

Review

Succinic Acid Production from Oil Palm Biomass: A Prospective Plastic Pollution Solution

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Abstract: Plastic pollution has placed a significant emphasis on the need for synthesising bioplastics, such as polybutylene succinate (PBS), which is derived from succinic acid. Furthermore, environmental concerns and the depletion of non-renewable fossil fuels have initiated an interest in exploring the biotechnological route of succinic acid production via fermentation. Consequently, oil palm biomass might be a prospective substitute for the costlier pure carbon source, which is more sustainable and cost-effective due to its abundance and high lignocellulosic content. The current review focuses on the potential of oil palm biomass utilisation to synthesise succinic acid and its associated bioplastics. The pretreatment and hydrolysis of various oil palm biomass and studies on bioplastics generation from oil palm biomass are also discussed. This review also identified the challenges of manufacturing succinic acid from oil palm biomass and included several recommendations.

Keywords: plastic pollution; oil palm biomass; succinic acid; polybutylene succinate



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1. Introduction

Plastic production from non-renewable fossil fuels has led to devastating environmental impacts as it releases greenhouse gases (GHG), which result in global warming [1]. Moreover, the number of plastic refuse has reached over 8.3 billion metric tons since its initial production [2]. The build-up from its employment in online commerce and personal protective equipment during the COVID-19 pandemic critically aggravated the situation [3]. Furthermore, the annual plastic production could increase to 1606 metric tons by 2050 [4]. The manufacture of plastic is predicted to emit 6.5 gigatons of carbon dioxide (CO₂) gas equivalent of GHG in the same year [4].

Although plastic is undoubtedly useful in everyday life, its decomposition is estimated to take at least 450 years [5]. Recycling strategies are only considered a stopgap measure as they do not aid in diminishing plastic accumulation. Only 15% of accumulated plastic is recycled globally, while the rest amass in various forms, such as microplastics in landfills and oceans, harming marine species before ending up in food chains [6].

Leslie et al. [7] revealed that microplastics were detected in human bloodstreams at 1.6 µg/mL mean concentration. The level is highly damaging as the substance could be transported to organs, resulting in severe health complications. Water, air, food, and personal care products are potential exposure routes of microplastics into the bloodstream [7].

Although the public's awareness has improved regarding the issue, approximately 50–75 trillion plastic pieces are still floating in oceans. Moreover, plastic flow into oceans is projected to triple by 2040 [8]. Currently, several legislations are imposing a ban on single-use plastics and some microplastic ingredients in specific products [9], but legislation alone is insufficient to overcome plastic pollution, necessitating more interventions. One of the prospective interventions is the production of bioplastics, which may serve as an environmentally benign substitute for commercial plastic due to its biodegradable properties, which may help to reduce the environmental impacts of plastic pollution [10].

Due to this reason, a considerable amount of attention is paid to bioplastics production. One of the types of bioplastics is polybutylene succinate (PBS), which is derived from succinic acid and 1,4-butanediol through polycondensation. Succinic acid has been identified as one of the top 10 most essential C4-building blocks that can be generated from by-products and biomass sources as well as converted into high-value commodities and specialty chemicals [11]. The present commercial manufacturing process of succinic acid is heavily driven by the consumption of fossil fuels through the catalytic hydrogenation of maleic anhydride [12]. Due to the depletion of fossil fuels and environmental deterioration, a more sustainable approach, such as fermentation-based succinic acid, should be explored. However, the biotechnological route of succinic acid production is plagued by the high cost of raw materials.

The oil palm industry has flourished in Malaysia as one of its main commodities, with a large amount of biomass generated. The biomass has lignocellulosic components including carbohydrates that can be valorised as the carbon source for the microorganism during the fermentation of succinic acid [13]. The approach of utilizing unattended biomass will assist in reducing the cost of succinic acid production to accommodate its mass production. This review highlights the potential of succinic acid from biomass in its application as the precursor for polybutylene succinate (PBS). It critically presents the recently developed pretreatment and hydrolysis technologies of the biomass, while paying attention to the technical aspects of the processes including the parameters involved. The research gaps and challenges in succinic acid and bioplastics production from palm biomass are identified and addressed with several recommendations, which serve great potential to contribute to the improvement in this area of research.

2. Bioplastics Production

The global search for a sustainable replacement for plastics led to bioplastic synthesis. Recently, biodegradable polymers are being widely applied in biomedical and packaging settings due to their biocompatibility and environmentally-friendly attributes [14]. Synthesising bioplastics as a more sustainable alternative to plastics would aid in diminishing the impacts of plastic pollution on the environment from the reduced fossil fuel resource usage. Furthermore, Spierling et al. [15] reported that the annual emission of CO₂ gas equivalent to GHG could be reduced by 241–316 metric tons if 65.8% of the global conventional plastic is substituted with bioplastics.

Bioplastics are materials derived from biological, biodegradable, or both sources [16]. Although bioplastics are not the solution to the plastic pollution issue, they are far less toxic than plastics as they do not contain bisphenol A (BPA), a hormone disrupter commonly found in conventional plastics [17]. Furthermore, the worldwide bioplastic market is estimated to reach a value of \$25.93 billion by 2029 due to its unique characteristics, namely biodegradability, and biocompatibility [18].

Examples of commercially available bioplastics include polybutylene succinate (PBS), polylactic acid (PLA), polyhydroxyalkanoates (PHA), and polyhydroxybutyrate (PHB). Bioplastics, such as PLA, are on par with conventional plastics, including polyethylene (PE), polypropylene (PP), and polystyrene (PS) due to their similar mechanical properties [19]. Nonetheless, PBS demonstrates the potential as a solution to solve plastic pollution.

The higher flexibility of PBS could cater to diverse applications considering its improved eco-efficiency to end-of-life (EOL) routes [20]. Furthermore, the attributes and

biodegradation rate of PBS are adjustable through copolymerisation with different comonomer unit types and contents [21]. Nevertheless, global bioplastic commercialisation is facing setbacks where its production is two to three times more expensive than conventional plastics [22].

3. Polybutylene Succinate (PBS)

PBS is a biodegradable polyester considering that succinic acid could be manufactured from renewable resources, a biogenic monomer, and can be biologically degraded. Moreover, polysuccinate hydrolysis rates are higher than polyesters derived from higher aliphatic dicarboxylic acids [21]. The melting (~ 115 °C) and the heat distortion (~ 97 °C) temperatures of PBS are also the highest of all polysuccinate derivatives [23].

The excellent mechanical properties of PBS enable its applicability in numerous end applications via conventional melt processing techniques, such as injection, extrusion, and blowing processes [21]. The polymer contains an ester group that degrades into low molecular weight polymers when exposed to water. The substance is a white crystalline thermoplastic with 1.25 g/cm^3 density, a melting point (T_m) within 90 – 120 °C, and a low glass transition temperature (T_g), from -45 to -10 °C. Furthermore, the rate of PBS depletion rises with rising temperature [24].

The melt processability and mechanical properties of PBS are excellent. Furthermore, due to its high crystallinity degree, the polymer exhibits a slow biodegradation rate in polymers or copolymers. The physico-chemical properties of PBS, mainly thermo-mechanical and barrier, could also be enhanced through copolymerisation with other monomers or by incorporating nanofillers into bio polyesters [25]. Moreover, PBS has a broad workable temperature range, 160 – 200 °C, similar to polyolefins [24].

The superior flexibility of PBS allows its utilisation in numerous applications involving film production. Recently, potential applications of PBS have been investigated, such as developing novel materials for ecological agricultural purposes. For instance, mulched non-woven materials and pots were reported as possible alternatives to polypropylene [24]. The findings enabled the exploitation of the polymer in several other sectors, including textile filaments, packaging, foaming, drug encapsulation and carriage systems, orthopaedic, bio-resorbable implants, wound dressings, coffee capsules, and the industrial fields [21].

The PBS is an aliphatic polyester that could be synthesised in various ways, including polycondensation of succinic acid and 1,4-butanediol, where both monomers are derivable from petroleum oil-based sources or bacterial fermentation. As illustrated in Figure 1, the most common method of processing petrochemical succinic acid is the catalytic hydrogenation of maleic anhydride to succinic anhydride, followed by hydration to succinic acid [26]. Conversely, the bacterial fermentation technology can be adopted to generate succinic acid from sustainable feedstock, such as biomass and waste materials from the agricultural sector [27]. The bacterial strains that have been evaluated and commercialised for the purpose include *Actinobacillus succinogenes* [28], *Anaerobiospirillum succiniciproducens* [29], *Mannheimia succiniciproducens* [30], and recombinant *Escherichia coli* [31], *Corynebacterium glutamicum* is another genetically-modified bacterial strain that demonstrated enhanced efficiency in succinic acid production [32].

Generally, PBS is produced by reacting succinic acid and 1,4-butanediol (BDO) via polycondensation. The synthesis of PBS includes two steps. First, the esterification of succinic acid and BDO to obtain PBS oligomers [33]. The second step is polycondensation of the oligomers to remove BDO to form high-molecular-weight (M_w) PBS. Typically, PBS is manufactured in a reactor furnished with a mechanical stirrer, an inert gas inlet (typically nitrogen to avoid oxidation during esterification), and a distillation column. Initially, the reactor is heated to 160 – 190 °C to start esterification while stirring under a controlled atmosphere [33]. Subsequently, polycondensation is further conducted at high temperatures (220 – 240 °C) under vacuum conditions when no more water (or alcohol) is distilled under normal pressure [34].

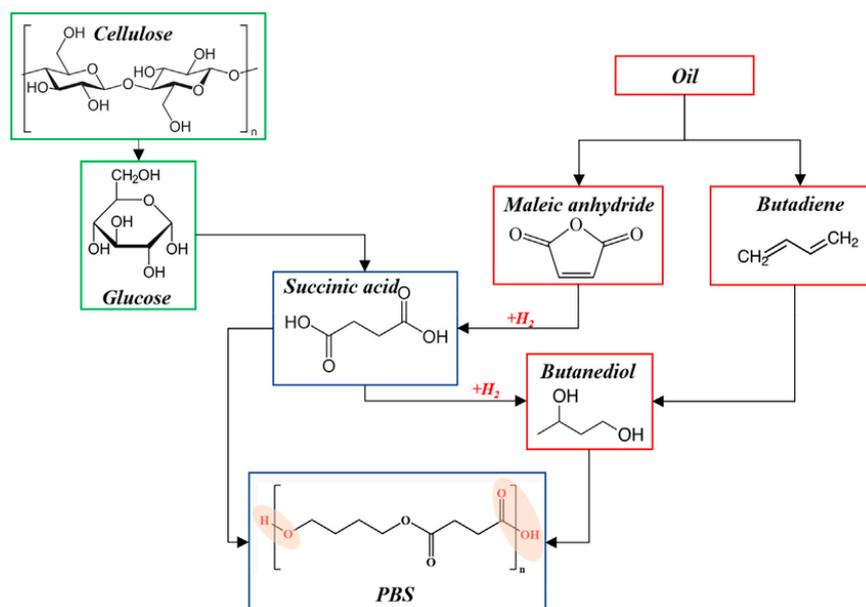


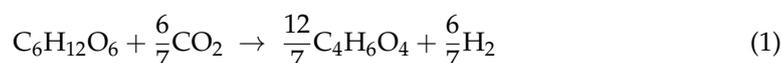
Figure 1. Flowchart of PBS synthesis via bio- or fossil-based sources. Reprinted with permission from [26]. Copyright 2022 Elsevier.

4. Succinic Acid and Its Associated Bioplastics

In 2021, the global succinic acid production market was valued at \$158.96 million and was predicted to reach \$315.89 million by 2030 given its broad application [35]. The acid also possesses a total addressable market of over \$7.2 billion [36]. Commercially available PBS is derived from chemically synthesised succinic acid [21]. Aside from PBS, the other succinic acid-based bioplastics includes poly(butylene succinate-co-butylene adipate) (PBSA), poly(butylene succinate-co-L-lactate) (PBSL), poly(ethylene succinate) (PESu), poly(butylene succinate-co-butylene-fumarate)s (PBSFs), poly(butylene succinate-co-ethylene succinate)s (PBESs), poly(butylene succinate-co-diethylene succinate)s (PBDEGs) and poly(butylene succinate-co-butylene-terephthalate)s (PBST) [10]. Succinic acid is employed in various applications as an essential C-4 building block or precursor for numerous critical speciality chemicals, including tetrahydrofuran, adipic acid, N-methyl pyrrolidinone, 2-pyrrolidinone, gamma-butyrolactone, succinamide, putrescine, succinonitrile, and succinate salts [37]. The acid is also a precursor for 1,4-butanediol production through hydrogenation and reduction [38].

Fossil fuels are the conventional raw material in the chemical synthesis of succinic acid on industrial scales. The fossil fuels are converted to succinic acid via the catalytic hydrogenation of maleic anhydride [12]. Nevertheless, the process is substantially expensive due to its high temperature and pressure requirements [39]. The process could harm the environment due to fossil fuel depletion, GHG emissions, and the toxic catalysts employed [40].

The awareness of avoiding further deterioration of the climate anticipated from current succinic acid production practise has led to the progressive transition towards green processes, thus leading to research associated with bio-succinic acid production through fermentation or biorefinery. Bio-succinic acid has been reported to contribute towards CO₂ emissions abatement, where 50% of GHG and 30–40% of energy consumption could be reduced, compared to conventional process [41]. Furthermore, CO₂ gas is utilised during the succinic acid fermentation process. One kg of succinic acid will utilize 0.37 kg of CO₂, as shown in the following stoichiometric reaction [39].



Large companies, including Bioamber, Reverdia (a joint venture between Royal DSM and Roquette), and Myriant technologies have invested actively in the bio-succinic acid production business on large scales considering that it is greener and possesses potential. Commodity chemicals, such as BDO, dominate the bio-based succinic acid (its precursor) market with over 45% global market share. Consequently, companies, including BioAmber and Mitsui & Co. (Sarnia, ON, Canada) have jointly planned to penetrate other BDO productions following bio-based succinic acid manufacture [42].

The PBS biosynthesis could be sustainable considering the amount of succinic acid generated and its derivable BDO. The significant attention on bio-succinic acid contributed to its high projected market size of USD 272.4 million by 2030 [43]. Nevertheless, mass bio-succinic acid manufacture is plagued by low yields and high costs [44]. The cost of manufacturing bio-succinic acid is USD 1.66–2.2/kg, while succinic acid synthesised from conventional petrochemical processes only requires USD 1.05–1.29/kg [40]. The costly production could result from its cultivation requirement during fermentation and downstream processing [44]. The downstream processing accounts for 60% of the total processing cost of bio-succinic acid synthesis, which is the direct result of numerous purification and recovery processes performed to achieve high succinic acid purity (>98%) for successful PBS production [45].

5. The Palm Oil Industry

The oil palm species primarily cultivated on an industrial scale is *Elaeis guineensis* Jacq., which originated in west Africa [46]. The palm oil industry has not only boosted the economy but provided job opportunities to local smallholders and farmers. Furthermore, the demand for palm oil is projected to grow at 1.7% annually until 2050 [47]. Palm oil is preferred among vegetable oils as it generates the highest oil yield per hectare [48]. Consequently, significantly smaller land areas are utilised to cultivate oil palm plantations than other oil-bearing plants. In 2021, the global oil palm plantation area was recorded at 21 million hectares [49]. The palm oil industry has become the primary commodity for countries such as Indonesia and Malaysia. Currently, palm oil production is 75 million metric tons worldwide, where Indonesia contributes 50% and Malaysia 30% [46]. In 2022, the total area of oil palm plantations in Malaysia is 6 million hectares, and for each hectare, the amount of oil palm fresh fruit bunches (FFB) produced from fully matured oil palm trees is 17.10 tonnes [50,51].

5.1. Palm Oil Production By-Products

The first FFB from each oil palm tree for crude palm oil (CPO) processing could only be harvested after 3 years of cultivation, while the maximum FFB yield from oil palm trees is estimated up to 12 to 15 years post-plantation [52]. The FFB are mechanically pressed after being sterilised via boiling at a high temperature to recover CPO. By-products, including empty fruit bunches (EFB), palm press fibre (PPF), palm oil mill sludge (POMS), palm oil mill effluent (POME), mesocarp fibre, palm kernel shell (PKS), and palm kernel cake (PKC), are generated from CPO extraction and decanting in palm oil mills. The harvesting of FFB also leaves pruned oil palm fronds (OPF) as mulch at plantation sites [53]. The anatomy of the oil palm tree is shown in Figure 2.

After approximately 20 years of cultivation, oil palm trees demonstrate lower productivity, while their heights would result in difficulty harvesting the FFB [54]. Consequently, approximately 10% of the total oil palm plantation area requires annual replantation, which generates oil palm biomass [55]. Loh [56] reported that the amount of oil palm biomass by-products generated are in tonnes per hectare. The total amount of oil palm biomass generated in Malaysia and globally was calculated based on the total area of oil palm plantation [46,48] and presented in Table 1. Unattended biomass at plantations would incur the risk of plant diseases due to the growth of the biodeterioration organisms [57]. Furthermore, improper management of unutilised waste contributes to GHG, which leads to environmental deterioration [58].

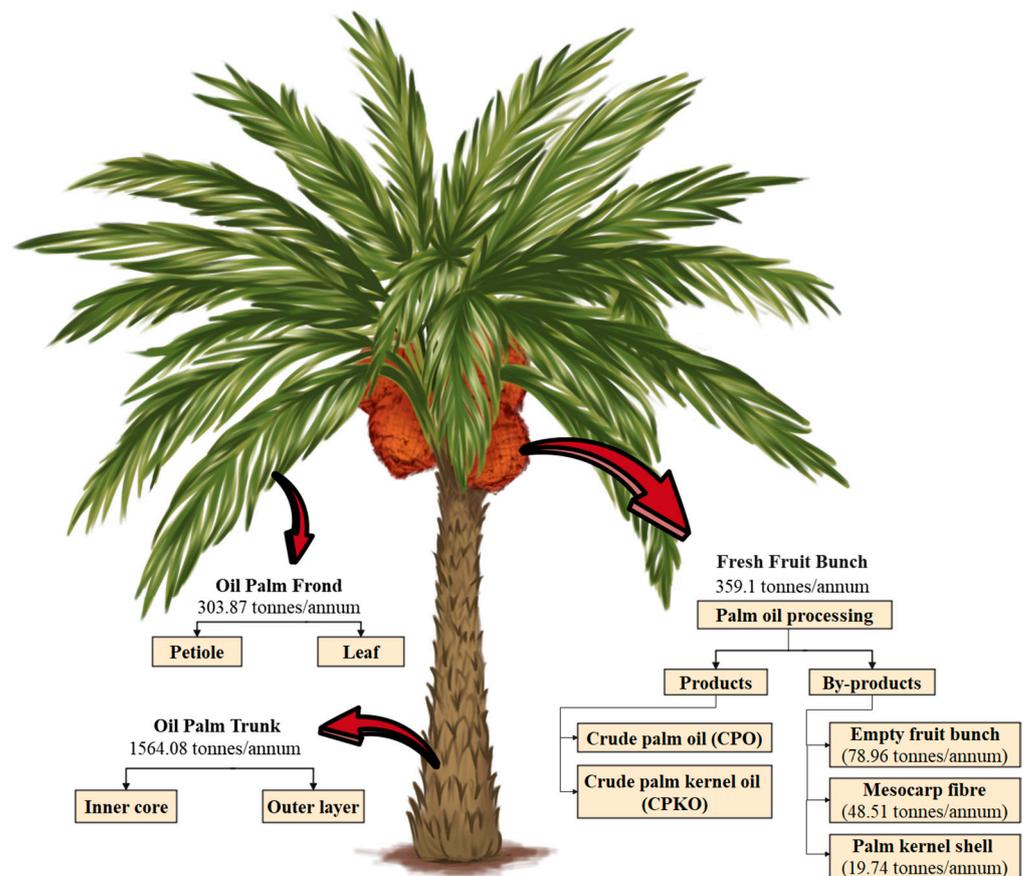


Figure 2. The anatomy of oil palm tree with the global amount of biomass generated.

Table 1. Amount of oil palm biomass generated.

Type of Biomass	Amount Generated (Tonnes/Hectare)	Amount Generated in Malaysia (Tonnes/Annun)	The Total Amount Generated Globally (Tonnes/Annun)
Oil Palm Frond (OPF)	14.47	86.82	303.87
Oil Palm Trunks (OPT)	74.48	446.88	1564.08
Empty Fruit Bunches (EFB)	3.76	22.56	78.96
Palm Kernel Shells (PKS)	0.94	5.64	19.74
Oil Palm Mesocarp Fibre	2.31	13.86	48.51
Total	95.96	575.76	2015.16

5.2. Oil Palm Biomass Compositional Attributes

The oil palm biomass is a lignocellulosic and primarily comprised of cellulose (30–50%), hemicellulose (20–35%), and lignin (10–15%) [40]. Generally, the lignocellulosic structural matrix is sturdy against natural or induced degradation and decomposition, thus requiring a pretreatment process to break down its components. Polymeric cellulose also necessitates hydrolysis into simple monosaccharides, such as glucose, before attempting to transform it into value-added products [59].

Typically, hydrolysate contains simple sugars that would be utilised during fermentation. Nevertheless, the sugar compositions of each oil palm biomass vary due to species, types, storage time, and seasonal differences [46,60,61]. Other nutrients, including proteins, lipids, and other organic and inorganic compounds are also found in oil palm biomass [62]. Moreover, the sap generated from oil palm trunk (OPT) contains amino acids, vitamins, and minerals, allowing it to become a potential feedstock for bacterial growth and cultivation [63].

5.3. Pretreatment and Hydrolysis of Fermentable Sugars from Oil Palm Biomass

Oil palm biomasses are typically subjected to size reduction, pretreatment, and hydrolysis as preparation as the carbon source in fermentation. Larger biomasses are cut into smaller chunks to ease the pressing process with a mechanical pressing machine to attain bagasse and sap (if any). During pretreatment, the heterogeneous structure of lignocellulose, which consists of lignin, cellulose, and hemicellulose, is disrupted [64]. Pretreatment is commonly followed by hydrolysis, which converts the cellulose and hemicellulose into fermentable sugars [46].

Lignin can be eliminated after loosening intact lignin-holocellulose matrixes through pretreatment [65]. Lignin is a complex non-carbohydrate phenolic polymer that is a potential inhibitor of the fermentation process [66]. The presence of lignin and other organic acids at high concentrations would reduce water content, resulting in environmental stress on the microorganisms involved in fermentation [67].

In studies employing OPT, the hard outer layer of the trunks is typically removed for plywood processing. The inner cores that are high in moisture are then pressed with a laboratory pressing machine at 80 MPa [61,63,68]. Conversely, only the petiole section of the fronds is extracted for pressing to obtain bagasse and sap from OPF [69–71]. Table 2 depicts the pretreatment and hydrolysis process conducted for the oil palm biomasses. It can be observed that a high amount of total fermentable sugar can be valorised in the liquid fraction of the hydrolysate generated.

Table 2. The pretreatment and hydrolysis process conducted to the oil palm biomasses.

Pretreatment Condition	Hydrolysis Condition	Findings	References	
OPF Bagasse	Alkaline Chemical: 3 M KOH Temperature: 40 °C Agitation speed: 400 rpm Time: 4 h Solid to liquid ratio: 1:10 (<i>w/v</i>)	Enzymatic Hydrolysis Enzyme: Xylanase (2 U/mL) Hemicellulase, HTec2 concentration: 2% (<i>w/v</i>) Temperature: 40 °C pH: 4.6, Time: 48 h Buffer: 0.05 M citrate buffer	(1) Carbohydrates component (% <i>w/w</i>) through Phenol-sulfuric acid method: <ul style="list-style-type: none"> • Xylose: 46.67, Arabinose: 11.01, Glucose: 9.01 (2) 23.8 g/L of Xylose was obtained in the liquid fraction. (3) Total Xylobiose and Xylotriose: 21.91 g/L (at 8 h of hydrolysis) After ultrafiltration, Xylobiose: 56.64% (<i>w/w</i>), Xylotriose: 45.80% (<i>w/w</i>)	[72]
OPF Bagasse	Alkaline Chemical: 4.42% (<i>w/v</i>) NaOH Temperature: 100 °C Pressure: 1 atm Time: 58 min Solid to liquid ratio: 1:10 (<i>w/v</i>)	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (15 FPU/g cellulose) and Hemicellulase, HTec2 (25 U/g) Temperature: 50 °C pH: 4.8, Time: 72 h Agitation speed: 150 rpm	(1) Solid fraction: 28.0% lignin removal (2) Liquid fraction: 50.3% fermentable sugars yield (3) 90.0% glucan conversion (4) Structural carbohydrates: 78.3% (<i>w/w</i>)	[73]
OPF Bagasse	Alkaline Chemical: 1.2 M NaOH Temperature: 100 °C Time: 58 min Autohydrolysis: 121 °C for 20 min	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (30 FPU/g cellulose) and Hemicellulase, HTec2 (50 U/g) 6% glucan loading	(1) Fermentable sugar (g/L): <ul style="list-style-type: none"> • Glucose: 54.1, Xylose: 13.4 	[74]
OPF Bagasse	Alkaline Chemical: 4% (<i>w/v</i>) NaOH Temperature: 100 °C Time: 58 min Solid to liquid ratio: 1:10 (<i>w/v</i>) Autohydrolysis: 121 °C for 20 min	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (50 FPU/g cellulose) and Hemicellulase, HTec2 (75 FXU/g) Temperature: 50 °C pH: 4.8, Time: 72 h Buffer: 0.05 M citrate buffer	Total fermentable sugar: 72.7 g/L	[28]
OPF Bagasse	Alkaline and water Chemical: 1.2 M NaOH Temperature: 100 °C Time: 58 min Autohydrolysis: 121 °C for 20 min	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (30 FPU/g cellulose) and Hemicellulase, HTec2 (7.5 FXU/g) 6% glucan loading Temperature: 50 °C pH: 4.8, Time: 72 h Agitation speed: 150 rpm	(1) Lignocellulosic components (% dry weight) through NREL: LAPs: <ul style="list-style-type: none"> • Cellulose: 47.3, Hemicellulose: 18.2, Lignin: 23.9 (2) Total fermentable sugar: 43.5% (<i>w/w</i>) <ul style="list-style-type: none"> • Glucose: 44.5 ± 0.9 g/L, Xylose: 13.8 ± 0.6 g/L 	[69]
OPF Bagasse	N/A	Acid Hydrolysis Chemical: 3 mL of 72% of sulphuric acid H ₂ SO ₄ into 0.3 g of dried samples Temperature: 30 °C Agitation speed: 150 rpm Time: 1 h Autohydrolysis: 121 °C for 1 h	(1) Total carbohydrate (% <i>w/w</i>) through NREL: 57.6 ± 5 <ul style="list-style-type: none"> • Glucan: 2.8 ± 3.1, Xylan: 12.5 ± 2.5, Arabinan: 2.3 ± 0.9 • Structural carbohydrate: 14.8 (2) Lignocellulosic components (% dry weight) through NREL: LAPs Lignin: 19.7 (3) Total fermentable sugar: 44.2 g/L <ul style="list-style-type: none"> • Glucose: 24.8 ± 2.5 g/L, Xylose: 15.2 ± 1.3 g/L 	[75]

Table 2. *Cont.*

	Pretreatment Condition	Hydrolysis Condition	Findings	References
OPF Bagasse	Acid Chemical: 4% (v/v) HNO ₃ Temperature: 130 °C Time: 20 min Solid to liquid ratio: 1:8 (mL/g) Autohydrolysis: 121 °C for 20 min	N/A	(1) Structural carbohydrate (% w/w): 61.4 ± 0.6 (2) Lignocellulosic components (% dry weight) through NREL: LAPs • Lignin: 20.5 ± 0.4 (3) Total fermentable sugar (g/L): • Xylose: 22.1, Glucose: 8.9	[70]
OPT Bagasse	Dilute acid Chemical: 5% (w/v) Oxalic acid Temperature: 120 °C Time: 3 h Solid to liquid ratio: 1:10 (w/v)	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (15 FPU/g cellulose) 0.5% (v/v) Triton X-100 Temperature: 50 °C Agitation speed: 150 rpm pH: 5, Time: 72 h	(1) Total fermentable sugars of 39.6 g/L • Glucose: 19.66 ± 3.17, Xylose: 19.91 ± 0.86	[76]
OPT Bagasse	Dilute acid Chemical: 5% (w/v) Oxalic acid Temperature: 120 °C Time: 2 h Solid to liquid ratio: 1:10 (w/v)	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (30 FPU/g cellulose) Temperature: 50 °C Agitation speed: 155 rpm pH: 4.8, Time: 48 h	Total xylose: 61.2% (w/w)	[77]
OPT Bagasse	N/A	Acid Hydrolysis Chemical: 500 mL of 1% (v/v) of sulphuric acid H ₂ SO ₄ into 50 g of OPT fiber Agitation speed: 150 rpm Time: 15 min Microwave heating: 700 W for 10 min	Total fermentable sugars of 30 g/L	[78]
OPT Bagasse	Acid Chemical: 1.0% (v/v) H ₂ SO ₄ Temperature: 120 °C Time: 90 min Solid to liquid ratio: 1:10 (w/v) Autohydrolysis: 121 °C for 20 min	Enzymatic Hydrolysis Enzyme: UKM-enzyme Formulation-3 (49.9 U/g) 0.123% (v/v) Triton X-100 Temperature: 50 °C pH: 5, Time: 48 h Buffer: 0.05 M citrate buffer Agitation speed: 155 rpm	(1) Maximum glucose 21.7% (w/w) (2) Removal of 59.5% hemicellulose and 13.3% lignin.	[79]
OPT Bagasse	Dilute acid Chemical: 5% (w/v) Oxalic acid Temperature: 120 °C Time: 2 h Solid to liquid ratio: 1:10 (w/v) Autohydrolysis: 121 °C for 4 h	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (15 FPU/g cellulose) Temperature: 50 °C Agitation speed: 150 rpm Time: 72 h	(1) Total xylose and glucose of 15.2% and 4.6 (w/w) For pretreated solids: • Cellulose digestibility of 86–87% • 22.1–22.2 g sugar per 100 g OPTB For the whole slurry: • Cellulose digestibility of 71–74%, • 43.2–44.9 g sugar per 100 g OPTB	[80]

Table 2. *Cont.*

	Pretreatment Condition	Hydrolysis Condition	Findings	References
OPT Bagasse	Dilute acid Chemical: 1% (<i>w/v</i>) Oxalic acid Temperature: 120 °C Time: 180 min Solid to liquid ratio: 1:10 (<i>w/v</i>)	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (15 FPU/g cellulose) Triton X-100 at 0.5% (<i>v/v</i>) Temperature: 50 °C Agitation speed: 150 rpm Time: 72 h	(1) Total fermentable sugar: 43.22 ± 3.87 g/L • Glucose: 24.21 ± 3.33 g/L, Xylose: 19.01 ± 0.54 g/L	[81]
EFB	Autoclave alkali (AA) Chemical: (20% <i>w/v</i>) 2.5 M NaOH Temperature: 121 °C Time: 2 h Pressure: 0.12 MPa Sequential dilute acid microwave alkali (DA-MWA) Chemical: 8.0% (<i>v/v</i>) H ₂ SO ₄ Temperature: 121 °C Time: 1 h Pressure: 0.12 MPa Followed by: Chemical: 10.0% (<i>w/v</i>) 2.5 N NaOH Microwave heating: Power of 900 W for 20 min	Enzymatic Hydrolysis Enzyme: Cellulase, CTec2 (25 FPU/g cellulose): Cellobiase (10 CBU/g) with ratio of 7:1 Buffer: 50 mL citrate buffer Temperature: 50 °C Agitation speed: 180 rpm Time: 48 h	(1) Lignocellulosic components (% dry weight) through TAPPI: • Cellulose: 53.3, Hemicellulose: 28.7, Lignin: 11.8 (2) 86.8% (<i>w/w</i>) cellulose was obtained from sequential DA-MWA pretreatment. (3) Maximum glucose accumulation of 31.4 g/L was achieved after hydrolysis using DA-MWA treated slurry.	[82]
EFB	Dilute acid microwave Chemical: H ₂ SO ₄ Temperature: 121 °C Time: 1 h Followed by: Chemical: 10.0% (<i>w/v</i>) 2.5 N NaOH Microwave heating: 900 W for 20 min	Simultaneous saccharification and fermentation Enzyme: Cellulase, CTec2 (40 FPU/g cellulose) Temperature: 36 °C pH: 5, Time: 48 h Agitation speed: 210 rpm	(1) Lignocellulosic components (% dry weight) through TAPPI: • Cellulose: 86.8 ± 1.4 • Hemicellulose: 3.4 ± 1.5 • Lignin: 5.3 ± 0.16 (2) Glucose accumulation of 4.3 g/L during fermentation.	[83]

Based on the studies presented in Table 2, the pretreatment for oil palm biomass employs alkaline [potassium, (KOH) and sodium (NaOH) hydroxides] and dilute acid (nitric, oxalic, and sulphuric acids) due to the simplicity and higher technological readiness level (TRL) of the technique compared to other available methods. The most frequently utilised parameters for the process are a temperature between 100 and 121 °C, a solid-to-liquid ratio of 1:10 (*w/v*), and a time range of 20–180 min.

It can be deduced from Table 2 that the lignocellulosic components extracted from the biomass depend on the pretreatment method. The autoclave alkali (AA) is a pretreatment technique utilising mild NaOH. Although the method requires an extended treatment period, it impedes sugar degradation to furfural while preventing lignin condensation and keeping it stable [82]. Nevertheless, (AA) only removes lignin and extracts hemicellulose rather than cellulose.

Sequential dilute acid and microwave alkali (DA-MWA) is a pretreatment method that recorded 72.9% lignin removal by breaking the seal of the lignocellulosic structure [82]. The microwaves penetrate the lignocellulosic structure and cause its molecules to oscillate. Akhtar & Idris [82] subjected EFB to both (AA) and (DA-MWA) pretreatment. A maximum cellulose increase of 53.3 g/100 g, a maximum hemicellulose decreases of 28.7 g/100 g, and lignin content at 11.8 g/100 g were achieved for (AA). Nevertheless, when (DA-MWA) was conducted, a higher cellulose content, 86.8% dry weight, with lower hemicellulose and lignin contents, 3.4 and 5.3% dry weight, were extracted.

Studies shown in Table 2 primarily explored hydrolysis employing acid (sulphuric acid) and enzymes (xylanase, hemicellulase, cellulase, and cellobiase). The optimum ratio of cellulase to cellobiase as 7:1 for hydrolysis, resulting in a 31.4 g/L glucose accumulation [82]. Most studies reviewed reported hydrolysis at 50 °C, a pH of 4.6–5, agitation speed between 150–210 rpm, and time from 48 to 72 h. Moreover, separate hydrolysis and fermentation processes are mainly conducted in the reviewed studies. Nevertheless, Akhtar et al. [83] reported a unique approach of simultaneous enzymatic hydrolysis of EFB with fermentation in a bioreactor operated at 36 °C, which is lower than the temperature adapted for separation hydrolysis, 50 °C, to accommodate for fermentation.

This study reported an improved cellulose hydrolysis rate post enzyme dosage increment during hydrolysis considering more active sites were available for cellulose molecules to be converted into glucose. The surge in glucose accumulation from 3.2 to 4.3 g/L when the enzyme load was increased from 10 to 40 Filter Paper Units (FPU)/g was observed in the study. Nonetheless, hydrolysis releases toxic by-products, including organic acids, furfural, hydroxymethylfurfural, and phenolic compounds, which inhibit cell growth and cultivation of microorganisms and succinate production [84,85]. The issue could be overcome via detoxification with activated carbon where the compounds are adsorbed onto the activated carbon surfaces.

Luthfi et al. [28] documented a maximum total fermentable sugar content of 72.7 g/L from OPF bagasse hydrolysate by adopting alkaline pretreatment with 4% (*w/v*) NaOH at a solid-to-liquid ratio of 1:10 (*w/v*) at 100 °C for 58 min. Subsequently, enzymatic hydrolysis employing cellulase and hemicellulase at 50 °C for 72 h was performed. The total reducing sugar yield in the study rose from 4.0 to 38.6 g/g when the cellulase load was increased from 0 to 10 FPU/g, while at 30 FPU/g of cellulase, maximum glucose and xylose were generated. Nonetheless, the total reducing sugar yield was only enhanced by 5.1% at 50 FPU/g, which is economically ineffective. The study also documented that an increment of hemicellulose load to 45 FXU/g resulted in a higher total reducing sugar yield of 63.7%.

Compared to the enzymatic hydrolysis of OPF bagasse by Luthfi et al. [28], the acid hydrolysis of OPF bagasse reported by Tan et al. [75] generated lower total fermentable sugar, 44.2 g/L. The study utilised the frond section 1.5 to 3 m away from the trunk, which recorded the highest sugar content compared to other portions of the frond. Before hydrolysis, the study samples were ground and sieved through a 0.5 mm mesh to remove powdery particles that could be over-hydrolysed with sulphuric acid.

The highest sugar content in OPT bagasse was valorised at 43.22 g/L, which consisted chiefly of glucose and xylose at concentrations of 24.21 and 19.01 g/L, respectively [81]. The bagasse, at 10% (w/v) solid loading, was pretreated with 1% (w/v) dilute oxalic acid at 120 °C for 180 min before subjecting it to enzymatic hydrolysis. The pretreatment method has efficiently solubilised hemicellulose to monomeric xylose with an excellent catalytic performance at low concentrations. Resultantly, 56–59% of hemicellulose solubilisation was achieved after 3–4 h of pretreatment.

Besides the pretreatment using acid and enzymatic hydrolysis, microbial hydrolysis of biomass for the valorisation of the fermentable sugars is one of the strategies worth exploring. Although the study on the microbial hydrolysis of oil palm biomass is still scarce, some research has been done to efficiently disintegrate the lignocellulosic substrate and release fermentable sugars from other types of biomasses, such as rice straw, sugarcane bagasse, rice husk, and banana pseudostem [86,87]. Fungal-derived cellulase can be used to hydrolyse cellulose in the biomass. For instance, in the research done by Aggarwal et al. [86], 12.0 U/gds of carboxymethyl cellulase, CMCase was produced by *Aspergillus niger* in 96 h, using 6.0% substrate concentration, 7.5% inoculum concentration, 1:2 solid to liquid ratio, at pH 5.5, and temperature 28 °C by supplementation of the fermentation medium with 0.1% carboxymethylcellulose and 0.1% ammonium nitrate. Prior to hydrolysis process, the rice straw is pretreated with alkali-assisted acid at 10% (w/v) concentration. The cellulase generated in this study has successfully conferred 23.78% sugars and 35.96% saccharification value after hydrolysis process with 2% (v/v) enzyme concentration for 2.5 h at 40 °C. Besides fungal-derived cellulase, the microbial hydrolysis of biomass can also be achieved using *Clostridium thermocellum*. This strain was acclimated and employed in the study done by Nisha et al. [87], to hydrolyse rice husk, sugarcane bagasse, and banana pseudostem in the standard media supplemented with cellobiose. As a result, reducing sugar of 672 mg/g, 636 mg/g, and 513 mg/g was generated from sugarcane bagasse, rice husk, and banana pseudostem, respectively.

5.4. Bioplastic Production from Oil Palm Biomass

Several studies had directly generated bioplastic products with oil palm biomass as the feedstock and various bacterial strains through fermentation. Nonetheless, based on studies presented in Table 3, only the production of bioplastics, such as PHA and PHB was explored, while only EFB has been studied for PHB production.

Table 3. Bioplastics production from oil palm biomass through the fermentation process.

Substrate	Bioplastics	Microorganism	Fermentation Conditions	Substrate Concentration (%)	Product Titer (g/L)	Reference
EFB Hydroly-sate	Polyhydroxy-butyrate (PHB)	<i>Bacillus cereus suaeda</i> B-001	Batch T: 30 °C P: 6.8 Speed: 150 rpm Media:Luria-Bertani (LB) broth	Sugar: 1.44	0.99	[88]
OPT sap	Poly-3-hydroxy-butyrate (PHB)	<i>Bacillus megaterium</i> MC1	Batch T: 30 °C Speed: 200 rpm Media: Mineral Medium (MM)	Sugar: 2.5	3.28	[89]
OPT sap	Polyhydroxy-alkanoate (PHA)	<i>Bacillus megaterium</i> MC1	Batch T: 30 °C Speed: 200 rpm Media: Mineral Medium (MM)	Sugar: 2.5	1.91	[60]
POME	Polyhydroxy-alkanoate (PHA)	<i>Comamonas</i> sp. EB 172	Batch T: 30 °C pH: 7 Speed: 200–800 rpm Time: 70 h DO: 30% of air saturation Aeration rate: 1 vvm	POME: 0.1	2.57	[90]
POME	Polyhydroxy-alkanoate (PHA)	<i>Bacillus licheniformis</i> HAs-007mutant M2-12	Batch T: 37 °C pH: 7 Speed: 150 rpm Time: 48 h Aeration rate: 1 vvm	POME: 3 Sugar: 4	7.35	[91]

Yustinah et al. [88] pretreated EFB via acid hydrolysis at 121 °C and 15 lb of pressure for 60 min in an autoclave. The hydrolysate was added as part of the fermentation media composition in the study as it consisted of 14.3 g/L total sugar, producing 0.99 g/L of PHB. Takamitsu et al. [89] documented the highest amount of PHB by employing oil palm sap and *B.megaterium* MC1. A maximum PHB concentration of 3.28 g/L with 2.5% *w/v* sap sugar concentration was generated during 16 h of fermentation at a carbon to nitrogen ratio (C/N) of 50 under optimum conditions, as shown in Table 3. Other reports also employed biomass from the oil palm industry, liquid by-products from OPT, such as OPT sap pressings, and oil palm processing, including POME.

Sangkharak and Prasertsan [91] documented the highest PHA titer from POME. The sequential mutation of *Bacillus licheniformis* PHAs-007 was conducted in the study to construct its mutants that performed better in PHA production. The mutagenesis process was initiated by placing the plates containing the strains under an ultraviolet lamp at a distance of 55 cm for 15 s to 30 min. The plates were then kept in the dark for 1 h. Only the strains that exhibited halozones were selected for the NTG treatment.

During the NTG treatment, 1 mL NTG solution was added to 1 mL cell suspension of the strain that comprised $5\text{--}8 \times 10^8$ cell/mL. The mutant M2-12 obtained could synthesise novel PHA copolymers, such as 3-hydroxyvalerate and 3-hydroxyhexanoate, which the wild type could not induce. Furthermore, its utilisation in the fermentation process has resulted in the highest PHA yield of 19.55 g/L, 3.18 times higher than the amount produced by the wild type. The value was achieved through cultivation under optimal conditions of 45 °C, pH 7, and 3% POME without additional trace elements.

5.5. Biomass By-Products from the Palm Oil Industry Utilisation for Bioplastic and Succinic Acid Productions

Biorefineries that utilise fermentation involve pure sugars derived from edible starch-based raw materials, which are not favourable as they belong to the food chain and could potentially lead to food resource competition [85]. Moreover, food resource utilisation is not feasible due to its depletion and the current global food crisis. Larger-scale fermentations are also economically ineffective due to costly traditional feedstock [37].

Numerous research has been conducted to establish an inexpensive fermentation process that employs abundant and renewable raw material sources [11]. A large amount of underutilised oil palm biomass with high lignocellulosic contents is a potential carbon source for fermentation [13]. The utilisation of oil palm biomass would result in economically palatable production of bioplastics and other value-added chemicals, such as succinic acid as it is regarded as a by-product with low economic demand. The process flow for the utilization of oil palm biomass for succinic acid and PBS production is shown in Figure 3. Moreover, oil palm biomass is renewable, constantly available, and comprises non-edible segments, and thus does not interfere with the food chain [40].

Amid the climate change issue, sustainability and the environmental impacts of waste generated by agricultural industries, including palm oil, are concerning. Accordingly, valorising oil palm biomass would aid in making the industry more sustainable by reducing waste accumulation and GHG emissions. Furthermore, the waste-to-wealth concept adapted by converting by-products into high-value-added products, including succinic acid, is advantageous. The effort also resonates toward achieving the zero-waste and circular economy aims. The extensive list of studies conducted in utilizing oil palm biomass for succinic acid production is represented in Table 4.

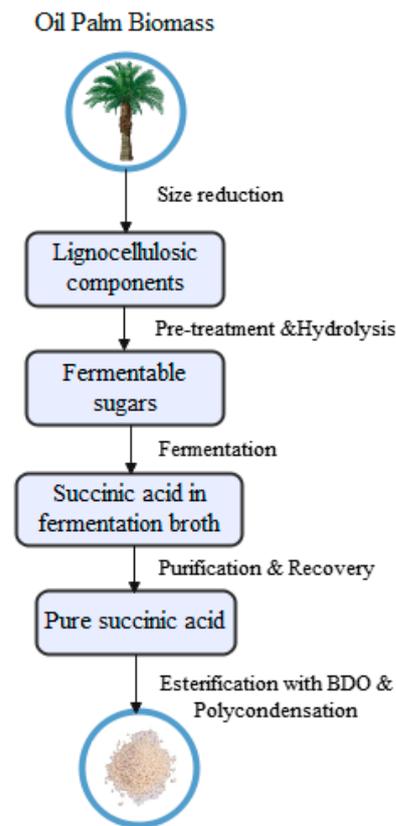


Figure 3. Process flow for the utilization of oil palm biomass for succinic acid and PBS production.

Luthfi et al. [74] recorded the highest succinic acid yield from OPF bagasse hydrolysate at 0.87 g/g. The study repeated the batch fermentation of succinic acid for five runs by adopting bacterial immobilisation with coconut-activated carbon as its carrier. Nonetheless, only 25–40% of the immobilised cells were retained during batches two to five as the non-viable cells were removed during washing with isotonic saline. Although the immobilisation strategy yielded a 23% improvement in succinic acid yield compared to fermentation with cell suspension, the performance was inconsistent due to fine carbon dust production.

Tan et al. [75] documented a 0.73 g/g OPF juice, the highest yield. The by-products formed in the study were maintained at low levels where only 0.7 g/L of formic acid, 0.6 g/L of acetic acid, and 0.4 g/L of ethanol were detected. The study also reported enhanced succinic acid production with carbonate loading from 100 to 400 mmol/L. Less by-products with a maximum concentration of 21 g/L succinic acid was achieved at 400 mmol/L carbonate load, which was four times more than the control. Incorporating CO₂ into phosphoenolpyruvate (PEP) during the anaplerotic reaction led to the aforementioned results.

Table 4. Succinic acid production from various types of oil palm biomass.

Oil Palm Biomass	Pretreatment Process	Mode	Microbes	Sugar Consumed (g/L)	SA Titer (g/L)	Yield (g/g)	Productivity (g/L. h)	Reference
OPF bagasse hydrolysate	Alkaline pretreatment, Enzymatic Hydrolysis	Batch	<i>A. succinogenes</i> 130Z	51.55	36.6	0.71	0.61	[73]
OPF bagasse Hydrolysate	Alkaline pretreatment, Enzymatic Hydrolysis	Repeated batch pH: 6.5 Temperature: 39 °C Agitation speed: 200 rpm Fermentation period: 32 h (batch), 180 h (repeated batch of 5 runs)	<i>A. succinogenes</i> 130Z	50.69	44.1	0.87	1.34	[74]
OPF bagasse Hydrolysate	Alkaline pretreatment, Enzymatic Hydrolysis	Batch pH: 6.8 Temperature: 39 °C Agitation speed: 200 rpm Fermentation period: 50 h	<i>A. succinogenes</i> 130Z	61.29	38.0	0.62	1.95	[28]
OPF juice	Alkaline pretreatment, Enzymatic Hydrolysis	Batch pH: 6.8 Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 50 h	<i>A. succinogenes</i> 130Z	51.36	33.9	0.66	6.58	[69]
OPF juice	-	Batch pH: 6.8 Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 50 h	<i>A. succinogenes</i> DSM 22257	43.24	30.7	0.71	n/a	[71]
OPF juice	Acid Hydrolysis	Batch pH: 6.8 Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 60 h	<i>A. succinogenes</i> 130Z	28.77	21	0.73	n/a	[75]
OPT bagasse Hydrolysate	Dilute acid pretreatment, Enzymatic Hydrolysis	Batch Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 72 h	<i>A. succinogenes</i> 130Z	39.77	17.5	0.44	0.36	[80]
OPT bagasse Hydrolysate	Acid, Enzymatic Hydrolysis	Batch Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 72 h	<i>A. succinogenes</i> 130Z	28.08	7.30	0.26	0.30	[79]

Table 4. Cont.

Oil Palm Biomass	Pretreatment Process	Mode	Microbes	Sugar Consumed (g/L)	SA Titer (g/L)	Yield (g/g)	Productivity (g/L. h)	Reference
OPT bagasse hydrolysate	Dilute acid pretreatment, Enzymatic Hydrolysis	Batch pH:6.8 Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 60 h	<i>A. succinogenes</i> 130Z	33.95	13.92 ± 0.85	0.41 ± 0.03	0.23 ± 0.01	[77]
OPT bagasse hydrolysate	Dilute acid pretreatment, Enzymatic Hydrolysis	Batch Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 72 h	<i>A. succinogenes</i> 130Z	36.82	13.99 ± 0.16	0.38	n/a	[76]
OPT bagasse Hydrolysate	Dilute acid pretreatment, Enzymatic Hydrolysis	Batch Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 72 h	<i>A. succinogenes</i> 130Z	36.38	21.1	0.58	0.37 ± 0.03	[81]
OPT sap	-	Batch Temperature: 37 °C Agitation speed: 200 rpm Fermentation period: 60 h	<i>A. succinogenes</i> 130Z	14.48	7.82	0.54	0.36	[63]
EFB	Dilute acid-microwave-alkaline pretreatment, Enzymatic Hydrolysis	Batch Temperature: 38 °C Agitation speed: 210 rpm Fermentation period: 48 h	<i>A. succinogenes</i> 130Z	71.06	33.4	0.47	1.69	[82]
EFB	Dilute acid-microwave-alkaline pretreatment, simultaneous saccharification and fermentation (SSF)	Batch Temperature: 38–40 °C Agitation speed: 210 rpm Fermentation period: 48 h	<i>A. succinogenes</i> ATCC 55618	70.33	42.9	0.61	n/a	[83]
EFB	Autohydrolysis and cellulase	Batch Temperature: 37 °C Agitation speed: 120 rpm Fermentation period: 38 h	<i>A. succinogenes</i> 130Z ATCC 5618	71.21	23.5	0.33	0.62	[92]

Glucose and xylose are the two main types of sugar that were commonly valorised from the oil palm biomass after undergoing saccharification, pretreatment, and hydrolysis, as previously presented in the studies included in Table 2. The direct link between the sugar valorisation of the oil palm biomass and the succinic acid production has been presented in Table 4, where most studies have reported the successful consumption of glucose and xylose by *A. succinogenes*. This finding is in line with the findings on the capability of *A. succinogenes* in utilising multiple types of carbon sources ranging from glucose, xylose, cellobiose, fructose, sucrose, lactose, maltose, mannose, arabinose, and glycerol [93].

For instance, Bukhari et al. [80] utilized OPT bagasse hydrolysate that comprised 43.22 g/L total sugar where 24.21 g/L is glucose, and 19.01 g/L is xylose. As a result, this study has reported the final succinic acid titer of 21.11 g/L with glucose and xylose consumption of 88.17 g/L and 72.19 g/L, respectively. The potentiality of OPT bagasse hydrolysate as the carbon source for succinic acid synthesis was highlighted by Bukhari et al. [81]. The report documented OPT bagasse hydrolysate utilisation at a reduced yeast extract dosage of only 5 g/L and without mineral supplement to generate a 0.58 g/g yield. The study also observed improved succinic acid yield from fermentation in a 5 L bioreactor compared to the serum bottles, which recorded 0.49 g/g. The findings were attributed to the improved capability of the bioreactor in terms of sparging CO₂ to promote its diffusivity into the cultures.

In another study by Bukhari et al. [63], raw OPT sap without yeast extract and nutrient supplements produced a slightly similar succinic acid yield of 0.54 g/g. The study proved the role of magnesium carbonate (MgCO₃) load in increasing succinic acid titer where a maximum of 18.68 g/L was recorded with 30 g/L MgCO₃. Nonetheless, the study reported that a CO₂ source of over 50 g/L was detrimental to the culture as it increased the osmotic pressure to an inhibitory level. The succinic acid concentration was at a maximum during the 36th hour of fermentation with 88% sugar consumption after which the succinic acid concentration and sugar residual remained constant until the end of fermentation.

Akhtar et al. [83] obtained succinic acid from EFB bagasse hydrolysate through simultaneous saccharification and fermentation. The parameters, including temperature, pH, and cellulase loading, were optimised via response surface methodology (RSM). Notably, all parameters significantly affected succinic acid production. The optimum conditions for simultaneous saccharification and fermentation (SSF) were 36 °C, pH 5, and 39.7 FPU/g cellulase load, which yielded a 42.9 g/L succinic acid and a conversion of 0.61 g/g. Nevertheless, considering that hydrolysis and fermentation were simultaneously conducted, glucose concentration throughout the fermentation period could only be detected at a lower level as the glucose generated during hydrolysis was continuously utilised for succinic acid production.

The utilisation of oil palm biomass has dramatically reduced the pressure on bioplastics and rendered succinic acid bio-based productions more economically practical. Nonetheless, critical characteristics, such as pretreatment procedures, suitable host microorganisms with well-studied biochemical pathways, fermentation strategies, and efficient downstream processing methods are the requisites for successful, cost-effective mass production of bio-based products [85,94].

6. Production Parameters and Strategies for Higher Yield of Fermentation-Based Succinic Acid

6.1. Succinic Acid Fermentation Biocatalysts

The type of microorganism is one of the primary factors influencing succinic acid production. Bacteria, fungi, and yeast could synthesise succinic acid. Although succinic acid fermentation from yeast is achievable at a low pH, the excretion of succinic acid in yeast and fungi is through two borders, mitochondrial and cytoplasm membranes, which is less favourable [95]. Consequently, bacterial strains, such as *A. succinogenes*, *Basfia succiniciproducens*, *A. succiniciproducens*, *M. succiniciproducens*, *C. glutamicum*, and *E. coli*, are more frequently utilised in succinic acid production under anaerobic condition studies [11].

Among the bacterial strains, the Gram-negative bovine rumen-isolated *A. succinogenes* is deemed one of the best succinic acid producers due to its high acid concentration tolerance and high succinic acid yield from a variety of carbon sources generating ability [96]. Moreover, its attribute as a facultative anaerobic has contributed to reducing bio-succinic acid fermentation costs as aeration is not required [85].

The production of succinic acid by *A. succinogenes* is possible through the Krebs cycle. The acid is specifically generated as the end product of the C4-succinate-producing pathway of phosphoenolpyruvate (PEP) during glycolysis. The C4 pathway is energy-consuming as it utilises CO₂ to push the flux in PEP towards oxaloacetate where it would be converted into malic, formic, and succinic acids [97].

Other than single cultures, co-cultures for succinic acid fermentation have also been explored as a strategy to improve succinic acid yield. The strategy could increase yield and product qualities as synergistic utilisation of the metabolic pathways of all involved microorganisms would result in the efficient employment of the substrates [98]. Salma et al. [98] co-cultured *A. succinogenes* with *S. cerevisiae* in a batch mode for succinic acid fermentation from glucose and fructose. The CO₂ generated as *S. cerevisiae* performs cellular respiration through the oxidative phosphorylation of pyruvate could be employed to drive the flux towards succinic acid production by *A. succinogenes*. Furthermore, the glycerol produced by *S. cerevisiae* could be metabolised by *A. succinogenes*. The study also reported a high succinic acid yield of 0.70 mol/mol substrate.

Genetic engineering technology could be employed to modify bacterial strains involved in succinic acid production to improve the yield. Choi et al. [99], genetically modified the *M. succiniciproducens* LK strain by deleting the *pta-ackA* genes encoding phosphotransacetylase and acetate kinase. The modification also disrupted the *ldhA* gene encoding the fermentative lactate dehydrogenase. Resultantly, the *M. succiniciproducens* PALK (DldhA and Dpta-ackA) strain constructed conferred high succinic acid titer, yield, and productivity of 66.14 g/L, 1.34 mol/mol, and 3.39 g/L/h, respectively.

6.2. pH

In succinic acid fermentations, pH is considered an essential parameter. The activity of key enzymes engaged in phosphoenolpyruvate carboxykinase (PEPCK) pathways is higher at low pH, thus contributing to higher succinic acid production [100]. Indirectly, low-pH fermentation would simplify and cut downstream processing costs as no additional acid is required to lower the pH to recover succinic acid [62,101]. Conversely, microorganisms grow better at higher pH values [100].

The optimum pH to maintain cell growth and succinic acid production at a maximum level is necessary. Consequently, a pH regulator to maintain the pH during fermentation is often necessary. The pH regulators play a role in increasing succinic acid production. In bioreactor scales, pH regulators are pumped when the pH sensor detects a pH lower than the set value. Choi et al. [99] reported that an ammonia and magnesium hydroxide solution employed as a pH regulator improved succinic acid concentration to 90.68 g/L.

Sparging CO₂ as a pH buffer during fermentation is another method of maintaining the pH [96]. Besides being a driving force that regulates PEPCK activities in the succinic acid production pathway, the technique also aids in enhancing the gas-liquid mass transfer and reduces by-product formation [71]. Nonetheless, in cases involving external CO₂ sources, MgCO₃ is added as a part of media formulation to maintain the anaerobic behaviour of succinic acid fermentation and suitable pH for cell growth [63].

6.3. Nitrogen Sources and Additional Supplementations

Besides carbon, nitrogen sources also significantly influence succinic acid fermentation costs [39]. A nitrogen source is vital for the production of enzymes, as it supplies the microorganisms with the building blocks of organic molecules, such as proteins. The significance of enzymes, such as PEPCK, in succinic acid synthesis is evident in the reverse tricarboxylic acid pathway. Enzymes push the carbon flux from PEP to oxaloacetate where

succinic acid is generated. Moreover, nitrogen supply is strictly required for auxotrophic bacteria, including *A. succinogenes*, that are incapable of synthesising the amino acids and vitamins necessary for growth and replication [96].

Vitamins essential for the growth of *A. succinogenes*, such as B1, B2, B6, and B12 are reportedly available in yeast extracts [102]. Furthermore, yeast extracts have been prominently utilised as an excellent nitrogen source for microbial growth and succinic acid production [103]. Nevertheless, the cost ineffectiveness of obtaining yeast extract pressures the economic viability of biotechnologically derived succinic acid, considering its already costly production process attributable to the downstream processes [39]. Accordingly, cheaper and more abundant complex nitrogen sources are targeted as alternatives to yeast extract. For instance, Akhtar et al. [83] investigated succinic acid production from EFB. The study incorporated 20 g/L of corn steep liquor as the nitrogen source. Resultantly, 42.9 g/L of succinic acid was obtained at the end of the fermentation process.

6.4. Strategies for Increasing Succinic Acid Yield via Fermentation

More strategies should be developed in succinic acid production through biorefinery to increase its yield and productivity given that a minimum productivity of 2.5 g/L/h is required for bio-based production to be economically competitive [99]. Furthermore, higher succinic acid yield in the fermentation broth is essential for efficient purification and recovery [104]. According to Sun et al. [105], high succinic acid levels in the fermentation broth have accommodated the crystallisation process by providing solubility distances when recovering the acid through crystallisation.

A strategy adopted to improve succinic acid yield is semi-simultaneous saccharification and fermentation (SSSF), which is superior to SSF. Although SSF possesses the advantages of maintaining low contamination, minimal osmotic pressure variation, high amount of water, nutrients, and dissolved oxygen [106,107], it confers slow growth rate and mixing, together with heat generation and environmental conditions regulation that serves as scaling up challenges [44]. The SSSF overcomes the issues by conducting presaccharification within a short duration that provides a faster hydrolytic rate before fermentation, thus producing higher succinic acid concentration and yield [108].

Shen et al. [108] conducted the presaccharification of duckweed (*Landoltia punctata*) by adding 5.90 Amyloglucosidase Units (AGU)/g dry matter glucosidase to 31.86 u/g dry matter pullulanase at pH 4.5 at 2, 4, and 6 h under agitation in a 60 °C water bath shaker. Subsequently, SSF with *A. succinogenes* was performed. A notable increase in succinic acid titer of 65.31 g/L was achieved post-fermentation in serum bottles compared to those attained via conventional separate hydrolysis and fermentation (62.12 g/L) and simultaneous saccharification and fermentation (52.41 g/L). The same strategy was adopted for SSSF albeit with subsequent fermentation in a 2 L bioreactor. After 56 h of cultivation, a higher concentration of succinic acid (75.46 g/L) with 82.87% yield and 1.35 g/L/h productivity was documented.

Immobilising microbial cells onto support materials is another technique for enhancing succinic acid production. In a study by Luthfi et al. [74], coconut shell-based activated carbon (CSAC) with an excellent natural structure and low ash content was employed as the support material for succinic acid production by *A. succinogenes* 130Z from OPF bagasse hydrolysate. A high succinic acid titer, yield, and productivity of 19.4 g/L, 0.85 g/g, and 5.27 g/L.h were achieved at a 0.4/h dilution rate. The improvement was attributed to the efficient lignocellulose sugars intake due to high cell retention [74].

Cao et al. [109] investigated succinic acid production from cane molasses by immobilising the same strain in luffa sponge matrices with NaOH as the neutraliser. After five cycles of the repeated batch culture under optimal pH 6.4, high succinic acid titer, yield, and productivity, 45.6 g/L, 0.76 g/g, and 1.9 g/L.h, respectively, were recorded. The immobilisation assisted in solving the cell flocculation issue at the end of fermentation. The technique also facilitated product separation from the cells, thus making them reusable.

Nevertheless, desorption might occur during immobilisation, which could result in cell loss and mass transfer, and diffusion limitations [44].

7. Conclusions, Challenges, and Recommendations

The present review exhibited an overview of the oil palm biomass utilisation to produce succinic acid and bioplastics. Several challenges still require attention despite the successful generation of succinic acid from oil palm biomass in the studies discussed. Before reaching the fermentation stage, the production of succinic acid from oil palm biomass involves a series of processes, such as saccharification, pretreatment and hydrolysis, which require more time and energy. Besides that, the exploration of cost effective and clean techniques for pretreatment and saccharification, such as microbial hydrolysis of oil palm biomass, is still scarce. Furthermore, Cimini et al. [84] and Ferone et al. [85] confirmed that the toxic by-products generated post-pretreatments posed inhibitory effects on succinic acid production, cell growth, and cultivation of the microorganisms employed due to environmental stress. Moreover, the immobilisation technique adopted as one of the strategies to increase succinic acid production during fermentation could be hindered by mass transfer and diffusion limitations, non-renewable properties of the spent matrix, and the possible loss of cells due to desorption [44]. The current review recommends the approaches listed as follows to address the identified challenges:

1. The potentials of microbial pretreatment and saccharification of oil palm biomass should be explored in great length, as it would be beneficial in establishing a cost effective and clean production process, due to the reduced amount of chemicals utilization;
2. More strategies, such as semi-simultaneous enzymatic hydrolysis and fermentation (SSSF) of oil palm biomass should be explored and optimized to increase time efficiency and minimize energy consumption. To date, a limited amount of research has been conducted to merge the processes prior to fermentation of succinic acid from oil palm biomass into a single unit operation. Proper optimization of the simultaneous process for hydrolysis and fermentation will help to confer a faster hydrolytic rate before fermentation in order to achieve a higher succinic acid yield and final titer;
3. Detoxification process optimisation by targeting to maximise the removal of inhibitors while minimising sugar loss should be considered to solve the issue of toxic by-product generation during pretreatment. Furthermore, alternative methods of detoxification besides activated carbon treatment require exploration.
4. This review only included the generation of PHA and PHB bioplastics through fermentation from OPT sap and EFB hydrolysate in oil palm biomass. Consequently, future studies should consider different types of bioplastic production from oil palm biomass. The possibility of directly generating PBS from the succinic acid downstream processing unit in tandem through process integration should also be examined.

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