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REVIEW

New developments in biological phosphorus accessibility and recovery approaches from soil and waste streams

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This article is dedicated to Prof. Thomas Bley on the occasion of his 70th birthday.

Abstract

Phosphorus (P) is a non-renewable resource and is on the European Union's list of critical raw materials. It is predicted that the P consumption peak will occur in the next 10 to 20 years. Therefore, there is an urgent need to find accessible sources in the immediate environment, such as soil, and to use alternative resources of P such as waste streams. While enormous progress has been made in chemical P recovery technologies, most biological technologies for P recovery are still in the developmental stage and are not reaching industrial application. Nevertheless, biological P recovery could offer good solutions as these technologies can return P to the human P cycle in an environmentally friendly way. This mini-review provides an overview of the latest approaches to make P available in soil and to recover P from plant residues, animal and human waste streams by exploiting the universal trait of P accumulation and P turnover in microorganisms and plants.

KEYWORDS

biological phosphorus recovery, phosphorus recovery, phosphorus recovery from soil and waste streams, resource recovery, soil and waste streams phosphorus content

1 | INTRODUCTION

Phosphorus (P) is an essential macronutrient for all living beings. It is a necessary component of the energy production machinery in cells, a component of membrane structures and of DNA and RNA. P is involved in many regulatory processes of metabolism in cells and can also be stored in cells as an energy reserve in the form of poly-P. Due to these essential functions, P is, next to nitrogen, the most important component of fertilizers in agriculture. Here, P cannot be replaced by other components.

At present, mankind is dependent on the naturally dispersed P, which is mined as P rock. Large sedimentary deposits of P rock are found in Africa (Jordan, Morocco, and Western Sahara), China, the Middle East and USA [1]. P rock is only finitely available and inaccessible to many countries, including the European Union [2], which has therefore classified P as a critical resource [3]. Due to fast depleting reserves, the future of P dependent industries is uncertain. The most prominent P dependent industry is agriculture, and food security therefore is directly linked with P scarcity reality [2,4]. P recovery must, therefore, be undertaken to ensure the future availability of P.

Abbreviations: DM, dry matter; EBPR, enhanced biological phosphorus removal; P, phosphorus; PSM, phosphorus solubilizing microorganisms; SSA, sewage sludge ash; WWTP, wastewater treatment plant

PRACTICAL APPLICATION

Soil and human waste as a source of phosphorus can mediate an imminent shortage of phosphorus. Currently, the main methods of phosphorus recovery are chemical methods, which in most cases have a significant impact on the environment. On the other hand, there are environmentally friendly biological technologies for phosphorus recovery, but these are still at an early stage of research and development, apart from a few traditional applications. Therefore, the practical application of this study is to identify possible sources of biogenic phosphorus and approaches to biological phosphorus recovery in order to facilitate and promote the development of novel biological phosphorus recovery approaches.

P is abundant in soil and human waste streams. These include biologically unavailable P in soil, agricultural runoff after over-fertilization, crop residues, animal manure, food and food processing waste, and wastewater and sewage sludge [1,2,4 5]. In addition to better economic and ecological management of P dispersion, which would conserve P resources, the P recovered from these waste streams or made available to plants in the soil represents an alternative to the use of P rock [6–8].

P from different wastes can be recovered with both chemical and biological technologies. Chemical P recovery technologies with already available full-scale application focus on production of struvite from P rich liquid waste streams or from solid wastes such as biochar, and from incineration ashes obtained by pyrolysis or incineration processes [9]. In Christiansen et al. (2020) [6] a recent review on the current situation of chemical P recovery technologies is covered.

An alternative to chemical P recovery is biological P recovery. Biological P recovery strategies focus on biomass production that can be converted into useful products such as fertilizers or on mobilizing P in soil, which is otherwise unavailable. Here, the ability of both prokaryotes and eukaryotes, such as fungi, bacteria, algae, higher plants and animals, is used to store P in their cells. In bacteria, fungi and algae P is stored in form of poly P [10], in plants P is organically bound as phytate, predominantly in seeds and grains [11,12], while in animals P can be found in tissue, bones and scales. Apart from storing P in their cells, living organisms can release P by dissolving poly-P or provide P by biomass degradation processes. In this mini-review, biological P-recovery is defined by the application of any biologically based process that reclaims P into end products valuable to humans. Different approaches for P recovery can range from traditional technologies that support formation of P rich products (fertilizer production, e.g. composting) to more unconventional approaches, which are still in early stage of development (e.g., new biosorbents for P recovery). Most traditional biological technologies that facilitate P recovery can be find at NUTRIMAN (Nutrient Management and Nutrient Recovery Network https://nutriman.net/farmer-platform/technology-categories).

Here, an overview of recent and currently most promising biological P-recovery approaches will be given, focusing on the source of P-containing soil andman-made waste, the scale of application, and the efficiency of the applied recovery strategies (Figure 1). All P values cited in the review were recalculated to g of elemental P per kg or per L if respective data were available in literature.

2 | AVAILABILITY OF P IN SOIL

P amount in soil ranges in a concentration of 0.4 g to 1.2 g P kg^{-1} soil. The soluble P fraction, which can be taken up by plants is less than 1% of these values [13,14]. Furthermore, due to the presence of iron, aluminum and calcium in the soil, about 75% to 90% of the added fertilizer is precipitated by complexation with metal cations and therefore no longer available [15]. Making most of the unavailable organic and inorganic P in soil available for agriculture could greatly reduce the use of industrially produced fertilizers [14,16]. Insoluble organic and inorganic P can be solubilized by P solubilizing microorganisms (PSM), which comprise bacteria, fungi, and cyanobacteria [16–19]. PSMs solubilize insoluble phosphates in the soil by acidification, chelation, mineralization and ion exchange reactions [15,20].

Potential PS bacteria such as the genera *Bacillus*, *Pseudomonas, Burkholderia, Serratia, Achromobacter, Agrobacterium, Micrococcus, Aerobacter, Flavobacterium, Acinetobacter, and Pantoea* were reported by Alori et al. (2017) [19], Kalayu (2019) [15], and Aliyat et al. (2020) [21]. Most promising PS fungi genera were *Penicillium* and *Aspergillus* [15,22], which potentially are even superior to bacteria [23]. The arbuscular mycorrhizal symbiosis between plants and fungi is another microorganism-driven process that increases phosphate availability [24]. Also, cyanobacterial genera, such as *Calothrix, Anabaena* and *Nostoc*, have been reported to contribute to P solubilization in the soil [15,25]. Furthermore, there are approaches to isolate and identify new PSM from desert soils [26], subtropical soils [27] or—interestingly—from phosphate mine sites [21,28], which showed a potential for P solubilization.

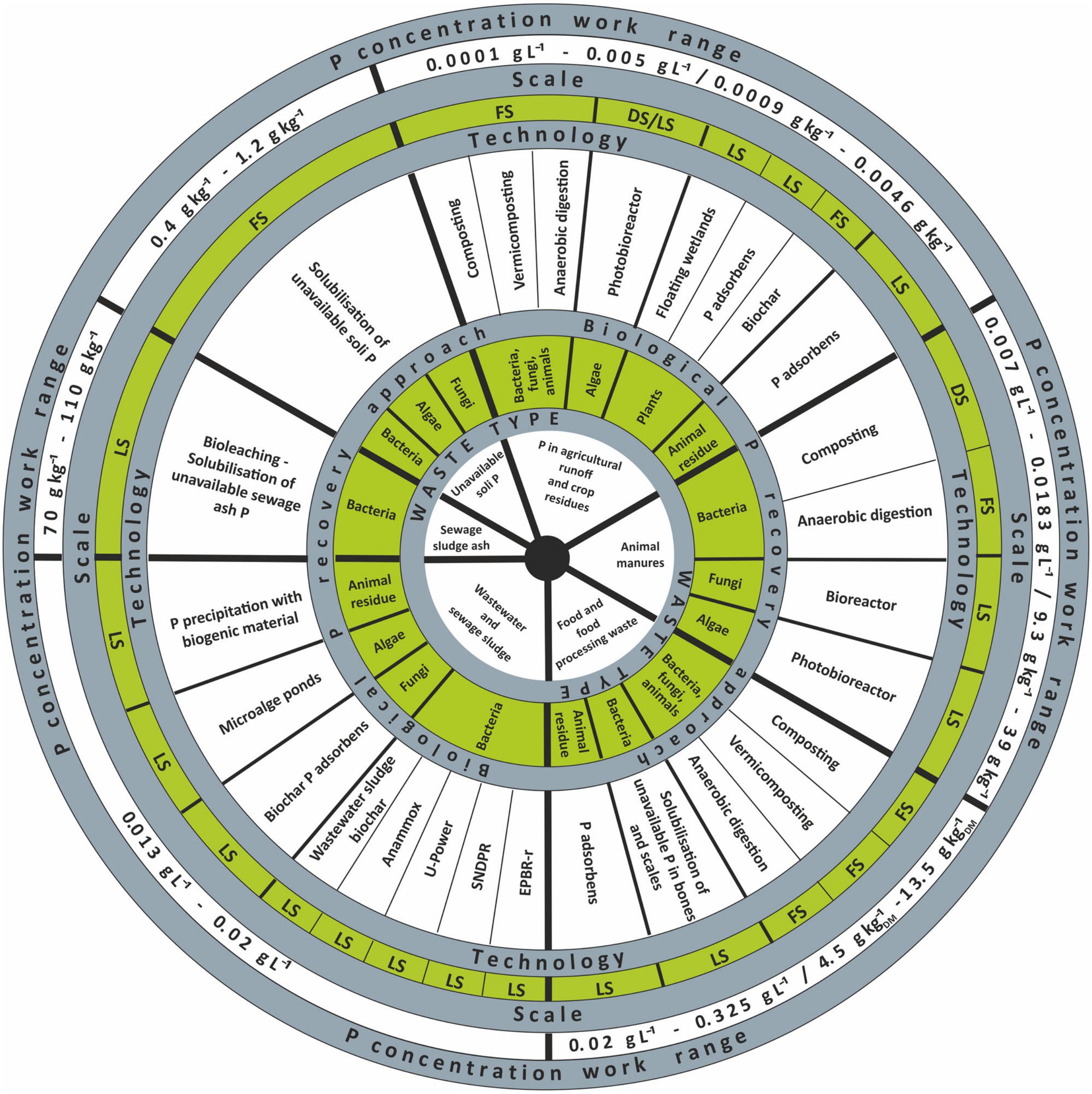

FIGURE 1 Overview of P-rich human waste streams together with possible biological P recovery approaches, application scales and P concentration work range. DM, dry matter; DS, demonstrator-scale; ERPB-r, enhanced biological phosphorus removal and recovery; FS, full-scale; LS, lab-scale; SNDPR, simultaneous nitrification, denitrification and P removal; U-Power, urine powered microbial electrochemical system

PSM were shown to improve plant resistance to pathogens, increase biomass yield and lower the need for industrially produced fertilizers [14]. The application of PSM has moved from lab-scale to greenhouses and fields with different crops [14] together with products based on PSM [29], which showed high crop growth promotion. Therefore, the supportive effect of PSM via the mobilization of unavailable P in soil must be considered as a valuable method for P recovery.

3 | P IN AGRICULTURAL RUNOFFS AND CROP RESIDUES

Agricultural runoff is defined as P leaching from farmlands through extensive fertilizer application [4], where the P concentration of 0.0001 g L^{-1} is considered high enough to cause eutrophication [30]. P loss in agricultural runoff is estimated to be around 8 Mt a^{-1} worldwide [31]. It causes eutrophication of water bodies evident in toxic algae growth [2,32] up until to the development of death zones in lakes and oceans due to low or no oxygen availability [33]. P enriched waters have been shown to be treatable with bacteria, algae or plants. Rueda et al. (2020) [34] demonstrated high P removal efficiencies (99% out of 0.00042 g P L^{-1}) from eutrophic water using microalgae and cyanobacteria in photobioreactors. *Chlorella vulgaris* was shown to remove P with an efficiency of 97% of initial 0.00063 g P L^{-1} on lab-scale [35]. A demonstrator-scale microalgae-bacteria photobioreactor (*Chlorella* sp., *Stigeoclonium* sp.) investigated seasonal impacts on the P recovery efficiencies showing underusage of P in summer and limited uptake in winter [36]. Here, the algae harvest can be supported in open ponds with algae scrubbers [37]. Generally, algae biomass can be used to obtain bioproducts such as biopolymers, pigments, food additives [34], pharmaceuticals, fertilizers [10] and bioenergy [36].

A valuable approach for the treatment of eutrophic waters seems to be biological composite material consisting of thermally treated marine mussel shells in combination with Al, La and Fe [30]. On lab-scale, P adsorption was up to 80% of the initial 0.005 g P L^{-1} from synthetic eutrophic water without any desorption being detected. A comprehensive overview of recent progress and current and future practice in agricultural run-off control can be found in Xia et al. (2020) [38].

New developments emerged in the context of applying plants either for the purification of run-off waters, for the storage of P in biomass and for the use of harvest residues as P source as well as new composite materials for P adsorption. On lab-scale the treatment of agricultural run-off using floating wetlands with suspended wetland plants is reported by Spangler et al. (2019) [39]. Wetland plant *Pontederia cordata* has removed 90% of P from an initial concentration of 0.00261 g L^{-1}. Macrophytes such as duckweeds (*Lemna minor, Landolita punctata, Spirodela polyrrhiza, Spirodela oligorrhiza*) and water hyacinths are other typical plants used in such applications [40,41]. Full-scale application of the floating wetlands is reported by Lavrnic et al. (2020) [42] but with inconsistent P removal efficiency due to the rain precipitation effect. A subsequent P-recovery from biomass is a valid option. The P content of crop residues gathered (3591 Mt a^{-1} of dry matter [DM]) from arable land is estimated to be around 4.35 Mt P a^{-1} with the P content range of 0.0009 to 0.0046 g kg^{-1} depended on the crop species [43,44]. Applied on arable land agricultural residues increase efficiency of P fertilizer uses and simultaneously provide P from plant biomass [45]. Furthermore, P uptake by plants, for example, in wetland buffer zones, facilitates water purification and returns the P to human processes, for example, through anaerobic digestion of the biomass, composting or as animal feed [46]. Bacterial and fungi activities and bacteria-earthworm synergies in compost are well known and widely applied. The current state of composting and vermicomposting of harvest residues and the transformations into products, which contain available P for plant growth was covered by Roy (2017) [41] and Ahmed et al. (2019) [8]. Harvest residues with a high P content can also be used as fertilizer in form of biochar products. Widespread waste substances such as aspen wood fibers and rice husks are further possible raw materials for the production of biochar [47]. Corn stalks converted to biochar and then impregnated with layered double hydroxides as P adsorbent showed a high adsorption capacity of 152 g P kg^{-1} on lab-scale [48]. Not only biochar can serve as an absorbent, but also pure fibers (e.g. from soybeans) can be mixed with fermentation residues, ensuring that P is first absorbed and then slowly released [49]. Biobased absorbents are efficient and sustainable. Perspectives, challenges and solutions for P recovery using sorbent can be found in an overview by Othman et al. (2018) [47].

4 | P IN ANIMAL MANURES

Approximately 15 Mt P a^{-1} is released from domestic animal manure worldwide [31]. In average the total P content in diary, poultry, and pig manure is 9.3 g, 18 g and 39 g P kg^{-1} manure, respectively [1]. Recovering all P from animal manure could satisfy 90% of the annual agricultural demand worldwide [50].

Firstly, P from manure is recovered by using manure directly as a fertilizer. However, due to high concentration of nutrients, nitrogen and phosphates can leach into the ground water together with high contents of antibiotics, which causes water pollution and leads to a spread of antibiotic resistant genes or bacteria into the environment [51–53]. Efforts to recover P from animal manures without causing over-fertilization and negative environmental effects can be made with the help of bacteria, fungi and algae. Depending of the liquid, semi-solid or solid states of manure, different approaches for P recovery can be applied accordingly. Solid and semi-solid manure is frequently added to large scale anaerobic digesters (AD) as a nutrient source [40,54], however, this routine does not recover P instantly from manure [55]. Integrating AD with composting plants presents attractive solutions to handle anaerobic digestate to yield a combined P rich environmentally friendly product [56] without risking the drawbacks coming from applying anaerobic digestate directly as a fertilizer [57]. Often unfavorable physical-chemical characteristics such as C:N ratio, moisture contents or pH values of manure digestate can be reduced when mixed with other substrates such as wood chips, zeolite or lignite [58,59]. Solid manure composting full-scale studies reported a low level of pathogens and the production of high quality compost [59].

P recovery from liquid manure is facilitated with diversified microorganisms. For swine manure bacteria and microalgae (*Chlorella* and *Coelastrella* strains) have been used [60,61]. Similarly, Lou et al. (2019) [62] reported a lab-scale application of the microalgae *Desmodesmus* sp. in a symbiotic interrelationship with bacteria (*Pseudomonas* and *Bacillus* strains) in a flat plate photobioreactor, where P was recovered with 0.007 g P L^{-1} d^{-1}. However, the high nutrient content, the dark color and the amount of suspended solids of raw manure slurries often inhibit photosynthesis and thus algae growth [61].

Fungi can also be applied in treatment of liquid manures. He et al. (2019) [63] used a 20 L demonstration-scale reactor where the filamentous fungal strain *Mucor circinelloides* was applied and accumulated 78% of the initial P concentration of 0.0183 g L^{-1}. The fungal biomass was reused in 7 batches for further P accumulation to produce a fertilizer with slow P-release. In summary, animal manure as a source of P has a great potential to return P to the human P cycle.

5 | P IN FOOD AND FOOD PROCESSING WASTES

Approximately 1300 Mt a^{-1} of food waste is generated worldwide [64] with P content in DM ranging from 4.5 to 13.5 g kg^{-1}[65]. P from organic food waste is frequently made available via composting or anaerobic digestion on full-scale. Both compost and anaerobic digestate are applied to the fields as fertilizer, although the digestate is questionable as an unstable and unhygienic fertilizer source [57,66].

New approaches have been developed for foods that are not readily biodegradable, such as egg shells [67], sea shells, and fish scales [68] that are rich in biologically unavailable P. PSMs, such as the bacterial strain *Acidovorax oryzae* can be used to treat hydroxyapatite powder from tilapia fish scales (*Coptodon rendalli*) where 40% of initial 0.325 g P L^{-1} were recovered on lab-scale [17]. Also, *Bacillus megaterium* was used on lab-scale to recover P from poultry bones and ash after incineration [69]. The bones were found to be the best source of P with 53.2 g of P solubilized from initial 77 g of P contained in the bone sample. Waste from fisheries such as crab shells can also be used to produce a chitosan calcium-rich adsorbent. Pap et al. (2020) [70] found an adsorbent capacity for P of 76.9% of the initially provided 0.02 g P L^{-1} on lab-scale. This capacity was even higher for modified mussel shell powder used for microalgae immobilization with P removal rates of 88% from initial 0.1025 g L^{-1} on lab-scale [71]. In summary, classical approaches such as composting and anaerobic digestion for biodegradable food waste are commonly utilized full-scale and provide P for fertilization. However, new technologies for use of less degradable food wastes are still scarcely available.

6 | P IN WASTEWATER AND SEWAGE SLUDGE

Around 3 Mt P a^{-1} is released in human urine and feces contained in wastewater worldwide [31]. Wastewater biological P removal is often coupled with chemical P recovery strategies on P containing wastewater streams to either reduce the P amounts in the streams or to recover it for further use or both [72]. Current wastewater P recovery technologies mostly rely on chemical based struvite production [73]. Therefore, there is lot of future potential for developing and applying biological P recovery approaches for wastewater treatment plants (WWTP) [74], and here technologies that show potential will be shown.

Wastewater is routinely treated with microbial communities to remove P, but currently there is no clear path for purely biological P recovery. Wong et al. (2018) [75] developed an approach to improve enhanced biological phosphorus removal (EBPR) towards a process of enhanced biological phosphorus removal and recovery (EPBR-r) process on lab-scale. A microbial biofilm containing polyphosphate accumulating organisms (PAOs) was used to take up P under aerobic conditions, which was even increased by extending the aerobic P uptake time. At the same time the hydraulic loading was increased by unchanged oxygen and carbon availability. In a subsequent step, P was released under anaerobic conditions and rose up to 0.09 g P L^{-1}. This approach improved the growth of phosphate accumulation organisms (PAO). Furthermore, this technology could be a good starting point for future full-scale EBPR-r designs with greater P removal and recovery capabilities. Another approach to produce P rich recovery streams was described by Salehi et al. (2019) [76], where a simultaneous nitrification, denitrification and P removal (SNDPR) process was facilitated on lab-scale by denitrifying polyphosphate accumulating organisms (DPAO) and denitrifying glycogen accumulating organisms (DGAO). High P concentrations were generated at the end of the anaerobic phase with 0.1 g P L^{-1}. Yang et al. (2017) [31] provided an overview on the main microorganisms serving as PAOs in WWTPs. Further new developments exploit microbial bio-electrochemical systems including microbial fuel cells (MFC) and microbial electrolysis cells, which are relatively new approaches for on-site energy generation and integrated nutrient separation [72,77]. An example of such a study demonstrates the utilization of human urine for nitrogen and phosphorus partitioning in a novel urine powered microbial electrochemical system (U-Power) on lab-scale [78]. Electrochemically active bacteria enable urea hydrolyzation and electrical potential driven NH_4^+ and PO_4^{3-} migration to subsequent nutrient recovery solutions. Bacteria responsible for this process belong to the genera *Enterococcus*, *Melissococcus* and *Tetragenococcus*, *Desulfovibrio*, *Pelobacteraceae* and *Geobacter*. The average total P recovery from fresh human urine was 0.1 g L^{-1}. A recent extensive overview on approaches for P recovery in wastewater using electroactive bacteria is given by Li at al. (2020b) [79]. Another interesting approach is using the anammox process together with phosphate precipitation [80]. On lab-scale the precipitation was found to take place inside of the anammox granules with 65.5 ± 6.1% of the initial 0.013 g P L^{-1}.

Also, fungi can be used for P recovery in wastewater. Jack et al. (2019) [81] reported the application of magnetic biochar produced from the fungi *Neurospora crassa*, which was grown on an iron-rich wastewater stream. The iron-rich magnetic biochar was tested in lab-scale and showed P adsorption capabilities of 23.9 g P kg^{-1}. Previously covered fungi application for wastewater P recovery can be found in Carrillo et al. (2020) [82].

P recovery from wastewater can also be achieved with algae by enriching biomass [55]. On lab-scale, microalgal polycultures in ponds were used to remove 50 to 90% of total P (out of 0.0032 ± 0.0006 g L^{-1}) from the secondary effluent [83]. Commonly used algae for these applications are from the genera *Chlamydomonas*, *Chlorella*, *Euglena*, and *Scenedesmus* [31,84,85]. Microalgae can also be applied in MFC for P recovery [86]. The harvesting of the wastewater algae biomass can be supported by filamentous fungi via a bioflocculation process [87].

The potential of biogenic materials that can be used for P precipitation is large and more promising than the use of, for example, magnesium, which is also on the EU list of critical raw materials. Bivalve seashells (mussels, scallops, oyster, Manila clam shell) can be used to obtain calcium hydroxide, which precipitates P as hydroxyapatite from diluted human urine with P recovery rate of 95% of initial 0.02 g P L^{-1} [68]. An extensive review on technologies used for wastewater treatment including biological P recovery is provided by Carrillo et al. (2020) [82].

The wastewater purification process leads to especially high concentrations of P in sludges [88], which were used directly as fertilizer for years. But this practice is no longer accepted due to the high risks caused by pathogenic and antibiotic resistant bacteria as well as the frequently high concentration of heavy metals and pharmaceuticals [10,89–91]. Sludge can be converted into biochar to mobilize P by supporting soil bacteria and fungi. But in general, new laws such as the German Sewage Sludge Ordinance [92] require all WWTP with a size >50,000 population equivalents to recover P from sludge within the next 12 years if the P values exceed 20 g P per kg DM of sludge. Other European Union states such as Sweden, Austria and Poland are also undertaking similar initiatives [93–95].

7 | P IN SEWAGE SLUDGE ASH

Sewage sludge ash (SSA) is produced during the incineration of sewage sludge and is rich in P making it a promising source for P recovery. The amount of SSA produced is 1.7 Mt a^{-1} worldwide, with the P content of SSA ranging from 7 to 11% which is 70 g P kg_{SSA} - 110 g P kg_{SSA} [96]. The common SSA P recovery method is the dry thermal or wet chemical method using strong acids or bases [12,97]. The wet chemical SSA extraction method performed with strong acids can be replaced by a P recovery via bioleaching. Semerci el al. (2019) [98] used unidentified sulphur oxidizing bacteria isolated from a WWTP on an SSA suspension with elemental sulphur to lower the pH and facilitate the leaching of the P from the SSA mineral form to the ionic form. A bioleaching experiment on lab-scale reached an amount of P released from SSA that was 1.3 g of originally 1.6 g P per 20 g ash. Most applied bioleaching microorganism are sulphur oxidizing bacteria such as *Acidithiobacillus thiooxidans*, iron sulphur oxidising bacteria *Acidithiobacillus ferrooxidans*, and iron-oxidizing bacteria, *Leptospirillium ferrooxidans* and *Leptospirillum ferriphilium* [99,100]. Therefore, bioleaching can be a possible alternative for the future extraction of P from SSA.

8 | BIOTECHNOLOGICAL APPROACHES TO ENRICH AND DOWNSTREAM P

Future approaches for enhancing biological P recovery include genetic engineering of microorganisms. Genetic engineering can produce more efficient PAOs using well characterized microbial chassis such as *Pseudomonas putida* or *Escherichia coli* with additional enzymes to maximize P uptake [10] for in house application. A further promising lab-scale application is the *Escherichia coli* enzyme AppA phytase, which was transferred via vector into *Pichia pastoris* for overexpression and subsequently the protein was applied to different plants pressed in cakes to free plant bound P [11]. The highest amount of phytase-released P was 9.97 g P kg^{-1} of pressed cake. Poly-P can also be enriched in *Saccharomyces cerevisiae* after a starvation procedure and extracted and purified as poly-P chains of different lengths [101, 102]. For plants, a bioengineering approach in conventional plant breeding can be steered into a direction where plants can produce higher amounts of organic acids and enzymes to facilitate better soil P immobilization and utilization [1].

9 | CONCLUDING REMARKS

Human waste streams are rich in P and, if recovered, can replace mined phosphate rock. P recovery technologies using biological processes can provide environmentally friendly fertilizers, industrial chemicals, increase the quality of the soil and even become a source of biogenic material. A further focus on the development of biological technologies for P-recovery can mobilize enormous valuable waste volumes, which are not only good for biological P-recovery but also have positive effects on environmental protection, resource management, degraded soils and water bioremediation. Therefore, there should be an urgent effort to guide and support current biological P-recovery technologies from lab- and pilot-scale towards an economically feasible P-recovery.

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CONFLICT OF INTEREST

The authors have declared no conflict of interest.

DATA AVAILABILITY STATEMENT

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

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