



# The Potential of Polyhydroxyalkanoate Production from Food Wastes

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## Abstract

**Background and objective:** Over 1 billion tons of foods are wasted every year (not consumed by humans or animals). Most of this waste ends up in landfills. As the global population increases, mankind must look for more sustainable means of living. A recently popular idea is the use of organic wastes as carbon feedstocks for fermentation that produces value added products. Polyhydroxyalkanoates are a family of bio-based, biodegradable polymers that can be produced in large quantities using food and food processing wastes as the main feedstocks. In many cases, biocatalysts have been engineered to efficiently use these waste compounds to produce large quantities of useful intracellular polyhydroxyalkanoates.

**Results and conclusion:** In the current study, various polyhydroxyalkanoates were produced; each with different thermal and mechanical characteristics useful for different applications. If polyhydroxyalkanoate production facilities are established next to food waste accumulation sites (e.g., large landfills), potentials for the economical and sustainable polyhydroxyalkanoate production sound promising.

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## 1. Introduction

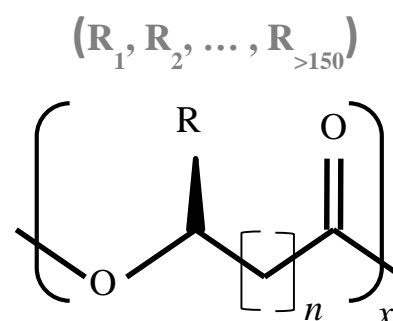
Polyhydroxyalkanoates (PHAs) are a family of biologically synthesized carbon-storage polymers. In microorganisms, PHAs are produced in response to stress conditions and provide protections from nutrient starvation and extreme environments [1]. These polymers are produced and stored intracellularly in the form of inclusion bodies called granules. The PHA granules, and indeed PHA homeostasis in general, have been thoroughly characterized in *Cupriavidus (C.) necator* (previously called *Ralstonia eutropha*). The bacteria are model microorganisms for the study of PHA synthesis-degradation cycle due to its vulnerability to genetic manipulation (gene knockouts and other DNA alterations). Furthermore, *C. necator* can produce up to 90% (w w<sup>-1</sup>) of their cell dry weight (CDW) of PHAs [2-4].

Polyhydroxyalkanoates are polyesters that include various monomers, largely depending on the producing microorganism and the carbon feedstock used by that

microorganism for polymer synthesis. These polymers are divided into at least four categories, including short chain length (*scl*) polymers composed of C3-C5 monomers, medium chain length (*mcl*) polymers composed of C6-C10 monomers, long chain length (*lcl*) polymers composed of C11 and longer monomers, and mixed chain length polymers (e.g., *scl-co-mcl* polymers) composed of monomers with wide variations in chain length [5-7]. Figure 1 illustrates example chemical structures of these categories. The most common PHA polymer is poly 3-hydroxybutyrate (PHB), which is a homopolymer that consists solely of 3-hydroxybutyrate (3HB) monomers; a C4 monomer. The PHB is widely described because many wild-type and environmental isolates produce this polymer when grown on most carbon sources [7-10]. However, some bacteria produce *mcl*-PHA heteropolymers [11-13]. Differences between the microorganisms that produce PHB and other *scl*-PHAs and those that produce *mcl*-PHAs are associated

to differences in monomer supply pathways and PHA synthase (polymerase) types expressed by the microorganism [5,14].

PHA	PHA Class	R <sub>1</sub>	R <sub>2</sub>	n
PHB	<i>scl</i>	CH <sub>3</sub>	.	1
P4HB	<i>scl</i>	H	.	2
P(HB-co-HV)	<i>scl-co-scl</i>	CH <sub>3</sub>	C <sub>2</sub> H <sub>5</sub>	1
P(HB-co-HHx)	<i>scl-co-mcl</i>	CH <sub>3</sub>	C <sub>5</sub> H <sub>9</sub>	1



**Figure 1.** General backbone structure of polyhydroxyalkanoate (right) and examples of PHA polymers (left). HB, hydroxybutyrate; HV, hydroxyvalerate; HHx, hydroxyhexanoate; HO, hydroxyoctanoate. No numbers in abbreviations indicate 3-HA monomers. Co-polymers of 3-HA and 4-HA monomers are possible as P (HB-co-4HB)

Polyhydroxyalkanoates have long been the subjects of biotechnological research. Many PHAs possess mechanical and thermal properties similar to those of petroleum-based plastics such as polyethylene, polystyrene and polypropylene [6,15,16]. Advantages of PHAs over these traditionally-synthesized plastics include that PHAs are bio-based, bio-compatible and biodegradable polymers [17,18]. Generally, PHAs are produced in microbial cultures which often use carbon-rich waste streams as feedstocks; therefore, PHAs produced in this way include more sustainable characters. Polymers of varying monomer compositions have been produced at pilot scales or larger to produce bioplastic raw materials for various used including medical, industrial, agricultural, packaging, cosmetic and household uses [18-22]. Polyhydroxy-alkanoates can be recovered and purified from their microbial hosts using a variety of methods [23,24]. Recently, bio-based methods have been developed using digestion of bacterial biomasses in guts of animals [25,26]. However, organic solvents, halogenated and non-halogenated, have been shown to include the most efficient recovery rate and highest yield and purity with a minimum of alteration to the PHA structure. However, only 50,000 t of PHAs were produced commercially in 2017, resulting in a low market share of 2.4% of the globally produced bioplastics [27-30]. One of the main reasons for the low PHA production is linked to high production costs due to the costs of carbon feedstocks and downstream processes [31]. Side and waste streams from the food processing industries, households and wasted foods in general include the potential to decrease the PHA production cost effectively since they are locally available at low prices in large quantities [32].

## 2. PHA production from food wastes

Food wastes can be divided into three major groups of (i) side streams of the food processing industries, (ii) homogeneous waste streams, and (iii) inhomogeneous food wastes. Various approaches have been described to use food wastes in PHA production (Figure 2). Depending on the composition, concentration, purity and the microbial biocatalyst used, food wastes can be used directly, after a pretreatment and/or a concentration step as feedstocks for the production of PHAs.

### 2.1 Side streams of the food processing industries

Side streams of the food processing industries provide several sugar-rich feedstocks such as whey, molass, starch and lignocellulosic biomass, which are favorable feedstocks for the *scl*-PHA production. Whey consists of 4-5% lactose and is produced in a 10<sup>8</sup>-t scale as byproduct of dairy industries each year [33]. Whey can be used directly or after hydrolyzing to glucose and galactose as feedstocks for the PHA production [34]. Wildtype strains of *Haloferax mediterranei* were used in a life cycle analysis study for the potentially industrial PHA production from whey hydrolysate. Results showed that reasonable high PHA contents were produced per CDW of 50% (w w<sup>-1</sup>). However, a long culture time (> 100 h) and a low conversion rate of whey to PHAs from only 0.8% at pilot plant scales were two fundamental problems that must be solved [35]. However, use of halophiles in bioproduction includes many advantages such as tolerance to high salt concentrations, which allows for a semi sterile bioprocess [36]. Up to date, use of recombinant *Escherichia (E.) coli* still results in the best PHA production with whey as carbon source. Moreover, hydrolysis steps are not necessary since *E. coli* produce enzymes for the breakdown of lactose (e.g.  $\beta$ -galactosidase). Ahn et al. used recombinant *E. coli* harboring the PHA production genes from *Alcaligenes latus* and showed a high PHB production of 2.6-4.6 g h<sup>-1</sup> from

concentrated whey solutions. High cell densities up to 200 g l<sup>-1</sup> were achieved in fed-batch cultivation systems. The

final PHA content reached up to 87% (w w<sup>-1</sup>), which facilitated downstream processing [37,38].

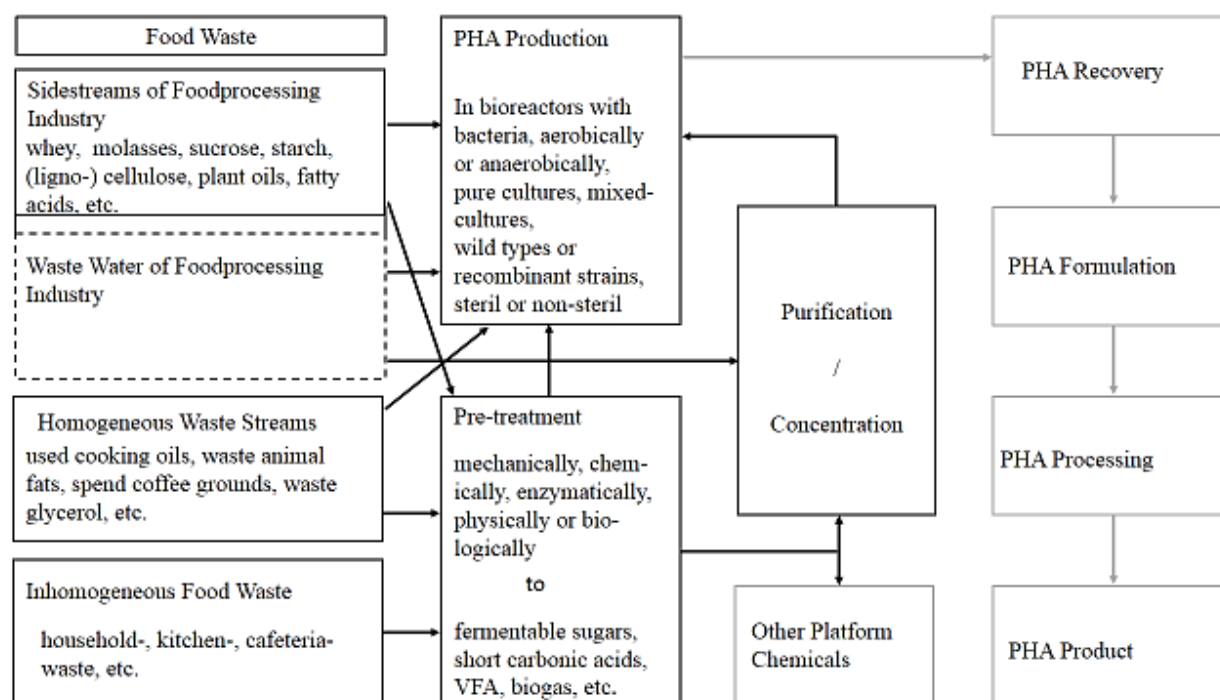


Figure 2. PHA production from food wastes

Molasses contain di and oligosaccharides as sucrose and raffinose and can directly be used as feedstocks [39]. However, a hydrolyzing step is necessary prior to culture for the plant-based materials such as starch to produce fermentable sugars [40]. Efficiency of this conversion is the main challenge for the production of value-added products such as PHAs [41,42]. Sugar concentration, as well as composition and concentration of inhibitors, in feedstocks (e.g., lignin and organic acids) can influence the PHA production. For an efficient PHA production, use of bacterial strains, which are capable of utilizing various carbohydrates in feedstocks, is important. Examples of these carbohydrates include sucrose, fructose, glucose and galactose from molasses and xylose, glucose, arabinose, mannose and glucuronic acid from hydrolyzed plant-based biomasses [39,43]. Saccharified waste potato starch was used to produce PHB in *C. necator* NCIMB 11599. Under phosphate limitation, a final biomass of 180 g l<sup>-1</sup> with a PHA content of 53% (w w<sup>-1</sup>) was produced. Similar results were reported when glucose was used as control substrate [44]. Using molasses, *scl*-PHA copolymer P (HB-co-HV) can be produced in fed-batch cultures by the halophilic bacteria, *Yangia* sp. ND199, with a productivity rate of 0.5 g h<sup>-1</sup>. Sucrose concentration was constantly maintained over 20% (w v<sup>-1</sup>) using two various feed solutions. The first solution used during the first 24 h included 130 g l<sup>-1</sup> of sucrose, yeast extract, peptone and salt. The second solution used up to 54

h included similar concentrations of salt and sucrose but did not include peptone and yeast extract. Furthermore, 50 g l<sup>-1</sup> of CDW with 53% (w w<sup>-1</sup>) PHAs were produced [45]. The highest *scl*-PHA production from sucrose was reported in *A. latus* with 4 g h<sup>-1</sup> of PHB and a total production of 68 g l<sup>-1</sup> during fed-batch culture [46]. The highest *mcl*-PHA production from sucrose was reached using a recombinant *C. necator* strain harboring *csc* genes from *E. coli*. A high PHA accumulation of 81% (w w<sup>-1</sup>) per CDW at the end of the fed batch culture resulted in a total PHA production of 113 g l<sup>-1</sup> P(HB-co-4mol%HHx) with a productivity of 1.7 g h<sup>-1</sup> and a yield of 0.4 g g<sup>-1</sup> [47]. Recently, Purama et al., used date molasses to control the *mcl*-HA concentration of P(HB-co-HHx) copolymer from 2-28 mol% HHx when culturing the recombinant *C. necator* Re2058/pCB113 with date seed oil [48]. The *C. necator* Re2058/pCB113 is able to incorporate HHx and HB monomers from plant oils but only HB monomers are formed from fructose [49]. Furthermore, the strain cannot use glucose or sucrose. However, molasses used by Purama et al. included mainly fructose and glucose and contained only traces of sucrose. Since glucose was not consumed during the culture, HHx content of the polymer was regulated only through the consumption of fructose [48]. The Italian company, bio-on, licenses production plants that use sugar feedstocks with an annual capacity of 5,000-10,000 t of PHAs (Table 1). In summer 2018, the company opened a special PHA production plant with an

annual capacity of 1,000–2,000 t, focusing on use of agricultural waste streams.

**Table 1.** Industrial polyhydroxyalkanoate production from food wastes

Company (founded)/country	Carbon source	PHA production capacity per year	PHA type	homepage
bio-on (2007)/Italy	Sugar beet and cane processing wastes	5,000–10,000 t per licensed plant	MINERV-PHA™ (scl-PHA)	www.bio-on.it
	Organic waste streams including food wastes	1,000–2,000 t		
Hydal (2012)/Czech	Using cooking oils	Starting end of 2018 with 1,000 t potential: 10,000 t	scl-PHA: PHB	www.hydalbiotech.com
Full Cycle Bioplastic (2014)/USA	Organic waste streams including food wastes	Commercial production has not started yet	scl-PHA: P(HB-co-HV), PHA (LA) <sup>1</sup> : P(HB-co-HV-co-LA)	www.fullcyclebioplastics.com

PHA=polyhydroxyalkanoate

## 2.2. Homogeneous waste streams

Homogeneous waste streams containing lipids (as oils and fats) are favorable feedstocks for the PHA production (especially *mcl*-PHA) due to the chain length of the fatty acids in oils and their high carbon contents [5]. Waste frying oils (also known as used cooking oils) has been used as feedstocks [50–53]. The highest production of 138 g l<sup>-1</sup> P(HB-co-8mol%HV) by *C. necator* H16 was achieved using waste rapeseed oil with propanol as HV-monomer precursor [51]. Stanislav Obruca is a part of the Czech/Chinese biotechnology company, Hydal, which produces PHAs from used cooking oils (Table 1). However, the feedstock price of used cooking oils is not much below the price of the available plant oils such as palm and soybean oils. This is seen due to the fact that the used oils can be used as starting materials in biodiesel industries. Used coffee grounds are byproducts of the coffee industries and contain nearly 15% (w w<sup>-1</sup>) coffee oils. Obruca et al. and Kovalcik et al. reviewed PHA production from the coffee waste lipid feedstock [54,55]. Using extracted coffee oils, 1.33 g h<sup>-1</sup> of PHB and a yield of 0.82 g of PHB from 1 g oil were produced by wild type *C. necator* H16 [56]. The *mcl*-PHA P(HB-co-HHx) with a high molar content of 22 mol% HHx was produced from coffee waste oils using a recombinant *C. necator* strain which resulted in a PHA content of 69% (w w<sup>-1</sup>) per CDW [57]. In addition to plant-based vegetable oils, waste animal fats can be used as carbon feedstocks. Riedel et al. produced P(HB-co-HHx) with a HHx content of 19 mol% using recombinant *C. necator* and waste animal fats with high contents of free fatty acids (> 50% (w w<sup>-1</sup>)) [58]. The high free fatty acid contents make these fats unattractive for the other industrial processes, including biodiesel production. This is a part of the research project, PHABIO APP. Another research project named ANIMPOL focused on the production of *mcl*-PHA from animal waste-

based FAMES and raw glycerol, both recovered from the biodiesel processes with *Pseudomonas citronellolis* and *P. chlororaphis* as biocatalysts. Results from both projects showed the possibility of producing 450,000 t of PHAs annually (300,000 t, PHABIO APP) from the 500,000 t available waste lipids of the animal processing industries in Europe [59].

### 2.2.1 VFA production from inhomogeneous food wastes

The most common volatile fatty acids (VFAs), acetic acid, butyric acid and propionic acid, are widely used as platform chemicals in chemical and food industries to produce various products such as adhesive agents, food additives and pharmaceuticals. Nearly 90% of the VFAs are produced synthetically from petrochemical resources. However, VFAs are intermediate products of many bacteria in anaerobic digestion of carbon substrates during the formation of biogas. Atasoy et al. reviewed the status of the bio-based VFA production from waste streams. In addition to the type of substrate and microorganisms used (including inoculum), operation conditions such as pH, temperature, C/N ratio and retention time greatly influence the VFA production yield and composition [60]. The key goals include increasing the efficiency of hydrolysis for the acidification processes and inhibiting the activity of methanogens to increase the yield of VFA (e.g., adjustment of pH during the fermentation) [61,62]. Wang et al. compared VFA formation from food wastes per volatile suspended solids (VSS) with activated sludge at various pH at 30°C. By the increase of pH from 4 to 6, VFA production increased from 124 to 918 mg (g VSS)<sup>-1</sup> under anaerobic conditions. Furthermore, the ratio of VFA species changed with the pH. At pH 4, 80% (w w<sup>-1</sup>) of acetic acid and 20% (w w<sup>-1</sup>) of propionic acid were formed. At pH 6, butyric acid was dominant with 70% (w w<sup>-1</sup>) followed by acetic acid with

15% (w w<sup>-1</sup>) and valeric acid with <5% (w w<sup>-1</sup>) [63]. Domingos et al. developed a continuous VFA production process from cheese whey using immobilized mixed acidogenic microbial cultures (*Lactobacillus*, *Olsenella*, *Actinomyces* spp. and unclassified bacteria) in a packed bed anaerobic bioreactor. The process was divided into two sequential phases of (i) lactose conversion to lactic acid and (ii) lactic acid conversion to VFA. Under steady-state conditions, up to 3 g d<sup>-1</sup> of VFA were produced, which consisted of C<sub>2</sub>-C<sub>8</sub> carboxylic acids with high contents of up to 33% (w w<sup>-1</sup>) of hexanoic acid and up to 25% (w w<sup>-1</sup>) of octanoic acid. The VFA yield was reported as up to 0.85 g g<sup>-1</sup> lactose [64,65].

Composition of VFA can affect PHAs produced from these feedstocks, depending on the biocatalysts used. Strains capable of *scl*-PHA synthesis alone produce mainly HB monomers from all VFAs, with the exception of HV monomers that are incorporated into the PHAs when VFAs such as valeric and propionic acids are present in the feedstocks. However, if microorganisms are used that are capable of *mcl*-PHA synthesis, hexanoic and octanoic acids (see above) present in the VFA feedstocks can be used as substrates for the incorporation of HHx and HO monomers. Therefore, a controlled formation to achieve specific ratios of VFAs can be used to tailor the PHA compositions and thereby the polymer properties.

In addition to increased bio-based VFA production from waste streams, many approaches of VFA recovery post fermentation have been described. Using electrodialysis, VFAs were recovered and concentrated up to 5-folds (12.6–63 g l<sup>-1</sup>) from a continuous production process [65]. However, investigations are focused on *In-situ* recovery methods to increase the total VFA production since VFAs inhibit growth of the producer microorganisms at certain concentrations [60]. Moreover, reduced fermentation times for the VFA formation have been reported when using *in-situ* recovery methods [66].

### 2.2.2 VFA from inhomogeneous food wastes for PHA production

As reviewed previously, concentrated VFA solutions, alone or mixed, have been used to produce *scl*-PHA with wild-type or recombinant strains of *C. necator*. Generally, PHB and P(HB-*co*-HV) (if valeric acid was present) with HV concentrations up to 99 mol% were accumulated. High productivities of >1 g h<sup>-1</sup> with PHA contents up to 73% (w w<sup>-1</sup>) were reached using pH controlled feeding of VFAs [5]. Concentrated VFA solutions produced from cheese whey as described above [64,65] have been used for the PHA accumulation using wild-type *C. necator* strains, which can only produce *scl*-PHA. A high yield of 0.6 g of PHA per gram of VFA with a high PHA content per CDW of 71% (w w<sup>-1</sup>) with a molar composition of 94 mol% of HB and 6 mol% of HV were produced after 52 h of fed-batch

culturing. However, the total biomass was low, only 15 g l<sup>-1</sup> [65].

Use of mixed microbial cultures (MMC) can help reduce the overall cost of PHA production due to the fact that sterile environments are not necessary. The general PHA production process using MMC is divided into four major steps of (i) acidogenic fermentation of organic substrates to form VFAs, (ii) enrichment of MMC biomasses able to produce PHAs using feast-famine cycling, (iii) accumulation of PHAs in MMC biomasses, and (iv) recovery of PHAs [67].

After oil separation, canteen wastes with organic loads of 18 kg of chemical oxygen demand (COD) (M<sup>3</sup> day)<sup>-1</sup> were used in a multistage process for the bio hydrogen and PHA productions. In the first stage, VFAs were produced during anaerobic fermentation with MMC acquired from a wastewater treatment plant for 48 h, resulting in a biohydrogen production of 60 l after 24 h, and 4 g l<sup>-1</sup> of VFAs after 48 h. Then, pH of the VFA solution was adjusted from 4 to 8 prior to use as feed for aerobic MMC obtained from a sequencing bioreactor (SBR) pilot reactor of the wastewater treatment plant under feast and famine conditions for the enrichment of PHA producing organisms. The VFA solution was supplemented with ammonium and phosphate to maintain a C:N:P ratio of 100:8:1 during feast conditions. After a 12 or 24-h cycle, nearly half of the biomass was transferred to another reactor, where VFA solution was fed without additional nutrient supplementation for the PHA production for 12–24 h. Shortening the cycle time resulted in increased PHA accumulation. A maximum PHA content of 24% (w w<sup>-1</sup>) per CDW was reached after 12 h in the PHA reactor with a storage yield of 0.17 g of PHA<sub>COD</sub> g<sup>-1</sup> WW<sub>COD</sub>. The storage yield was calculated as the ratio of stored polymer quantities at the end of the cycle to COD quantities consumed in the wastewater [68].

In general, PHAs from MMCs can vary in quality due to the various PHA producers in mixed cultures. Furthermore, when inhomogeneous food wastes from households or restaurants are used as carbon sources, their changed contents of carbohydrates, lipids and proteins can result in PHA product variability. After the recovery, a blend of PHAs with varying monomer contents and molecular weights is resulted, which influences properties and processing behaviors of the polymers [69]. However, the company, Full Cycle Bioplastics LLC, claims to produce a consistent polymer output from heterogeneous mixed organic waste streams under non-sterile, industrial operating environments with non-GMO bacteria (Table 1). The co-founders of the company, Dane and Jeff Anderson, patented the production of *scl*-PHA polymers P(HB-*co*-HV) and P(HB-*co*-HV-*co*-LA) with controllable monomer contents from various organic waste streams [70]. The carbon sources may include algae, vegetable fats and oils, corn

starches, agricultural wastes, food wastes, trash sluices and other biomass processing by-products such as glycerol. The highlight of the bioprocess includes a system to generate VFA mixtures with a desired composition to reach consistent polymer outputs. Organic waste streams are separated or mixed with each other, depending on the composition, before they are used for the VFA production. Prior to VFA production, waste streams can undergo a liquefaction process through enzyme and/or bacterial digestion (e.g., cellulose hydrolysis for starches). The VFA production process may be carried out using acid-phase anaerobic digestion, hydrolysis, bacterial fermentation or combinations of these techniques. Obligatory anaerobic acidogenic bacteria of the following genera are used, including *Pseudomonas*, *Bacillus*, *Clostridium*, *Micrococcus* and *Flavobacterium*. Depending on the individual acid composition, VFA-rich liquids are further fermented in polishing tanks to shift fermentation process to produce shorter-chain VFAs or allow longer-chain VFA molecules to remain. This can be occurred using control of fermentation time and pH adjustment. A relatively low-pH set point of 4-5 inhibits VFA producing bacteria to produce feedstocks with larger proportions of longer-chain VFAs. However, with an extended fermentation time at higher pH set points of 5–6.5, all acids are metabolized to acetic acids. Fermentation time is stopped when approximate target mixtures of VFA concentrations (or ratios) are reached. Desired VFA ratios (or concentrations) can be reached using separation of the target VFAs through dilutions or additions of previously separated VFAs (or lactic acids). Separation of a target VFA is carried out using several filtration systems, including membrane and electromagnetic filtrations.

Described VFA mixtures of 10-30 g l<sup>-1</sup> are then used for the PHA production from MMCs. A C/N ratio of 6-10 is used in SBR tanks under feast-famine cycles to enrich PHA production biomasses. A cycle time of 6-36 h is used under controlled pH set points of 7-9. Nitrogen is fed as ammonia over the pH control. After enrichment, the biomass is transferred into the PHA production tank, which is operated for approximately 4 h with a C/N ratio of 10-20 under aerated conditions. Nitrogen and/or VFAs are fed for further cell growth and PHA production. With depletion of nitrogen, the PHA production phase starts. Generally, PHA accumulation is triggered by stopping the airflow into the reactor, causing oxygen limitation. After PHA production, the biomass is separated (e.g., through centrifugation) and dried by heating. Intracellular PHA contents of 50–90% (w w<sup>-1</sup>) are reached. The dried biomass is transported to a processing facility, where they undergo various levels of extraction and purification processes tailored to various end uses. Normally, P(HB-co-10wt%HV), P(HB-co-wt22%HV), P(HB-co-wt33%HV), P(HB-co-wt6%HV-co-1wt%LA), P(HB-co-25wt%HV-co-25wt%LA) and P(HB-

co-10wt%HV-co-40wt%LA) are some examples of biopolymer portfolios. One of these examples, P (HB-co-33wt%-HV) is produced from a VFA mixture of 58% (w w<sup>-1</sup>) of acetic acid, 33% (w w<sup>-1</sup>) of propionic acid and 9% (w w<sup>-1</sup>) of isobutyric acid, where the HV content is dictated by the quantity of propionic acid in the VFA feed. In addition to PHA production, side-streams of VFA production or organic waste separation are used in other processes such as biofuel, biogas and biodegradable road salt (calcium magnesium acetate) productions. [70].

### 3. Food wastes as resources for PHA production

Any food processing end-product that is not consumed, recycled or used for other purposes is considered as waste. Over one billion tons of food wastes are generated worldwide every year [71,72]. In Europe, nearly 89 million tons of foods are wasted each year; with over 80% of this coming from the manufacture/production and household sectors (municipal wastes). Therefore, food wastes are produced at every stage of the supply chain [73]. In 2013 in USA, approximately 37 million tons of foods ended up in municipal waste systems [74]. These wastes include high moisture and biodegradability and can make significant disposal problems and adverse environmental impacts if landfilled [75]. While composition of food wastes can vary, it is clear that abundant usable carbons are present in these waste streams. An estimation of organic household wastes was shown to contain nearly 550 kg t<sup>-1</sup> of carbohydrates, 120 kg t<sup>-1</sup> of lipids and 55 kg t<sup>-1</sup> of proteins [71]. Carbohydrates and proteins in wastes can be converted into sugars and amino acids, respectively. These recovered nutrients can be used as feedstocks in industrially relevant microbial cultures for the production of value added products. Bioconversion processes have been developed for the microbial production of various small-molecule bioproducts such as ethanol, butanol and biocolorant compounds [76-78].

It is worthy to point out differences between industrial and household food wastes, especially if the purpose is to use these wastes as feedstocks in bioprocesses. Agricultural and food processing wastes are further predictable in volume and composition than household wastes are. Waste from agricultural and food processing industries are generated in a further concentrated manner and hence are easier to collect and use as feedstocks in valorization [73]. As described earlier, many types of food processing wastes have been used for the production of PHA bioplastics. Household food wastes and disposals are challenging and costly, especially in dense urban areas. As landfilling becomes a less attractive option for the disposal of food wastes, alternative methods such as local and household recycling and incineration for the energy production are

become further popular. According to the former fact, two types of social recycling systems have been suggested to solve the food waste problem. One includes translocation of food waste recycle streams to regional facilities and the other includes in-household food waste recycling machines [79]. While both methods include advantages and disadvantages, products from these processes (or the food waste powders) can be collected and their nutrients used for fermentative valorization. Production of these recycled food waste powders result in streams that are significantly reduced in overall masses and thus more transportable and less susceptible to rapid degradation (rotting) and odor production in the absence of water. In recent years, many city managers have attempted to subsidize the household food waste recycling processes to minimize the overall mass and volume of the wastes in metropolitans [72,79].

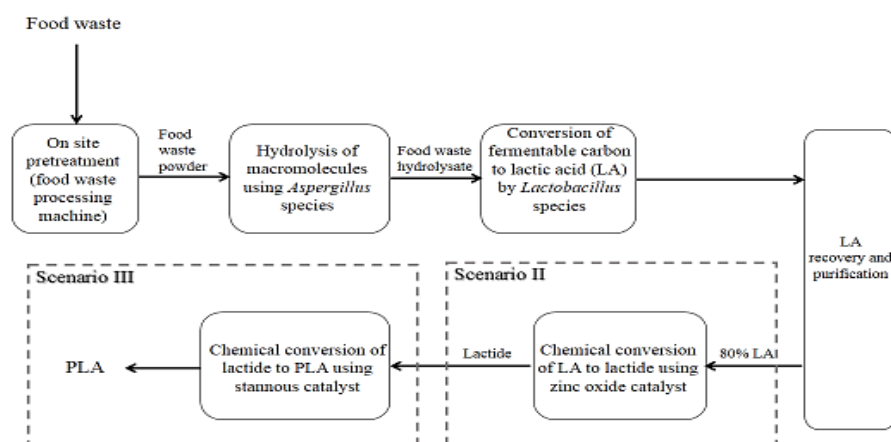
#### 4. Feasibility of PHA production from household food wastes

For a successful biorefinery, the process must be adapted to the market needs and bulk biochemical productions driven by supply/demand issues as well as scales of the economy. Production of commodity products from food waste streams may not be feasible in some cases because the low market prices of these products (or competing products) require large production capacities in industrial-scale plants. This issue is seen in PHA production since PHAs are competing with relatively inexpensive, traditional petroleum-based polymers as bio-based, biodegradable thermoplastics. Therefore, development of scalable, inexpensive PHA production processes is necessary to compete with these traditional thermoplastics. There are two significant problems that hinder the cost competitiveness of PHAs, including cost of the carbon feedstocks for microbial

culture and downstream process of the polymers [74]. Use of food wastes as carbon feedstocks can address the former problem as long as waste collection and transportation costs are not a hindrance.

The PHA production from organic municipal solid wastes has been demonstrated using conversion of wastes into organic acids and subsequent conversion of the organic acids into PHAs, specifically poly (hydroxybutyrate-*co*-hydroxyvalerate) [P (HB-*co*-HV)]. The best case yield from this process has been shown as nearly 33 g of PHAs (kg waste)<sup>-1</sup> [75]. Similar bioconversions have been carried out, using palm oil mill effluents to produce organic acid feedstocks [80]. Huschner et al. developed a high-cell density cultivation method to produce P(HB-*co*-HV) from mixed organic acids using a dual acid/acid salt feeding method [81]. To the best of the authors' knowledge, a technoeconomic analysis of PHA production from municipal food wastes has not been published yet.

Fermentative lactic acid (LA) and polylactate (PLA) production strategies have been described using municipal food wastes as carbon source. Venus et al. investigated LA synthesis using two different production approaches of a one-step approach using simultaneous hydrolysis of macromolecules found in food wastes and fermentation of hydrolysates and another two-step approach that separated the hydrolysis of waste carbons and subsequent fermentation of bioavailable carbons into separate steps. A one-step LA production scheme involving simultaneous saccharification and fermentation (SSF) is suggested for onsite use of waste carbon sources (e.g., potentially at food processing facilities where large amounts of wastes are collected). The two-step LA production method is more appropriate for offsite production facilities (e.g., when municipal food wastes must be transported from elsewhere) [71].



**Figure 3.** Lactic acid/lactide/polylactate production scenarios studied by Kwan et al. [72]. Municipal food wastes are pretreated onsite (at homes) using a food waste processing machine. Food waste powders are transported to a processing facility for further pretreatment and breakdown of polysaccharides, proteins and other macromolecules by *Aspergillus* spp. Then, hydrolysates are used as carbon feedstocks for fermentation by *Lactobacillus* spp. to produce lactic acid. Streams consisting of 80% of lactic acid are recovered and purified (Scenario I). Then, lactic acids are converted chemically to lactide using zinc oxide catalysts (Scenario II). Lactide can be a special product or further converted to polylactate. Lactide is converted chemically to polylactate using stannous octoate catalysts (Scenario III).

Kwan et al. analyzed production of LA, lactide and PLA using municipal food wastes. The three products represented three scenarios in the technoeconomic analysis used. Food wastes were first pretreated using commercial waste processing machines to produce food waste powders. These food waste powders were then hydrolyzed in cultures of *Aspergillus* spp. to produce fermentable carbon sources. The LA was produced by *L. casei* using food waste hydrolysates by the *Aspergillus* spp. as the major carbon source. In other scenarios, LA was chemically converted to lactide and then lactide to PLA (Figure 3). Mass balances showed yields of 3.1 Mt of 80% LA, 1.7 Mt of lactide and 1.3 Mt of PLA. The authors showed that all scenarios were economically feasible but the scenario producing only LA from municipal food waste carbons was potentially the most profitable [72].

## 5. Conclusion

Challenges for the large-scale industrial PHA production have always been centered on the selection of carbon feedstocks (e.g., costs) and polymer recovery methods. Use of food wastes, food processing wastes, treated processing wastes and municipal wastes as carbon feedstocks helps to address one of these challenges. Many studies have been carried out to find applicable solutions to decrease PHA production costs. Indeed, companies proclaim to solve the cost problem with patented methods. However, these proclaims remain to be practically verified.

What have been pointed out in this review include technology and innovation which are motivating the bioeconomy. As we strive to become cleaner, greener societies with lesser wastes and further recycles, processes such as those described here can help achieve these purposes. Relatively, PHAs are useful biodegradable raw materials which can be used for the recycle of waste streams. Similarly, VFAs are valuable materials which play multiple roles in industries, especially as feedstocks for the PHA production. These feed materials can help food industries to transform wasted carbons to value added products.

The future of bio-based production industries is promising which needs innovations. Implementation of processes and products require good knowledge of industrial strategy and ecology. Wastes should be used as feedstocks, but must be eliminated from the bio-based processes. For example, it is more efficient to process cells filled with PHAs onsite, than to ship them to other locations via roads or railroads. Outlooks for the PHA production must be (or at least should be) focused on providing a central environment for all steps of the production process, as well as closing the production loop and using waste materials.

## 6. Conflict of interest

The authors declare no conflict of interest.

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## تولید بالقوه پلی هیدروکسی آلکانوات‌ها از ضایعات مواد غذایی

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### تاریخچه مقاله

دریافت ۲۵ آگوست ۲۰۱۸  
داوری ۷ سپتامبر ۲۰۱۸  
پذیرش ۱۸ اکتبر ۲۰۱۸

### واژگان کلیدی

- پل زیست‌بسپارها
- منبع کربن
- تخمیر
- ضایعات مواد غذایی
- پلی هیدروکسی آلکانوات

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### چکیده

**سابقه و هدف:** هر سال، بیش از یک بیلیون تن مواد غذایی (آنچه توسط انسان یا حیوان مصرف نشده است) ضایع می‌شود. مسیر اغلب این مواد ضایع شده به گورستان زباله‌ها منتهی می‌شود. با افزایش جمعیت جهان، بشر باید به دنبال وسایلی بادوام‌تر برای زندگی باشد. ایده استفاده از ضایعات آلی به عنوان منبع کربن برای تخمیر به منظور تولید فرآورده‌هایی با ارزش افزوده‌ای در دهه‌های اخیر محبوبیتی به دست آورده است. با استفاده از ضایعات مواد غذایی و ضایعات فرآوری مواد غذایی، به عنوان ماده اولیه، می‌توان پلی هیدروکسی آلکانوات‌ها، که (بسپارهایی)<sup>۱</sup> (زیست‌تجزیه‌پذیر)<sup>۲</sup> بر پایه ترکیبات زیستی می‌باشند را در مقادیر زیاد تولید کرد. در بسیاری از موارد، برای تولید مقادیر زیاد بسپار خارج سلولی و مفید پلی هیدروکسی آلکانوات‌ها با استفاده از مواد ضایعاتی، از زیست‌کاتالیست‌ها در طراحی کاراً استفاده می‌شود.

**یافته‌ها و نتیجه‌گیری:** انواع بسیار گوناگون از پلی هیدروکسی آلکانوات‌ها تولید شده‌اند، هر بسپار با ویژگی‌های مکانیکی و حرارتی مناسب برای کاربردهای متفاوت کارایی دارد. در صورت فراهم شدن شرایط تولید پلی هیدروکسی آلکانوات و بهره‌برداری از مناطق تجمع ضایعات مواد غذایی (مانند مراکز بزرگ دفن زباله)، توانایی تولید اقتصادی و کافی پلی هیدروکسی آلکانوات‌ها به نظر امید بخش می‌باشد.

**تعارض منافع:** نویسندگان اعلام می‌کنند که هیچ نوع تعارض منافی مرتبط با انتشار این مقاله ندارند.

<sup>1</sup> Polymers

<sup>2</sup> Biodegradable