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Technical review

Microbial PolyHydroxyAlkanoate (PHA) Biopolymers – Intrinsically Natural

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Abstract: Global pollution from fossil plastics is one of the top environmental threats of our times. The end-of-life scenarios of fossil plastic, including recycling, incineration and disposal result in microplastic formation, elevated atmospheric CO_2 levels, and littering of terrestrial and aquatic environments by plastic waste. Currently thought-out regulations centered around banning plastics production and use and only recycling on a regional, national and global level are impeding efforts to rapidly replace fossil plastics through the use of natural alternatives. In particular, this review demonstrates how microbial polyhydroxyalkanoates (PHA), a class of intrinsically natural polymers, can easily remedy for the fossil and persistent plastic dilemma. PHA are bio-based, bio-synthesized, bio-compatible, bio-degradable, and home- and industrially compostable. Therefore, they are one of the perfect replacements for our fossil plastics pollution dilemma, providing us with the benefits of fossil plastics and meeting all requirements of a truly circular economy.PHA biopolyesters are natural and green materials in all aspects of their life cycle. The review elaborates in detail how PHA's production, consumption, and end-of-life profile are perfectly embedded in the current topical 12 Principles of Green Chemistry, which constitute the basis for sustainable product manufacturing.It is shown that it is time for a paradigm shift in plastics manufacturing, use and disposal. Humankind needs alternatives to fossil plastics, which, as recalcitrant xenobiotics, contribute to the increasing deterioration of our planet. Natural PHA biopolyesters represent that paradigm shift!

Keywords: biopolymers; green chemistry principles; natural polymers; polyhydroxyalkanoates

1. Introduction

Increasing production and use of synthetic and recalcitrant polymeric materials of petrochemical origin for packaging, personal care and others are currently responsible for several global threats: accumulation of plastic waste in aqueous and terrestrial environments, formation of highly recalcitrant xenobiotic microplastic particles causing the death of ocean animals, deteriorating ecosystems and negatively affecting human health.

As reaction to these threats, policies worldwide are striving to replace unsustainable linear economy approaches ("take-use-dispose") like production and use of fossil and persistent plastics with circular alternatives ("take-use-recycle"). Legislations such as California Assembly Bill (AB) 1201 on "Solid waste: products: labelling: compostability and biodegradability" [1], Senate Bill (SB) 54 on "Solid waste: reporting, packaging, and plastic food service ware" [2], the European Commission´s Single Use Plastics Directive or SUPD (EU 904/2019) [3], the Plastics and Packaging Waster Directive (PPWR) [4], and the REACH Amendment on Intentionally Added Microplastics [5] go in this direction. The overarching goal is to reduce virgin plastics use, their leakage into the environment, and the resulting pollution. By encouraging plastics recycling the legislations seem to believe that the effects of persistent fossil plastics can be reduced or even eliminated. Recycling needs to be encouraged and improved; the sheer volume of fossil plastics used today (annual production of about 400 Mt) has no immediate replacement. However, recycling only postpones the negative effects of plastics by postponing the end of life of the materials. Chemical recycling is in its infancy, and mechanical recycling increases microplastics formation that are inherent in all plastics handling, processing, and use. These policies and legislations while restricting fossil plastics, are also restricting introduction of innovative alternatives. Policies and legislations are redefining words like “plastics” and “natural polymers” that are restricting introduction of innovative alternatives that are both circular and inherently sustainable. Instead, these policies need to focus on the circularity and sustainability of materials, through a) renewable carbon use and from the b) end-of life perspective of the material and not just the packaging. Some examples of restrictive legislations have been mentioned above. Many of these legislations such as California SB 54 [2] and the European Union’s Single Use Plastics Directive (SUPD) [3] are focused on defining materials using terminologies like “plastics” and “natural polymers” to restrict the use of these persistent fossil plastics for cutting harmful plastic pollution. Instead, we believe that legislation needs to focus on the circularity and end of life profiles and options rather than focusing on redefining already established terminologies. For example, renewability, biodegradability and compostability should be specified, along with standards and certifications that allow for such materials to be used. A second area that these legislations should focus on are encouraging and mandating increased separation and collection, and composting infrastructure to facilitate increased organics recycling. Examples of such legislations, which allow for biodegradable and compostable materials are California Assembly Bill 1080 – “Solid waste: packaging and products” [6], or California SB 270, which imposes a statewide ban on single-use plastic bags at large retail stores [7]. Here, it should be noted that no nation-wide regulations exist for the US. Since December 20th, 2022, the manufacture and import for sale in Canada of checkout bags, cutlery, foodservice ware, stir sticks and straws (i.e., straight straws), as defined in respective Regulations, are prohibited [8]. In the People’s Republic of China, a policy document jointly issued by the National Development and Reform Commission (NDRC) and the Ministry of Ecology and Environment on January 16th, 2020, postulated the plastic ban that prohibits restaurants throughout the country from providing single-use plastic straws and stores in the major cities from providing plastic shopping bags; this ban took effect on January 1st, 2021 [9]. In April 2022, the Act on the Promotion of Resource Circulation for Plastics was enacted in Japan to improve the circularity of plastics [10]. In summer 2022, India imposed a ban on single-use plastic to tackle pollution, including straws, cutlery, ear buds, packaging films and cigarette packets, etc. [11].

These legislations have also restricted materials that are renewable, biodegradable, and home and industrially compostable, attributes that would reduce and eventually eliminate the accumulation of persistent and fossil polymers and the dangers that they cause. Indeed, many of these legislations consider plastic recycling an appropriate strategy to overcome today´s plastic pollution predicament [12]; however, plastic recycling rather postpones the problem, and does not provide a solution. Instead, real “Natural Polymers” should be forcefully promoted by legislation.

In this position paper we clarify: i) what “Natural Polymers” are, and ii) how they comply with the terminus “plastics” in its true sense of the meaning. In addition, we highlight attributes in materials that enable circularity and sustainability without the additional burden of fossil carbon and plastics thereof.

2. Definition of “natural polymers” – clarifying existing ambiguities

When it comes to sustainable and circular materials, nature is the best teacher. What nature makes, nature unwinds and remakes. Therefore, the prefix “bio” is frequently used to connect an object or a product to nature. Several such words exist:

“**Bio-based**” – A chemical compound or material produced from renewable carbon sources. “Bio-based” does not specify whether the product or material is synthesized chemically or biologically in plants, organisms, and animals.

“**Bio-synthesis**” - The synthesis of a chemical or material through the actions in living organisms (plants, microorganisms, or animals) or parts thereof (enzymes). A biosynthesized polymer implies having been produced in plants, microorganisms, or animals through the action of enzymes *in vivo*.

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The word “polymerization” implies a synthetic or an anthropogenic process for generation of polymers from building blocks that are either bio-based, bio-synthesized or chemically synthesized.

“**Bio-degradation**” This implies the breakdown of a chemical or material through the actions of naturally occurring enzymes *in vivo* or extracellular enzymes that are secreted by plants, microorganisms, or animals. Generally, breaking down chemicals or materials involves eventual transformation to carbon dioxide (CO_2) and water and complex biomass called humus, also known as organic fertilizer. Hence, “biodegradation” can be understood as nature’s way of material recycling.

“**Bio-compatibility**” Implies chemicals or materials that do not exert harmful effects on the environment and on living systems including humans.

Materials that fulfil these criteria specified above are well-known such as cellulose, starch, chitin, alginates, proteins, and nucleic acids (DNA, RNA). All these are also macromolecules and are excellent examples of “Natural Polymers”, also called a “Biopolymers”. Therefore, “Natural Polymers” are defined as those that, at the same time,

a. Derived from renewable carbon such as sugar from sugar cane, fatty acids like canola oil or carbon dioxide or methane from biogas or the atmosphere.
b. Are biosynthesized in plants, microorganisms, and animals.
c. Biodegrade or turn into CO_2, water, and humus, and
d. Are biocompatible.

3. PolyHydroxyAlkanoates are Natural Polymers

PolyHydroxyAlkanoates (PHA) fulfil every criterion set out in the definition of a “Natural Polymer” or a “Biopolymer”. PHA biopolymers, a group of biopolyesters, are found in nature, and they are biosynthesized using renewable carbon in microbes. PHA are biodegradable, because nature has the tools to convert them into CO_2 and water and about 10% organic fertilizer or humus, the same as in the biodegradation of cellulose or cotton. PHA are biocompatible, they are harmless to all living beings and can be used *in vivo* as medical devices such as scaffolds, stents or as meshes. They will eventually be absorbed by the body with no traces of the PHA remaining [13].

Evidence: PHA is a Natural Polymer

We spoke of “Nature knows best” when it comes to circularity of chemicals and materials that are found in nature. For what nature creates, nature also develops ways to degrade and recycle. This is especially true for cellulose as well as PHA. After their end of life, PHA gets mineralized or biodegraded by natural hydrolytic depolymerase enzymes to exactly those chemical building block from which they were once made in nature. They include CO_2 and water, which then through photosynthesis get converted into sugar (sugarcane or corn) or lipids (canola oil or palm oil), used today to produce PHA. This demonstrates that the formation and degradation of PHA is inherently circular!

Figure 1 illustrates the fact that PHA biopolyesters are entirely embedded in nature´s closed material cycles, which is vividly exemplified by the old quote attributed to Lavoisier, a pioneer in the field of chemistry, when he explained: In Nature “*Nothing gets lost, nothing gets created, everything gets transformed.*” [14].

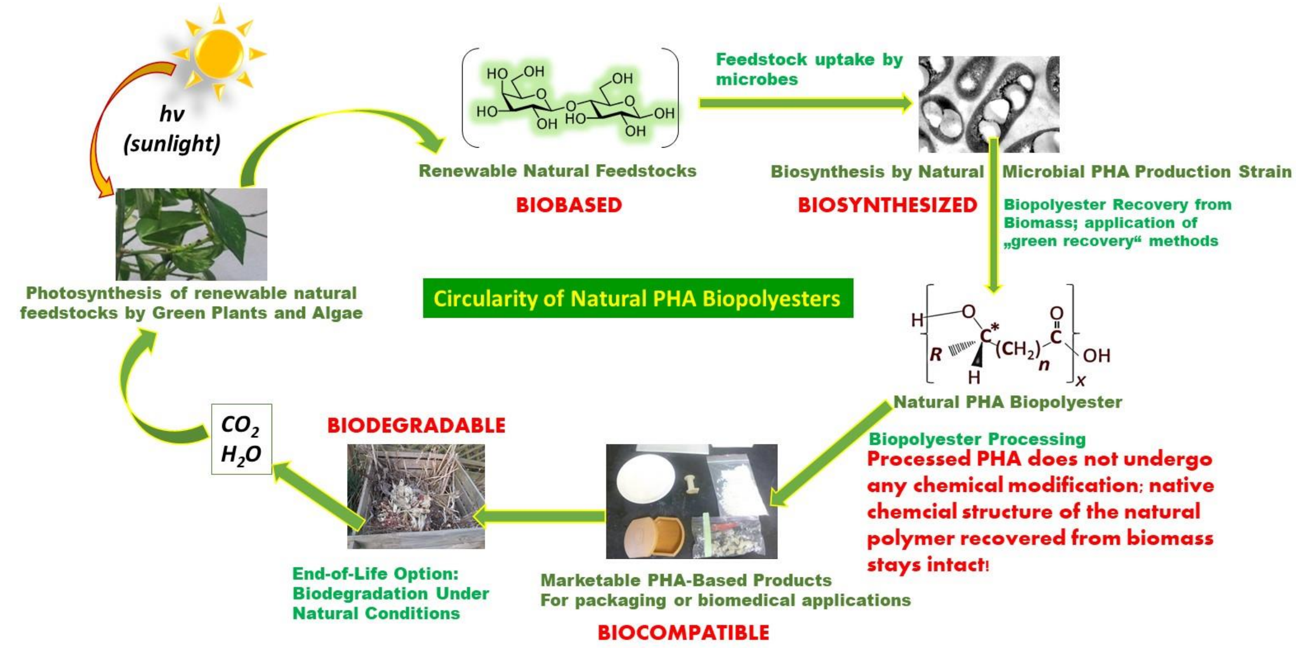

Figure 1. Circularity of natural PHA biopolyesters.

4. PHA are produced by natural microorganisms

Production of PHA occurs in naturally occurring prokaryotic microorganisms, which can be isolated from natural sources as diverse as the ocean, estuaries, soil, salt rocks in the mountains, plant surfaces, the rhizosphere, or the guts of insects. Hundreds of microbial species from both prokaryotic domains, Bacteria and Archaea, are reported to produce PHA. These microbial "biopolymer cell factories" thrive under most diverse ecological conditions regarding their optima of temperature, pH-value, salinity, substrates, or concentrations of organic or inorganic toxins, hence, as well mesophilic organisms as extremophiles are reported as PHA accumulators. The best described PHA production strain, *Cupriavidus necator* [15], or the strain *Bacillus* sp., in which PHA was detected for the very first time by Maurice Lemoigne [16], were originally isolated from soil samples. Other strains, which are currently heavily studied due to their robust nature and convenient cultivability under low-sterility conditions, such as *Halomonas* sp., are marine isolates [17]. From polluted habitats like oilfields, versatile PHA-producing species like *Aneurinibacillus thermoaerophilus* were isolated [18]. PHA biosynthesis is observed in both heterotrophic (conversion of organic carbon sources for biomass, product and energy generation) and (photo)autotrophic (CO_2 as inorganic carbon source) species isolated from diverse natural samples [18].

5. Biosynthesis of PHA in natural strains occurs via natural biocatalysts

PHA biopolyesters are accumulated in natural organisms as intracellular granules; they serve the harboring cells as reserves for energy and carbon, electron sinks, and stress protectants, and provide PHA-containing cells survival advantages under conditions of starvation due to lacking exogenous carbon source. They are predominantly produced by the cells under conditions of high intracellular energy charge, ample availability of exogenous natural carbon sources, combined with deprivation of additional growth factors, such as nitrogen or phosphate sources. Such conditions inhibit the formation of catalytically active biomass but boost PHA productivity. Hence, PHA are synthesized as typical intracellular products of the cell´s secondary metabolism [20]. As a precondition for PHA biosynthesis, a given strain needs to possess the key biocatalysts (enzymes) for PHA biosynthesis. The first is 3-ketothiolase (E.C. 2.3.1.9), which condenses two molecules of acetyl-CoA, the central catabolite of the breakdown of natural resources, to acetoacetyl-CoA that in turn gets reduced by the action of a reductase enzyme (E.C. 1.1.1.36), generating *R*-3-hydroxybutyryl-CoA, the substrate for PHA synthase (polymerase) enzymes (E.C. 2.3.1.304), which link the individual *R*-3-hydroxybutyryl-

CoA building blocks to form polymeric PHA chains *in vivo* [21]. Therefore, the catalysts needed for PHA biosynthesis (the enzymes) match Principle 9 of Green Chemistry, which says: New catalysts shall be non-toxic, selective, and efficient [22].

This is vastly different from poly(lactic acid) (PLA), where polymerization of biologically produced lactic acid occurs via chemical processes (dimerization, ring opening polymerization), which requires non-natural, expensive, and often hazardous catalysts [23]. Importantly, PHA biosynthesis occurs under mild conditions, reflecting the optima of the strains´ enzymes for pH-value, temperature, salinity, or pressure. Production of petroplastics, on the contrary, needs harsh, energy-demanding reaction conditions, and typically expensive and hazardous catalysts. For illustration, the production of poly(ethylene terephthalate) (PET, a polyester) requires temperatures above 200°C, and typically antimony oxide as a catalyst [24]. Hence, PHA biosynthesis follows Principle 3 of Green Chemistry, which demands safe chemical reaction for the production of safe products from safe educts [22].

6. PHA biosynthesis is based on natural feedstocks

Principle 7 of Green Chemistry postulates the replacement of fossil feedstocks by renewables [22]. This is perfectly valid for PHA: Carbohydrates stemming from the photosynthetic fixation of CO_2 are the predominant natural feedstocks used for PHA biosynthesis (Figure 1). This goes especially for monosaccharides such as the hexoses glucose or fructose and, for a lower number of microorganisms, the pentose xylose. These monomeric sugars can conveniently be generated from polysaccharides like starch, cellulose, or even abundantly available lignocellulose via well-established hydrolysis techniques (enzymatic or chemical). Notably, these polysaccharides frequently stem from agro-industrial waste; upcycling of such waste to feedstocks integrates PHA biosynthesis into biorefinery concepts, which perfectly matches current circular bioeconomy paradigms. In many cases, hydrolysis of said polysaccharides is not even needed: A cohort of strains is reported to directly convert di-, oligo-, and polysaccharides to biomass and PHA, provided the strains have the adequate hydrolase enzymes available. Examples are the expedient direct starch converter *Haloferax mediterranei*, which has high amylolytic activity [25], the invertase-excreting strain *Paraburkholderia sacchari* [26], which readily converts the disaccharide sucrose, or *Hydrogenophaga pseudopflava*, a well-described consumer of the disaccharide lactose [27]. Other strains are excellent PHA accumulators from lipids produced by natural organisms (microbes, plants, or animals), such as *Pseudomonas putida* [28]. Emerging PHA production processes are based on gaseous substrates; here, a range of hydrogen oxidizers like *C. necator* [29] and many phototrophic cyanobacteria [30] can grow and accumulate PHA by using CO_2 as the sole carbon source; CO_2 in turn is the final product of the bio-mediated aerobic breakdown of biomass and also of disposed items made of PHA. Moreover, several Type II methanotrophic bacteria can be cultivated on CH_4, a product of the composting and anaerobic degradation of natural products like organic waste and PHA. It is also the main component of natural gas [31]. This again nicely illustrates the circularity of PHA: After their life span, bioplastic products made of PHA undergo natural degradation to exactly those starting materials, which, in a next cycle, fuel PHA biosynthesis (Figure 1). Finally, the microorganism class Rhodospirilli are known to produce PHA even from CO-rich syngas, a substrate accessible from pyrolysis of most biological resources, particularly waste biomass [32].

7. PHA are biodegraded in nature – marine, fresh water and soil in every environment around the world

Principle 10 of Green Chemistry is about “biodegradability”: Products should be able to degrade naturally (or “biodegrade”) after use without harming the environment [22]. As has been mentioned above, aerobic (oxidative) degradation of PHA by microbes like bacteria or fungi generates CO_2 and water, while anaerobic PHA consumption by living organisms, e.g., in biogas plants, results in generation of CH_4 in addition to water and CO_2. While biodegradability of PHA *per se* has been long established, decisive factors influencing biodegradability of PHA, such as shape and thickness of polymer specimens, crystallinity, composition on the level of monomers, environmental factors (humidity, pH-value, temperature, UV-radiation), and surrounding microflora, though comprehensively studied and reviewed, are currently in status of scrutinization. While more studies are needed to further refine biodegradation timelines for various types of PHA, the fundamental fact that those types of PHA produced to date at reasonable quantities are biodegradable has already been established [33]. This variability in biodegradation of PHA follows all other natural materials including starch, cellulose, proteins, or chitin.

Biodegradability and compostability of PHA biopolyesters have been scrutinized under diverse environments and test conditions, i.e., soil, water, marine, simulated body fluids, activated sludge, as well as industrial and home composting. PHA producing companies have tested their PHA products according to corresponding standards by certain certification organizations to verify the claims of biodegradability and compostability of each product [34-37]. Standards and specifications have been developed by several authorities such as the European Committee for Standardization (EN), the American Society for Testing and Material (ASTM), the International Organization for Standardization (ISO), the British Standard Institution (BSI), etc.

Even the highly crystalline poly(3-hydroxybutyrate) (P(3HB)) homopolymers, the best studied type of PHA, is biodegradable and compostable in nature. The P(3HB) producer Biomer confirms that P(3HB) is "fully biodegradable" and compostable [38]. Previously, commercially available P(3HB) homopolyester from Imperial Chemical Industries (ICI), UK, even outperformed biodegradability of Novamont´s thermoplastic starch (TPS)-based composite material Mater-Bi. At different temperatures (28, 37, and 60°C), biodegradability of these materials was studied by burying thin sheets made of these materials in forest soil, sandy soil, activated sludge soil, and in farm soil. P(3HB) showed almost 100% degradation (average mass loss of five parallel samples: 98.9%) in activated sludge soil at 37°C after only 25 days, while degradation in farm soil (68.8% mass loss after 25 days) sandy soil (10% mass loss) and forest soil (7% mass loss) was considerably slower, again showing the high impact of the environment and microflora on PHA biodegradability. Remarkably, no complete degradation was achieved for TPS (72.1% degradation in activated sludge after 25 days at 60°C as highest degradability) at any condition studied [34]. Regarding PHA biodegradability in seawater, degradability of P(3HB), poly(3-hydroxybutyrate-*co*-3-hydroxyvalerate) (P(3HB-*co*-3HV)) and poly(3-hydroxybutyrate-*co*-4-hydroxybutyrate) (P(3HB-*co*-4HB)) films was studied in marine environment over one year. It was shown in this study that degradation rate highly depends on the temperature of seawater; moreover, mechanistically, all PHA samples were degraded via surface erosion, where depolymerase enzymes first attack the amorphous P(3HB) regions on the surface of a polymer sample; after that, crystalline P(3HB) regions are depolymerized [39].

8. PHA does not create recalcitrant microplastics

Typically, fossil plastics that are disposed as macro-plastic waste in landfills and in the environment, turn into micro- and nanoplastic particles ("secondary microplastic") sized between 1 nm and 5000 µm through erosion and abrasion. Such nano- and microplastic particles are also generated during the use of the fossil plastics by abrasion during diverse industrial processes, from vehicle tires, and even from shoe soles. [40, 41]. Nanoplastic and microplastic particles have the appropriate size to enter the food chain starting with planktons which ingest them. Plankton serves to nourish higher animals like fish, and the nanoplastic and microplastic particles finally end up on our table [42]. Plastic recycling, a strategy often promoted by legislative regulations, is a key source of microplastic formation. This was clearly shown by a study on mineral water sold in PET-bottles: Recycled bottles released tremendously more microplastic particles into the water than bottles made of virgin PET did [43].

At this point, it needs to be emphasized that small-sized PHA particles are not resistant in nature, they undergo biodegradation and do not leave any recalcitrant remnants behind. Hence, "secondary microplastics" consisting of PHA biopolymers simply do not exist. This matches the first principle of Green Chemistry, which postulates that the generation of precarious waste shall be avoided; it is "better to prevent than to cure" toxic waste like fossil microplastic, which is realized by switching to PHA [22]. The circularity of PHA biopolyesters is already being commercially exploited in many uses, including for replacing intentionally added fossil microplastics (“primary microplastic”) in cosmetics to offer UV protection (sun screen), and in skin peeling and scrubbing products [44]. Intentionally added, PHA microparticles that are added in shower gels, cosmetic peeling and scrubbing agents are released into the environment such as in sewage and waste water treatment plants, lakes, rivers and in the marine environment undergo biodegradation, unlike fossil microparticles, which remain recalcitrant. These persistent microplastics cause excessive microplastic loads in sewage sludge generated in wastewater treatment plants. Wastewater treatment plants filter up to 99% of the microplastic from domestic wastewater, but with PHA microparticles this would not be needed. The presence of PHA microparticles in sewage and wastewater treatment plants have an added benefit. PHA while undergoing biodegradation converts the high nitrogen content chemicals such as nitrates and nitrites present in sewage sludge into nitrogen gas, a process also called “denitrification”, thus aiding in improved sewage treatment [45]. Sewage sludge is an excellent organic fertilizer; however, currently they contain significant microplastics which end up in agricultural fields when used as fertilizer [46]. Replacing fossil microparticles with natural PHA microparticles would allow the PHA microparticles in sewage sludge to readily biodegrade in the sewage system or in the fields by the microflora present there.

Products containing natural PHA microparticles have already been commercialized, e.g., “Naturetics™” products by Nafigate Cooperation, which was launched in the Czech Republic in 2021 and are based on the Hydal technology to produce PHA from waste cooking oil [47]. In the most recent Microplastics Amendment (to Annex XVII) within the REACH Regulation, the European Union has again introduced language that restricts the use of biodegradable materials by tying the amendment to a definition of “Natural Materials” that the European Union (ECHA) created within the SUP Directive (EU 2019/904) [48]. This, even though under the new amendment any new intentionally added microplastic must meet specific biodegradability standards, where one of those positive controls (standards) is P(3HB), a type of PHA biopolymer. This type of action by the European Union is being picked up by other nations creating a patchwork of legislations that are negatively affecting investments, commercialization, and adoption of these very molecules that nature created and humans have mastered to make and break down in a circular way.

9. Industrial PHA production is analogous to PHA produced in nature

PHA is produced by microorganisms *in vivo*, as an energy source that the organism itself utilizes to survive in nature when it cannot obtain an external carbon source [49]. Hence PHA production can be compared to plants producing cellulose when they grow, although cellulose is a structural part of the plant, not a reserve compound like PHA. In industrial-scale manufacturers use these same microorganisms to produce PHA, the industrial process being referred to as “fermentation”. Many of our food products are manufactured using microorganisms such as grapes are fermented to produce wine, milk is fermented to produce cheese and yogurt, and cabbage transforms into sour kraut through fermentation. The PHA manufacturer also resorts to the same natural process of renewable carbon substrate uptake by their microorganism of choice to produce PHA; i.e., the microorganisms carry out their natural processes in adequate vessels or containers, called “bioreactors” or “fermenters”. On an industrial scale, the manufacturer produces PHA using microorganisms of their choice under controlled cultivation just like in the case of producing fermented food products. They do so in “bioreactors” or “fermenters” of different size and geometry, where cultivation conditions (temperature, pH-value, dissolved oxygen tension, actual substrate concentration, etc.) have been optimized through research and development to produce the PHA biopolymer economically [50]. Prime examples other similar processes are the aerobic propagation of baker’s yeast biomass for making bread [51], high-throughput vinegar manufacturing in well-aerated bioreactors (“acetators”) [52], production of penicillin antibiotics [53], or citric acid production by *Aspergillus niger* [54]. Specifically, penicillin and citric acid production processes are based on submerged microorganism cultivations in bioreactors similar to PHA production: In all three cases, two cultivation phases are observed: first, high densities of active biomass are produced under nutrient-rich conditions, which, provoked by limitation of a growth-essential nutrient, generates the desired bioproducts in a second cultivation phase as typical secondary metabolites. The two processes run sequentially sometimes in the same bioreactor. The only major difference is that penicillin and citric acid are excreted by the production strains as extracellular products [53, 54], while PHA is stored in cells intracellularly and needs to get released from the cells. Recovery of PHA from cells is increasing based on application of natural techniques, such as enzymatic disintegration of non-PHA biomass, or application of biogenic "green" extraction solvents [55]. Coming back to the 12 Principles of Green Chemistry, industrial PHA production matches Principle 8 (reduction of intermediate production stages and by-product formation, efficient and intensified processes [22]), Principle 11 (real time process monitoring by modern bioreactor equipment, in-line analytics and digitalization [22]) and Principle 12 (general risk mitigation by selection of safe raw material, safe process management, avoidance of explosion, fire, release of toxins [22]). Moreover, Principle 6 talks about energy efficiency in manufacturing processes: Processes shall be carried out under mild process conditions, such as at room temperature and atmospheric pressure [22]. This is exactly the case for biotechnological processes like PHA production, which take place at the biologically optimum, mild conditions for a given production strain. Finally, Principle 5 refers to the use of safe and non-toxic solvents in chemical processes [22]: The solvents used in bioreactors for PHA production is the safest solvent imaginable, namely water.

10. Products made of PHA are natural and biocompatible

PHA can be processed by established machineries to vendible biopolymeric products of high utility, e.g., via injection molding, blow molding, extrusion molding, compression molding, additive manufacturing (3D-printing), electrospinning, etc. [56]. During melt processing, PHA retains its basic chemical structure and is not modified such as with crosslinked or thermoset materials. [57] Hence, PHA undergoes no chemical modification from the time it is biosynthesized to its melt forming except that heat treatment of all polymers generally lowers their molecular weight. However, the end-of-life scenario of products made of biopolymers like PHA differ from their fossil competitors: According to Peng *et al.*, biodegradable polymers such as PHA are materials that can work for a limited time before degrading into "*readily discarded products*" (end products of mineralization) through a "*regulated procedure*" (enzymatic biodegradation) [58]. Hence, the circularity of products made of PHA is completed through their end-of-life options: They are biodegradable both under aerobic conditions (composting in both home- and industrial composting conditions) and without oxygen (biogas and anerobic digestion plants). Therefore, no persistent macro- and microplastic waste remains from leftovers of PHA-based materials, in contrast with fossil plastics which are persistent. Again, this meets the definitions of "Natural Polymers".

This pronounced propensity to biodegradability is illustrated by numerous biodegradation studies carried out worldwide in academic and industrial laboratories and at the laboratories of certification bodies. The most diverse types of PHA produced in small quantities at research laboratories as well as at on a large scale by various companies were already studied to assess their biodegradability. These industrially produced types of PHA include the homopolyester P(3HB) and the copolyesters of *R*-3-hydroxybutyrate (3HB) and the co-monomers *R*-3-hydroxyvalerate – 3HV, 4-hydroxybutyrate – 4HB, or *R*-3-hydroxyhexanoate – 3HHx). All these materials, although being of different crystallinity and processability, were shown to be readily biodegradable under most diverse test conditions, such as in various temperatures and in soil, fresh water, or marine environments [59]. Importantly, for several types of PHA, biodegradation, hence, the conversion of the carbon in the PHA into CO_2 outperformed the biodegradation performance of cellulose, the prototype natural reference material. Remarkably, compostability of PHA, hence, the disintegration into tiny fragments, which is essentially the recycling of carbon, does not only occur under industrial compositing conditions characterized by elevated temperature, as it is also the case for competing "bioplastics" like PLA; beyond that, PHA can even be composted at home, which allows for a simply and efficient management of polymeric household waste, when considering disposal of PHA-based packaging materials [60].

Ultimately, the high biocompatibility of PHA constitutes the final "brick in the wall" built by the rest of the arguments demonstrating that PHA is natural. This is shown by the huge number of eco- and bio-toxicity tests carried out with PHA, and their successful application as drug carriers, scaffold materials, nerve repair conduits, artificial blood vessels, surgical pins and wires, and stents. *In vivo* applications of PHA-based implants do not exert inflammation reactions, because PHA gets metabolized in the tissue to metabolites constitutively present in the human body, such as 3HB. This again is in contrast to PLA-based implants, which can cause inflammation due to lactic acid formation [13]. Most of all, it should be noted that resorbable surgical meshes made of poly(4HB) and P(3HB-*co*-4HB) are FDA approved, and already successfully commercialized by the company Tepha, Inc., owned by BD (Becton, Dickinson and Company) since 2021. These materials constitute macro-porous, monofilament, fully absorbable scaffolds with long term strength retention and defined absorption time [61]. The biocompatibility of PHA addresses Principle 4 of Green Chemistry, which is about the development of safe products, which, while exercising beneficial performance are of no or minimal toxicity [22].

PHA is demonstrably biodegradable and compostable when subjected to sustained enzymatic or microbial treatment, however, as a material in use they are extremely durable in numerous applications. Just like the cellulosic papyrus and old scrolls have survived many millennia when appropriately preserved, PHA articles would be long-lasting, until they experience sustained enzymatic or microbial action in nature, at waste water/sewage treatment plants or during industrial composting. This durable attribute of PHA makes them attractive as a replacement for many fossil plastics.

11. Definition of plastics and how PHA is also a plastic

Defining “plastics”: The terminus “plastic” stems from the Greek word πλαστικός (*plastikos*), which means "can be shaped or molded," while πλαστός (*plastos*) means "molded". And “plasticity” explicitly refers to the deformability of materials, which in this case refers to the polymers used in plastics manufacturing. Plasticity of materials enable molding, extrusion, or compression into specimens of various shapes: films, fibers, meshes, plates, bottles, tubes, containers, etc. Plasticity also has a technical definition in materials science, referring to the non-reversible change in form of solid substances, such as in the case of thermoplastics that are materials that can be shaped through the application of heat and pressure but retain their shape at ambient or lower temperatures below their melting point.

This definition of thermoplastics has been transported to popular culture and vocabulary from the scientific vocabulary over the last 50 years to refer to polymers made from fossil carbon. The reference has a rationale, almost all polymers that can be shaped like thermoplastics have originated from fossil plastics since the 1950’s. Therefore, “plastics” today generally refer to fossil derived plastics, however, if we refer to the original definition of plastics, meaning a material that can be shaped and reshaped using heat and pressure, PHA would fall in that definition.

What makes PHA “plastics”: PHA polymers are also thermoplastics as described above; they can be processed on conventional processing equipment in which fossil plastics are melt processed to manufacture marketable products.

This thermoplastic characteristic of PHA is an important distinction relative to other biopolymers such as cellulose. It is this characteristic of PHA that allows it to be processed like fossil plastics. PHA are a class of biopolymers with around 150 different building blocks allowing for numerous combinations of polymers with properties that cover a significant range of properties that mimic the properties of the top 7 best-selling fossil plastics including polyethylene, polypropylene and polyesters such as PET. They differ in their properties which are dependent on their chemical composition (homo-or copolyesters, type of hydroxy alkanoic acids making up the polyester): PHA made up of building blocks with 3 to 5 carbon atoms (short-chain-length PHA) are typically thermoplastics with amorphous and crystalline moieties (up to 70% crystallinity); possessing melting temperatures ranging from 100 °C up to 180 °C [62]. The best described PHA, the thermoplastic homopolyester P(3HB) is similar in its material properties to PP, has a good resistance to moisture and excellent gas and aroma barrier properties; they can be used in applications where high hardness or creep-resistance are needed. P(3HB) do not change their properties over a broad temperature range even when stored for several years. UV-resistance and mechanical stability of P(3HB) even outperforms many competing petrochemical thermoplastics [63]. Processability, impact strength and flexibility of PHA grades improve with the introduction of co-monomers such as 3-hydroxyvalerate, 4-hydroxybutyrate, or 3-hydroxyhexanoate. Such PHA co-polymers or "copolyesters" are already being commercially produced [64]. In contrast to short-chain-length PHA, those PHA consisting of building blocks with at least 6 carbon atoms (medium-chain-length PHA) are more amorphous, have low melting temperature, and display elastomeric to latex-like properties [62]. Industrial PHA biopolymers are already being used in packaging applications where food contact and contamination renders recycling difficult and such products are easily composted [64]. Once PHA volumes increase, they could also be recycled like fossil plastics, due to their thermoplastic nature, this making PHA recyclable, as well home- and industrially compostable [59].

11. Conclusions

This technical review presented the criteria which need to be considered in classifying polymeric materials as "natural materials" or "natural polymers". Nature uses renewable carbon as starting raw material and nature's workhorses (enzymes in living organisms) or "bio-synthesis" to produce natural materials or natural polymers. In addition, nature has the tools to unzip or "bio-degrade" them. PHA is one of those natural materials or natural polymers. They are products of natural living organisms produced from renewable carbon raw materials. Their chemical structure remains unchanged during or after melt processing them into diverse biocompatible consumables, that do no harm to living systems, and they do not disintegrate into persistent microparticles. Their beneficial end-of-life options, namely recyclability and biodegradation via composting or anaerobic digestion to biogas, are analogous to all other known biopolymers, such as carbohydrates, proteins, or nucleic acids. Table 1 summarizes these aspects, which are key for defining polymeric materials as "natural", while Table 2 brings PHA manufacturing in context with the 12 Principles of Green Chemistry, mentioned in individual sections of the text.

Table 1. Comparison of different natural/non-natural polymers.

Material:	**PHA**	**Carbohydrates (starch, cellulose, alginates, chitin, etc.)**	**Proteins**	**PLA**	**PCL**	**Bio-PE**	**Fossil plastics (PE, PP, PVC, PET, etc.)**
Criterion:							
Production depletes fossil resources	NO	NO	NO	NO	YES	NO	YES
Biobased (produced from renewable natural resources or via photosynthesis)	YES	YES	YES	YES	NO	YES	NO
Biosynthesized (polymerization taking place in nature by nature´s enzymatic toolbox)	YES	YES	YES	NO	NO	NO	NO
Biodegradable (degradation in nature by enzymes present in living organisms)	YES	YES	YES	YES	YES	NO	NO
Biocompatible (not harmful to the biosphere)	YES	YES	YES	NO	YES	NO	NO
(Natural) chemical structure after (bio)synthesis can be maintained during processing	YES	YES	YES	YES	YES	YES	YES
Formation of persistent microparticles	NO	NO	NO	NO	NO	YES	YES
Incineration generates surplus atmospheric CO_2, thus fueling global warming	NO	NO	NO	NO	YES	NO	YES
Summary: **Natural polymer or not??**	**YES**	**YES**	**YES**	**NO**	**NO**	**NO**	**NO**

PCL: poly(ε-caprolactone); PE: poly(ethene); PP: poly(propene); PVC: poly(vinyl chloride); PET: poly(ethylene terephthalate)

Table 2. PHA in the context of the 12 Principles of Green Chemistry [22, 65].

Principle	Description	Context to PHA?	Explanation of the context
Principle 1: Prevention	"It is better to prevent waste than to treat or clean up waste after it has been created."	YES	Neither PHA biosynthesis nor PHA biodegradation generates any waste, which needs to be treated, in contrast to production and disintegration of fossil plastics. This is especially true for formation of microplastic particles, which, in the case of fossil plastics, are recalcitrant and endanger the eco- and biosphere, while micro-sized PHA particles undergo biodegradation.
Principle 2: Atom Economy	"Synthetic methods should be designed to maximize incorporation of all materials used in the process into the final product."	NO	No! As natural, aerobic process, PHA biosynthesis creates intrinsically CO_2 as side product; however, generated CO_2 is embedded into the natural carbon cycle
Principle 3: Less Hazardous Chemical Syntheses	"Wherever practicable, synthetic methods should be designed to use and generate substances that possess little or no toxicity to human health and the environment."	YES	PHA biopolyesters are highly bio-compatible; they do not exert any risk to the environment or human health
Principle 4: Designing Safer Chemicals	"Chemical products should be designed to preserve efficacy of function while reducing toxicity."	YES	PHA biopolyesters can function as replacement for plastics, but, in contrast to fossil plastics, do not generate any toxic compounds when disposed or recycled.
Principle 5: Safer Solvents and Auxiliaries	"The use of auxiliary substances (e.g., solvents, separation agents, etc.) should be made unnecessary wherever possible and, innocuous when used."	YES	PHA biosynthesis occurs in aqueous phase, hence, water as the most sustainable and innocuous solvent is used. For PHA recovery from biomass, ecologically benign "green solvents" of natural origin can be used.
Principle 6: Design for Energy Efficiency	"Energy requirements should be recognized for their environmental and economic impacts and should be minimized. Synthetic methods should be conducted at ambient temperature and pressure."	YES	PHA biosynthesis *per se* typically occurs at room temperature and under ambient pressure conditions, which makes it a process of low energy requirement. Required energy supply is mainly due to upstream processing (e.g., sterilization of the bioreactor and media compounds), aerating and stirring of the bioreactor, and downstream processing. In case of using extremophilic organisms, energy for sterilization can even be avoided.
Principle 7: Use of Renewable Feedstocks	"A raw material or feedstock should be renewable rather than depleting whenever technically and economically practicable."	YES	PHA originate from renewable feedstocks.
Principle 8:	"Unnecessary derivatization (use of blocking groups,	YES	Derivatization is not needed during PHA biosynthesis due to the efficiently

Reduce Derivatives	protection/deprotection, temporary modification of physical/chemical processes) should be minimized or avoided if possible, because such steps require additional reagents and can generate waste."		coordinated enzymatic sequence in the intracellular biocatalytic cascade.
Principle 9: Catalysis	"Catalytic reagents (as selective as possible) are superior to stoichiometric reagents."	YES	A highly efficient enzymatic cascade, starting from the biocatalysts responsible for substrate catabolism towards acetyl-CoA, propionyl-CoA, etc. (glycolysis, KDPG pathway, β-oxidation, oxidative pyruvate decarboxylation, etc.), via enzymes generating the PHA building blocks (3-ketothiolase and reductase) until PHA synthases catalyze PHA biosynthesis. Activities of enzymes involved in the entire pathway from substrate to bioproduct (PHA) are highly specific, and efficiently coordinated.
Principle 10: Design for Degradation	"Chemical products should be designed so that at the end of their function they break down into innocuous degradation products and do not persist in the environment."	YES	Products made of PHA degrade naturally after use without harming the environment into exactly those innocuous compounds they derive from: water and CO_2 under aerobic conditions (composting), plus methane under anaerobic conditions (biogas plants).
Principle 11: Real-time analysis for Pollution Prevention	"Analytical methodologies need to be further developed to allow for real-time, in-process monitoring and control prior to the formation of hazardous substances."	YES	Modern bioreactor equipment used for PHA production encompasses online analytical tools to monitor temperature, pH-value, dissolved oxygen concentration, redox potential, foam formation (via conductivity sensors), substrate consumption (e.g., glucose sensors), biomass formation (via turbidity sensors), and the CO_2 level and remaining oxygen in exhaust gas. These parameters are processed by high-level digitalization, and control the process in real time by adjusting substrate supply, oxygen input, temperature, pH-value, etc.
Principle 12: Inherently Safer Chemistry for Accident Prevention	"Substances and the form of a substance used in a chemical process should be chosen to minimize the potential for chemical accidents, including releases, explosions, and fires."	YES	PHA production is based on safe, renewable feedstocks. No risk of fire or explosion exists during PHA production, and no toxins are released during the process.

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