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Review Article

Microbial Metabolic Engineering for Biopolymers Production

Priti Regmi, Tribikram Bhattarai

Central Department of Biotechnology, Tribhuvan University, Kirtipur, Kathmandu, Nepal

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CORRESPONDENCE

Priti Regmi

Central Department of Biotechnology
Tribhuvan University
Kirtipur, Kathmandu
Nepal

Email: priti.regmi@cdbt.tu.edu.np

0000-0001-9795-4123

Abstract

This review delves into microbial engineering strategies for biopolymer production, which is crucial for tackling contemporary environmental challenges. It surveys microbial species capable of synthesizing biopolymers and examines genetic engineering techniques to enhance biopolymer yields. For the genetic manipulation approach, mainly the over-expression of the genes responsible for the production, the downregulation/knock out of competing branches, or the abundant supply of cofactors needed for the reaction are mostly tested, among others. This study provides insights into a few applications of metabolic engineering endeavors towards the production of some biopolymers as for poly-γ-glutamic acid (PGA), polyhydroxyalkanoates (PHAs), starch-based materials, and polylactic acid (PLA). This review highlights the importance of genetic manipulation in optimizing microbial strains and fermentation processes for sustainable biopolymer production.

Keywords: Biopolymers, genetic manipulation, microbial engineering

Introduction

The increasing focus on natural biodegradable bio-based polymers in the scientific and industrial realms suggests an urgent imperative for sustainable development. With the environmental toll of petroleum-derived polymers becoming increasingly evident, there is a growing shift toward renewable alternatives such as proteins, polysaccharides, and lipids. This transition addresses pressing environmental concerns and meets the rising demand for eco-conscious materials. Biotechnology plays a pivotal role in this transformation, offering innovative solutions like genetic manipulation of metabolic pathways, heterologous gene expression, and alternative expression systems. This review explores key applications of genetic engineering in the development of biodegradable materials, highlighting its potential to drive sustainable innovation.

For decades, bacteria have emerged as an up-and-coming alternative for synthesizing polymers due to their metabolic versatility and efficiency. Their ability to produce biopolymers such as polysaccharides, polyamides, polyesters, and polyhydroxyalkanoates (PHAs) has been extensively explored as highlighted. This review also highlights the potential of different fungal genus such as *Aspergillus*, *Trichoderma*,

Penicillium, *Absida* etc. to produce various polysacchrides. Furthermore, besides bacteria and fungi, the role of algal species in producing different types of biopolymers such as polylactic acid, polyhydroxyalkanoates and polyhydroxybutyrates is also noted by the authors (Akinsemolu & Onyeaka, 2023). Other polymers, such as polyesters, polyamides, and polysaccharides, are frequently produced using microbes, including wild-type strains, mutated variants, or genetically modified organisms (GMOs). To elaborate, wild-type microbes are natural, unmodified strains found in the environment. Mutations can be introduced by inducing random changes in their DNA, often through exposure to mutagens like UV light or chemicals, in order to improve specific traits. In contrast, GMOs are engineered through precise genetic modification, where particular genes are inserted, deleted, or altered to achieve desired functions. These microbial approaches offer versatile and sustainable pathways for producing biopolymers. Xanthan gum, the first commercial polysaccharide—a polymer composed of long chains of carbohydrate (sugar) molecules—was initially produced using bacteria (Kumar et al., 2018). Through genetic manipulation, researchers produced a highly white-colored xanthan gum while reducing ethanol usage, thereby lowering downstream processing costs. This improvement was achieved by introducing the *Vitreoscilla* globin gene (vgb) into *Xanthomonas campestris* to enhance its metabolic capabilities (Dai et al., 2019). This review explores microbial-based polymer production and genetic engineering techniques aimed at enhancing synthesis, with a primary focus on key polymers such as PGA (poly-ϒ- glutamic acid), PHAs, starch, and PLA (polylactic acid).

Improvement of PGA yield

One of the most widely recognized polypeptides, frequently discussed for its biopolymer potential, is poly-γ-glutamic acid (PGA). This versatile biopolymer and its derivatives have numerous applications, including uses in the food (Tanimoto, 2010), and medical industries as a flocculant in water treatment (Campos et al., 2016), a chelating agent in wastewater treatment (Mark et al., 2006), an ingredient in cosmetics (Serra et al., 2024), and a carrier molecule for drug and gene delivery (Schlechter et al., 1989). PGA is a polymerized product of L- and/or D-glutamic acid by means of ϒ -amide linkage. A common genus used to produce PGA is *Bacillus* and its strains, such as *B. licheniformis* and *B. subtilis* (Xu et al., 2005). These are well reported to be a PGA producer by microbial fermentation. Genetic engineering techniques were employed in *Bacillus* species to increase the PGA yield (Li et al., 2022). Despite the potential of *Bacillus* sp. for PGA production, the presence of hydrolytic enzymes such as *pgdS* (formerly known as *YwtD*), *ggt* and *CWLO* poses a challenge as these enzymes can degrade the synthesized PGA (Yao et al., 2009; Scoffone et al., 2013; Mitsui et al., 2011) (Figure 1). To address this, some of the engineering strategies have been employed to increase the PGA yield, including the plasmid based γ-PGA expression, deletion of genes *pgdS* and *ggt*, and expression of glutamate racemase (Scoffone et al., 2013; Ashiuchi et al., 2006). Expression of the glutamate racemase (*glr*) gene positively impacts PGA yield, likely due to increased uptake of L-glutamic acid, a precursor of PGA. The *glr* gene encodes an enzyme responsible for converting L-glutamate to its D-isomer and vice versa, thereby enhancing the availability of substrates for PGA synthesis (Jiang et al., 2011).

Counterintuitively, overexpression of the PGA degradation gene *pgdS*, increased γ-PGA yield by 54%. Higher *pgdS* expression positively influences the glutamate transporter (glutamate being a precursor of PGA) and increases the expression level of the PGA synthetase complex. Together, these factors likely contributed to the increased product yield (Tian et al., 2014).

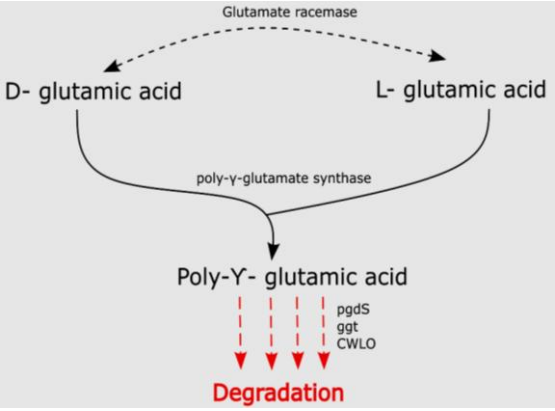

Figure 1: Microbial synthesis of poly-ϒ- glutamic acid (PGA). D- glutamic acid can be converted to L- form and vice versa using glutamate racemase. Polyglutamate synthetase complex can produce poly-ϒ- glutamic acid from its monomer. The degradation of PGA is mediated by the enzymatic activity of genes *pgdS*, *ggt* and *CWLO*.

Production of PHAs

Polyhydroxyalkanoates (PHAs), as naturally occurring biopolymers, have attracted considerable attention in various industries due to their biodegradable nature, biocompatibility, and physicochemical properties resembling those of petroleum-based counterparts (Sharma et al., 2021). Wang and their colleagues provide a thorough review of polyhydroxyalkanoates (PHAs) production, covering substrate roles, fermentation methods, and genetic engineering advances. Some notable examples listed include *Escherichia coli (E. coli)* modifications for ethanol-based poly(3-hydroxybutyrate) production and optimization of PHAs synthesis through propionate utilization (Wang et al., 2023). Propionate assimilation is crucial for generating propionyl-CoA, a precursor for medium-chain-length PHAs with odd-chain monomers (Zhuang & Qi, 2019). Microbes, particularly bacteria, are widely regarded as the most efficient systems for producing PHAs due to their natural ability to synthesize and accumulate these biopolymers. However, the high production cost remains a major barrier to scaling up PHA production (Tsang et al., 2019). Since substrate cost is the main expense, it can account for up to 50% of the total production cost (Pérez et al., 2020). To enhance the economic feasibility of PHAs commercialization, the utilization of low-cost carbon sources, as opposed to pure reducing sugar substrates, has been explored. For instance, studies have demonstrated that PHAs production can exceed 80% of the cell dry weight when waste rapeseed oil and propanol are used as substrates (Obruca et al., 2010). Among the various types of PHAs, polyhydroxybutyrate (PHB), which is composed of 3-hydroxybutyrate (3HB) monomer units, is one of the most extensively studied. PHB is produced by a wide range of microbial species (Markl et al., 2018).

There are numerous genetic modifications approaches tried to boost up the level of PHAs. The deletion of glucose dehydrogenase gene in *Pseudomonas putida* increased the titer of PHAs by 60%. The deletion of glucose dehydrogenase, an enzyme that directs glucose to gluconate, likely redirected more carbon toward PHA biosynthesis, resulting in increased production (Poblete-Castro et al., 2013). Similarly, a study from Salvachúa and the coworkers found that the knockout of several genes (*phaZ* and, β- oxidation related genes *fadBA1* and *fadBA2*) combined with overexpression of PHAs synthesizing genes (*phaC*, *phaG*, and *alkK*) was able to increase the yield of PHAs by using *P. putida.* Here, Salvachúa and the colleagues report a significant increase in PHAs yield by using solubilized lignin (derived from corn stover) as a primary carbon source for PHAs production. Genes *phaG, alkK,* and *phaC* catalyze forward reactions to produce 3-hydroxy acyl ACP and direct them towards mcl (medium chain length) – PHAs, while PhaZ reversely catalyzes this conversion (Ren et al., 2009; De Eugenio et al., 2010). Similarly, β-oxidation genes were deleted in this study to decrease 3-hydroxyacyl-CoA flux towards fatty acid β-oxidation and to increase the flux of 3-hydroxyacyl-CoA towards mcl- PHAs (Tsuge et al., 2000). Altogether, this genetic manipulation (Figure 2) done in *Pseudomonas putida* was able to give the highest titer and resulted in about a 200% increase in the yield of mcl polyhydroxyalkanoates (Salvachúa et al., 2020). This underscores the importance of a detailed understanding of metabolic pathways, where strategic interventions—such as overexpressing genes in the forward direction, downregulating competing branches, and minimizing reverse reactions—can significantly enhance the titer of a desired product, provided that the metabolic network is carefully balanced and optimized. In the context of other microbes, it has been shown with the over expression of *phbC,* P3HB yield was increased 1.4 times in *Ralstonia eutropha* (Barati et al., 2021). *phbC* is the gene encoding the main enzyme PHB synthase for PHB biosynthesis in *Ralstonia eutropha.* Another study carried out on Purple non-sulfur bacteria *Rhodobacter sphaeroides,* strain modified with the overexpressed PHA genes and deleted *phaZ* exhibited 1.7–3.9 folds higher production than the parent strain (Kobayashi & Kondo, 2019).

P3HB4HB is a biodegradable polymer within the PHAs family having good mechanical properties and thereby possess wide range of potential applications including tissue engineering (Wee et al., 2017, Luo et al., 2007, Crétois et al., 2015, Li et al., 2015). Acetyl CoA is converted to P3HB4HB in 3 steps. PhaA converts two acetyl CoA to acetoacetyl CoA. This acetoacetyl CoA is then converted to 3- hydroxy- butyryl CoA using PhaB. 3- hydroxy- butyryl CoA finally gets converted to P3HB4HB by PhaC. Chen and the colleagues

engineered *E. coli* to enhance P3HB4HB production by introducing *phaA*, *phaB*, and *phaC* from *Ralstonia eutropha* while deleting sad and *gabD* to increase succinate semialdehyde levels. Additionally, *sucD*, *4hbD*, and *orfZ* from *Clostridium kluyveri* were expressed to synthesize the key precursor, 4-hydroxybutyryl-CoA, leading to a titer of 0.85 g/l. Further optimization by expressing *ackA* and *pta* boosted acetyl-CoA levels, increasing P3HB4HB production to 1.71 g/l—nearly a 100% improvement. This study demonstrates how metabolic engineering can enhance biosynthetic pathways in *E. coli* for higher yields (Chen et al., 2018).

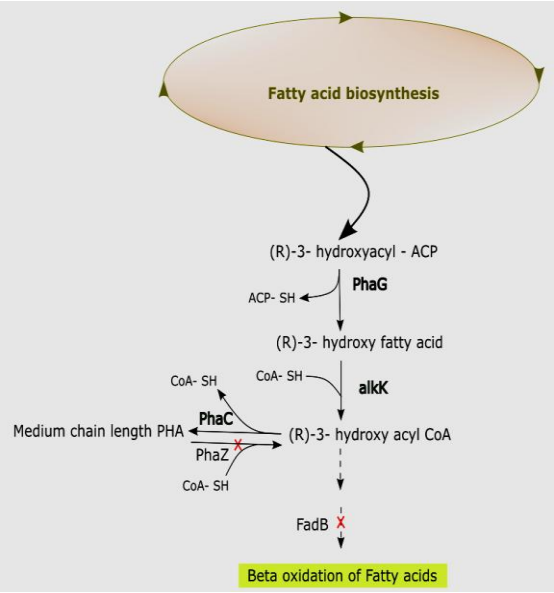

Figure 2: PHA metabolic pathway existing in *Pseudomonas putida*. Dotted lines and multiple arrows indicate multistep reactions. Red cross mark symbol in the arrow indicates deletion of the respective genes (*phaZ* and *fadB*), and bold letters shown for genes *phaG*, *alkK*, and *phaC* are the over expressed genes for high yield of PHA as done by Salvachúa et al., 2020.

The genetic modification of *Halomonas bluephagenesis* to enhance the xylose metabolic pathway, enabling the strain to utilize xylose as a substrate and achieve a poly-3-hydroxybutyrate (PHB) titer of 0.39 g/l (Tan et al., 2022). Further engineering of the strain, including optimization of the ribulose-5-P pathway and phosphoketolase pathway, resulted in a final PHB titer of 5.37 g/l. This highlights the potential of engineering diverse metabolic networks in microbial hosts to achieve high PHB yields. *Halomonas bluephagenesis*, a halophilic PHA-producing bacterium, has also been optimized in shake flask systems, where the expression of the *phaCAB* operon led to a PHAs content of up to 94% (Ren et al., 2018). Similarly, in *Pseudomonas putida*, genetic modifications such as the deletion of the *phaZ* gene, combined with DO-stat fed-batch fermentation, enabled the strain to utilize crude glycerol as a substrate and achieve a 48% increase in PHAs titer (Borrero-de Acuña et al., 2021).

Beyond the manipulation of metabolic pathways, studies have highlighted the importance of engineering cofactors, regulatory elements, promoter systems, and cell morphology to enhance PHAs synthesis (Wang et al., 2023). As an additional strategy, increasing the supply of cofactors required for PHAs synthesis has been explored in various studies. For instance, PHAs synthesis in most microbial strains relies on NADPH-dependent acetoacetyl-CoA reductase, making NADPH availability a critical factor for efficient production. To increase the supply of this cofactor, overexpression of the NAD salvage pathway genes *pncB* and *nadE* was employed, enhancing the NAD(P)H pool through improved NAD^+ biosynthesis. In *Cupriavidus necator*, overexpression of *pncB* and *nadE* not only increased PHB production efficiency but also improved tolerance to lignocellulose-derived inhibitors (Lee et al., 2022). Similarly, another study done with transhydrogenase gene overexpressed was shown to increase the NADPH pool for P3HB production. Transhydrogenase facilitates the conversion of NADH to NADPH, thereby boosting the NADPH pool, an essential cofactor for P3HB biosynthesis (Jung et al., 2019; Tadi et al., 2021).

Indeed, factors beyond pathway gene expression, such as cell structure, size, and intracellular space modifications, also influence cell morphology and thereby impact PHAs production, as discussed by Wang and the co- authors (Wang et al., 2023). Another study (Zhao et al., 2019) investigated *minC* and *minD*, known as FtsZ inhibitors (Bi & Lutkenhaus, 1993), and found that their deletion resulted in elongated cells with improved PHA titers. FtsZ is a bacterial protein that has a crucial role in coordinating cell wall and membrane growth, hence plays a key role during bacterial cell division Furthermore, in this engineered elongated strain background, it was shown that the individual overexpression of morphology related genes *mreB* and *ftsZ* made an increased titer of mcl- PHAs. However, when these genes *mreB* and *ftsZ* were simultaneously overexpressed, it resulted in the cells

with diverse shapes. In this scenario, the production went down, possibly due to adverse effects on cell division (Zhao et al., 2019). In another investigation, *E. coli* expressing PHB synthase underwent engineering, resulting in a notable increase in polyhydroxybutyrate (PHB) production, with the PHB titer rising from 5.72 g/l to 9.29 g/l. This enhancement was achieved by modifying the size of *E. coli* cells through engineering strategies, which included disrupting the *mreB* gene followed by its overexpression. The larger cell size was identified as a contributing factor to the higher accumulation of PHB granules (Jiang et al., 2015). Beside *mreB*, *sulA* was identified as another target to improve PHB titre. In *E. coli*, overexpression of *mreB* using a medium- copy number plasmid in a *mreB*-deleted background, combined with arabinose-induced expression of *sulA*, nearly doubled the PHB titer (Jiang et al., 2015). This demonstrates the potential for enhancing PHB accumulation in *E. coli* by targeting genes involved in cell morphology. MreB, an actin homolog, plays a critical role in peptidoglycan synthesis and maintaining cell shape (Rueff et al., 2014), while *sulA* inhibits FtsZ, leading to filamentous cell formation, increased cell size, and greater intracellular space for PHA accumulation (Bi & Lutkenhaus, 1993; Fenton & Gerdes, 2013). These examples highlight the importance of cell morphology-related genes in PHAs production, emphasizing how cell shape and internal structural space can significantly impact PHAs yields.

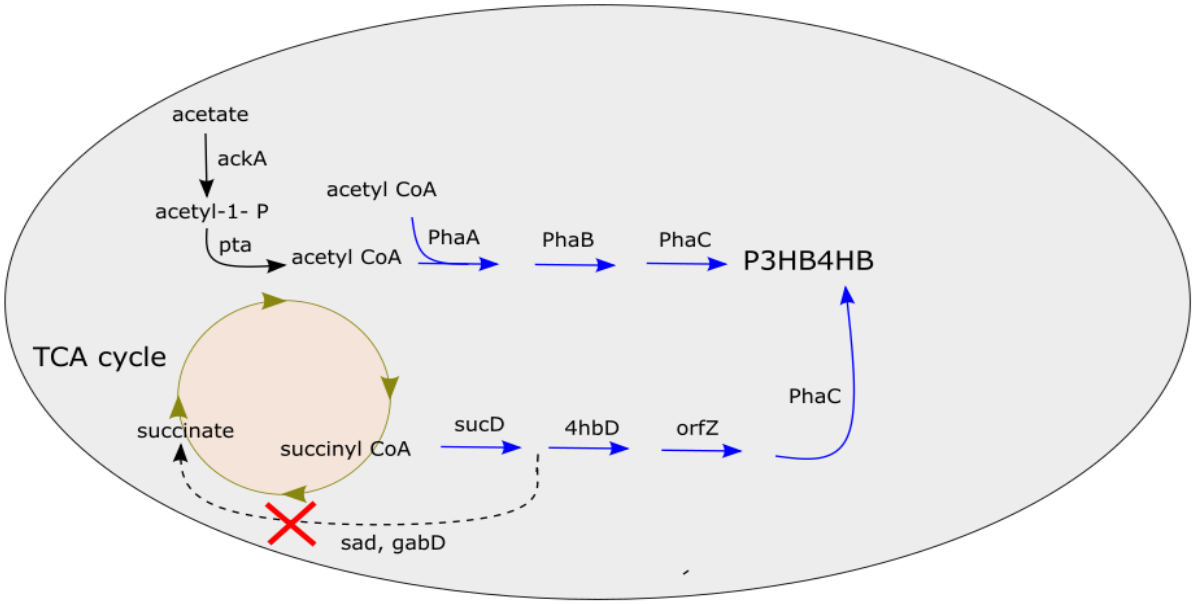

Figure 3: Metabolic engineering of *E. coli* as performed by Chen et al., 2018. Blue coloured lines indicate the steps converted by foreign genes introduced, where genes *sucD*, *4hbD*, *orfZ* originated from *Clostridium kluyveri* and *phaA*, *phaB* and *phaC* are derived from *Ralstonia eutropha*. A red cross mark symbol represents the step deleted as seen for *E. coli* native genes *sad* and *gabD*. Some intermediates are not shown and are denoted with multiple arrows representing multistep reactions. Genes responsible for the expression of respective enzymes are shown above arrows.

Starch based polymers

Another alternative in the biomaterials sector could be starch. Starch is a natural polymer and is thus used as a biodegradable material. Starch is a polysaccharide produced mainly by plants with an aim for energy reservoir system within. Starch granules, which are intracellularly localized, typically exhibit a variety of shapes, with spherical granules being the most common. These granules, ranging in size from 2 to 100 μm in diameter, are primarily derived from edible crops such as corn, rice, and wheat, as well as from tubers like potato and cassava (tapioca) (Chakraborty et al., 2020). Among these, corn, rice, wheat, potato, and cassava starches are the most commercially used due to their widespread availability and functional properties. There are several starch-based products commercialized, such as films, capsules, sheets, foam, etc. available in the market. A review study by Jiang and the co-authors illustrated challenges and potential opportunities for using starch-based materials to meet downstream. One of the key challenges described was moisture sensitivity, which arises due to the inherently hydrophilic nature of starch (Jiang et al., 2020).

A significant challenge in the starch industry is the extensive modification required for plant-based starch to meet industrial needs. Various physical and chemical treatments are employed for starch modification (Kaur et al., 2022), with the most common chemical modifications including esterification, etherification, or oxidation of hydroxyl groups. However, the application of genetic engineering techniques for starch production in microorganisms remains limited. Additionally, since starch is an insoluble polysaccharide at room temperature, it poses considerable challenges for enzymatic treatment and limits access to its functional properties. As a result, native starch is often unsuitable for direct industrial use. This limitation can be addressed by modifying starch to create starch derivatives, which can be tailored to meet specific industrial processing requirements. Microorganism has immense importance due to the versatility of enzymes obtained from them. One such microbial enzyme is a thermostable pullulanase from *Bacillus naganoensis*, having the ability to hydrolyze amylopectin to amylose in starch (Chang et al., 2016). This highlights a promising alternative to traditional physical and chemical methods, leveraging large-scale production of recombinant enzymes through genetically modified microorganisms for starch processing and modification. Such approaches offer innovative solutions to overcome the challenges associated with conventional starch modification techniques.

One example in the published literature is utilizing the bacterium *Klebsiella* enzyme cyclodextrin glycosyl transferase to be expressed in potato tubers to generate cyclodextrins (Oakes et al., 1991). Starch or starch derivatives as a source acted upon by cyclodextrin glycosyl transferase produce cyclodextrin. This starch-derived cyclodextrin is described for its advantageous application in food, pharmaceuticals, cosmetics, chemicals, agriculture, etc. (Del Valle, 2004; Szente & Szejtli 2004). Hence, enzymes related to starch modification or starch derivatives production hold significant promise in biotechnology for nutritional applications.

Polylactic acid synthesis

Another polymer shown to be produced microbially and has a high industrial application is named - Polylactic acid, mainly used to form biodegradable plastic, hence, addressing the environmental concerns. Review literature from Shah and coworkers reports the different biopolymers that can be used for bioplastic synthesis, such as starch, cellulose, PHAs, and polylactic acid (Shah et al., 2021). Among a range of biodegradable plastics, one of the most commercially used and successful bioplastics is polylactic acid (PLA), which has good processability and mechanical properties.

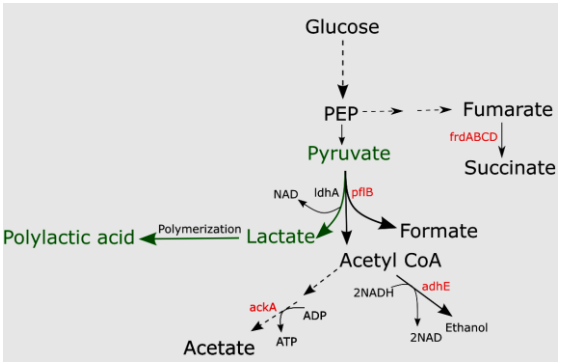

Figure 4: Metabolic pathway for production of polylactic acid in *E. coli*. Monomer lactic acid is produced via pyruvate with the enzyme lactate dehydrogenase (*LdhA*). Competing branches encoded by genes such as *frdABCD*, *ackA*, *adhE*, *pflB* are deleted to accumulate more pyruvate, a precursor of polylactic acid. Letters in red (*frdABCD*, *ackA*, *adhE*, *pflB*) indicates deleted genes. The solid arrows represent a one-step reaction, and the dotted arrows with multiple arrows indicate a multistep reaction. This figure is modified from Zhou et al., 2003.

Apart from the ability of some microbes and plants to produce PLA naturally (Paswan et al., 2023; Guo et al., 2011; Marzo-Gago et al., 2023; Lahtinen et al., 2011), some genetic engineering insights show promising approach for increasing the productivity. Polylactic acid is made up of its monomer lactate, which is produced from pyruvate by the action of lactate dehydrogenase (LDH) using NADH as a cofactor (Figure 4). Different competing branch to over-accumulate pyruvate was experimented on to increase the lactate yield (as shown in Figure 4). PflB directs some pyruvate towards formate as a byproduct. And, *frd* genes product catalyzes the last step in formation of succinate from PEP as a main source. Similarly, AdhE converts the pyruvate derived acetyl CoA to ethanol. The formation of acetate from acetyl-CoA is carried via *ackA*. All of these genes catalyze a reaction that competes with the accumulation of pyruvate, as this is the main precursor of lactate. Via knocking out of these genes (*pflB, frdABCD, adhE* and *ackA*) together with the replacement of native *E. coli* LDH (lactate dehydrogenase) with the heterologous gene L-LDH

from *Pediococcus acidilactici* (ldhL), authors were able to elevate the titer of D-lactic acid by ~ 2.5 times with the engineered *E. coli* strain named SZ63, and the product achieved 99% optical purity (Zhou et al., 2003). Similarly, in another prokaryotic engineering endeavor, the metabolic engineering of *Corynebacterium glutamicum* was undertaken, where the organism was engineered to utilize cellobiose and xylose through the expression of cellobiose and xylose utilizing genes. This engineered strain demonstrated the ability to consume cellobiose and xylose, resulting in the production of approximately 450 mM lactic acid with the consumption 29.3 mM cellobiose, 134 mM xylose, and 222 mM glucose (Sasaki et al., 2008).

In *Saccharomyces cerevisiae*, heterologous expression of the *ldh* gene from *Leuconostoc mesenteroides* resulted in an improved D-lactic acid titer of 2.3 g/l. To enhance the titer and minimize side product formation, efforts were made to increase the availability of pyruvate, a precursor for lactic acid, while reducing carbon loss. This was achieved by deleting genes *GPD1*, *GPD2*, and *ADH1*, which are responsible for glycerol and ethanol production. The engineered strain with these deletions exhibited improved lactic acid production, showing a 6.3-fold increase in titer. Notably, Adh1 in yeast catalyzes the conversion of acetaldehyde to ethanol, while *GPD* genes are involved in converting the glycolytic intermediate dihydroxyacetone phosphate (DHAP) to glycerol (Sornlek et al., 2022).

In another study, metabolic engineering was conducted in cyanobacteria by heterologous expression of *ldh* from *Bacillus subtilis* and co-expressing a transhydrogenase, resulting in an improved yield of lactic acid production. Expression of *ldh* derived from *Bacillus subtilis* in the cyanobacterium *Synechocystis* led to the accumulation of approximately 0.7 M lactic acid. Furthermore, the additional expression of transhydrogenase showed a five-fold increase in titer (Angermayr et al., 2012). In addition to the rational approaches mentioned above for increasing PLA (lactic acid) yield, some studies also explore non-rational approaches such as evolutionary engineering for strain improvement, aiming to achieve high-purity yields. Tian and his group report attaining highly pure lactic acid (99.1%) in a temperature-resistant strain capable of withstanding up to 45 °C. It was achieved through an adaptive laboratory evolution approach applied to an engineered strain of *Lactobacillus paracasei* (Tian et al., 2021). These improved strains can offer significant advantages for the polymer industries.

Taken together, these examples illustrate the potential of both rational and non-rational strain engineering methods to achieve high production of polylactic acid or its monomers. This represents a sustainable approach to biopolymer production using biotechnology and reveals the future of biotechnology in the production of biodegradable plastic as demanded by our modern society.

The biodegradable plastic market is projected to grow USD 20.9 billion by 2028, as noted by Biodegradable Plastics Report (2023); it reflects a substantial expansion of degradable plastic industries driven by the escalating demand for environmentally friendly materials. Similarly, in the market research survey 2022, the Fact.MR (Fact.MR, 2023) reports a significant increase in the value of biodegradable package materials, expected to rise from 88 billion dollars in 2022 to 169 billion dollars over the next decade. Key players from various countries, including Germany, the US, and Japan, are leading the innovation and adoption of biodegradable materials, fostering economic growth and job opportunities. Notably, industry giants like Coca-Cola and PepsiCo have committed to adopting 100% biodegradable and recyclable packaging materials by the end of 2025, further propelling the market forward. Metabolic engineering stands as a crucial driver in this bioplastic revolution, optimizing microbial pathways to efficiently produce biopolymer precursors through genetic modifications. The diverse array of bioplastic monomers, such as polylactic acid (PLA), polyhydroxyalkanoates (PHAs), and Polyethylene terephthalate (PET), highlights the versatility and potential of bioplastics in addressing environmental concerns and advancing sustainable development on a global scale.

Conclusion

To conclude, this review elucidates the significant potential of microbes in addressing environmental concerns through the production of biodegradable polymers. Examining various microbial species and their reported biopolymers, alongside genetic manipulation techniques such as heterologous gene

expression, gene knockout, and overexpression, reveals promising avenues for enhancing product yields. By showcasing the applicability of genetic engineering in elevating high-value product production, this review highlights its pivotal role in industry and environmental sustainability. Looking forward, optimizing microbial strains and refining fermentation technologies can offer promising directions for advancing biopolymer production.

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