification, sugar improvement, and sequencing strategies in dissecting key biofuel traits to improve the biomass composition. In sugarcane, the genetically diverse sugarcane germplasm offers promising opportunities for crop improvement. This genetic variation is found in biomass yield, fiber content, and sugar composition (Hoang et al. 2015).

Saccharum officinarum has a genome size of 7.50–8.55 GB, *S. robustum* has a genome size of 7.56–11.78 GB, and *S. spontaneum* has a genome size of 3.36–12.64 GB, whereas the other three species *S. sinense*, *S. barberi*, and *S. edule* and modern sugarcane are interspecific hybrids whose genome size varies with each cross (Zhang et al. 2012). Fiber (heterogeneous organic solid fraction), soluble solids (sucrose, waxes, and other chemicals), non-soluble solids (inorganic compounds), and water are the four main sections of sugarcane biomass, the composition of which is determined by the industrial process (Canilha et al. 2012; Shi et al. 2013). Sugarcane lignin quantity and composition vary depending on tissue types and stem positions, according to a recent study by Bottcher et al. (2013). With new

Table 7.2 Breeding strategies in tree-borne oilseeds

TBOs	Targeted traits	Breeding techniques/strategies
Jatropha	Agronomic traits: clonal multiplication, component traits for seed yield, sustainable seed yield, flowering duration, Functional Branch Analysis (FBA), plant height, pest and disease resistance Quality traits: seed oil content, kernel protein content, fatty acid profile of oil, toxicity-phorbol esters Reproductive behavior and/or physiological traits: male sterility, floral architecture, chlorophyll content in leaves	Molecular diversity at DNA level, mutation breeding, RNAi techniques, QTL identification for seed and kernel traits, in vitro culture
Moringa	Agronomic traits: pest and disease resistance, traits for dwarf stature Quality traits: seed oil and phytochemicals, increased phytochemical levels in leaves Reproductive behavior and/or physiological traits: synchoronized flowering, pollen biology	In vitro propagation, outcrossing and molecular diversity, genetic diversity Genome-wide genetic marker discovery and generation sequencing, in vitro mutagenesis
Pongamia	Agronomic traits: yield and oil seed productivity traits, traits for seed development mechanism, growth and development traits Quality traits: oil content and oil quality traits, early flowering, carbon sequestration Reproductive behavior and/or physiological traits: traits related to nodule formation and nitrogen fixation, extended shelf life of seeds	Selection and clonal propagation of elite varieties, tissue culture and genetic transformation, genomics and gene discovery, organellar gene identification, high-throughput siRNA profiling, identification of candidate plus trees, molecular diversity, mapping genes, whole genome sequencing and functional genomics
Alexandrian-laurel	Agronomic traits: traits for cold flow properties, physico-chemical characteristics, traits for chemotoxicity, morphophysiological characters for seed and fruit yield, pest and disease resistance, drought tolerance, circadian clock genes Quality traits: oil composition Reproductive behavior and/or physiological traits: in vitro regeneration, seed viability improvement	Genetic diversity, allele mining, mutation breeding, marker aided selection, in vitro genetic conservation, functional genomics and transcriptomics, small RNA for oil biosynthesis, deep sequencing, chloroplast DNA characterization
Rubber	Quality traits: seed yield improvement, seed and oil quality for biodiesel production Reproductive behavior and/or physiological traits: shortening of breeding cycle	Novel gene discovery for seed oil biosynthesis, genetic transformation and Candidate Expressed Sequence Tags for oil biosynthesis, allelic diversity, in vitro propagation of elite candidate plus trees
Neem	Agronomic traits: yield, clear bole, canopy diameter, outcrossing rate for biomass Quality traits: high limonoid and oil content, seed yield and oil quality Reproductive behavior and/or physiological traits: photosynthesis rate, straight bole and crown form, early flowering	Selection of candidate plus trees, hybridization, progeny testing, in vitro techniques, function genomics for oil synthesis

varieties combining these features, genetically modified sugarcane has a lot of potential to contribute to biofuel production.

7.6.2.2 Genetic Improvement of Maize for Biofuel Production

Maize originated in tropical regions and is an impressive crop with tremendous diversity and economically significant characteristics. It is a C4 plant recognized as a prospective source of bioenergy as it features desirable traits like wide adaptability, efficient carbon sequestration, and nitrogen utilization (Hufford et al. 2012). Moreover, unlike other potential biofuel crops, maize is a source of both starch (seed) and cellulosic (stover) material (Torney et al. 2007). With present biotechnological interventions like cell wall functional genomics, protein engineering, and quantitative trait loci (QTL) mapping, maize has great potential as a sustainable second-generation feedstock for meeting bioenergy requirements (Choudhary et al. 2020).

In light of the potential of maize as the future bioenergy crop, there is also a need to dissect genes involved in the biogenesis of the key cell wall components (cellulose, hemicellulose, and lignin) and their regulation in order to improve biomass yield and composition. Research indicates that variation in lignin content responsible for enhanced bioconversion efficiency is heritable and can be promoted and exploited to identify promising maize feedstocks (Torres et al. 2013). A number of QTLs for traits related to cell wall digestibility and lignification have been identified, but a meta-analysis of QTL research is needed to identify the meta-QTL (consensus chromosomal areas) that can characterize actual trait variation (Veyrieras et al. 2007). The potential of maize bioethanol programs using omics platforms has been explained by expression studies supported by microarray research. This reveals a deeper understanding of tissue or stage-specific expression of multigene families linked to metabolic cell wall metabolism genes such as the brown midrib (bm) genes (Guillaumie et al. 2007).

In bioenergy crop research and breeding efforts, accurate phenotyping or screening of genetic variations for the mapping of novel biomass-enhancing features is a major difficulty. Fourier transform mid-infrared (FTIR) spectroscopy has developed as a novel tool for tracking the dynamic changes in cell walls that occur throughout the growth and development of maize coleoptiles. Pyrolysis molecular beam mass spectroscopy is a new method that aids in assessing cell wall chemistry-related qualities based on analytical pyrolysis and thus further advancing the phenotyping of cell wall composition attributes (Sykes et al. 2009). TILLING (Targeting Induced Local Lesions in Genomes) is a reverse genetics technique in which the mutagen Ethyl Methane Sulfonate causes mutations to arise randomly throughout the genome. TILLING populations in maize are currently available therefore they can be utilized to map genes related to bioenergy feedstocks like total biomass and conversion efficiency (Massman et al. 2013). Furthermore, functional genomics plays an important role in gaining a full understanding of the genetic and pharmacological pathways that drive cell wall formation. In this context, maize gene expression research has been investigated for the identification and annotation of several cell wall biosynthesis genes (Bosch et al. 2011). Gene-editing tools like CRISPR-Cas9 can help in the usage of large genetic diversity in maize for biofuel production, avoiding the issues that come with GM technologies (Young et al. 2019). Multipurpose maize hybrids with much higher energy efficiency are better candidates for serving as geared supplements to the current ethanol bioprocessing scenario for long-term lignocellulose-based biofuel production.

7.6.2.3 Genetic Improvement of Sorghum for Biofuel Production

Sorghum bicolor has evolved as a potential candidate for sugar and lignocellulosic biofuel production. Due to its high sugar content and easily extractable nature, sorghum is one of the outstanding feedstock crops. It has comparatively low input needs with capabilities to grow on marginal and arable lands. Energy sorghum, including biomass and sweet type varieties, has recently evoked interest as bioethanol feedstock. Several desirable agronomic traits of sorghum include a short life cycle, adaptability to poor environmental conditions, low input and cost of cultivation, and C4 photosynthetic mechanism. The term “biofuel syndrome” in sorghum refers to a group of sweet sorghum traits associated with biofuel production such as plant architecture, phenology, and biomass conversion efficiency (Anami et al. 2015). Evaluating the biology of specific attributes in sorghum involves the identification of polymorphic genetic loci governing these traits, dissecting them into specific genomic regions, and interpreting the expression profiles, regulation, and functions of the genes (Mathur et al. 2017). Identification of polymorphic genetic loci in sweet sorghum through various marker systems is also gaining importance. Genetic diversity of sorghum has been assessed by several researchers using marker technology: simple sequence repeat (SSR) markers (Mofokeng et al. 2014), amplified fragment length polymorphism (AFLP) markers (Gerrano et al. 2014), and random amplified polymorphic DNA (RAPD) markers (Ruiz-Chután et al. 2019).

Moreover, compared to corn, sweet sorghum utilizes water and nitrogen more efficiently and requires less than 50% total nitrogen to produce equivalent yields of ethanol (Umakanth et al. 2019). The composition of mature sorghum biomass is affected by genotype, environmental conditions, genotype × environment interaction, and photoperiod sensitivity (Byrt et al. 2016). Genetic variation for stalk biomass, Brix and stalk weight has been studied as the result of both additive and nonadditive effects (Felderhoff et al. 2012). Heterosis breeding can be employed for developing high-biomass sweet sorghum hybrids with desirable biofuel-related genetic features. Sweet sorghum bagasse is a lignocellulosic feedstock for the production of ethanol. Brown midrib (BMR) mutants were first induced in sorghum in 1978 (Porter et al. 1978) for enhancing sorghum forage quality. These mutants are characterized by reduced lignin concentration (51% lower in stems and 25% lower in leaves) which improves cellulosic ethanol conversion efficiencies by mitigating any recalcitrance due to lignin (Xu et al. 2018). The highest biomass production in sorghum is ascribed to photoperiod-sensitive genotypes with long vegetative growth. In subtropical and temperate regions, these genotypes remain in the vegetative phase until the day length is shorter than 12 h. There is however limited information about the biomass composition of such photoperiod-sensitive genotypes with prolonged vegetative phases (Byrt et al. 2016).

7.7 Fuel Crops for a Cleaner Environment

The rising human population, global per capita expenditure and dietary requirements while alleviating malnutrition and inequity, are putting tremendous pressure on our agroecosystems. With agriculture reliant on some of the most vulnerable natural resources such as soils, water, and biodiversity, it is important to identify sustainable pathways for holistic resource management. Additionally, with society's reliance on fossil fuels, atmospheric CO_2 levels have risen to dangerously high levels, contributing to global warming and ensuing climate change. Other greenhouse gases such as methane and nitrous oxide are released from the agriculture sector. Simultaneously, rising food and feed demand have put increasing pressures on forest ecosystems. Deforestation results in the loss of carbon sinks, the release of CO_2, as well as biodiversity loss, land degradation, and loss of soil fertility. The fossil economy is characterized by machinery for boosting agricultural productivity, manufacture of fertilizers and pesticides. To reduce, prevent, or even negate the harmful impacts of the fossil economy, civilization must transition to a post-fossil society based on more resilient biological techniques and processes. Plants become the principal source of all organic matter, textiles, food, and nutrition in such a "bio-society," as well as contribute to clean fuel and energy demands without net CO_2 emissions (Harbinson et al. 2021). The Sustainable Development Goals (SDGs), the Paris Climate Agreement (COP21), and the European Green Deal targets will all be easier to achieve if plant traits which are a key part of agriculture are exploited to meet the fundamental needs of societies and economies in terms of food, feed, and biofuel systems (Fig. 7.3).

Our current agricultural, feed, food, fiber, and fuel systems are both propelling and vulnerable to a number of significant risks that jeopardize their long-term viability. Similarly, in order to reach the SDGs, our agri-food/fiber/feed/fuel systems must produce more and do it more sustainably. Plant trait innovation is a way of future-proofing plants and near-term agriculture against risks so that they can contribute to achieving the SDGs.

Among the 17 SDGs, biofuel production is directly related to SDG 13 and SDG 7 (Nazari et al. 2021). SDG 13 aims at taking urgent action to address climate change and its impacts by developing the capacity of countries to mitigate risks and work towards adaptation. SDG 7 aims to ensure access to affordable, reliable, sustainable, and modern energy through specific targets by 2030. These include enhancing the share of renewable energy in the global energy mix, doubling the rate of improvement of energy efficiency, expanding international cooperation, infrastructure, and technology development to increase the supply of modern and

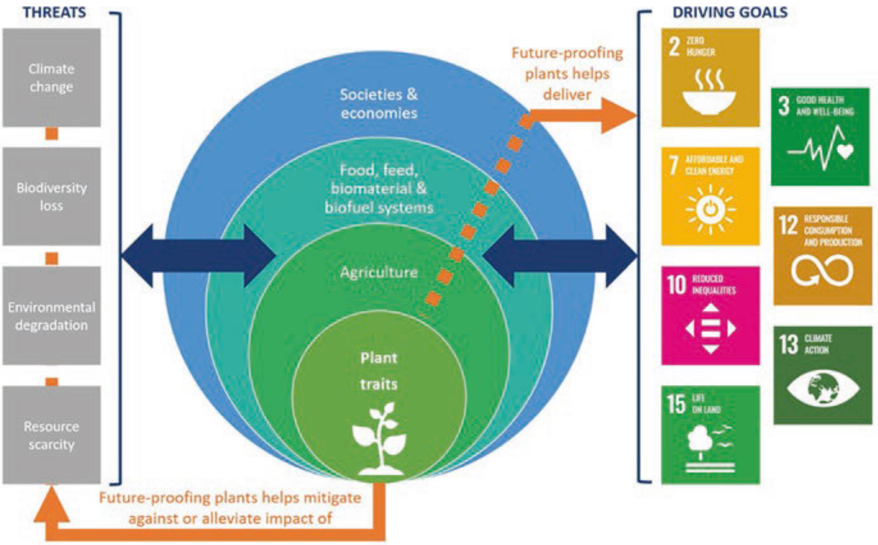

Fig. 7.3 Impact of fuel crops on sustainable development goals. (Reproduced from Harbinson et al. 2021 under the creative commons attribution (CC BY) license privilege)

sustainable energy services in developing countries. Since biofuels are an important alternate source of energy, they will play a key role in the pursuit of these goals.

7.8 Conclusion

The accelerating demand for food, fuel, and fiber in parallel with a growing world population is set against a backdrop of predicted decreasing access to readily available fossil fuels. There is an urgent need to secure future fuel resources through the development of economically viable and environmentally sustainable fuel sources that can meet the demands of industries while mitigating global climate change. Realizing SDG targets related to climate action and clean energy also calls for large-scale, commercial production of alternate fuel sources. Biofuels are increasingly being recognized as valuable renewable sources of energy. However, the key limiting issue for the mass deployment of biofuels across the energy sector is the identification and commercialization of suitable feedstock species. These feedstocks should yield commercially significant amounts of biomass capable of the transformation of cellulosic biomass and fatty acids to relevant biofuels (e.g., ethanol, biodiesel, aviation jet fuel). At the same time, this has to be accomplished with better land-use management strategies that do not pose challenges to food crop production.

Several plants can be used for biofuel production including food and feed crops, trees, and perennial grasses. In order to avert competition with food production, the diversion from food-based biofuels to biofuels produced from crop residues, grasses, and TBOs is rapidly gaining momentum. Moreover, TBOs can be cultivated in a wide range of agroclimatic conditions both in forest and non-forest areas including wastelands. However, most of the emerging biofuel crops are rarely domesticated or are encumbered with problems related to low yield and productivity. Biotechnological interventions can reengineer plants to improve growth, yield, flowering, and insect, pest, and disease resistance in potential biofuel crops. Alteration of the characteristics and properties of nonfood lignocellulosic biomass and oil, and enhancing feedstock conversion efficiency is also possible through genetic manipulation of biological resources. Plant breeders therefore have a pivotal role to play in ensuring that biofuel feedstocks are produced at reasonable costs and in competitive quantities.

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Chapter 8
Microbial Approaches for Bioconversion of Agro-Industrial Wastes: A Review

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Abstract The fact that high levels of agro-industrial pollution are produced due to the linear economy is well established. The circular economy is appropriate for meeting human requirements by converting excessively generated agro-industrial trash into various value-added products (VAP) such as bioenergy, biofuels, biopolymers, and bioactive. Because of its environmentally benign character, the microbial mediated transformation of agro-industrial waste has gained much interest in recent years. This chapter discusses microbial and their enzymatic ways for producing these bioproducts from agro-industrial food wastes as a long-term solution. Pretreatment technologies have been found in recent research to increase enzyme production several-fold. This chapter provides an overview of the agricultural waste utilized as source material, the extraction methods for obtaining VAPs from waste conversion. This chapter emphasizes the environmental benefits of waste utilization by minimizing the amount of garbage disposed of and increasing the generation of green fuels and other green materials; thus, the possibility of achieving a circular economy through environmentally friendly processes that engage agro-industrial wastes.

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8.1 Introduction

Every year, agricultural-based companies generate a large number of wastes. If these wastes are discharged into the atmosphere instead of being properly disposed of, they potentially pollute the ecosystem and threaten animal and human health. Because most agricultural wastes are unprocessed and unused, they are considerable discretion by dumping, burning, or unintentional landfilling. These unprocessed materials impact the environment by raising the emissions of greenhouse gasses produced. Aside from that, the use of fossil resources impacts greenhouse gas emissions (Sharma et al. 2020). As a result, it has become a worldwide threat to mandate the exploration of alternative, better, and sustainable bioenergy supplies. Agro-waste residues are difficult to dispose of; one such scenario is that the juice industry generates a large quantity of waste in the form of agro-residues; the coffee sector contributes residues in the form of pulp husks from the cereal factory. Around 147 MMT of fiber sources are found around the globe; other agro-waste such as rice straws and wheat straw waste were projected to be 709 and 673 MMT in the previous century. Because the constitution of these agricultural and agri-wastes has a higher nutrient potential, these are being given increased attention for quality standards and are being classified as agricultural and agri-byproducts (Beltran-Ramírez et al. 2019; Ma et al. 2019).

Multiple agro-wastes, such as green walnut husks, lemon peels, and pomegranate peels, could be used as antimicrobial substances. While organic chemical residues pose a threat to the environment, they may be a substrate source for mushrooms production and essential bio-based value-added products such as biofertilizers and bioenergy. Animal feed is made from some of the agricultural waste. Furthermore, such wastes include a wide range of compositions, including significant carbohydrates, proteins, and minerals. Because of their high nutritive value, these wastes are not regarded as “trash” but rather “natural resources” for the design and growth of other products (Adámez et al. 2012; Katalinic et al. 2010).

Continuous research is being carried out worldwide on agro-industrial waste for their conversion to VAPs. Bibliography studies carried out on the Scopus database on the keywords “Agro-industry waste” provided a matrix of 3274 documents. Surprisingly, the Top 10 contributed countries are from developing nations, with Brazil and India holding the top two positions (Fig. 8.1). The outcome infers that developing nations focus more on the agro-industrial waste conversion since they would like to solve the issues associated with generating more agro-industrial waste in their nations.

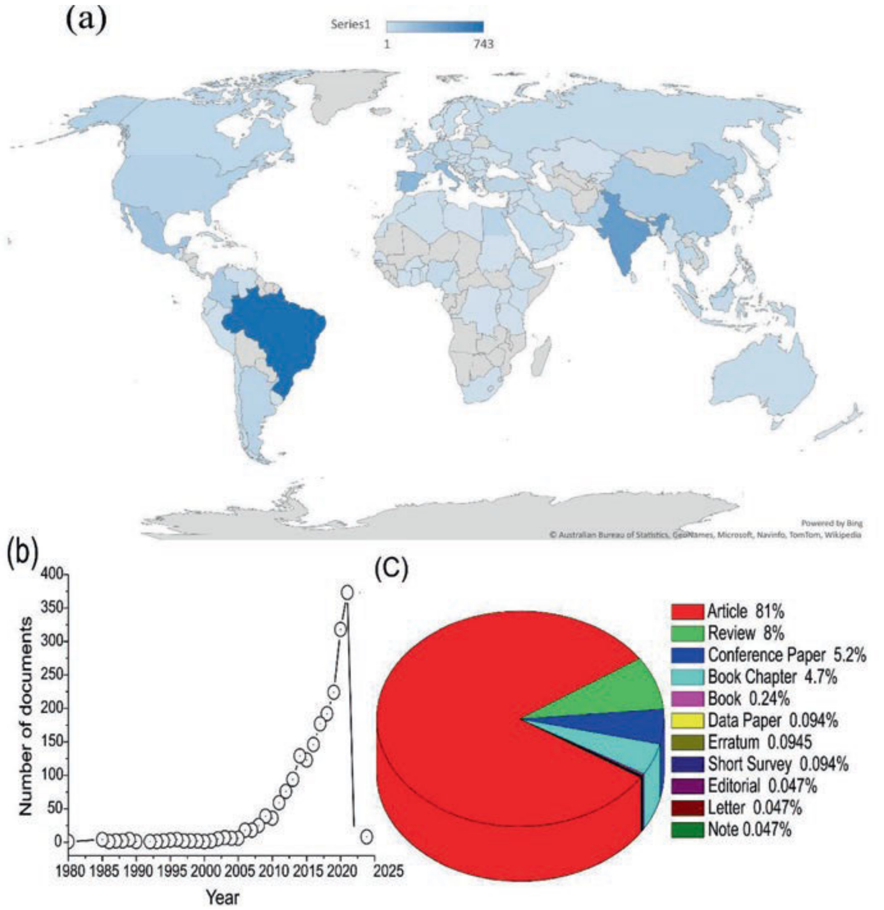

Fig. 8.1 Graphical representation for the Scopus database document for the keywords [TITLE-ABS-KEY (agro-industrial AND waste)]. The number of documents by country-wise (**a**), Number of documents produced by TOP 20 developing countries (**b**), and type of document produced by TOP 20 developing countries (**c**)

8.2 An Overview of Current Conversion Strategies

One-third of the world's most considerable agro-sourced food substance (about 1.3 billion tonnes) is squandered each year. Vegetables and fruit and tubers and roots have the largest percentages of wastage of any agricultural residue and food waste, contributing 40–50% (520–650 MTPA) of global food waste each year. Food waste in the EU totals 89 MTPA (39% of which occurs during production methods), whereas overall agricultural waste accumulation (crop residues or parts of harvested plants that are not consumed as food) is 367 MTPA (Searle 2013). Furthermore, most of the waste is used on farms as livestock feed and feed for horticultural purposes.

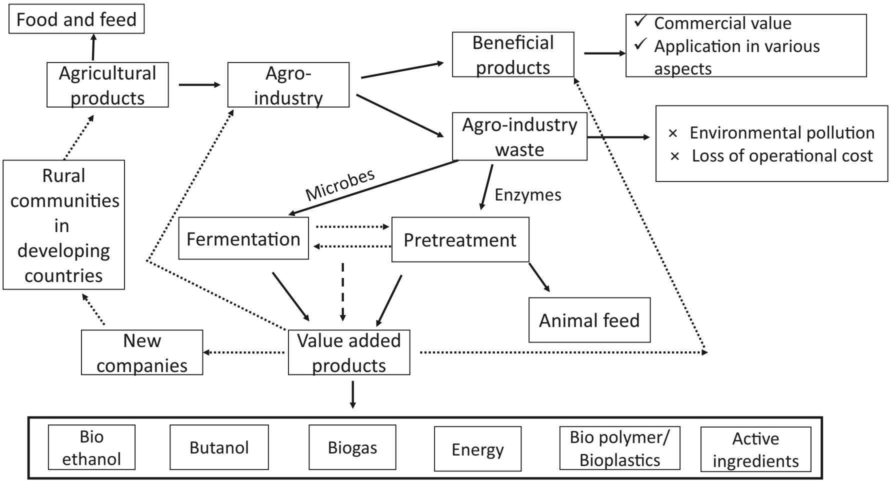

Fig. 8.2 Overview of microbial bioconversion of agro-industrial waste

Much research is needed to assess the effective usage of agricultural residues that lead to possible waste resources for product development during the last several decades. Most research has been carried out in nations where agriculture plays a significant role in the economy. Liquid-fuel-based manufacturing is presently the only commercially viable operation that uses agricultural production residue as raw resources; this is performed using microbial fermentation such as waste digestion followed by production or enzymatic hydrolysis and fermentation both at the same time (Guan et al. 2016). The schematic view of microbial bioconversion of agro-industry waste is depicted in Fig. 8.2. Despite this, researchers began exploring potential alternatives for converting agricultural lignocellulosic wastes into raw resources to manufacture various enzymes (Ravindran and Jaiswal 2016). The conventional techniques of landfilling or incinerating food waste do not alleviate the environmental or economic impact; valorization is increasing. Biogas, ethanol, hydrogen, butanol, biodiesel, and methane are all examples of energy molecules or biofuels generated from food waste (Sharma et al. 2020).

8.3 Bioconversion of Agro-Industrial Wastes for Biofuel Production

8.3.1 Biodiesel

The interest in alternative fuels has grown in response to rising environmental contamination, rising fuel demand, and the exhaustion of fossil resources. Biodiesel is an alternative fuel produced from fats and oils, a form of renewable fuel whose manufacture is costly due to the expense of source; therefore, the prospect of using food waste as a lost cost raw material has been examined to lower costs (Karmee et al. 2015). Direct transesterification with an acid and alkaline catalyst or microbial-based oils generated by oleaginous bacteria can convert food waste to biodiesel (Kiran et al. 2014). Since microorganism-generated oils source has a comparable fatty acid makeup to vegetable oils, they can be used as a biodiesel source material. For the expansion of microalgal species for biodiesel manufacturing, food waste hydrolyzate can be employed as a nutrients source and culture medium. The food waste hydrolyzate was made from *Aspergillus awamori* and *Aspergillus oryzae*, and it was employed as a growth medium for *Schizochytrium mangrovei* and *Chlorella pyrenoidosa* culture, giving 10–20 g biomass. *C. pyrenoidosa* and *S. mangrovei* generated fatty acids that might be used to make biodiesel (Pleissner et al. 2013).

Penicillium expansum and five distinct species of *Aspergillus* sp. have been found to use waste cooking olive oil as a medium for the formation of lipid-rich biomass. *Aspergillus* sp. yielded the highest quantity of fat (0.64 g/g DCW) (Papanikolaou et al. 2011). All around the planet, biodiesel production from animal fats, butter, and plant oils is predicted to yield 24.5 GJ of energy each year (Kiran et al. 2014). Biodiesel manufacturing using the corona discharge plasma reactor method was described by Cubas et al. (2016). Improved esterification, fast biodiesel extraction, and no co-product formation are just a few of the benefits of this method.

8.3.2 Ethanol

Ethanol's significant commercial use has boosted its interest around the world. It is a common feedstock chemical for ethylene synthesis and an essential raw resource for polyethylene manufacture. Formerly, bio-based ethanol was made from starchy crops and cellulose such as sugar cane, potato, and rice. Industrial catalysts can transform starch to glucose, which *S. cerevisiae* can ferment into ethanol, but cellulose degradation is complex (Kiran et al. 2014). As a result, food waste with a low cellulosic concentration is a preferable option for bioethanol production.

Considering severe or hot processing potentially induces substantial destruction of nutritive value and carbohydrates, food waste is appropriately sterilized before fermentation to increase ethanol quality and productivity (Sakai and Ezaki 2006). Because dried food waste has a relatively low surface area, which leads to decreased reaction efficiency between enzyme and substrate, clean and moist food waste was proved to be an excellent source for ethanol synthesis. Furthermore, because yeast cannot convert cellulose or starch to ethanol effectively, the conversion efficiency depends on carbohydrate saccharification (Kiran et al. 2014). A combination of enzymes such as pullulanase, alpha-amylase, beta-amylase, and glucoamylase will be added for high molecular weight sources. In 48 h of incubation, Hong and Yoon (2011) observed that 100 g of food waste produced 36 g ethanol and 60 g reducing sugar. Kitchen trash and non-diluted food waste were simultaneously saccharified and fermented, yielding 17.7 g/L/h and 0.49 g ethanol/g sugar (Ma et al. 2019).

Several nations had established pilot facilities to convert food waste to bioethanol, including Finland, Spain, and Japan (Kiran et al. 2014).

8.3.3 Biohydrogen

Hydrogen is a promising sustainable energy source because it has 142 MJ/kg (Jarunglumlert et al. 2018). Although hydrogen is not available in large quantities, it can be produced utilizing primary renewable resources. Hydrogen is extensively used as a substitute for fossil fuels all around the globe since it is carbon-free, has the maximum power generation, and releases water when used (Nikolaidis and Poullikkas 2017). Biohydrogen is hydrogen gas that is created through biological activities. Biohydrogen generation by fermentation utilizing food waste uses less energy and reuses waste; hence, it became the most used approach.

Carbohydrate-rich food residue is favored for biohydrogen energy production since it has a 20-fold significant possibility than fat and protein-based food waste. Multiple fermentation techniques have been documented for producing hydrogen from food waste, including batch, continuous, semi-continuous, multiple stages, and single stages (Kiran et al. 2014). Aeration tank anaerobic digester blanket and anaerobic sequencing batch processors were used with high methane-producing fractions due to their high biomass concentration in the reactors (Karthikeyan et al. 2018). solid retention time (SRT), organic loading rate (OLR), and Hydraulic retention time (HRT) all influence production of hydrogen in such operations. In scenarios to reduce methane yields, the fermentation process is controlled to a low HRT with extreme acidic environments, similarly for organic material breakdown. In the dark fermentation process, biohydrogen production from the butyrate, glucose, and acetate systems utilize one sugar molecule to create two and four molecules of biohydrogen, respectively (Karthikeyan et al. 2018). Minimizing microbial biohydrogen consumption is prudent; heat is applied to seed biomass while inhibiting lactate formation and increasing biohydrogen production. Lactobacilli are common in unprocessed food waste, but biohydrogen producers predominate in pretreated food waste (Kiran et al. 2014).

8.3.4 Methane

Production of methane using anaerobic conditions is preferred due to its use of alternative energy sources, little residual waste output, and low cost. This method produces bioavailability and nutritive-rich agro-waste that can be utilized as a natural fertilizer or fertilizer (Kiran et al. 2014). Methane has a 55.5 MJ/kg energy content. Production of methane involves anaerobic treatment by decomposing organic waste and decreasing it. Nutrients, reactor type, alkalinity, pH, operation temperatures, ammonium ions, carbon/nitrogen ratio, organic loading rate, nutrients, volatile fatty acids, reactor type, and substrate properties all influence methane synthesis in this process (Park et al. 2018). The methane gas generation is reduced during the anaerobic process due to a decrease in pH and the aggregation of volatile fatty acids (Chen et al. 2018). Compared to solely using an anaerobic fermentation reactor, Park et al. (2018) found that integrating a microbial fuel cell with anaerobic digestion (AD) reactor boosts the methane production rate by 1.7 times. Microbial electrolyte cell enhances methane generation by boosting the speed of degradation of volatile fatty acids, nondegradable organic detritus, and concentrated organic residues. When electrolytes pass a voltage drop in the reactor throughout this process, electrons produced by exoelectrogenic bacteria form methane at the cathode.

Alkali treatment, acid, grinding, biological treatment, heat, microwave-assisted, high-pressure treatment, high-voltage pulse discharge, and micro-aeration are typical pretreatment methods (Karthikeyan et al. 2018). The optimum pretreatment procedures for anaerobic fermentation of food waste are thermal treatment followed by alkali treatment. Alkali pretreatment increased methane yield by 25%, and when paired with heat processing, the yield increased by another 32% (Naran et al. 2016). Agro-waste sources of 54 fresh vegetables and fruits residue can produce 180–732 mL/CH_4/g volatile substance. In two different steps of an anaerobic digester, fruit and vegetable wastes yielded 530 mL/g volatile solid utilizing 95% of volatile materials (Kiran et al. 2014). Zhao et al. (2017) looked at how varying salt (NaCl) contents affected agro-waste. Low salt levels boosted acidification and digestion while suppressing methanogenesis, whereas high salt levels stifled both mechanisms.

8.3.5 Biobutanol

Term butanol is four-carbon alcohol, a well-refined biofuel source than ethanol because it has significant benefits over ethanol, including better lower vapor pressure, higher energy density, and combustion efficiency the solubility in vegetable oils (Girotto et al. 2015). Biobutanol is conventionally made from sugarcane molasses and other starch (wheat, corn, and potato) by a fermentation process. The cost of the agro-waste substrate source accounts for nearly half of the cost of manufacture and has a significant impact on the economic sustainability of biobutanol production via fermentation (Ujor et al. 2014). The effectiveness of species for generating biobutanol utilizing food waste as a substrate source has been documented in most researches (Ujor et al. 2014; Girotto et al. 2015). *Clostridium acetobutylicum* used food waste to create biobutanol during the fermentation process. Butanol provided 0.3 g/g of carbs in lactose-rich waste whey (Girotto et al. 2015). Biobutanol concentrations were similar in carbohydrate-rich industrial food waste such as inedible dough, bread liquid, and batter liquid (Ujor et al. 2014).

8.3.6 Electric Power Generation

The majority of the food waste produced was managed using inefficient methods such as incineration, composting, and landfilling, harmful gas emissions, and groundwater pollution (Paritosh et al. 2017). As a result, using food waste to extract energy or electricity is regarded an environmentally benign, cost-effective, pollution-reducing, and long-term solution. Benefits can be accomplished by processing the agro-waste anaerobically in devices like microbial fuel cells (Li et al. 2017). Microbes are used as catalysts in MFC to recover power generated from various wastes, including surplus sludge, industrial effluents, and domestic wastewater. In an MFC, microbes oxidize organic compounds, giving electrons to the anode, while the oxidized chemicals or oxygen are reduced at the cathode, either by microbial or abiotic processes (Cercado-Quezada et al. 2010). “Electricigens” in Microbial fuel cells consume organic agro-waste as an alternative fuel, and hydrolysis of the organic fraction is the rate-limiting step in generating power energy (Li et al. 2017). Microwave and sonication preprocessing of food waste have been shown to improve substrates degradation, improving power generation efficiency (Yusoff et al. 2013).

8.3.6.1 Strategies for Conversion of Agro-Waste for Bioelectricity Generation

In the hydrolysis reaction, essential characteristics of biomass residue components such as hemicellulose, lignin, and cellulose are crucial. Cellulose is a part of lignocellulose with a linear polysaccharide with monomers glucose range of 7000–15,000 molecules and 1–4 beta glycosidic connections. Hemicellulose, on the other hand, is a branching polysaccharide composed of polymers range of 500–700 molecules such as mannose, glucose, xylose, arabinose, and galactose linked by 1–4 beta glycosidic bonds in the framework (mainly xylose) and 1–2,1–3,1–6 alpha bonds in the branches (primarily arabinose). Hemicellulose is amorphous, whereas cellulose is crystalline, and the beta bond is stronger than the alpha bond (Khodayari et al. 2021).

Lignin organic molecule is a non-polysaccharide cross-link polymer that supports the other two by incorporating phenyl propane units such as conniferyl, synapyl, and *p*-coumaryl alcohols. Hemicellulose is quickly metabolized by a weak acid or base, and lignin is exceedingly resistant to breakdown (Shrotri et al. 2017). The fundamental issue with the anaerobic digestion process for producing biogas or the fermentation reaction for producing bioethanol is the need for preprocessing before digestion of lignocellulosic biomass. Because the intricate character of the bonding and interlinking of lignocellulose constituents makes it extremely difficult to digest it into monomers, it is required to utilize a pretreatment approach before the hydrolysis process. The pretreatment method is an effective way to (1) weaken bonds, (2) reduce cellulose crystallinity, (3) eliminate lignin from the complex mixture, (4) remove hemicellulose, (5) swell biomass, (6) increase internal surface area, and (7) increase cellulase accessibility to cellulose fibers (Yu et al. 2019).

Pretreatment of lignocellulosic biomass is required, as previously stated; the four types of pretreatment methods commonly used are (1) physical, (2) chemical, (3) physicochemical, and (4) biological. The merits and demerits of these general pretreatment methods are summarized in Table 8.1.

8.3.6.2 Biological Pretreatment

Primary treatment criteria: temperature 25–30 °C, which varies depending on the type of microorganisms used; incubation period varies depending on the type of biomass and microbial strain used, and incubation timing hours to 6 days; Water and pH is specific to the type of microorganism; for example, white-rot fungus require a moisture content of 70–80% and a pH of 4–5. Regulated aeration is vital for increased delignification if the microbes utilized are aerobes; inoculum concentration—a tiny quantity of inoculum may take longer to colonize, while a big lot may be needless; this can be accomplished by determining the correct amount of inoculation to be used.

Some microorganisms, such as fungi and bacteria, have been found to break down lignocellulosic material. Although slow, the biological technique has the benefits of being ecologically friendly, low energy-intensive, and effective. It is a frequently used process as a result of these exemplary aspects. Before anaerobic codigestion with cow dung, Akyol et al. (2019) utilized *Trametes versicolor* (white-rot fungi) to pretreat rye straw, wheat, and barley. For aerobic processing of organic wastes and manure waste in a 1:1 ratio with suspended *T. versicolor* spores, 250 mL conical Flask flasks were employed with distilled water to keep 75% of moisture content. pH was 4.2 adjusted and the medium was maintained for 6 days at 25 °C with vigorous agitation at 135 rpm. The entire pretreatment residue was fed to a bioreactor for additional anaerobic digestion after fungal processing. Cow manure is used in both the pretreatment and anaerobic digestion stages of this process. The methane yield was enhanced by 10–18%, depending on the feedstock.

On rice straw, examined the efficacy of biochemical preprocessing state. Their experiment used five alternative preprocessing conditions: CaO, AS (ammonia solution), CaO-AS, LFD (liquid fraction of digestion that contains microorganisms), and CaO-LFD. The effect of anaerobic digestion on the physical and chemical characteristics of rice straw after pretreatment was examined. The Cao-LFD biochemical processing strategy was found to be the more influential among the various pretreatment conditions. Cao-LFD preparation of rice straw resulted in a 21% increase in lignin removal compared to the control sample. Rice husk that had been pretreated with CaO-LFD produced 58% more biogas than rice husks that had not been pretreated. In mixture CaO-LFD preprocessing samples, the lignin removal efficiency was 102% and 37% higher, respectively, than in simply CaO or LFD pretreated samples which suggests that combination preprocessing for lignin removal has a high specificity (Guan et al. 2018).

Table 8.1 General pretreatment methods for agro-waste

Pretreatment type	Advantages	Disadvantages
Physical method		
Mechanical comminution	Decrystallization of cellulose transpires	Lignin and hemicellulose were retained. Higher quantum of energy usage
Hydrothermal	Noncorrosive procedure	Ineffective for biomass with maximal lignin
Irradiation	Depolymerization of cellulose and solubilization of lignin. Easy operation with maximal effectiveness	High economic cost
Chemical method		
Acid pretreatment	Functioned at modest state with maximal pentose sugar generation. Depolymerization of cellulose with solubilization of hemicellulose up to 90%	Lignin not digested; corrosive and hazardous, generation of unwanted residues like furfurals
Alkaline pretreatment	The ester bonds in the lignin complex are detached sufficiently and with 50% lignin solubilization	Reagent expensive, extended pretreatment time, creation of inhibitory composites, involvement of higher water usage
Ozonolysis	Effective delignification at trifling circumstances	Extended dosages of ozone required
Organosolv pretreatment	Effective lignin elimination with maximal xylose production	Reagent expensive, not appropriate for commercial scale
Physicochemical method		
Stream explosion	Causes swelling of biomass; hence hemicellulose solubilization could be up to 80–100% High-energy efficiency	Depolymerization of cellulose is not significant. Due to xylan degradation, undesirables are formed
Ammonia fiber explosion (AFEX)	Decrystallization of cellulose, 20% solubilization of lignin, 60% solubilization of hemicellulose. Xylan reserved without inhibitors creation	Ineffective for high lignin biomass. Corrosive chemicals used
Supercritical CO_2	Lower temperature, no residues formed, cost-effective, upsurges the accessible surface area feasible for enzymatic hydrolysis	Expensive, very high pressure is compulsory, effective for high lignin biomass.
Wet oxidation	Lignin (40%) and hemicelluloses (90%) are effectually detached	Not operative for high lignin biomass
Biological method		
Microbial Delignification Soft-rot fungi Brown-rot fungi White fungi	Lower energy usage, feasible lignin removal, no unwanted residues formed, environmentally benign	Procedure is trifling, sluggish proportion of hydrolysis with forfeiture of cellulose and other monosaccharides.

8.3.7 Enzymatic Bioconversion of Agro-Industrial Wastes

Natural microorganisms are frequently used to hydrolyse and biologically transform organic wastes to turn them into value-added goods. These bacteria can break down the refractory structure of most biomasses by using enzymes like hydrolytic enzymes (which depolymerize lignin) and hydrolytic enzymes (such as hydrolases) (Masran et al. 2016). As a result, the preprocessing phase in the extraction and purification process is crucial because it prepares the residual biomass for the purification step, such as hydrolyzing. In microbial fermentation, microorganisms that hydrolyze hemicellulose and cellulose to create monomeric glucose are called cellulolytic and hemicellulolytic microbes. Enzymes secreted by these microbes can produce reducing sugars, or pure catalysts can convert preliminary materials to microbial fermentation molecules. Subsequent degradation of these items produces bioproducts such as lactate, acetate, organic acids, enzymes, and biofuels such as methane, hydrogen, and ethanol (Tsegaye et al. 2019).

Microbial pretreatment uses bacteria to break down biomass, whereas enzymatic bioconversion uses isolated enzymes generated from the same microorganisms. Hydrolases, which belong to the EC 3.2 class of enzymes, have been successfully employed to break down waste biomass such as corn cob, paddy straw, maize straw, sugarcane bagasse, and rice husk. Several pieces of research on the hydrolysis process of diverse biomass sources, such as laccases, xylanases, peroxidases, glucosidase, and cellulases, are all members of this group. Hydrolytic enzymes account for 75% of all microbial enzymes manufactured today, and they are utilized to make paper, feed, deinking, biofuels, pulp, food, and wastewater treatment, among other things (Usmani et al. 2021). Some of the enzymes and the corresponding substrates utilized for the transformation are highlighted in Table 8.2.

8.3.7.1 Cellulases

Exoglucanase, endoglucanase, and beta-glucosidase are the three enzymes that make up cellulase. Exo- and endoglucanases hydrolyze the glycosidic linkages in cellulose to liberate cellobiose. The cellobiose is then cleaved into glucose by the bet-glucosidases. Trichoderma, Aspergillus, Cellulomonas, Clostridium, and Thermomonospora are among the microbial origins of these enzymes (Yadav 2017). *Clostridium acetobutylicum* produced biobutanol/g glucose (0.16 g) when digested glucose-rich hydrolysate (Rahnama et al. 2014). Rice husk was pretreated with NaOH and then processed with enzyme production from *Trichoderma harzianum* to create a glucose-rich hydrolysate. *T. reesei* RUT produces cellulase, which can catalyze the hydrolysis rice straw and convert it to glucose, and it flourishes on rice husks media. A bioethanol yield of 0.093 g/g of processing rice husk is produced (Sukumaran et al. 2009). Delignifying rice straw with industrialized pectinase and cellulase resulted in higher total reducing sugars generation.

Table 8.2 Microbial strains used in biomass degradation and enzyme secretion

Enzyme	Fungal strain	Bacterial strain	Substrates
Laccase	*Trichoderma reesei*	*Pseudomonas* sp.	Wood waste
Manganese peroxidase	*Trichoderma longibrachiatum*	*Bacillus subtilis*	Paper waste
Lignin peroxidase	*Phanerochaete chrysosporium*	*Azospirillum lipoferum*	Winery biomass waste, water hyacinth
Versatile peroxidase	*Merulius tremellosus*	*Raoultella ornithinolytica*	Aspen wood waste, sawdust, wheat straw
	Ganoderma applanatum	*Pseudobutyrivibrio* sp.	Wheat straw, brewery spent
	Dichomitus squalens	*Cupriavidus basilensis*	Wood waste, rice straw
β-Glucosidase	*Penicillium helicum*	*Streptomyces* sp.	Citrus waste, sugarcane bagasse
Endoglucanase	*Trichoderma viride*	*Thermomonospora* sp.	Wastepaper, food waste
Glycosyltransferases	*Trichoderma reesei*	*Bacillus* sp.	Wood waste
Exoglucanase	*Aspergillus niger*	*Cellulomonas* sp.	Sugarcane bagasse, date seeds
	Fusarium merismoides	*Clostridium* sp.	Maize waste, clavel leaves, oats lamella
	Aspergillus awamori	*Ruminococcus flavefaciens*	Biomass waste, grape pomace
Xylanase	*Pleurotus ostreatus*	*Ruminococcus flavefaciens*	Tomato pomace, rice bran, wheat straw
β-Glucosidase	*Aspergillus oryzae*	*Bacillus* sp.	Paper waste, wood waste
Acetyl esterase	*Aspergillus fumigatus*	*Fibrobacter succinogenes*	Rice straw, wheat straw
α-Galactosidase	*Piptoporus betulinus*	*Pseudobutyrivibrio xylanivorans*	Birchwood, rapeseed
Endoglucanase	*Aspergillus niger*	*Mucilaginibacter* sp.	Sugarcane bagasse
Mannanase	*Magnaporthe grisea*	*Pedobacter* sp.	Plant biomass waste

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8.3.7.2 Laccases

Laccases are commonly employed in a variety of industries to process organic wastes and produce value-added goods. Copper-based oxidases enzyme laccases that catalyze the oxidation of the aromatic compound and also phenolic amines. Laccase-mediated phenol remediation boosts microbial multiplication, increases digestion capacity, and speeds up the whole mechanism (Oliva-Taravilla et al. 2015). The lignin source used in conjunction with other industrial enzymes has been shown to degrade lignocellulosic biomass successfully. The interplay between hydrolytic and oxidative enzymes in maize cob degradation utilizing *Sporothrix carnis* (Ogunyewo and Olajuyigbe 2016).

Researchers discovered that the early biomass hydrolysis (96 h) was caused by the production of essential hydrolytic enzymes and that this was proceeded by subsequent hydrolysis (144 h) caused by laccases and peroxidases. Combining cellulosomes with dockerin-infused laccase enzyme produced by *Thermobifida fusca* invigorates the effect on wheat straw hydrolysis (Davidi et al. 2016). In comparison to not utilizing laccase in preprocessing, they observed a two-fold increase in resulting reducing sugars. Using dockerin pumped laccase in conjunction with miniCbpA complex on wheat straw waste resulted in a 2.6-fold increase in decreasing manufacturing of sugar molecules (Hyeon et al. 2014).

8.3.7.3 Xylanases

Humicola insolens, *Bispora* sp. *Thermoactinomyces thalophilus*, and *Bacillus* sp. based xylanases enzyme are used in many sectors, including removing xylans from producing pentose, kraft pulp, converting hemicellulose resource waste into energy products, and brewing (Yadav 2017). When molasses is processed with a commercial xylanase enzyme source isolated from *Trichoderma viride* species, it yields XOS (1.15 mg/mL). Alternatively, the xylan recovered from biomass can be used as a substrate to create XOS. Purified form of xylanase enzyme derived from *Aspergillus foetidus* can likewise be used to make XOS from maize cob xylan (Chapla et al. 2012).

8.4 Other Value-Added Products

8.4.1 Biopolymers/Bioplastics

Still now, polymeric substances such as exo- and endopolysaccharides, polyphosphates, and polyhydroxyalkanoates were produced from various agro-industrial waste by the action of various bacterial and fungal species (Moradali and Rehm 2020). All those biopolymers derived from agro-industrial waste have specific characteristics such as biodegradability, biomimicry, biocompatibility in food, feed, and biomedical-based industrial applications (Philippini et al. 2020). Few recent works of literature show promising outcomes for the production of biopolymers from various agro-industrial waste. Chitosan was produced from Corn steep liquor and papaya peel juice mixture after bioconversion by Mucorales fungi (Berger et al. 2018). Similarly, *Aureobasidium pullulans* LB83 was used to convert sugarcane bagasse for production polymer called pullulan (Hilares et al. 2019). Similarly, *Xanthamonas campestris* NCIM 2956 and *Xanthamonas campestris*, xanthan gum produced from Cassava bagasse, Chicken feather peptone, and Sugar beet molasses, respectively (Sujithra et al. 2019; Ozdal and Kurbanoglu 2019).

In recent times, by using various agro-industrial waste such as wheat bran, corn cob, rice bran, etc., used for the production of bioplastics such as polylactic acid (PLA), polyhydroxybutyrate (PHB), polybutylene succinate (PBS), Polyhydroxyalcanoates (PHA) by the microbial fermentation (Nayak and Bhushan 2019).

Likewise, high biocompatible PHB was produced by using *Bacillus* sp. C1 via Solid-state fermentation (SSF) (Pati et al. 2020). Similarly, hemp hurd feedstock is utilized to produce PHB by *Ralstonia eutropha* (Khattab and Dahman 2019). Arumugam et al. (2020) produced PHA from Cashew apple juice by the action of *Cupriavidus necator*. A microbial strain *Cupriavidus necator* strain A-04 produced PHB from Pineapple residue as feedstock (Sukruansuwan and Napathorn 2018). Likewise, microbial conversion of waste such as paper hydrolysate (Al-Battashi et al. 2019), Cider byproducts (Urbina et al. 2018), Brewery's spent grain (Mendez et al. 2018), Sugarcane molasses (Rathika et al. 2019), and Spent coffee grounds (Kovalcik et al. 2018) were used as feedstock for production PHA. Various byproducts derived from agro-industrial waste are reported in Table 8.3.

Table 8.3 Value-added products from agro-wastes and their applications

S. no.	Value-added products	Sources	Applications	References
1.	Antioxidant	(a) Potato peel (b) Grape's stalk, grapes marc, grapes seeds	Used as preservatives in foods and cosmetics	Amado et al. (2014) Barba et al. (2016)
2.	Bioethanol	Leaf waste of pineapple	Alternate fuel	Chintagunta et al. (2017)
3.	Biobutanol	*Amorphophallus konjac* waste	Used as fuel for internal combustion engine.	Shao and Chen (2015)
4.	Biodiesel	Palm oil waste and coconut meal residue	Alternate fuel	Thushari and Babel (2018)
5.	Biogas	Food waste	Alternate fuel	Deepanraj et al. (2017)
6.	Bioelectricity	Orange peel waste	Alternate fuel	Miran et al. (2016)
7.	Biopolymer—poly (vinyl alcohol-co-ethylene)	Tomato plant powder and food waste from urban areas	Supporting films for packaging material	Nistico et al. (2017)
8.	Pectin	Tomato peel waste	Used as corrosion inhibitors	Grassino et al. (2016)
9.	DHA (docosahexaenoic acid)	Byproduct from brewery industry and potato chip processing industrial liquid residues	Dietary supplement	Quilodran et al. (2010)
10.	Glucose	Potato peel waste	Dietary supplement	Kumar et al. (2016)
11.	Xanthan gum	Kitchen waste		Li et al. (2017)
12.	Collagen	Residues such as fish waste, broiler chicken processing waste	Biomedical applications	Munasinghe et al. (2015)

(continued)

Table 4.1 (continued)

S. no.	Value-added products	Sources	Applications	References
13.	Epoxy resin blends and composite	Waste vegetable oil	Biomedical applications	Fernandes et al. (2017)
14.	High fructose syrup	Beverage waste	Nutrient supplement	Haque et al. (2017)
15.	Gluconic acid	Sugarcane molasses	Broader application in food, pharmaceutical, textile, and leather industries	Sharma et al. (2008)
16.	D-Tagatose	Onion waste and onion juice residue (OJR)	Food application	Kim et al. (2017)
17.	Vinegar	Pineapple waste	Food preservation	Roda et al. (2017)

8.4.2 *Bioactive Ingredients*

Many bioactive compounds have been extracted from agro-wastes in recent years, especially in the fruits and vegetables processing industry. By comparing agro-waste, these two industries generate a considerable volume of solid waste, which contains a variety of diverse molecules with a wide range of biological activities. For example, dry citrus peel waste consists of approximately 3.8% D-limonene (Pourbafrani et al. 2010) and flavonoids, like hesperidin, naringin, nariturin, and eriocitrin (Chen et al. 2017), and these bioactive ingredients widely used in various sectors of food, pharmaceutical, and cosmetic industries.

Likewise, grape skins have significant amounts of tannins (16–27%) and bioactive polyphenolic compounds (2–6.5%), such as catechins, anthocyanins, proanthocyanidins, quercetin, ellagic acid, and resveratrol (Martinez et al. 2016). Total polyphenols in coffee byproducts (1.5%), silver skin (25%), spent coffee grounds (19%) were reported (Campos-Vega et al. 2015), and extracts of coffee byproducts contain chlorogenic acids, and it serves as an excellent antioxidant, anti-inflammatory, and anti-allergenic activities (Zuorro and Lavecchia 2012).

8.5 Emerging Methods for Extraction of Byproducts from the Microbial Conversion of Agro-Industrial Residue

8.5.1 *Ultrasound-Assisted Extraction*

The ultrasonic wave creates microbubbles inside the material's cell wall in Ultrasound-Assisted Extraction (UAE). The disintegration of the cell wall is associated with the production and collision of these massive bubbles, which increases the extracting process' mass transfer (Chuyen et al. 2018). Food waste can be turned into high-end items using ultrasound, which also helps the environment. Banozic et al. (2019) employed UAE to recover biologically active compounds from commercial tobacco residue (trash, midrib, and dust). Both the leaves and the tobacco residue were used to separate caffeic acid (10.8 and 2.34 g/L), Solanesol (598.9 and 294.9 g/L), rutin (93.7 and 11.56 g/L), and chlorogenic acid (804.2 and 3.64 g/L).

Ultrasonic separation has evolved into a fast, rapid, wide distribution, and successful process for separating antioxidants from discarded coffee grinding in a small amount of time and with the lowest energy use. From coffee waste, 33.84 ± 0.59 mg gallic acid equivalent per gram, total phenol content, protocatechuic acid, 0.53 ± 0.02 mg/g, 5.04 mg quercetin equivalent per gram, 1.43 mg/g chlorogenic acid, and 0.03 mg/g total flavonoid were extracted under solid/liquid ratio at 40 °C and perfect conditions of 244 W ultrasonic frequency, 1.17 g/L in 34 min separation. UAE has been identified as a technology with the greatest potential to change traditional extraction techniques at similar input costs (Al-Dhabi et al. 2017).

8.5.2 Microwave-Assisted Extraction

Electrostatic interaction and dipole-dipole spin of inner particles generate heat within the substrates during Microwave-Assisted Extraction (MAE). The cellular membranes lyse due to the heat produced, enabling biomolecules to flow opposite ends of the cell and into the separation process (Chuyen et al. 2018). Polyphenols can be extracted from the skin of a ripe mango using MAE and deep eutectic solutions. Under optimized condition (436.45 W power, 59.82 mL/g liquid-to-solid ratio, 19.66 min), 2,2-diphenyl-1-picrylhydrazyl scavenging was found to be 82.64 mmol DPPHs ferric reducing antioxidant power was found to be 683.27 mmol ascorbic acid equivalent gram per dry weight, total antioxidant content activity was observed to be 56.17 mg gallic acid equivalent per gram dry weight (Pal and Jadeja 2020). Under optimum conditions, Plazzotta et al. (2020) were able to extract anthocyanins (8 mg CGE), antioxidant activity (2.1 mg Trolox equivalent), flavonoids (94 mg quercetin equivalents), polyphenols (309 mg GAE) from 100 g dry matter frozen peach wastes (540 W, 50 s).

Especially compared to specific other extraction techniques, MAE extracts 108 mg of vitamin C from fruit peels in half of that time (50 s). The carotenoid-rich Granular activated carbon fruit oil company manufactures *Momordica cochinchinensis* skin as waste material. Chuyen et al. (2018) discovered that a microwave pretreatment of 120 W for 25 min was the most effective in removing carotenoids from Gac citrus fruit. Since MAE has a short processing time, it has been developed to remove various bioactive components from a range of wastes. Garrido et al. (2019) employed MAE to extricate phytochemical compounds and polyphenols from Chardonnay grape debris throughout a 10 min separation utilizing 48% alcohol as the solution and 1.77 g of solid mass. Epicatechin, procyanidin catechin accounted for the proportion of the world phenolic synthesis, which was 1.21 mg galacturonic acid equivalent per milliliter. Pectin is a polysaccharide that acts as a cement among plant cells and the middle lamella. It has a variety of uses in food manufacturing, pharmaceuticals, and cosmetics. When compared to traditional procedures, MAE extraction of pectin is a potential strategy for obtaining higher yields.

8.5.3 Homogenizer in Food Waste Valorization

Technology in homogenization pressure is a more environmentally friendly and long-term separation method that uses less energy and emits fewer pollutants. It is mainly utilized to extract bioactive chemicals from plant tissues using homogenizer aided extraction (HAE) or turbolysis (Mesa et al. 2020). For solid-liquid extraction in homogenization, a high rotation speed is used, which generates strong shear force for a short duration with increased temperature to tear cell walls (Zuin et al. 2020). High-energy processing processes include elevated homogenization, high-pressure homogenization (HPH), ultrahigh-pressure homogeneity (UHPH) (400 MPa or more), high dynamic pressure (50–300 MPa), and conventional homogenization (0–50 MPa). It can be used for (a) having to release subcellular substances into the system, (b) regulating the design and composition of bioactive substances (polyphenols) and biological macromolecules (proteins, fibers, enzymes), (c) crystallite size reduction, and (d) homogeneous particle (droplets, globules, aggregates) allocation throughout the fluid system, all while maintaining the nutritive benefits of heat-labile food (Mesa et al. 2020).

8.5.4 Bioreactor Assisted Valorization

A fermenter is a dynamically stimulated container/vessel that supplies biochemical and biomechanics demands of various microorganisms in a bioactive condition, allowing optimum bacteria to be grown in a controllable environment at a commercial scale while producing a high output (Musoni et al. 2015; Pino et al. 2018). The bacterium can utilize organic waste as a source for optimum growth and development and bioactive compound production when the humidity, temperature, water activity, and pH parameters are fulfilled (Singhania et al. 2009).

Newly constructed bioreactor mediums have become widely present in recent years and are widely used in cultivation. Microbial-assisted valorization of agricultural solid waste into products with high value is of great interest since it contributes significantly to the long-term control of food waste and the bioeconomy life cycle. Microbial growing in a bioreactor medium is an exciting prospect that has the opportunity to convert food waste into profit. On the substrate, multistage techniques are used to turn food residue into a product with high value. Researchers used *Caldicellulosiruptor saccharolyticus* to produce biohydrogen and biomethane from food waste utilizing a two-stage method that included mesophilic anaerobic digestion and hyper-thermophilic dark fermentation (Abreu et al. 2019).

The conclusion is that converting agro-waste into product energy yields of 24.4 MJ/kg, similarly to other residual organic waste and gases, necessitates additional treatment. As a result, a three-stage method (Hydrolysis method, Acidogenesis method, Methanogenesis method, and Composting method) was developed, promising to improve resource extraction efficiency. In multiple approaches, codigestion was performed in a Leach Bed Reactor (LBR) before hydrogenation and acidogenesis. Airlift bioreactors are effective in the methanogenesis process since they utilize off-gases from LBRs and solid waste to boost methane recovery. Zero waste is generated through a three-stage procedure (Chakraborty and Mohan 2019).

8.5.5 *Enzyme Immobilization-Assisted Valorization*

Food residue can be used as a resource for enzyme conversion to high-value products such as biodiesel, pectin, biofuel, biosurfactants, prebiotics, oligosaccharides, and food stabilizers because it is abundant in natural materials. On a large scale, this process has traditionally been based on chemical catalysts, which consume much energy, have limited selectivity, and are not environmentally friendly (Bilal and Iqbal 2019; Andler and Goddard 2018). As a result, microbial enzyme catalysts are now often used in bioproducts manufacture; in contrast to chemical catalysts, Enzyme catalysts have various advantages, including minimal energy consumption, catalytic power, and high specificity. On the other hand, Enzyme catalysts have several drawbacks, such as limited stability, recovery, and temperature sensitivity. As a result, the approach of enzyme immobilization was used to improve biocatalyst qualities. Enzyme immobilization is the trapping of protein catalysts onto or inside a matrix, which increases the matrix's stability, adaptability, reusability, and resistance to environmental changes (Bashir et al. 2020).

In addition to its thermal and physicochemical stability, enzyme immobilization provides excellent efficiency and is cost-effective (Ng et al. 2020). Food waste converted using enzyme sources into value-added products such as biodiesel, sweeteners, biofuel, nutraceuticals, food supplements, antioxidants, prebiotics, food colorants, stabilizers, thickeners, and fat replacers has been carried out using mechanisms such as oxidation, phosphorylation, esterification, acylation, deamination, glycosylation, hydrolysis, and others (Chen et al. 2019). To enhance the possibility of biocatalysts to produce significant products, an appropriate and site-directed immobilization, low-cost support matrix, sensitivity to solvents and inhibitors, the use of several enzyme systems, and cofactor demands should all be carefully considered. As a result, the technology of immobilizing enzymes source for bioconversion of agro-based food residue into valuable goods appears to be a viable option (Sharma et al. 2021).

8.6 Technologies for Improving Production Efficiency and Yield of Value-Added Products from Agro-Industrial Wastes

Inadequate soil condition due to decreasing organic material application and non-conservational activities that disrupt topsoils is critical for diminishing agriculture sustainability (Kibblewhite et al. 2008). Maize stover, orchard, rice straw, and animal waste pyrolysis and biochar studies suggest that a large quantity of agricultural leftovers in farmer's fields delivers environmental and economic benefits (Kung et al. 2015). Agricultural residues coupled with cattle manure for biogas production after anaerobic digestion using microbial cells (acidogenic and acetogenic bacteria) are feasible (Muthu et al. 2017). Biodegradation can give value to the result of anaerobic digestion after waste if the production of biogas remains. Notable microbial population modifications were discovered when anaerobic biogas from municipal food leftovers and agricultural and domestic pollutants were composted under natural composting conditions (Franke-Whittle et al. 2014).

Understanding microbiological processes at numerous recycling steps enabled better control of bio-oxidative reactions, accompanied by stabilization and development phases, performed in a substantial static reactor (up to 600 L) (Villar et al. 2016). Enhanced composting with beneficial microbes has created new paths for better exploitation of anaerobic digestate, the byproducts of which might be used directly in farms to boost soil organic content. Such composts have proven to be adequate field substitutes for farmyard organic manure. Recycling methods are farmer-friendly, repeatable, and simple to employ, and they generate valuable agricultural inputs and methane for bioenergy (Achinas et al. 2017). Agricultural waste has continued to be a primary component of biofuels all around the world.

Microorganisms serve a critical role in sustaining nutrient transfer from farm leftovers to farm soils (Erickson et al. 2009). Plant materials, agro-based leftovers, have crystal structures linked with silica, lignin, suberin, and other polymeric components that inhibit the efficient microbial degradation rate essential for recycling. Pretreatment of lignocellulosic materials using steam, acid, urea, alkali, and hydrolases is indicated for significant complex elements degradation and smooth the composting process. The use of biologically defined microbial inoculants with significant enzymatic activity for lignocellulosic breakdown can speed up the biological conversion process (Choudhary et al. 2016). For the production of *Agaricus bisporus* mushrooms, industrialized composting is a well-established biological method. Mushrooms are one of the most exciting fungal species that can be exploited as preliminary degraders of lignocellulose elements in crop wastes and perform better enzymatic monosaccharide extraction for biofuels. It also facilitates the transformation of crop residues into high-protein, high-nutrient-value edible fruits (Jurak et al. 2015).

Large-scale animal manufacturing systems significantly increase agricultural waste biomass resources of organic manure and sludges that can be applied to the land to increase productivity. Composting ingredients such as pig sludge and organic poultry manure have remained relevant (Pampuro et al. 2016). The polyphenolic content, organic matter, soluble carbon, pH, electrical conductivity (EC), humification properties, and plant germination index of wastes from wine distilleries composted with poultry and livestock manure in a static pile composting technique were all measured. Agriculture and food residues make good composting resources because they can be converted into decomposed cattle manure that can be utilized to grow high-value crops (Rubio et al. 2013).

8.6.1 Strain Improvement

In metabolic regulation, strain characterization, and strain creation, genetic approaches are becoming increasingly important. Even though gene cloning and expression have produced a significant set of genes, most of them have yet to be given a function. However, microorganism genetics is started to develop, although great work is currently being put into it. The development of expression vectors and techniques for transferring plasmids and transposons were the primary goals. Young et al. (1989) studied microbial cell genetics utilizing several techniques such as conjugation, natural/protoplast transformation, transduction, transposons, and electroporation.

Several genes producing fermentative metabolic enzymes have been identified and characterized. The development of a physical map of a single organism's chromosome and the formation of genetic manipulation of homologous recombination in bacterial chromosomes have also been established. The majority of organism genome sequences have been sequenced, and numerous DNA transfer techniques have been developed and implemented in various research (Thomas et al. 2014a).

Microorganism has several advantages, including the ability to synthesize a wide range of metabolic products and enzymes. It does however have a branching fermentation mechanism, which results in decreased ethanol production and the generation of other undesired byproducts such as acetate and lactate. Knocking down the genes producing lactate dehydrogenase, acetate kinase, and phosphotransacetylase, which are essential for the branched metabolic pathways, is one technique to deal with it. These target genes' DNA sequences can be found by examining existing genomic sequences or cloning the appropriate genes into *E. coli*. The electroporation approach is a good and dependable approach that is widely utilized for various bacterial species (Thomas et al. 2014b). Electrotransformation with other plasmids has also been successful in the transformation of several other stains. It is hoped that electroporation will become more widely utilized in clostridial genetic modification and that, in conjunction with conjugative methods, it will contribute significantly to strain creation and understanding of clostridial physiology. Clostridia have also been transformed using protoplast transformation methods. An array of strain improvement techniques are available to increase the yield of value-added products from agro-industry-based waste (Table 8.4).

Table 8.4 Strain improvement methods employed on *Clostridia* sp.

Organism	Method of strain improvement employed
C. thermocellum	Gene knockout
C. beijerinckii NCIMB 8052	Tn 1545 transfer
C. thermohydrosulfuricum DSM 568	Transformation
C. beijerinckii NCIMB 8052	Electrotransformation
C. pasteurianum	Electrotransformation
C. acetobutylicum	Protoplast transformation
C. perfringens	Protoplast transformation
C. thermohydrosulfuricum	Protoplast transformation
C. thermocellum	Sonoporation
C. difficile	Conjugative transposon Tn5397
C. perfringens	Tn4451 and Tn4452 transfer
C. difficile	Tn4453a and Tn4453b transfer
C. difficile	Tn5398 transfer
C. perfringens	Tn5 transfer
C. cellulyticum	Tn1545 transfer
C. difficile	TA mariner-based transposon system that involves Himar 1 element
C. acetobutylicum NCIB 8052	Transformation with plasmid pAMfll
C. acetobutylicum	Transfection by bacteriophages, CA1 and HM3
C. thermocellum	Thermotargetron technology
C. cellulolyticum	Metabolic engineering
C. cellulolyticum	Metabolic engineering
C. thermocellum	Gene knockout
C. cellulolyticum	Targeted mutagenesis
C. cellulolyticum ATCC 35319	Tn1545 transfer
C. acetobutylicum ATCC 824	Spontaneous mutation
C. acetobutylicum ATCC 824	Transformation with plasmid pCAAD
C. beijerinckii NCIMB 8052	Mutagen NTG
C. acetobutylicum DSM 1731	Genome shuffling
C. acetobutylicum ATCC 824	Chemical mutation by nitrosoguanidine
C. acetobutylicum ATCC 824	ClosTron technology
C. acetobutylicum Rh8	Overexpression of adh gene

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8.7 Challenges in Value-Added Product Synthesis from Agro-Industrial Wastes

Agricultural residues is a massive source of nutrients that can have both financial and environmental implications. Crop residues are turned into biofuels and bio-based goods within the circular economy, taking into account residual materials. Numerous significant problems must be overcome during the process. The first significant challenge is related to the research on agricultural waste management's environmental and economic repercussions. In this research study, there are inadequate and premature prediction tools. As a result, it would be unable to give governments and end users clear guidance. The approach of evaluating the potential environmental impacts and services is known as life cycle analysis (LCA). Concerning its usability, the LCA has a resource constraint. As a result, inventory data on biomass residues are either lacking or difficult to get. The second difficulty is that the context of agricultural waste conversion technologies is relatively ineffective. For instance, the anaerobic digestion process is inefficient, making it unsuitable for converting lignocellulose wastes into methane. Efficient and inappropriate nutrition supply, pollution development, and agricultural waste conversion are significant challenges.

Agricultural waste conversion obstructs the incorporation of other sources of energy, contaminants, minerals, and pathogens. As a result, the third challenge is to expand employees' awareness and understanding throughout all farming sectors. Polychlorinated dioxins, Furans, lead, mercury, and other undesirable and dangerous byproducts are released during the conversion of agricultural residues to biofuels. As a result, efficient steps to avoid the generation of dangerous substances should be considered and adequate control technologies. For sustainable growth and better awareness of residue source's positive roles, processing, and recycling operations, other controlling agencies and infrastructure capitals are required for commercial manufacturing of these value-added products. The majority of the microorganisms that served a crucial role in various conversion processes were wild kinds. These wild-type strains can mutate and modify their expression levels quickly.

As a result, strain evolution is a big challenge, as wild-type strains may generate the required product but convert the predicted products in too little a volume, causing fermentation to take longer. As a result, conquering these obstacles is critical to moving forward. In most cases, thorough research in the sector ultimately closes the technological gaps. There is a critical shortage of economically feasible, creative, and environmentally friendly technology. Other agro-waste recycling solutions should be offered. Research has shown that nanotechnology may be effectively used to develop potential pretreatment techniques and processing techniques when particular nanomaterials are used (Ingle et al. 2020).

Similarly, problems with biofuel production during waste transformation in hydrolysis using microbial enzymes can be solved using nanotechnology. Magnetic nanoparticle complexes significantly reduce the cost of treatment when the same enzyme is intended to be used multiple times (Rai et al. 2019). As a result, such a creative and worthwhile technological implementation might be advantageous in resolving such challenges in every aspect of agro-waste utilization, and it could be accomplished via significant research. Other issues such as the cost of agro-waste, transportation can be adequately addressed by implementing proper supply chain management and related analyses. Furthermore, progressive policies and execution evaluations reduce the challenges connected with political, financial, and market opposition in the long run.

8.8 Future Prospects

Different types of microbial enzymes that favor efficient fermentation processes are discussed and advanced technologies of agro-waste bioconversion; microorganisms participated, the advantages of microbial enhanced and enriched compost, and options for adopting such microbial technologies eco-enterprising models. All of these actions are straightforward to implement by farming communities. In addition, the farmers' ingredient resources are frequently available at their residences; the process aids in the reintroduction of organic compounds to soils and beneficial microbes that aid in the improvement of soil nutritional status for the development and growth of plants.

Farmers who adopt such techniques can not only enhance rural sanitation on the ground and promote government cleaning campaigns around the world, but they can also boost soil fertility. The process produces high-value, low-cost agriculture inputs from agricultural farm leftovers that would otherwise be thrown away. It produces harmful greenhouse gases (GHGs), fog, and haze when burned in farmer's fields. Microbial consortia of plant growth stimulating and biocontrol microorganisms have been added to these products. Microorganisms can benefit from these biologically rich bio-farm supplies.

Agricultural crop wastes can be used effectively to benefit farms and rural communities. Microbe-based bioconversion of agricultural residues, when established as an eco-enterprise model, can significantly assist rural populations in generating rural subsistence through commercially valuable products (Naresh 2013). For various reasons, mushroom growing in rural areas of several nations has taken on the form of an eco-business; it is based on locally sourced farm waste resources that would otherwise go to waste. Second, it can be carried out with technical skills that can be instilled in agricultural sectors through learning-by-doing approaches. Many types of research contributed to developing farmer-friendly and versatile composted products that satisfy standards of quality for commercialization and entrepreneurship. These models should be created to introduce multi-enterprise support for innovative farming systems (Pramanik et al. 2013).

8.9 Conclusion

Agro-industry produces a considerable amount of trash, which has significant environmental consequences. These issues have compelled us to devise techniques that would aid in the reduction of waste volume and environmental contamination. Agricultural waste offers much potential as a medium for value-added bioproducts. Biofuels like biodiesel, butanol, bioethanol, biogas, biobutanol, bioenergy, biopolymers, bioplastics, and other active components have recently gotten much attention. Microorganisms can successfully handle biodegradable agro-industrial waste during fermentation by using the waste as a carbon source. Pretreatments that allow for higher saccharification kinetics at lower enzyme loadings can enhance the profitability of enzyme production even more. Although progress has been achieved in improving processes in the laboratory, research-based on bioprocess scale-up are needed. The cost of operations while managing microorganisms limits the scale-up of microbial waste transformation. As a result, there is a rising need to create novel microbial processes that use low-cost carbon sources while addressing the other shortcomings. In addition, to get these value-added items to the commercial market, substantial planning permission and capital investments are required. Converting agro-industrial leftovers to vital compounds could not only give academics a new perspective, but it could also help to mitigate current environmental risks.

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Chapter 9
The State-of-the-Art Reverse Logistics for e-Waste Management: A Scenario Specific to India

K. Arun Vasantha Geethan, S. Jose, Rinaldo John, I. Aadil Ahmed, Prashanth Rajan, and Anand Prem Rajan

Abstract Original equipment manufacturers (OEMs) are required to adhere to the Waste from Electrical and Electronic Equipment (WEEE) standards in their material and product management. EPR (Extended Producer Responsibility) is implemented, and producers are required to take back the items when they reach the end of their useful life (EOL). The OEM has hired a third-party reverse logistics provider (3PRLP). Data on sales from each OEM are maintained in a single database that contains extensive information on the items and system participants. After its EOL, the product is monitored using the unique product code etched in the Radio-frequency identification (RFID) tag, and appropriate credits are provided to customers who return the goods. After registering the product in the integrated information system's returns database, the collected returns are returned to the OEM's remanufacturing organization. As a result, the information system makes it easier to integrate the quantification of sales and returns, as well as waste management system evaluations based on location, OEM, or product.

The existing closed-loop supply chain model has been modified to align with sustainability standards. The legal measures imposed by the European regulation

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were approved and established as the standard for OEMs. In this chapter, it is recommended that manufacturers in European nations implement the mandate of expanded producer responsibility. As a result, by overcoming the unpredictability of returns, the e-waste management system may achieve long-term development.

Keywords e-Waste · Lead-free solders · Reverse logistics · Remanufacturing

9.1 Introduction to e-Waste

Waste Electrical and Electronic Equipment (WEEE) is the formal designation for electronic items that have reached or are reaching the end of their "useful life" (UNEP 2007a). "e-Waste" is a common and informal term for these goods. e-Waste is defined as "the reverse supply chain that takes items that are no longer desired by a specific customer and refurbishes for other consumers, recycles, or otherwise processes garbage." There is no universal definition of e-waste, and it varies by country (UNEP 2007b).

e-Waste can also be defined as "*The reverse supply chain which collects the End of Life (EOL) products no longer desired by a given consumer and processes (Remanufacture) the waste.*"

Faster obsolescence and subsequent upgradation of electronics devices force customers to abandon outdated products, resulting in massive e-waste accumulation in the solid waste stream (Pathak et al. 2017). The fast expansion of the electronics industry, as well as the current consumer culture of growing rates of electronic device use, have had devastating environmental effects. The problems associated with e-waste in India began with the initial phase of economic liberalization that happened after 1990 (Adhia 2013). A shift in economic policy occurred that year, which enhanced spending habits. This transition is characterized by the widespread use of information technology in all domains. These advances, together with indigenous technology improvement, must result in the inclusion of a diverse range of e-waste generated by Indian homes, business organizations, industries, and public sectors to the waste stream (Pathak et al. 2017).

Computers, entertainment devices, mobile phones, goods used for data processing, telecommunications, and other items that are now regarded as old, damaged, or irreparable and destroyed by their original users are examples of electronic trash (Rathore 2020). Despite its traditional label as trash, discarded electronics constitute a substantial category of secondary resource due to their high potential for direct reuse (for example, many completely functioning computers and components are discarded during upgrades), refurbishment, and material recycling (Arya and Kumar 2020).

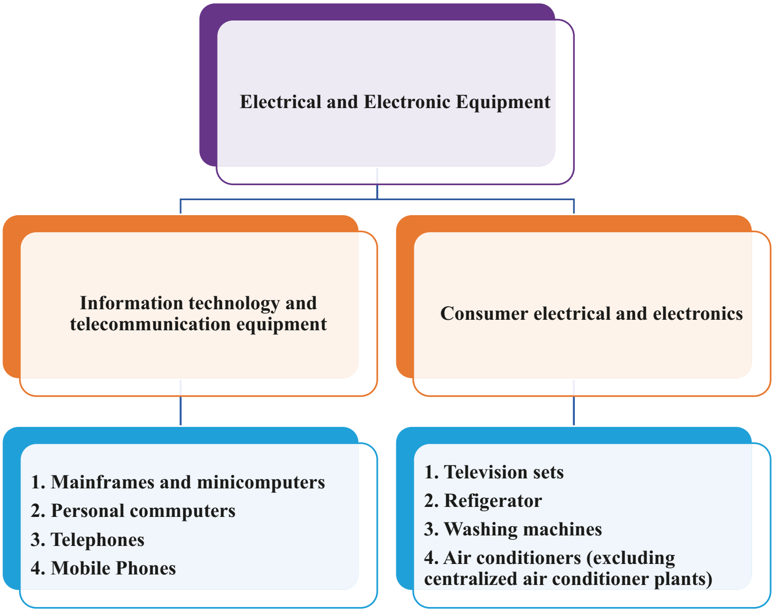

Fig. 9.1 Classification of electrical and electronic equipment

9.2 Classification of Electrical and Electronic Equipment

The categorization of electrical and electronic equipment under the Ministry of Environment and Forests (MoEF) standards is depicted in Fig. 9.1.

9.3 e-Waste Inventory Evaluation in India

In 2019, India produced 3.23 million Metric Tonnes (MT) of e-waste, and over 1,60,000 MT of municipal solid waste generated daily, it is anticipated that the e-waste produced will rise to roughly 2.0 million MT by the year 2025 (Arya and Kumar 2020).

The yearly rise in the volume of e-waste created in India is seen in Fig. 9.2a, Table 9.1 lists the top e-waste producing countries and Indian states (Biswas and Singh 2020; Tiseo 2021).

The top states in terms of e-waste contribution are Maharashtra, Andhra Pradesh, Tamil Nadu, Uttar Pradesh, West Bengal, Delhi, Karnataka, Gujarat, Madhya Pradesh, and Punjab (Jog 2008). Mumbai, Delhi, Bangalore, Chennai, Kolkata, Ahmadabad, Hyderabad, Pune, Surat, and Nagpur are the leading cities of the creation of electric trash. More than 2000 cars are carrying e-waste, mostly at the Turkman Gate, Shastri Park, Seelampur, and Mandoli in Delhi's rubbish yards. This

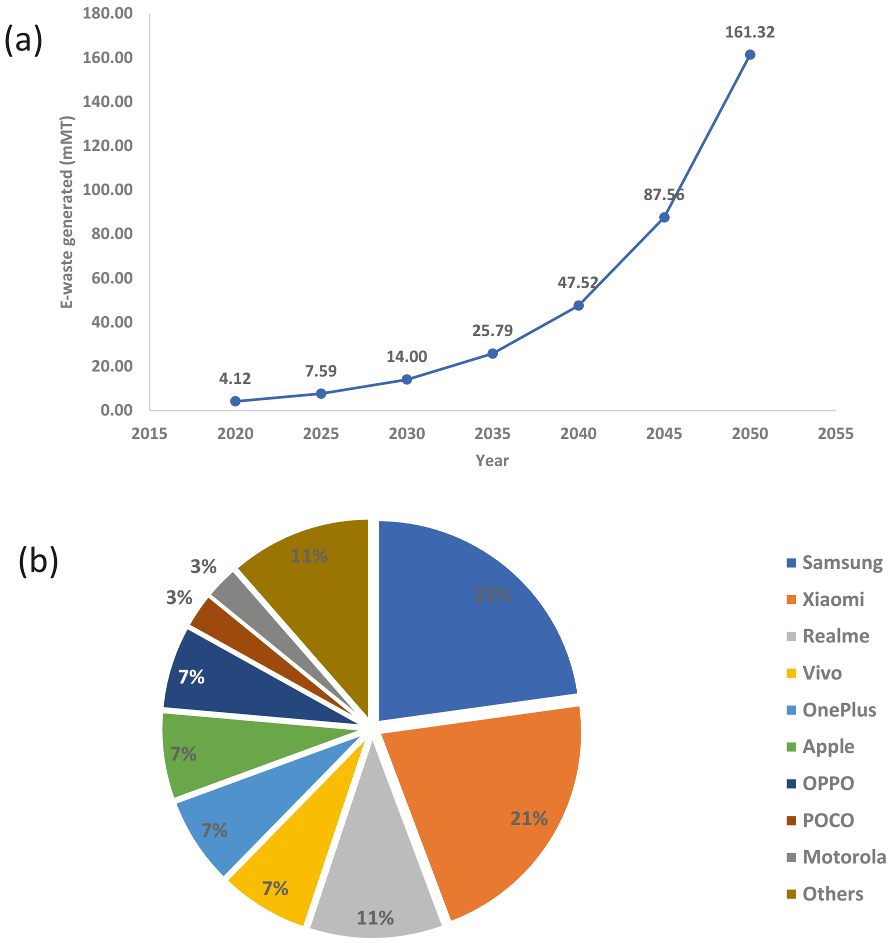

Fig. 9.2 India's projected rise in e-waste (**a**) and ownership of smartphones in India as of March 2021 (**b**)

garbage emanates primarily from Maharashtra, Tamil Nadu, and Karnataka and must be effective regulation if Delhi wishes to defend itself against this harmful waste. Three states that send trash to Delhi create over 25,000 tonnes of e-waste through various industrial processes (Borthakur and Govind 2018).

They dump around half of this in a discreet deal with transporters at different places throughout Delhi, Mumbai, Chennai, and Bangalore are usually presented with e-waste since there are glass and plastic ready markets in the national capital region. Every day at Delhi's waste yards, the bulk of 60–70 tonnes of abandoned gadgets comes from Mumbai. Delhi alone receives 25% of the e-waste generated in the industrialized world, which is imported at a reduced cost, is also expected. The threat has increased to the level of a branch with over 30,000 individuals working in

Table 9.1 Top countries e-waste generation and contribution of different Indian states

S. no.	Country	e-Waste (in 1000 MT)	Indian states	e-Waste (%)
1	China	10,129	Maharashtra	13.9
2	United States	6918	Tamil Nadu	9.1
3	India	3230	Andhra Pradesh	8.7
4	Japan	2569	Uttar Pradesh	7.1
5	Brazil	2143	West Bengal	6.9
6	Russia	1631	Delhi	6.7
7	Indonesia	1618	Karnataka	6.2
8	Germany	1607	Gujarat	6.1
9	United Kingdom	1598	Madhya Pradesh	5.3
10	France	1362	All other states	30

Table 9.2 Direct effect of increase in domestic production of LCD and mobile handsets on year wise e-waste generation in India

Type of electronic equipment	Number of units produced in 2014–2015 (in crores)	Number of units produced in 2018–2019 (in crores)
LCD and LED TVs	0.87	1.20
Cellular mobile handsets	6.00	32.00
Year	e-Waste generation (Million Metric Ton)	
2015	1.97	
2016	2.22	
2017	2.53	
2018	2.86	
2019	3.23	

different scrap yards and illegal recycling. Investment in scrap yards is required for states that relocate to Delhi. Since the NCR has more than 40,000 waste-generating industries and medical institutions, the Government of Delhi is supposed to prepare around 20 lakh saplings annually (Agoramoorthy and Chakraborty 2012). The following two e-waste categories represent the largest proportion of trash creation as stated in Table 9.2.

Because of changing configurations, technology, and tempting manufacturer offers, consumers prefer to purchase a new mobile phone rather than upgrade an old one. Table 9.2 shows the yearly e-waste generation in India (Forti et al. 2020). With over 1.38 billion wireless mobile subscribers across the nation, India is currently the world's second-largest market behind China, and rural India outperformed urban India in mobile growth rate in 2010–2020 (Sun 2021a). Top brands' ownership of smartphones in India is represented in Fig. 9.2b (Sun 2021b). Based on a total sale of 33,171 crores and 175 million phones, the average smartphone return in India is 1895. The e-waste collected from one domestic house in Coimbatore District of Tamil Nadu India is shown in Fig. 9.3.

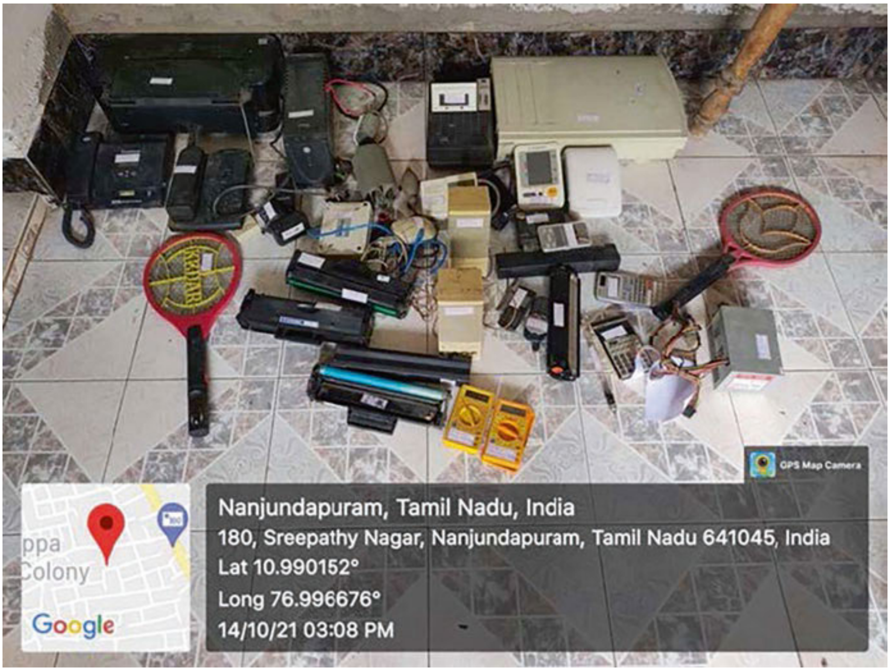

Fig. 9.3 Domestic e-waste collected from Coimbatore

9.4 Extended Producer Responsibility (EPR) Status in India

Following the Supreme Court's policy in Writ Petition (C) No. 657 of 1995 (with S.L.P. (C) No. 16175 and C.A. No. 7660 of 1997) by Research Foundation for Science, Technology, and Natural Resources Policy vs. Union of India and others (Pasayat 2007). The policy recognizes EPR as a key component of the management system for e-waste. The EPR is a strategy to protect the environment that accounts for the whole life cycle of its product, including its "end of life management" through takebacks, processing and final disposal, to the producer of electrical or electronic equipment. The producer must ensure the manager does not affect human health and the environment by manufacturing e-waste from the goods of the manufacturer.

9.4.1 The e-Waste Management Legislation in India

The area of South Asia has started to recognize the need for good management of e-waste. India is the only country having e-waste laws in South Asia, while numerous other countries consider such legislation. In India, e-waste management rules have been in effect since 2011, which require only dismantlers and recyclers who are authorized to collect e-waste (Dwivedy et al. 2015). The 2016 E-Waste Management Regulations included the refurbisher, supplier, fabricator, and producer responsibility organization (PRO). The National Resources Policy also provides for an important role for manufacturers to recover secondary resources from electrical waste (Central Pollution Control Board 2016).

e-Waste management in India is based mostly on informal collecting, dismantling, and recycling operations in the industry (Prithiviraj and Chakraborty 2019). The Indian legislation has driven the establishment of legal recycling installations and in India, there are more than 312 authorized recyclers with an annual treatment capacity of about 800 kt. However, there is a lack of official recycling capacity, since the vast bulk of the garbage is still handled in the informal sector. About 31 authorized PROs provide conformity services, including e-waste collection and channeling to official recycling plants, as well as the management of awareness campaigns. Enforcement regulations remain an issue, like other elements including lack of adequate collection and logistical infrastructure, low consumer knowledge of risks to unsuitable e-waste disposal, absence of collection requirements, disassembly and treatment of e-waste, and wasteful and laborious reports (Forti et al. 2020).

9.5 Options Available for Treating e-Waste

The rising environmental cost of a “throw-away” society has highlighted the need for trash alternatives to landfilling and incineration. Opportunities to reintegrate used goods and materials into industrial manufacturing processes have been sought. There are four primary options for recovering used goods. They are reuse, recycle, remanufacturing, and repair.

9.5.1 Need for Remanufacturing

For millennia, remanufacturing has occurred, generally for high-value, low-volume products such as locomotive engines and airplanes. The fundamental challenge in remanufacturing large, complicated products is the problem’s size. Products are frequently made up of tens of thousands of individual components and pieces. The disassembly, remanufacturing, and reassembly of such goods presents a technological difficulty in terms of shop flow control, appropriate testing of essential components, and part coordination at the reassembly point. Lund’s work at the World Bank saw remanufacturing as a means for poor countries to gain technical know-how while also touting the energy savings from remanufactured items. Remanufacturing is one of several product recovery methods (the others being repair, refurbishment, cannibalization, and recycling) that are categorized according to the degree of disassembly and the quality of the recovered product (Dekker et al. 2004).

Remanufacturing is described as “an industrial process in which goods are restored to their original function by replacing worn-out parts that do not correspond to their purpose.” Remanufacturing lays the groundwork for closed-loop supply chains by emphasizing value-added recovery (Guide and Wassenhove 2001). Remanufacturing can provide both economic and social possibilities. Remanufacturing should be favored over recycling since it restores consumer returns to the market, whereas recycling simply decreases user returns to the raw material condition. The processing of this recycled raw material is a time-consuming procedure, and remanufacturing allows us to save time, money, and energy. Because they had not been retouched after use, the reused items are untrustworthy in the eyes of the buyer. Furthermore, refurbishing cannot be a viable choice in poor nations due to the low quality of the returns. So, for our analysis, we chose to remanufacture since we determined it was the best reengineering method among the options. Furthermore, remanufacturing contributes to environmental balance by extending the life of a product’s parts (Singhal et al. 2019).

The remanufacturer faces several problems both before and after the remanufacturing procedure. Some of the most typical issues that remanufacturers encounter are—a lack of a comprehensive reverse logistics (RL) network; product information is scarce accessible at the collecting node; uncertainty in terms of time and quantity of returns; there is a need to increase market demand for remanufactured items; providing financial incentives to the remanufacturer; and to ensure system sustainability by generating socioeconomic advantages.

9.6 Overview of Reverse Logistics (RL)

The planning, implementation, and control process for the transfer of raw materials, in-process stocks and finished products from manufacturing, distribution, or use point to an adequate disposal site is known as RL. The RL definition is the most commonly used. To define RL or the characteristics it is used by various authors, for example, reverse flow logistics, reverse distribution, RL, reverse supply chain (RSC) (Santhanam 2006). The definition of RL differs depending on the point of view from which it is seen. Some of the other definitions are:

- RL is the process of relocating things from their usual end location to capture value or dispose of them properly. RL is the transfer of products from a consumer to a manufacturer through a distribution channel.
- RL refers to a supply chain that has been structured to efficiently handle the flow of items or components destined for remanufacturing, recycling, or disposal while also maximizing resource use (Dowlatshahi 2000).
- RL refers to all reuse methods of items and materials. This defines the RL as “The processes in which raw materials, stocks in process, completed items and related information may be planned, implemented or regulated efficiently and cost-effectively moved from point of consumption till the point of origin to recapture or appropriately disposes of the value” (Dekker et al. 2004).

- More specifically, RL is the act of relocating items from their usual end destination to capture value or dispose of them properly (Hawks 2006).

It can be defined as "*A supply chain modified to efficiently handle the movement of items or components intended for remanufacturing and to use resources efficiently*."

9.6.1 Drivers of RL

RL begins with items being returned to the supply chain for value recovery or reclamation. The following are the driving forces behind RL:

1. *Economics*: It refers to any recovery activities in which the firm receives direct or indirect economic benefits. Companies may become involved in recovery as a proactive measure to prepare for future laws.
2. *Legislation*: It refers to any jurisdiction suggesting that a firm should recover or accept the return of its items.
3. *Extended Producer Responsibility (EPR)*: RL operations are carried out following the EPR concept. Because of the EPR plan, the obligation for collecting items such as consumer electronics has shifted to the producers in Europe. EPR is a strategy to safeguard the environment that holds the company responsible for the whole life cycle of the product, including recovery, recycling, and final disposal. The responsibility of the manufacturer therefore extends to the stage of the after-consumer life cycle. EPR aims at encouraging manufacturers to design at the source, i.e., at the product design stage, environmental concerns to decrease waste management costs as much as feasible. The EPR approach is based on the polluter pays principle and is based on shifting responsibility to include treatment and disposal expenses in the price of the product from municipalities, reflecting the environmental impact of the product (Garlapati 2016).

National collecting and processing of end of life (EOL) systems have been established throughout Europe in partnership with or under pressure from national governments. Because of the high level of uncertainty in returns, RL operations may be extremely complicated to handle. Furthermore, demand might be difficult to forecast, making product and information flow difficult to manage. Firms that respond too slowly or ineffectively to RL are likely to face large cost increases, whereas businesses that respond in a timely and organized manner might expect not just a competitive advantage but also possible cost savings. This appears to be the result of the Waste Electrical and Electronic Equipment Directive (WEEE Directive), which went into effect at the end of 2005 (Dwivedy et al. 2015).

Most firms, who had never spent much time or effort before, have started to pay attention to RL management and comprehension. These firms compare their return operations with the best in the class. Some companies are even certifying for their return procedures by the International Organization for Standardization (ISO). The

Table 9.3 Differences between forwarding and reverse logistics

Forward logistics	Reverse logistics
Forecasting relatively straightforward	Forecasting more difficult
One to many distribution points	Many to one distribution points
Product quality uniform	Product quality is not uniform
Destination/routing clear	Destination/routing unclear
Disposition options clear	Disposition options unclear
Distribution costs are easily visible	Reverse costs less directly visible
Inventory management consistent	Inventory management is not consistent
Product life cycle manageable	Product life cycle issues are more complex
Visibility of processes more transparent	Visibility of processes less transparent
Negotiation between parties straightforward	Negotiations complicated
Product packaging uniform	Product packaging is often damaged

need for their services for third-party returns experts has increased significantly. In general, significantly more work has been expended in enhancing return procedures in businesses where the product's value is highest or when the return rate is highest. An excellent example is the information technology and telecommunications business.

9.6.2 *Main Differences in Forward and Reverse Logistics*

Reverse flows are significantly distinct from forwarding flows, and therefore RL is quite different from forwarding logistics, as illustrated in Table 9.3. The most significant distinction is that all RL operations lead in the other direction.

9.6.3 *Common RL activities*

When a product is returned to a corporation, the company has several disposal choices from which to select. Table 9.4 summarizes some of these efforts.

The product may be sold as restructured, but not as new, as a result of these measures. A wide variety of operations might include RL. These operations are ranked by determining if the product in reverse flow originates from the end-user or another part of the distribution channel, such as a store or a distribution center (Jenkins 2021).

Table 9.4 Common RL activities

Material	Reverse logistics (RL) activities
Products	Return to Supplier
	Resell
	Sell via Outlet
	Salvage
	Recondition
	Refurbish
	Remanufacture
	Reclaim Materials
	Recycle
	Landfill

9.6.4 Types of Flows

The RL method handles three types of flows. Products, as well as knowledge and money, go backward. The major fluxes, according to (Dekker et al. 2004), include:

1. *Material flow*: The physical items that are returned are included in the material flow. Consumer goods have a backward flow, from the client to the merchant or seller, then to the collecting point, and lastly to the site of origin or disposal. Factors such as the amount and qualities of the items are considered.
2. *Information flow*: The intangible flow that accompanies the material flow is information flow. It is critical to the process since it gives information on the items, their locations, market time, condition, and reusability.
3. *Financial flow*: The flow of money involved in the process is referred to as financial flow. It covers all costs associated with the backward movement, such as transaction, handling, storage, transportation, disposal charges, and certain firms' return money policies.

9.6.5 Types of Returns

Four primary sources of product returns in the RL are:

1. Supply chain return: Products returned by numerous different operators than the end customer through the supply chain. Unsold items returned by the store are a common example.
2. Warranty returns: Items returned by end-users within the manufacturer's stipulated time frame, as faulty products or owing to consumer dissatisfaction with the product.

3. End-of-lease equipment returns: Goods returned the end of the lease period, results in the shift from selling items to offering services based onneeds of the resource requirments.
4. End of life products: Products left by users after their useful lives have been reached, as evaluated by the user. The items can still work in some conditions. It is evident that around two-thirds of disposable gadgets still work.

EOL may alternatively be defined as the moment at which a product no longer meets the needs of the first user. This definition takes into account the quickly changing consumer preferences, which often occur before the product wears out. The end of life of a product may also be defined as the moment at which it no longer fulfills its intended functions. EOL treatment is concerned with recovering the value of goods, including the activities connected with strategic planning and implementation of used product collecting and processing, as well as the associated societal and environmental consequences (de Campos et al. 2017).

9.7 Research Scope Depicted in a Closed-Loop Supply Chain

Figure 9.4 depicts the breadth of RL research from the standpoint of remanufacturing, which gives a schematic picture of the operations in a typical forward and reverse supply chain in general.

Because of the uncertainties involved with the quality, quantity, and timeliness of returns, the complications associated with handling RSC activities are multifaceted. However, there are several chances for for-profit and company image growth that should not be overlooked. The regulations and economic benefits of the reverse supply chain have compelled businesses to examine their operations more closely. As a result, it is critical for businesses to efficiently manage their reverse supply chain system.

9.7.1 The Necessity of RL/RSC in India

The actions of a company that employs returned items owing to the product recall, surplus inventory, salvage, undesired or obsolete products, and so on are examples of typical RL operations. It also addresses programs for recycling and hazardous waste, the disposal of obsolete equipment and recovery of assets. The reverse flow of the substance begins at numerous locations and focuses on a few destinations. The many tasks performed under the RL activities include gatekeeping, re-fit-up and renovation, asset recovery, negotiation, outsourcing, financial administration, and customer assistance. RL thus focuses on the management of value-added materials, information and connection flows, and the disposal of products. The need for RL stems mostly from the broad issues of environmental economics (Wagner 1997).

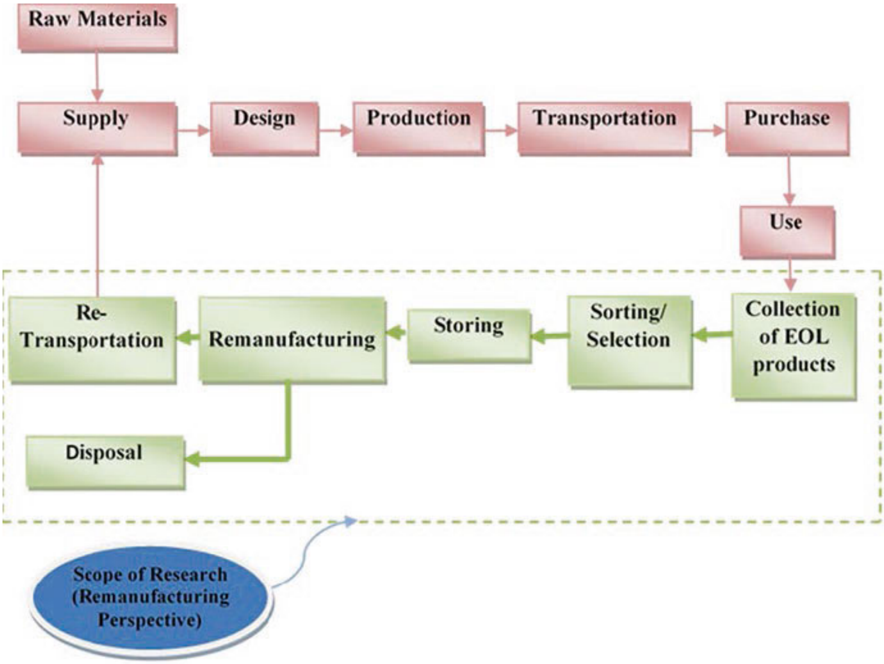

Fig. 9.4 Research scope depicted in a closed-loop supply chain

The following are a list of the benefits that may be obtained by using RL:

- A significant number of raw materials are conserved and partially substituted with commodities from the closed-loop supply chain.
- Because trash is being processed, less landfill capacity is required. Furthermore, as disposal prices skyrocket, RL has emerged as a feasible option.
- RL has become a feasible alternative for industries as environmental authorities have strengthened their stance on waste handling.
- RL is divided into two dimensions: the "green" dimension and the "value reclamation" dimension (Santhanam 2006). Green dimensions have been used to address environmental problems as well as value reclamation to provide economic and energy advantages.

9.7.2 Barriers in RL

1. *Lacking RL awareness*: A key obstacle to RL deployment is a lack of understanding about its benefits. Consumers have benefited from the market's wide range of items. This has resulted in a significant rise in the number of returns, unsold items, packing materials, and trash. This has increased the number of

goods returns via RL. RL may result in economic advantages by reclaiming returned products and enhancing the value of the product through reengineering alternatives such as reuse, remanufacturing, recycling, or a combination of all of these options.

2. *Management inattention*: The widely held belief has been that in recent years, many firms have used RL mostly in response to regulatory demands from government and environmental authorities, rather than for economic gain. As a result, little attention is placed on this nonprofit endeavor, and emphasis is placed on the onward flow of products.
3. *Financial constraints*: Expense issues have been a significant obstacle for commercial recycling. RL also necessitates significant expenditure. It is critical to creating management information systems and technologies since product tracking, tracing, and recovery cannot be accomplished without such an information platform. However, its implementation is more expensive, which is not feasible in the current situation. To make it viable, the staff must be trained in RL disciplines. However, financial resources are necessary to carry out all of these concepts.
4. *Issues with product quality*: The product quality in RL is not as constant as it is in the forward supply chain. Customers expect the same high-quality items from the producer, regardless of whether the product is returned. However, the returned goods may be damaged, defective, or partially faulty, and their quality may vary significantly. As a result, the prices of secondary market items may differ.
5. *Inadequate performance management system*: The product quality in RL is not as constant as it is in the forward supply chain. Customers expect the same high-quality items from the producer, regardless of whether the product is returned. However, the returned goods may be damaged, defective, or partially faulty, and their quality may vary significantly. As a result, the prices of secondary market items may differ.
6. *Insufficient information and technology systems*: An information platform is critical for aiding RL throughout the product life cycle. Management information systems must identify and trace unique returns and associate them with prior sale reports. Information technology, software, and hardware are critical for a reverse chain's overall organization and clarity.
7. *Company policies*: Corporate schemes are concerned with RL and the management of returns and non-saleable products. Companies often develop techniques that make it difficult to deal efficiently with returns and recover considerable secondary value from returns.
8. *Legal issues*: Excise-paid products vended by the manufacturer may not be fetched back to the factory without prior paperwork and approval from excise authorities. This is a highly laborious and time-consuming process, and disobedience may result in legal action being taken against the company. Many businesses see this as a barrier to the successful deployment of RL.
9. *The financial and administrative burden of the tax*: Appropriate planning and administration of direct and indirect taxes are critical financial considerations

within the reverse chain. The convoluted (and cross-border) flow of products and the wide range of services bought-in services indoctrinate in the reverse chain, resulting in unexpected tariff disclosures and charges.

10. *Limited forecasting and planning*: Precise return estimates are difficult to come by. This is an implacable impediment to both strategic and operational planning. Due to the large volume of product and flow changes, many companies are unable to forecast and plan the reverse chain.
11. *Chain members' cooperative conduct*: Cooperative behavior of chain members is necessary for communication division. The absence of assistance from RL distributors, retailers and was emphasized as the main hindrance.

9.8 Proposed Sustainable e-Waste Management Model

The closed-loop supply chain concept was modified to align with sustainability standards. The legislative requirements imposed by the European regulation were implemented and established as the benchmark for OEMs. It is also recommended that extended producer responsibility, which is a specified practice followed by manufacturers in European nations, be mandated in our system as well. Because manufacturers are under pressure to keep the environment clean, lead, which is hazardous and threatens the system's sustainability, should be eradicated during the product's design phase. Conventional lead solders are replaced with lead-free solders, for this reason, minimizing the impact of pollution produced by EOL electronic returns. It also attempts to extend the life of electronic items, reducing the number of EOL products entering the RSC.

As a result, from a production standpoint, the study recommends the use of lead-free solders to mitigate the negative effects of lead. In the process to extend the life of plumbing solder joints, ZrO_2 nanoparticles over Sn/Cu plumbing solders via nanostructured coating thickness 814 nm, clearly increase mechanical properties such as tensile strength, micro-hardiness, yield strength, and shear strength to 50% by average. This results in strong resistance to flaws in the lead-free solder connection caused by mechanical failures such as breakage, fracture, shear, cracks, wear, and so on. The life of the solder joint is enhanced as a result of this distinctive feature, and it has been verified that the life increase is up to 50% of the original life. This significant increase in mechanical strength and longevity impacts the solder's aging, which eventually improves the product's life.

In addition, hierarchical workflow monitoring is systemized, with the MoEF having the most power. The MoEF is subject to the national pollution control board, which oversees the operations and practices of original equipment manufacturers and other subordinate parties. The MoEF assesses electronic waste every year, and the reports are documented and stored in a database for future reference. The diagram depicts the product flow, monitoring flow, information flow, and credential flow inside the system. Original equipment manufacturers (OEMs) are required to follow WEEE standards in their material management. The OEM yearly contracts a third-party collector and RL provider based on their comparable criterion ratings. In the forward loop, sales data from each OEM is merged and kept in a single database. This database serves as a treasure trove of comprehensive information about the system's goods and participants. The usage of RFID, which aids in the tracing of the correct goods, facilitates the information system. After its EOL, the product is monitored using the unique product code etched in the RFID tag, and appropriate credits are provided to customers who return the goods. Credits are granted based on the date of return from the date of purchase and the value of the item. In this approach, the information system allows the integration of sales and return quantification.

Furthermore, the information system's sorting capability aids in waste management system evaluations based on location, OEM, or product. Credit also has an impact on increasing the number of returns. The 3PRLP collects the EOL product and returns it to the manufacturer's manufacturing base. The items go through many processes here, including gatekeeping, sorting, and testing before arriving at the remanufacturing facility. The remanufacturing rate is continually monitored, and completed goods from remanufactured product inventories are quantified to produce the remanufacturing index. Annually, the integrated information system retrieves the sales, returns, and remanufacturing index for each OEM and generates system reports. The produced reports are compared to the MoEF reports from the previous year, and appropriate credentials, such as eco-friendly certifications, are granted to the OEMs, which can improve the manufacturers' corporate reputation in the commercial sector. As a result, the model depicted in Fig. 9.5 attempts to

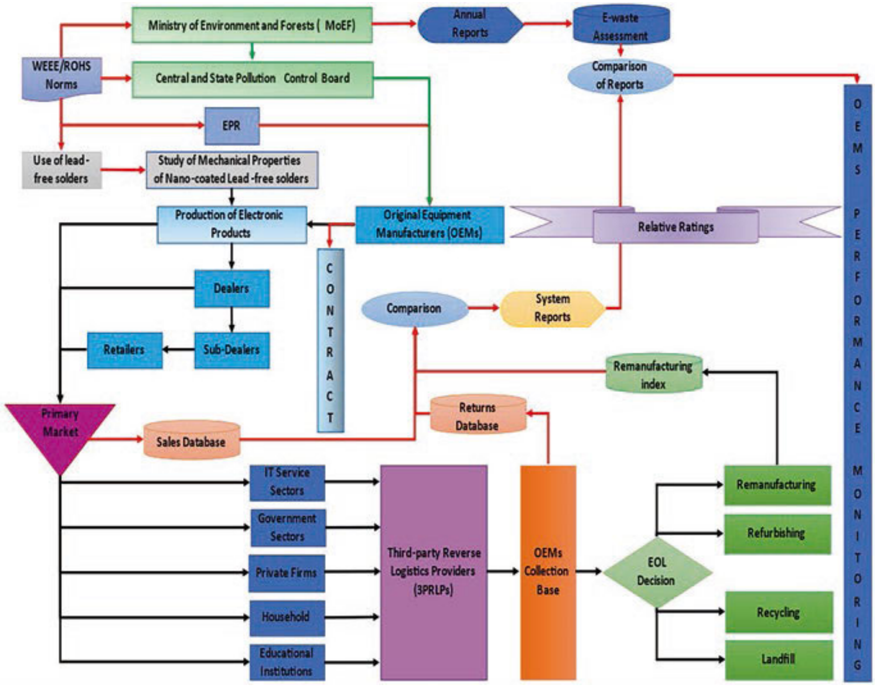

Fig. 9.5 Proposed sustainable e-waste management model

generate long-term development for the e-waste management system by overcoming the unpredictability of returns.

9.9 Future Scope and Conclusion

To reduce time consumption, the suggested multi-criteria approach may be automated. The case study may be carried out in various mobile phone manufacturing sectors to discover the significant elements impacting the outcome. This research might be expanded with a full study examining its application to different sectors. Once the model is in operation, the suggested integrated information model may be fine-tuned and performance evaluation can be performed. High-end software can be used to model the proposed collecting system. Various tests may be used to compare the dependability of different lead-free solder formulations.

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Chapter 10
Environmental Friendly Technologies for Remediation of Toxic Heavy Metals: Pragmatic Approaches for Environmental Management

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Abstract Contamination of different environmental matrices (air, soil, and water) by toxic heavy metals is a widespread problem that disturbs the environment as an outcome of many anthropocentric practices. Heavy metals exceeding the permissible limits exert deleterious impacts on human beings, causing life-threatening health manifestations and detrimental effects on the environment. This has alarmed the dire need to explore various modern remediation techniques that can be utilized to lower excessive concentrations. Owing to their high-cost effectiveness, unsatisfactory output, environmentally unfriendly, complicated procedure, and high operational costs, these technologies failed to find any practical utility in remediation. On the other hand, plants and associated microorganisms are receiving more consideration as a means of remediating or degrading environmental pollutants. This chapter provides us insights into the various environmental friendly techniques that will improve our environment's quality. Among which, phytoremediation is considered an effective technique which is known for its esthetic benefits and endless applicability. Furthermore, metal-resistant bacteria (plant growth-promoting rhizobacteria) are also reported to play a pivotal role in the phytoremediation and solubilization of

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minerals. Thus, this chapter critically reviews the phytoremediation technology and the efficient exploitation of microbes to alleviate the environmental burden of toxic heavy metals.

Keywords Phytoremediation · Remediation · Metal-resistant bacteria · Heavy metals · Contaminants

10.1 Introduction

Frequent emissions of pollutants from several industrial, commercial, and agricultural sectors have been the main topic of concern, as these are detrimental to human health and the entire planet. The dispersion of industrial and urban wastes caused by anthropocentric activities has had a detrimental effect on our ecosystem by releasing solid, liquid, and gaseous wastes containing heavy metals, inorganic and organic compounds (Miri et al. 2016; Sharma et al. 2020). Excessive deposition of toxic substances like heavy metals or hydrocarbons in marine and soil habitats fosters environmental degradation (Peng et al. 2015; Xi et al. 2018). The mobility method of heavy metal in the environment depends upon ores extraction and diverse processing purpose, resulting in releasing these elements in the environment. Biologically, “heavy metals” refer to those metals and metalloids that can pose detrimental effects on living organisms when exceeding the permissible limit. They are known for their detrimental effects on human beings, animals, and plants.

Though, heavy metals are reported to cause various health manifestations among living organisms. However, excessively absorbed heavy metals have been shown in numerous studies to alter cell membrane permeability, disrupt mineral nutrition, disturb the photosynthetic apparatus, and cause oxidative stress, all of which affect plant morphology and growth, and photosynthetic processes (Sharma and Kaur 2019). Elevated levels of heavy metals in plants induce cellular damage primarily through the formation of reactive oxygen species (ROS), including superoxide radical (O_2^-), hydroxyl radical (·OH), and hydrogen peroxide (H_2O_2). Unnecessarily increased synthesis of ROS is one of the organisms’ immediate reactions to various stressful events.

ROS can cause irreversible oxidation of lipids, proteins, chloroplast pigments, DNA, and RNA, which can compromise cell viability (Sharma et al. 2019; Rani et al. 2021). Furthermore, high levels of various harmful and toxic metals make the soil unsuitable for plant growth and deplete biodiversity. As a result, implementing efficient and eco-friendly remediation technologies is imperative for sustainable development. Regulation of heavy metal contamination in the soil can be achieved in many ways, including biological and physical, and chemical approaches.

Mechanical or physical pollutant isolation, acid leaching, electrocoagulation, electrokinetics, chemical treatment, thermal or pyrometallurgical separation, and biochemical methods can be used to treat contaminated soil. On the other hand, these techniques are costly and technically challenging to implement (Rajput et al. 2019). Furthermore, these chemical technologies could cause secondary pollution issues, as well as the generation of a large volume of sludge, raising the cost of sludge management. As a result, for heavy metals to be removed, an alternative solution is needed.

Bioremediation is a novel and promising technology available to exclude heavy metals and their revival from polluted and environmental matrices. This technique offers a clean, safer, low-cost, and environment-friendly method which requires microbes and plants to detoxify, degrade, transform, or mineralize pollutant concentration to a nontoxic (Azubuike et al. 2016). Furthermore, phytoremediation is a highly effective bioremediation tool that can be an alternative for removing heavy metals. Also, the phytoremediation technique is a cheap and environmentally sustainable treatment system that employs the utilization of plants/hyperaccumulators in order to eliminate toxic contaminants from the ecosystem (Ali et al. 2013). Since the contaminant-removal plant has no impact on the topsoil, this process is environmentally friendly and does not damage the ecosystem (Cristaldi et al. 2017). Because of its cost-effectiveness and unique characteristics, phytoremediation has been a promising technology for the remediation of contaminated soil in recent years. Thus, this chapter aimed to review the role of phytoremediation technology and the efficient exploitation of microbes to alleviate the environmental burden of toxic heavy metals. Furthermore, other integrated approaches for environmental management are also discussed. Our main goal is to emphasize more straightforward and economical remediation approaches with efficient results and easier preparation procedures.

10.2 Sources of Heavy Metals in the Environment

Naturally, in soil, metals are a very common component. Nevertheless, in high levels, metal for living organisms can be toxic and harmful. There are certain heavy metals that are primarily found in soil such as mercury (Hg), zinc (Zn), chromium (Cr), selenium (Se), cadmium (Cd), lead (Pb), arsenic (As), copper (Cu), uranium (U), cesium (Cs), and strontium (Sr). Many of them are micronutrients essential for plant growth and enlargements, such as nickel (Ni), cobalt (Co), Cu, manganese (Mn), and Zn, while others have a poorly understood biological function, like Hg, Cd, and Pb (Aksu 2015).

Elements with atomic numbers >20 and metallic properties are categorized under heavy metals. These are not soluble in the soil and are present in the form of colloids and ions. They are nonperishable, unlike organic pollutants (Bitew and Alemayehu 2017). As a result, they persist in the soil for a long time, with a half-life of more than 20 years (Sidhu 2016). Heavy metals concentration in soil ranges from 1 to 100,000 mg/kg (Karami and Shamsuddin 2010). One of the most severe threats to

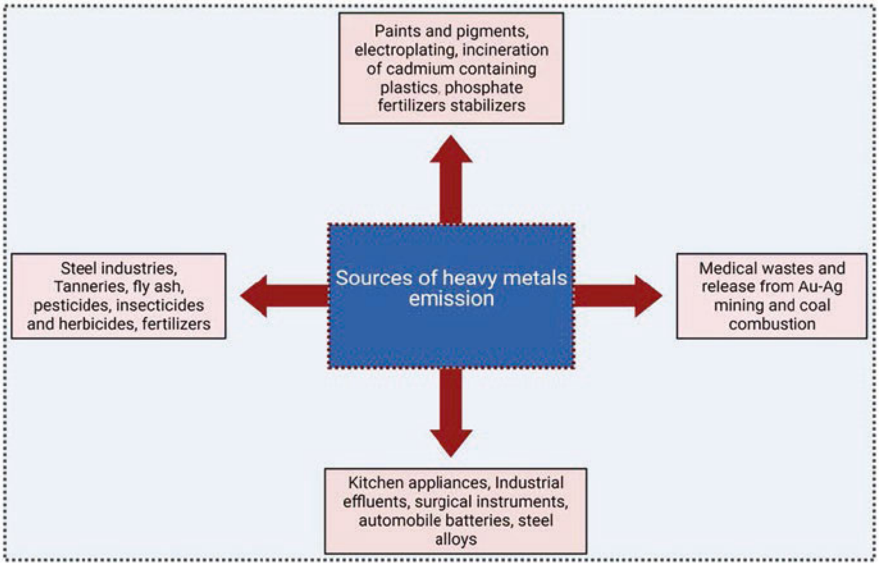

Fig. 10.1 Anthropogenic sources of heavy metals in the environment

human health is transferring these pollutants into the food chain (Singh 2012; Rostami and Azhdarpoor 2019).

Natural sources—Minerals, Volcanic activity, weathering, and erosion are the most significant natural sources.

Anthropogenic sources (human intervention sources)—Common anthropogenic sources are electroplating, mining, smelting, fertilizers and biosolids, pesticides in agriculture, industrial discharge, sludge dumping, etc. (Suvarapu and Baek 2017; Liu et al. 2018). Various anthropogenic sources of heavy metal emissions in the environment are given in Fig. 10.1.

10.3 Bioremediation

Hazardous waste material from industries releases organic and inorganic pollutants resulting in environmental pollution (Ojuederie and Babalola 2017). Accumulation of these metals in soil, water, and sediments has led to various environmental and human health concerns. There have been numerous reports, which confirm the presence of toxic heavy metals in soil, sediments, and groundwater (Rajput et al. 2019).

Numerous chemical and physical approaches have been developed to reduce the contamination level, but these techniques have many disadvantages of adding more chemicals to the polluted soil and water (Ayangbenro and Babalola 2017). Moreover, these methods are not effective in low metal toxicity areas (Akinci and Guven

Table 10.1 Different types of bioremediation techniques

Bioremediation	In situ	Land farming	Brown et al. (2017)
		Bioreactor	Talha et al. (2018)
		Biopile	Álvarez et al. (2017)
		Windrow	Jaain and Patel (2019)
	Ex situ	Bioslurping	Kim et al. (2014)
		Biosparging	Eslami and Joodat (2018)
		Bioventing	Krishna and Philip (2011)
		Phytoremediation	Eid et al. (2020)

2011). In recent years nonconventional techniques bioremediation has been developed, which is based on microorganisms' usage.

Bioremediation is a clean, safer, cheaper, environmental friendly, and sanguine technology which involves biological mechanisms like detoxifying, degrading, transforming or mineralizing pollutant concentration to a harmless state by making use of the integral biological mechanism of microorganisms and plants (Ekperusi and Aigbodion 2015; Azubuike et al. 2016; Verma and Kuila 2019). Microorganisms can degrade heavy metals more rapidly due to microbial enzyme activities. Bioremediation reduces about 50–60% of the cost when used to treat lead polluted soil compared to some conventional approaches like landfill and excavation (Yang et al. 2017). However, various environmental conditions like pH, temperature, and moisture are important factors for the growth and metabolism of microorganisms to degrade pollutants (Chibuike and Obiora 2014; Verma and Jaiswal 2016). Bioremediation involves the basic principle of solubility reduction of these environmental contaminants by changing the pH, inducing redox reactions, and contaminants adsorption from the polluted site (Jain and Arnepalli 2019). The biosorption capability of *Aspergillus niger* and *Mycobacterium chlorophenolicum* to remove pentachlorophenol is widely reported in the literature (Bosso et al. 2015).

The bioremediation technique is mainly of two types: In situ (Natural attenuation and enhanced) and ex situ (Biopile, windrow, bioreactor, and land farming) (Table 10.1). Enhanced In situ is further divided into bioslurping, bioventing, biosparging, and phytoremediation (Azubuike et al. 2016).

10.3.1 Ex Situ Bioremediation

Biopile: This method involves long-term accumulation of contaminated soil followed by nutrient variation, and further aeration process increases the microbial activity thus enhancing bioremediation. This technique's main components are nutrition, aeration, irrigation, treatment bed, and leachate collection (Whelan et al. 2015).

Windrows: In the windrows technique, the polluted soil is turned periodically to enhance microorganisms' activities, mainly hydrocarbonclastic bacteria (HCB).

This helps in the increased aeration and uniform distribution of nutrients and pollutants as well as increased enzymatic activities. This leads to an increase in bioremediation rate through various processes like absorption, biotransformation, and mineralization (Barr et al. 2002).

Bioreactors: As the name indicates, the raw material is converted into valuable products through a chain of biological reactions in bioreactors. Batch, fed-batch, continuous, sequencing batch, etc., are the different types of modes of bioreactors.

Land farming: Because of its cost-effectiveness and minimal equipment requisites for operation, land farming is one of the simplest ex situ remediation techniques. Polluted sites are frequently mined or cultivated through inland farming. There is a debate going on whether land farming should be placed in in situ or ex situ bioremediation techniques, probably due to the site of remediation. Depth of pollutants plays a crucial role in the determination of ex situ or in situ land farming.

10.3.2 In Situ Bioremediation

Bioslurping: To achieve water and soil remediation, this method involves three techniques, namely soil vapor extraction, vacuum boosted pumping, and bioventing (Gidarakos and Aivalioti 2007). This method is premeditated to recover light nonaqueous phase liquids, semi-volatile, and volatile organic compounds. This method utilizes a slurp that pulls up liquids from the free products layer to the surface in a similar manner to a straw entice liquid from the vessel. Following product removal, bioventing can be applied to the complete remediation process (Kim et al. 2014).

Bioventing: Bioventing comprises airflow stimulation in a controlled manner so that oxygen can be delivered to the polluted site's unsaturated zone to enhance the indigenous microorganism's activities. The addition of nutrients and moisture enhancement helps transform pollutants into a less harmful form (Philp and Atlas 2005). This method has attracted worldwide attention due to the restoration of petroleum spilled sites (Höhener and Ponsin 2014).

Biosparging: To stimulate the removal of pollutants, the air is introduced into the polluted site's saturated zone to cause an upward movement of volatile organic compounds to the unsaturated zone for enhancing the activities of microbes resulting in the degradation of pollutants. Soil permeability and pollutant biodegradability are the two main important factors determining the efficacy of biosparging (Philp and Atlas 2005).

10.4 Phytoremediation

Phytoremediation is another technique that relies on the physical, biological, chemical, and microbiological plant interactions to remove the detrimental effects of these present pollutants. This technique exploits higher plants for the removal of heavy metals (Table 10.2). Phytoremediation is a plant-based technique that improves polluted land and water supplies using either raw or genetically modified plant species. Phytoremediation with hyperaccumulator plants is now commonly accepted as a cost-effective and environmentally safe technique of removing pollutants from the environment (Rai et al. 2020).

Filtration, stabilization, accumulation, extraction, volatilization, etc., are the various mechanisms through which plants degrade pollutants. Heavy metals are mostly degraded through the process of removal, conversion, and sequestration. In contrast, organic pollutants such as hydrocarbons and chlorinated compounds can be removed through volatilization, stabilization, rhizoremediation, and degradation. However, plants like alfalfa (*Medicago sativa*) and willow are also used for phytoremediation and mineralization (Kuiper et al. 2004). Metals are indispensable for plants' biological functions, but they obstruct the organism's metabolic system at

Table 10.2 List of the various plant species screened for their heavy metal removal ability

Plant species	Metals	Method of degradation	Accumulation/removal	References
Vetiver zizanioides	Fe, Zn, Pb, Mn, Cu	Bioaccumulation	Maximum removal of Fe (98%) followed by Zn, Pb, Mn, and Cu	Sharif et al. (2016), Ng (2017)
Hordeum vulgaris	Pb, Zn	Rhizoaccumulation	The accumulation capability of *Hordeum vulgaris* was 38 mg/kg	Yang et al. (2017)
Spartina maritima	As, Cu, Pb, Zn	Rhizoaccumulation and bioaugmentation	19–65% pollutant removal from the initial concentration of 2153 mg/kg	Mesa et al. (2015)
Arundo donax	Zn, Cd	Rhizofiltration	100% removal from the initial concentration of 66.6 for Zn and 783.9 kBq/dm^3 for Cd	Dürešová et al. (2014)
Water hyacinth	Al, Pb, As, Cd, Cu	Phytoaccumulation	73–82.4% removal rate for all the heavy metals	Aurangzeb et al. (2014)
Salvinia natans	Zn, Cu, Ni, Cr	Phytoaccumulation	41–84.8% removal rate for all the heavy metals	Dhir et al. (2011)
Eichhornia crassipes	Cu, Mn, Pb, Cd	Bioaccumulation	Bioaccumulation factor for sediments was maximum for Mn and minimum for Cu whereas the bioaccumulation factor for water was maximum for Cu and minimum for Cd.	Prasad and Maiti (2016)
Pistia stratiotes	Cd, Cu, As, Al, Pb	Phytoaccumulation	73–82.8% removal efficiency for all the heavy metals	Aurangzeb et al. (2014)

higher levels. Metals like Hg, Zn, Cr, Se, Cd, Pb, and As, etc., are not expedient to plants. Moreover, they reduced photosynthetic activities, enzymatic activities, and mineral nutrition (Nematian and Kazemeini 2013). Depth and toxicity of the pollutant, plant adaptability, growth and survival rate, plant root system, biomass, resistance to pests and diseases, time requirement, etc., are crucial factors for choosing a plant that can be used for phytoremediation (Lee 2013). Miguel et al. (2013) reported that in some polluted sites, phytoremediation takes place through uptake and translocation of pollutants aided by xylem vessels and gets accumulated in shoots of the plant. Accumulation and transpiration are further dependent on segregation between xylem sap and adjacent tissues and the transpiration rate of the plant. Plant type and nature of the pollutant are other prominent factors on which the phytoremediation process depends. Most of the native plants growing on the polluted site can act as good phytoremediators. Thus, the pace of phytoremediation mainly relies on increasing the remediation capability of native plants by augmenting them with exogenous or endogenous plant rhizobacteria or by biostimulation. The role of plant growth-promoting bacteria (PGPB) in phytoremediation has been widely reported in the literature (Yancheshmeh et al. 2011; Maqbool et al. 2012; de-Bashan et al. 2012; Almansoory et al. 2015; Tiecher et al. 2016; Grobelak et al. 2017; Ramakrishna et al. 2020). Some valuable metals can be bioaccumulated in certain plants that are recuperated/recycled by the phytomining technique. Other relevant advantages of phytoremediation include cost-effectiveness, easy operation, low maintenance costs, soil structure preservation, and mitigation of soil erosion and metals leaching in the ecosystem (Ali et al. 2013). Owing to the organic matter input, there might be an improvement in soil fertility after phytoremediation (Mench et al. 2009).

10.5 Mechanisms of Phytoremediation

The phytoremediation mechanism is influenced by many factors, such as the nature and type of pollutants, concentration, and soil characteristics (Sharma and Kaur 2020). Mineral and nutrients as well as other nonessential elements or contaminants in the soil are absorbed by the plant root, which offers an enormous surface area for absorption (Yang et al. 2017). In order to effectively remove heavy metal contaminants from the ecosystem, plants have many mechanisms, which includes phytoextraction or phytoaccumulation, phytodegradation, phytovolatilization, phytostabilization, and phytofiltration or rhizofiltration (Dhir 2013; Ashraf et al. 2019; Sharma and Kaur 2020; Rai et al. 2020) (Fig. 10.2).

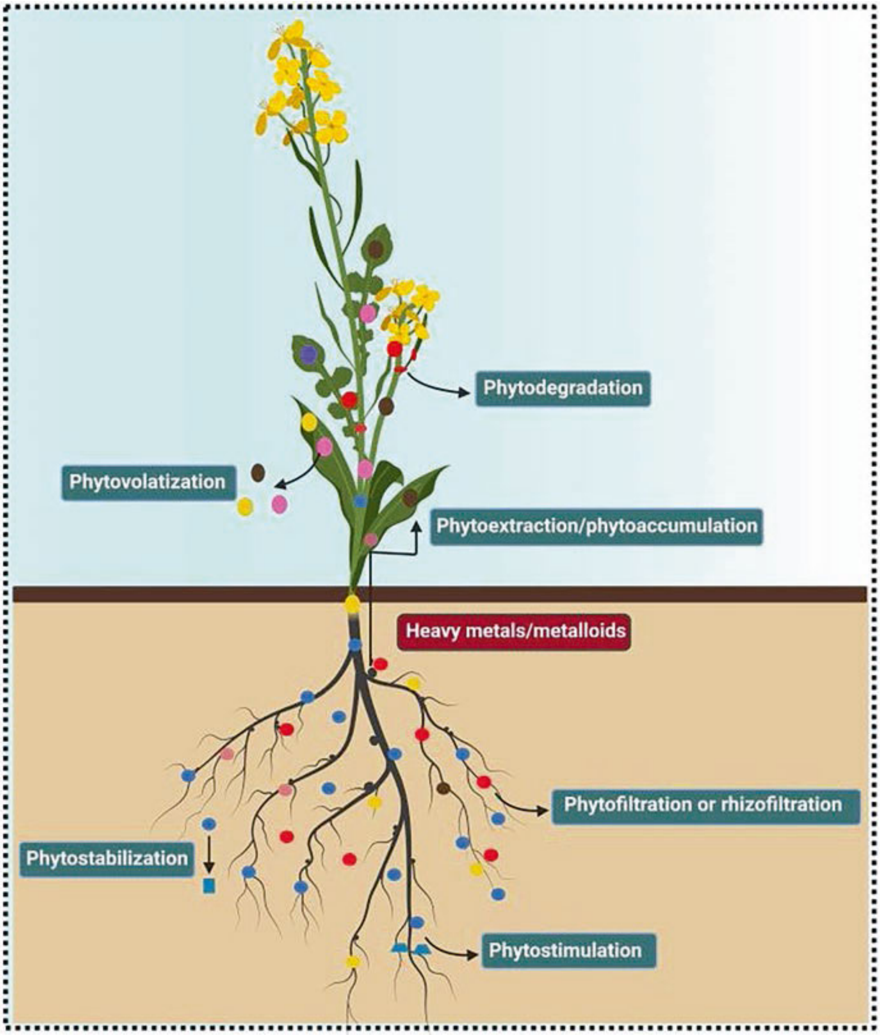

Fig. 10.2 Schematic mechanism of different processes involved in phytoremediation

10.5.1 *Phytoextraction/Phytoaccumulation*

It is defined as the acceptance of contaminants via plant roots from the soil and translocation to the aboveground portions of the plant (Pajevic et al. 2016). This method applies to metallic and radionuclides. This type of phytoremediation is reported to remediate Cd from soil, water, and sediments (Van Nevel et al. 2007). Furthermore, harvestable parts of the plants, where the metals are deposited, can be recycled from the ash that remained after drying, ashing, and composting (Singh and Bhargava 2017). Moreover, crops having high-biomass content can be grown in the contaminated soil to bio harvest and recover heavy metals easily. The concentration of contaminants in the soil can be decreased with successive cropping and harvesting (Pajevic et al. 2016).

Moreover, phytoextraction is a cheaper method compared to other conventional methods. It has an application in mineral industries to commercially produced metals by cropping (Sheoran et al. 2009). Also, it is advantageous when rapid immobilization is required to conserve drinking water resources.

10.5.2 Phytodegradation

Phytodegradation is also known as phytotransformation. It is defined as the uptake of contaminants from the soil and degradation of complex organic components resulting in the formation of simpler molecules or incorporating these molecules into plant tissues (Dhir 2013; Kumar et al. 2018; Yadav et al. 2018). The plants utilize the broken byproducts of the contaminants for their development and growth. Fertilizers, pesticides, chlorinated solvents, and other organic compounds can be degraded by phytodegradation (Sharma et al. 2019; Sharma and Kaur 2020). Phytodegradation has been earmarked to fix a few of the organic pollutants, which include chlorinated solvents, pesticides, and herbicides (EPA 2000). There are various plant enzymes such as nitroreductase, lactase, dehalogenase, peroxide, and nitrilase, which helps in the breakdown of complex inorganic and organic contaminants into simpler forms in plants (Dhir and Srivastava 2013). For example, *Myriophyllum aquaticum* secretes nitroreductase, which helps in the degradation of TNT (trinitrotoluene) (Rajakaruna et al. 2006). *Brassica juncea* and yellow poplar *Liliodendron tulipifera* are genetically modified plants focused by biotechnologists because of their practical phytoremediation abilities (Karami and Shamsuddin 2010; Ashraf et al. 2019).

10.5.3 Phytovolatilization

It is defined as the uptake of pollutants by plants from the soil, conversion into volatile forms, and releasing them into the atmosphere via transpiration process (Zhao et al. 2016). Thus, contaminants are discharged into the atmosphere in a less toxic form (Sharma and Kaur 2020). Selenium (Se), Arsenic (As), and Mercury (Hg) are some of the hazardous metals which can be transformed into volatile forms such as dimethyl selenide and mercuric oxide and then transpired into the environment (Table 10.3). The transformed dimethyl selenide and mercuric oxide are less harmful to living organisms, so this is an efficient phyto technique for fixing contaminants (Sakakibara et al. 2010). Also, there is no trace of the transfer of contaminants to other matrices (Kumar and Gunasundari 2018). The advantage of

Table 10.3 List of various plant species reported for their phytovolatilization ability

Plant	Contaminants	Reference
Festuca rubra L.	Cu	Radziemska et al. (2017)
Athyrium wardii	Pb	Zhao et al. (2016)
Agrostis castellana	Cu, Pb, and Zn	Pastor et al. (2015)
Typha latifolia L.	Zn, Mn, Co, Cd, Cr, Ni, and As	Varun et al. (2011)

Table 10.4 List of various plant species reported for their phytostabilization ability

Plants	Contaminants	References
Pennisetum sinese, *Setaria pumila*, and *Elsholtzia splendens*	Cu, Cd	Cui et al. (2020)
Impatiens glandulifera	Cd	Coakley et al. (2019)
	Cd	Zeng et al. (2018)
Spinacia oleracea L. and *Solanum nigrum* L.	Cd, Cr, Cu, and Pb	Dinesh et al. (2014)
Clerodendrum indicum L.	Fe	Mukherjee et al. (2013)
Amaranthus spinosus L.	Pb, Cd, and Cu	Chinmayee et al. (2012)

phytovolatilization in converting toxic contaminants such as mercuric ion into a less toxic form, i.e., elemental mercury, is also reported in several studies (Kramer 2018).

10.5.4 Phytostabilization

Phytostabilization is also known as in-place inactivation and is primarily utilized to fix contaminants in soil, sediments, and sludges (Singh 2012; Zeng et al. 2018). In this process, contaminants are immobilized in the rhizosphere by agglomeration by roots via root hairs, adsorption onto the root surface, or precipitation (Khalid et al. 2017). Phytostabilization decreases contaminants' mobility, and hence intercepts contaminants' migration to groundwater and therefore enters into the food chain (United States Protection Agency 2000). This technique successfully helps in reestablishing vegetation in polluted sites (Kohler et al. 2014). Besides, this type of phytotechnology has been employed for the remediation of severe toxic metals such as As, Pb, Cu, Zn, Cr, and Cd and is also helpful in reducing soil erosion and runoff via fixation of soil by plant roots (Bandara et al. 2017; Ramanjaneyulu et al. 2017). Various plant species screened for their capability to phytostabilization of heavy metals are enlisted in Table 10.4.

10.5.5 Rhizofiltration

Rhizofiltration is also called phytofilteration and is primarily used to remediate groundwater, wastewater, and surface water contaminated with a low concentration of pollutants (Ashraf et al. 2019; Javed et al. 2019). It is defined as the uptake of contaminants by plant roots and this technique helps alleviate contaminants present in natural wetlands. It involves aquatic as well as terrestrial plants for absorption and extraction of contaminants from contaminated water sources because of their extensive root mass (Uddin et al. 2016; Dhanwal et al. 2017; Yan et al. 2020). Toxic heavy metals which are adjourned within the roots are fixed by the technique of rhizofiltration. Some plants such as Indian mustard, tobacco, corn, spinach, and rye have been reported to remove Pb from the contaminated water resources. Among these, sunflower has the most remarkable ability to remediate contaminants (da Conceição Gomes et al. 2016; Yan et al. 2020).

10.6 Role of PGPRs in Phytoremediation

Plant growth-promoting rhizobacteria (PGPR) are mainly used to promote plant growth by assisting plants in uptaking nutrients from soils. However, incorporating the potential PGPRs with plants has been extensively used to remediate heavy metals (e.g., Cd, Cu, Cr, Hg, Pb, Zn, and Al) and organic pollutants in contaminated soils (Zhuang et al. 2007; Manoj et al. 2020). The potential PGPR strains used for heavy metal removal belong to the genus of *Acinetobacter*, *Azospirillum, Azotobacter, Bacillus, Enterobacter, Klebsiella, Paenibacillus, Pseudomonas*, *Rhizobium, Serratia,* and *Variovorax* (Abdelkrim et al. 2020).

PGPRs facilitate metal mobility and increase bioavailability to the plant by acidification, phosphate solubilization, and redox changes. They also produce iron chelators and siderophores that further aid in iron mobilizing and increase availability to the plant (Zhuang et al. 2007). They release numerous natural chelating agents and organic acids such as citric, oxalic, acetic, malic, succinic, gluconic, and 2-ketoglutaric acids, which reduces the soil's pH and sequester soluble ions (Khan and Bano 2018) (Fig. 10.3).

Recently, Manoj et al. (2020) reviewed the molecular mechanisms excreted by PGPRs to promote plant growth and heavy metals remediation. Abdelkrim et al. (2020) studied the in situ effects of *Lathyrus sativus* PGPR to remediate Pb and Cd polluted soils. They use PGPR inoculum (*Luteibacter* sp. + *Pseudomonas fluorescens* + *Variovorax* sp. + *Rhizobium leguminosarum*) and found that the Pb accumulation in the aboveground tissue was 1180.85 mg/kg DW, at the same time, the total reduction in Pb (46%) and Cd (61%) was also noted in the contaminated soils. Zand et al. (2020) investigated the effects of joint application of TiO_2 nanoparticles and PGPRs and reported that TiO_2 NPs and PGPRs increased *Trifolium repens* growth and Cd accumulation in Cd-contaminated soil. Recently, Guo et al. (2020) showed

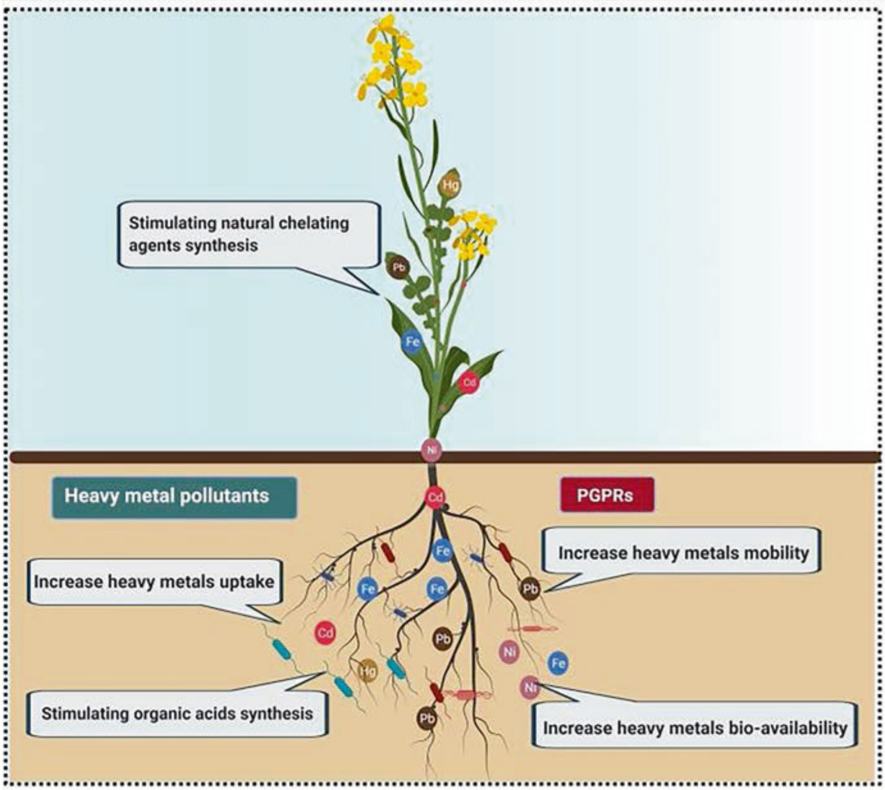

Fig. 10.3 PGPRs mediated heavy metals remediation mechanism

the effect of EDTA given in joint combination with PGPRs (*Burkholderia* sp. D54 or *Burkholderia* sp. D416) on the growth and metal uptake potential of *Sedum alfredii* plant. It was revealed that the EDTA decreased shoot and root biomass by 50% and 43%, respectively, meanwhile the uptake of heavy metals (Zn, Cd, and Pb) was also reduced to some extent. Wu et al. (2019) investigated the combination of biochar (BC) and PGPR strain with vetiver grass (*Chrysopogon zizanioides*) and observed that the Cd content was enhanced by 412.35% and bioaccumulation factor of accumulator 403.41%, as compared to control.

Moreover, Asad et al. (2019) reviewed the current status of PGPR with hyperaccumulator plants in improving the growth and remediation of heavy metals in contaminated environments. Furthermore, Mishra et al. (2016) and Mousavi et al. (2018) reported that siderophore-producing PGPR strains increase Fe, Zn, and Pb accumulation in their host plants. However, nowadays, genetically engineered PGPR strains associated with hyperaccumulator plants are used to remediate the heavy metals from the contaminated soils. These PGPRs strains are responsible for metal uptake, chelation, transport, degradative enzymes, homeostasis, and biotic-abiotic stress regulation (Ullah et al. 2015).

10.7 Role of Chelators in Phytoremediation

Phytoextraction is a promising tool for extracting heavy metals from soil and decontaminating a wide soil area (Zeng et al. 2018; Liang et al. 2019). However, the lack of availability and restriction on the translocation of certain heavy metals to plant shoots is a limiting factor in the process of phytoextraction. As a result, various chelating agents are used to increase metal solubility in soil thus increasing their availability to plants (Liang et al. 2019). Chelators and acidifying agents are added in the soil polluted with less concentration. This will help in increasing the solubilization in soil, and, thus, readily absorbed by plants. By forming a complex of metals, ethylenediaminetetraacetic acid (EDTA) introduces metals into the soil solution system and is eventually absorbed by the plant roots and translocated to the plant's aerial organs (Rostami and Azhdarpoor 2019). The most widely used phytoremediation agents are EDTA and citric acid (Lesage et al. 2005; Rostami and Azhdarpoor 2019). According to Lesage et al. (2005), the application of both chelating agents raised heavy metal concentrations in the soil, such as Cu, Zn, Cd, and Pb. In contrast to citric acid however EDTA is reported to increase the bioavailability of Cu and Zn to plants.

Chelating agents mainly follow the apoplastic pathway in plants (Duarte et al. 2011). Also, chelating agents can raise the concentration of soluble metals, adjust their transition pathway from symplastic to apoplastic in plants, and make it easier for them to transfer across the plant (Rostami and Azhdarpoor 2019). Additionally, the use of chelating agents improves a plant's drought tolerance (Rostami and Azhdarpoor 2019). High levels of toxicity in plants, leaching of these metals into groundwater, and intervention in metal transport from the root to the plant shoot are some of the negative consequences of these chelating agents (Rostami and Azhdarpoor 2019). Ethylenediamine disuccinate (EDDS) is secreted naturally by microbes and facilitates the uptake of heavy metals (Sidhu et al. 2018; Ashraf et al. 2019). EDTA is reported to show an excellent potential to absorb Pb accumulated in corn and pea, and further improves Pb accumulation ability in corn shoots. However, the application of chelates to improve metal bioavailability has raised so many queries, such as increment in the formation of a metal-chelate complex in the soil. Some studies have also pointed out the risk of groundwater contamination (Balakrishnan and Velu 2015). The effect of EDTA toxicity on microbes present in the rhizosphere is an indicator of environmental stress. Moreover, EDTA has household and industrial applications, such as cosmetics, detergents, perfumes, pharmaceuticals, water purification, agro-industries, paper, and pulp industry, is widely present in the environment, raising concerns about its role in heavy metal mobilization (Oviedo and Rodrıguez 2003).

Furthermore, despite their lower efficacy than other chelates such as phosphate and amino acids, EDTA-Zn and Zn sulfate chelates are widely recommended (Wang et al. 2013; Laware and Raskar 2014). Some chelates aid in absorbing more zinc; citrate, for example, has been documented to assist *Thlaspi caerulescens*, to absorb more zinc (Singh and Bhargava 2017). Citrate is also a metal chelator in the xylem tissue, indicating that it can play a pivotal role in the translocation of absorbed heavy metals in aerial regions (Singh and Bhargava 2017). Besides this, amino acids (AA), amino polycarboxylic acids (APCAs), and organic acids with low molecular weight (LMWOA) are the organic compounds used in phytoremediation of several heavy metals. For example, nicotianamine increased Cu presence by *Lycopersicon esculentum*, histidine increased Ni uptake by *Brassica juncea*, phytochelatins upregulate Cd uptake by *Helianthus annuus*, and cysteine is reported to increase the uptake of various metals by *Helianthus annuus.* These are some of the most widely used amino acids as per the reports (Meers et al. 2005). When plants are under extreme heavy metal stress, then free AA acts like nitrogen donor ligands and help the plant to adapt well in such condition by accelerating tolerance and detoxification mechanisms. Also, a significant increase in AA in plant tissues helps in adjusting osmotic stress and maintaining water potential in plants. Accumulation of heavy metals such as Uranium (U) by *Berberis vulgaris*, Cu, Zn, U, Pb, Cd, Zn by *Brassica juncea,* Cu by *Nicotiana tabacum*, *Eubleekeria splendens*, and *Trifolium repens* is assisted by LMWOA. These organic acids are exudates from plant roots that can control ion dispersibility and absorption owing to their metal-chelating features, indirect influence on microbial action, and alteration in rhizosphere region (Ashraf et al. 2019). Numerous genera of rhizobacteria (*Pseudomonas, Rhizobium, Sinorhizobium*, *Bacillus,* etc.) are used to enhance the pace of metal accumulation when phytoextraction is assisted by microbes (Meers et al. 2005; Ullah et al. 2015). Many symbiotic and nonsymbiotic bacterium can enable the absorption of heavy metals by plants such as *Pityrogramma calomelanos*, Cu by *Elsholtzia splendens*, Ni by *Eichhornia crassipes,* Pb by *Alyssum murale,* and Se by *Hordeum vulgare* (Ullah et al. 2015). Several factors such as pH, microbes, and the oxidation state of the metal (Ullah et al. 2015) affect the accumulation potential of plants. For example, a microbial strain has been reported to reduce the mobility and toxic nature of Cr(VI) to the nontoxic and immobile Cr(III), as well as minimize the mobility of other toxic ions in the soil system (Ullah et al. 2015). Furthermore, many beneficial bacteria in the rhizosphere are capable of improving plant growth, suppressing the activity of phytopathogens, and synthesizing plant hormones even under stress (Ullah et al. 2015). The metallophytes, which can withstand and survive in toxic and metal-polluted soil, are the most commonly used plant to stabilize metals. In nonsaline and saline soils of semiarid and arid areas, several studies have shown that halophytes are ideal for heavy metal phytoremediation. Experimental evidence indicates that resistance to salt and heavy metals depends on common mechanisms of physiology (Przymusiński et al. 2004).

10.8 Other Pragmatic Approaches for an Environmental Clean-up

Besides phytoremediation, there are some other approaches employed for the management of the environment. The electrokinetic assisted microbial approach is utilized efficiently to convert the organic substances to useful products electrochemically. The generation of bioelectricity can be made possible via different microorganisms' activities (Selvi et al. 2019). Literature survey revealed the use and successful operation of this integrated approach in the remediation of various heavy metals such as Hg, Co, Fe, As, Cu, As, Pb, etc., by various researchers (Azhar et al. 2016; Sharma et al. 2018; Cai et al. 2019). Likewise, to obtain efficient phytoremediation results, an electrokinetic assisted phytoremediation approach can be used. This integrated approach has been reported to alleviate the increased levels of Cr, Cd, and Cu in contaminated soil (Bhargavi and Sudha 2015: Azhar et al. 2016). Furthermore, the electrokinetic remediated soil was used to grow various plants.

A phytobial remediation approach is an integrated approach vastly explored and involves the simultaneous use of plants and microorganisms to remove excessive toxic metals from groundwater and soil. In this method, plants are used to uptake contaminants from soil or water, and microbes are utilized to break down these contaminants into less toxic forms (Selvi et al. 2019). Unlike other invasive technologies, this phytobial remediation approach is considered an environment-friendly and cheap process. Nevertheless, certain microbes (bacteria and fungi) are present in the plant's rhizosphere region and play a pivotal role in the immobilization of certain toxic metals by secreting enzymes. This remediation technique can further be investigated for proteomics, genomics, and metabolomics (Rai et al. 2020).

Practical application of hairy root biotechnology was also reported to remediate the areas contaminated with the mixture of organic and inorganic pollutants. However, this technique has the potential to remove the contaminants present at a very lower concentration. Thus, the technique has limited scope for large-scale phytoremediation (Ibañez et al. 2016). According to Song et al. (2019), the pace of phytoremediation can further be enhanced using nanoparticles to treat various contaminants. It was revealed that the remediation potential of ryegrass to remediate Pb from contaminated sites could be accelerated by the addition of Nano-hydroxyapatite (Huang et al. 2018; Song et al. 2019). Similarly, nanoparticles' addition augmented the phytoremediation ability of various plants such as *Phragmites australis, Salix alba,* etc. (Fernandes et al. 2017; Mokarram-Kashtiban et al. 2019).

Similarly, nanosized zerovalent iron, TiO2 nanoparticles, and salicylic acid nanoparticles are reported to enhance the phytoremediation of Trinitrotoluene (TNT), Cd, and As, respectively (Gong et al. 2017; Souri et al. 2017; Song et al. 2019). Also, the presence of humic acid can further enhance nanoparticles assisted phytoremediation (Le et al. 2019). Moreover, some algae, fungi, and aquatic plants are also reported for their tolerance, sequestration, and heavy metals' detoxification (Sharma et al. 2019). Literature survey unravels that those integrated technologies that involve the use of electrokinetics and bioremediation and phytoremediation had shown better results in alleviating the deleterious consequences of heavy metals. These techniques can be further utilized for their practical applicability in affected regions. Furthermore, there is an urgent need for a hyperaccumulators database to achieve site-specific phytoremediation (Reeves et al. 2018). For the proper recognition and acceptance of environmental friendly technologies, an integrated forum for ecologists, private/government agencies, environmental scientists, and engineers should be made (Rai et al. 2020; Sharma and Kaur 2020).

10.9 Advantages and Disadvantages of Phytoremediation

Plants and microbes have the potential to extract toxins from the environment and accumulate it in their bodies. Researchers compiled a list of the benefits, drawbacks, and limitations of phytoremediation technologies. These have shown the advantages of being ideal for a range of pollutants (organic compounds, metals, and metalloids), being cheap, and not requiring energy (energy is obtained from solar radiation) (Selvi et al. 2019). Phytostabilization is the first mechanism of phytoremediation, in which pollutants in the soil and groundwater were immobilized by using plants to avoid their movement by adsorption or accumulation onto the roots or precipitation within the root region (Awa and Hadibarata 2020). Through the process of phytostabilization, migration of heavy metals in the aerial parts of the plant can be prevented in those plants that have shown excellent tolerance potential under heavy metal stress (Sumiahadi and Acar 2018). When rapid immobilization is needed to save land and surface waters, it is highly effective. The deterioration or decay of heavy metals and organic contaminants in the soil is known as rhizodegradation. The application of microorganisms further aided this method (Mahar et al. 2016; Awa and Hadibarata 2020). It has a low start-up and maintenance rate.

The main benefits of phytoremediation technology are its low-cost equipment requirements, low labor costs, and cost-effectiveness. This technology may be used in situ, or on-site, to remove pollutants, whether in the soil, groundwater, or elsewhere. They are esthetically appealing and widely accepted by the general public. It has a relatively low environmental effect on soil and water because it is nondestructive, nonintrusive, and highly biologically active. They reduce soil erosion, make inorganic soils thinner, reduce particulate matter leaching, and disperse toxicants. Contaminants can be extracted from plant tissues, and plant biomass can be used for generating thermal energy and biogas. This technology shows the best results in the regions contaminated with low levels of heavy metals. It can be used for phytoremediation of non-agriculturally productive soils (Muthusaravanan et al. 2018). On the other hand, phytoremediation is slow and depends on soil profile composition, pH, salt concentration, and other toxins. There is limited applicability to various types of wastes, especially wastes with high levels of toxicity (Dhanwal et al. 2017).

10.10 Conclusion and Future Recommendations

Although it is evident that phytoremediation delivers as an economic tool for in situ mitigation of polluted sites, it is not feasible to get it into practice due to certain constraints. Moreover, field studies should be conducted in vivo rather than in vitro to analyze plants' efficiency to alleviate the environment's toxic metals. More investigations are required to understand better interactions existing among metals, soil, microbes, and plant roots and their mechanisms to degrade or detoxify the toxic metals. The application of various transgenic plants in the remediation of heavy metals can also help the elucidation of the phytoremediation mechanism adopted by the plants. Recent advances in plant biotechnology in genome editing can further play a pivotal role in accelerating phytoremediation. Also, elucidation of detoxification processes adopted by plants under heavy metal stress must be explored and explicitly studied. Exploring various hyperaccumulators, endophytes, algae, and their potential to remove high concentrations of toxic metals present from soil/water is required.

Lastly, in perspective to expand the horizon of phytotechnologies and other approaches for environment management, nanoparticles' role in enhancing the pace of phytoremediation needs to be understood. Other international agencies should be developed to conduct regular meetings to address the challenges and barriers in the path of the evolving technology of phytoremediation.

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