

Microbial polymers, natural pesticides, and environmental protection from chemical pollutants

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ABSTRACT

Through this article, we offer you to highlight the important and vital role of microbial polymers by identifying them, their types, and their different uses in industry and agriculture, and how to extract them from microbial environments in different ways. The interest in this topic comes from the global concern based on preserving the environment and not using chemicals represented in pesticides and chemical fertilizers and their harmful effects on the environment, climate changes and global warming, and among them comes the interest in using natural materials produced by microbes, which have a good effect on the environment and at the same time disposal Security from harmful pests using environmentally safe natural pesticides.

1. Introduction

1.1. Polymers definition

Polymer, any of a class of natural or synthetic substances composed of very large molecules, called macromolecules, that are multiples of simpler chemical units called monomers. Polymers make up many of the materials in living organisms, including, for example, proteins, cellulose, and nucleic acids. Moreover, they constitute the basis of such minerals as diamond, quartz, and feldspar and such man-made materials as concrete, glass, paper, plastics, and rubbers.

The word polymer designates an unspecified number of monomer units. When the number of monomers is very large, the compound is sometimes called a high polymer. Polymers are not restricted to monomers of the same chemical composition or molecular weight and structure. Some natural polymers are composed of one kind of monomer. Most natural and synthetic polymers, however, are made up of two or more different types of monomers; such polymers are known as copolymers.

1.2. Types of polymers

There are several types of polymers. Among the main ones are: natural, synthetic, addition, condensation and rearrangement. For more detailed information about each, check out the descriptions below!

1.2.1 Natural polymers

Natural polymers are all those found in nature. Among the main examples are rubber, polysaccharides, starch, glycogen and proteins.

1.2.2 Synthetic polymers

Synthetic or artificial polymers are manufactured in the laboratory and generally have petroleum-derived ingredients. The best-known examples of this option are: polystyrene, methyl polymethacrylate (acrylic), polypropylene, polyethylene and polyvinyl chloride (PVC).

1.2.3 Addition polymers

This compound is obtained by successively adding monomers. As examples of these polymers, we have polysaccharides, which are formed by monomers of monosaccharides, and proteins, which are produced by amino acid monomers.

1.2.4 Condensing polymers

The condensing polymers are obtained by adding two different monomers with the elimination of a molecule of acid, alcohol or water during the polymerization process.

1.2.5 Rearrangement polymers

The rearrangement polymers are the result of the reaction between the monomers that undergo rearrangement and their chemical structures throughout polymerization. An example of this is polyurethane.

1.2.6 Biodegradable polymers

Finally, biodegradable polymers degrade into biomass, water and carbon dioxide as a result of the action of enzymes or living organisms. Under favorable conditions, they can be degraded in a few weeks.

1.3. History of bacterial polymers

The first discovery of a bacterial polymer dates back to the mid nineteenth century, when Louis Pasteur discovered dextran as a microbial product in wine¹²⁴. Van Tieghem¹²⁵ then identified the bacterium (*Leuconostoc mesenteroides*) that is responsible for dextran formation. This discovery was followed by the finding, in 1886, that cellulose is produced by bacteria¹²⁶. Shortly after the discovery of these exopolysaccharides, the first intracellular reserve polymers were discovered, such as the polyamide cyanophycin in cyanobacteria¹²⁷ and, 40 years later, the polyester polyhydroxybutyrate in *Bacillus megaterium*¹²⁸. Most other industrially and medically relevant bacterial polymers were found in the early to mid twentieth century, such as alginate¹²⁹, xanthan¹³⁰, poly-g-glutamate¹³¹ and polyphosphate¹³². Shortly after the discovery of the various biopolymers, the activities of their biosynthesis enzymes (either purified or in cell extracts) were described, and radioisotope-labelled precursors were also used to elucidate some details about the metabolic pathways for biopolymer formation^{133–140}.

Between 1970 and 2000, the advent of gene-cloning techniques and DNA-sequencing methods enabled the identification of biosynthesis genes, such as the cyanophycin synthetase gene (*cphA*)⁵⁸, and gene clusters, such as those found in the *Pseudomonas aeruginosa* genome^{76,77,141–144}. It is striking that around two decades after the identification of genes and gene clusters involved in the biosynthesis of well-established polymers (for example, cellulose and alginate) the functional assignment of essential genes is still lacking^{10,145}. Moreover, the reaction mechanisms of key enzymes, including various synthases, synthetases and polymerases, as well as the functions of co-polymerases and polymerase subunits and of proteins involved in polymer export and secretion (such as polysaccharide transporters, secretins and translocons) are still poorly understood.

1.4. Production of Microbial Polymers for Food Industry

Natural polymers can be classified as microbial-, plant-, and animal-derived based on their sources. High cost of downstream processes of plant and algal gums drives the polymer industry toward microbial derived polymers. Furthermore, microbial polysaccharides generally have higher molecular weights than plants, which affect their properties (Oner et al. 2016).

Current discoveries in microbial polymer biosynthesis have initiated new areas for medical and industrial applications. Novel molecular mechanisms and production processes have been discovered. These molecular mechanisms have formed

important tools for process engineering and applications, which are getting popular in pharmaceutical, agriculture, and particularly, in food industry.

Microbial polymers are long-chained, natural, biodegradable, biocompatible, nontoxic materials and are easy to handle compared to synthetic polymers. Xanthan gum, dextran, alginate, bacterial cellulose, gellan, curdlan, levan, pullulan, glycogen are important microbial polysaccharides that can be of bacterial or fungal origin (Vijayendra and Shamala 2014).

Generally, water-soluble polymers are used as suspending, gelling, and thickening agents in food industry. Polymers can also add characteristics such as sweetening, cryoprotection, antioxidant, anticaking, flavoring, antifoaming, chelating, stabilizing, preservative, and coating (Rosalam and England 2006).

1.4.1 Production Processes for Levan

Levan is an unusual fructan homopolysaccharide composed of β -(2,6)-fructofuranosyl residues with a terminal d-glucopyranosyl unit. Levan is synthesized from various bacteria such as *Acetobacter*, *Aerobacter*, *Azotobacter*, *Bacillus*, *Erwinia*, *Gluconobacter*, *Pseudomonas*, *Streptococcus*, *Zymomonas* and *Halomonas* sp. (Kazak Sarilmiser et al. 2015). Extremophilic and gram-negative *Halomonas* sp. was reported as first levan producer in 2009 by Poli et al. This system is very promising compared to mesophilic producers because it enables unsterile, low-cost production (Oner et al. 2016). *Halomonas* levan and its derivatives can be used as bioflocculating agent (Sam et al. 2011), adhesive nanostructured multilayer films (Costa et al. 2013), heparin-mimetic bioactive material (Erginer et al. 2016), and temperature sensitive hydrogels for drug-releasing systems with pNIPAA (Osman et al. 2017) among many others.

1.4.2 Production Processes of Pullulan

Pullulan is a natural, water soluble, linear homopolysaccharide composed out of maltotriose units. Maltose molecules are linked by α (1 \rightarrow 4) glycosidic bonds, while consecutive maltotriose units are linked by α (1 \rightarrow 6) glycosidic bonds. Pullulan was discovered in the late 1950s and isolated from a polymorphic fungus called *Aureobasidium pullulans*. It has been commercially produced since 1976. This homopolysaccharide has been used in many studies and applications involving cosmetics, pharmaceuticals, and food industries.

Pullulan is a biopolymer that is synthesized within the cell and then excreted on the outer layer after production. Like many biopolymers, the main disadvantage is a high production cost. Therefore, the research has been shifted to the use of for the production process (Prajapati et al. 2013; Wang et al. 2015; Wu et al. 2016).

Characteristics of pullulan are highly dependent on fermentation parameters, fungal strain, and its morphology. Even though many studies have been carried out to find a relationship between the morphology of the fungus and the characteristics of pullulan, no definitive evidence has been found yet. The content of the fermentation medium is crucial for the optimal polymer yield. Commercial fermentation media are composed of peptone, phosphate, and basal salts.

1.4.3 Production Processes for Alginate

Alginate is a polysaccharide composed of β -D-mannuronate and α -L-guluronate linked by 1,4 glycosidic bonds. Alginate was initially collected from brown seaweeds and has been commercially available since the beginning of the twentieth century. Alginate is produced by several different species of brown seaweed and two different species of bacteria; *Pseudomonas* and *Azotobacter*.

Microbial production has benefits over algal production such as low cost, ability of production in small scales and applied in different fields. As mentioned previously, bacterial alginate can be obtained using *Pseudomonas* and *Azotobacter*;

for commercial alginate production, human pathogen *Pseudomonas aeruginosa* and soil bacteria *Azotobacter vinelandii* are most widely preferred (Sabra and Zeng 2009; Hay et al. 2013; Ahmad et al. 2015).

Microbial production of alginate can be obtained through batch, fed-batch, and continuous fermentation. Epimerases lyases and acetylase enzymes are the important alginate modifying enzymes that were reported previously (Høidal et al. 2000).

1.4.4 Production Processes for Curdlan

Curdlan is a linear bacterial exopolysaccharide and classified as (1, 3) β -glucans. Curdlan is a special polysaccharide due to its rheological properties, solubility, and biomedical characteristics. Curdlan is named after its “curdle” competence when heated. Parameters such as pH, nitrogen, carbon, oxygen, and phosphate levels affect the production yields of curdlan. Curdlan is an extracellular polymer and biosynthesis occurs in three different steps. Substrate utilization, followed by intracellular metabolism of utilized substrate and finally excretion of polymer out of the cell membrane (Sutherland 1977, Zhang and Edgar 2014).

1.4.5 Production Processes for Gellan Gum

Gellan is a bacterial polysaccharide produced by *Sphingomonas elodea*. It belongs to a group of polysaccharides called sphingans, named by the bacteria from which it is produced. This biopolymer is an anionic, linear polysaccharide with high molecular weight composed out of D-glucose, L-rhamnose, and D-glucuronic acid in molar ratios of 2: 1: 1 (Tako 2015). Production of gellan begins with the isolation of the bacterium from the surface of a plant belonging to *Elodea* genus. Gellan production is accomplished via fermentation with immersion method. The medium used for incubation contains nitrogen, carbon sources, and some crucial trace minerals.

1.4.6 Production Processes of Xanthan Gum

Xanthan is a complex exopolysaccharide synthesized by plant-pathogenic bacterium *Xanthomonas campestris*. Exopolysaccharides produced by these pathogenic bacteria have a characteristic feature of protection against adverse environmental conditions such as drying, temperature oscillations, radiation, and adhesion (Luvielmo et al. 2016).

Xanthan gum is commonly applied as a thickening and stabilizing agent in different types of food and industrial products. The process of production of xanthan includes several steps. First, the chosen microbial is grown on solid media or in liquid media and used to inoculate the culture in large bioreactors. The mode of operation, medium composition, type of bioreactor, temperature, pH, and dissolved oxygen concentration influence both the microorganism growth and xanthan production. At the end of the fermentation, cells are usually removed via filtration or centrifugation operations from the culture broth that contains xanthan, bacterial cells, and numerous other chemicals. Next step is purification, where precipitation can also be included by using water-miscible nonsolvent, followed by the addition of certain salts and pH adjustments. The product is then mechanically dewatered and dried. The dried product is milled and packed into containers with a low permeability to water.

1.4.7 Production Processes of Dextran

Dextran is a group of homopolysaccharides composed of a linear chain of α -(1, 6) glycosidic linkages that may form branches on the main chain. It was first observed by Louis Pasteur, but this biopolymer's potential in food industry was not investigated until the 1950s. Dextran is one of the oldest bacterial polysaccharides with a multitude of functions.

Dextran is an exopolysaccharide synthesized by *Streptococcus*, *Lactobacillus*, and some *Weissella* species and is very sensitive to environmental conditions like substrate concentration, pH, temperature, and salinity. Because different strains of bacteria belonging to the same species can produce dextran with varying structures, it is, in theory, possible to produce dextran

according to specific needs. For example, keeping the substrate levels low tends to give dextran a higher molecular weight (Das and Goyal 2014; Zannini et al. 2016; Baruah et al. 2017).

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