Introduction

Vinyl polymers are derived from vinyl monomers containing double bonds C=C. The main chain of polymer is made from carbon atoms containing single bonds, to which other substituents can be added such as chlorine, fluorine or hydroxyl group. Some polyolefins, polyvinyl acetates, polycrylates and others [1, 2] belong to vinyl polymers. Due to their high molecular mass and considerable hydrophobic properties, they are classified as stable polymers (e.g. biodegradation time of polyethylene is assumed to be ca. 300 years [3]).

In the environment, synthetic polymers undergo degradation influenced by abiotic agents and microorganisms [4]. Biodegradation of materials is impeded as microorganisms have not formed suitable mechanisms of their degradation. Nowadays, environmental pollution caused by vinyl polymers originated, inter alia, from packaging poses a serious problem. Therefore, a great pressure is put under the acquaintance with biodegradation pathways of polymers for which this process is possible [5].

Poly(vinyl alcohol) is acknowledged to be one of few vinyl polymers that can have higher biodegradation rate. This is possible owing to the presence of hydroxyl groups which condition hydrophilic nature of this material [6].

Material biodegradation in various environments

Worldwide research into poly(vinyl alcohol) degradation in the environment was initiated for its application in pulp and paper as well as textiles industry which has resulted in generating significant quantities of wastewater containing PVA [7]. Poly(vinyl alcohol) degradation was studied in, e.g. compost, soil, water, under aerobic and anaerobic conditions.

Experiments conducted by Chellini et al. in compost from solid municipal waste [8] demonstrated that biodegradation degree of PVA foil containing 12% of acetate groups was 7% during 48 days. In other studies [9], the same polymer was incubated for 300 days in the presence of compost extract as the source of microorganisms. It was shown that its 25% underwent biodegradation, whereas PVA with 2% content of acetate group was degraded only in 15%.

The incubation of PVA foil in soil conducted by Chellini et al. [8] demonstrated that 8-9% of this material undergoes biodegradation within 74 days, and the level of this process is not affected by such factors as polymer concentration or its physical state. Similar results were obtained by Solaro et al. [10] during 120 days of PVA degradation with 88% hydrolysis degree. The studies on the susceptibility of PVA without acetate groups to degradation caused by soil microorganisms were carried out by Sawada [11]. For this purpose, foil samples were dug in 18 places representative of various types of soil and ambient conditions. After two years of samples incubation, in spite of high microbiological activity of soil, no traces of material colonization by microorganisms was found and only slight decrement of foil mass (less than 10%) was observed. Contrary to PVA, other polymers such as PHB/HV or PCL were intensively degraded under the same conditions [12].

Also, the studies into degradation of poly(vinyl alcohol) in aquatic environment are conducted. The considerable degree of material biodegradation was shown only in the presence of previously acclimatized microorganisms [13]. Relatively low level of PVA biodegradation (13% after 21-day period of incubation) was observed under aerobic conditions in liquid cultures inoculated by sludge taken from municipal wastewater. However, biodegradation degree of PVA performed in the presence of sludge taken from wastewater generated in a paper-mill reached values comparable with the ones for cellulose. The authors suggest that this can be caused by high selective pressure resulting from the presence of significant quantities of poly(vinyl alcohol) in wastewater from paper factories. This pressure allows microorganisms present in such wastewater to gain abilities required for degradation of this polymer [8].

The studies on PVA biodegradation under anaerobic conditions were performed with the application of microorganisms from river sludge and municipal wastewater. Foil samples with average (14 kDa) and low (2.2 kDa) molecular mass were analysed. The important impact of polymer molecular mass on its degradation degree by microorganisms was reported. Microorganisms from river sludge caused a 75% decrement of mass of polymer with lower molecular mass, and a 50-60% decrement of mass for samples characterized with higher molecular mass in comparable incubation periods. Slightly lower biodegradation degrees of the polymer were observed for microorganisms originating from municipal wastewater [14].

Mechanisms of polymer biodegradation

Phytopathogenic fungi Fusarium lini were the first known microorganisms capable of PVA degradation. They synthesised an exoenzyme, previously known as “dehydratase”, which action resulted in the release of carbon dioxide and water [15]. First microorganisms capable of degrading this polymer as the only source of carbon were isolated from soil samples and identified as bacterial strains called Pseudomonas [16]. During further studies also other bacteria, such as Alcaligenes and Bacillus, were isolated from environments polluted with PVA [17]. They are able to degrade this material as well.

The studies conducted by Watanabe et al. [18] and Morita and Watanabe [19] demonstrated that Pseudomonas strain releases two exoenzymes which degrade poly(vinyl alcohol). They were identified on the basis of hydrogen peroxide production during the reaction and their ability to degrade some low molecular mass secondary alcohols as secondary alcohol oxidase [EC 1.3.18 – SAO) and β-diketone hydrolase [EC 3.1.1.7 – BDH]. SAO is highly specific to 1,3 – hydroxyl groups and catalyses oxidation of polymer molecules to adequate β-diketones. Then, they are attacked by a specific β-diketone hydrolase which requires β-diketone chain of length at least equal to 6 carbon atoms for its complete activity.

This reaction results in products terminated with carboxyl and methyl groups. Picture 1 presents the biodegradation mechanism through SAO and BDH [17].

Sakai et al. [20] proposed a biodegradation mechanism of poly(vinyl alcohol) containing acetate groups by Pseudomonas vesicularis PD strain. It is based on the activity of SAO and BDH exoenzymes. However, it was suggested that their degradation products containing acetate groups can be uptaken and assimilated by bacterial cells. Specific esterase present both in cytoplasm and cytoplasmic membrane turned out to be able to hydrolyse acetate groups. Its highest activity was observed for poly(vinyl alcohol) molecules with low molecular mass and high content of acetate groups. Deacetylation cata-
lysed by esterase leads to the formation of such products as acetic acid or hydroxy fatty acids. Then, they are metabolised within Krebs cycle and β-oxidation. Picture 2 presents a biodegradation mechanism of poly(vinyl alcohol) containing acetate groups.

Matsumura et al. [21] were investigating the pathway of poly(vinyl alcohol) degradation by specific dehydrogenase of PVA observed in Alcaligenes faecalis KK314 strain isolated from river water samples. This enzyme catalyses the formation of β-hydroxyketone groups along polymer chains, without their further oxidation to β-diketones. They also isolated an enzyme responsible for dissection of formed β-hydroxyketones. Comparison of molecular masses with gel filtration profiles of both enzymes demonstrated that examined proteins are the only enzyme which has two active places: dehydrogenase and aldolase, whereby CaCl2 and Pyrroloquinoline Quinone (PQQ) as coenzyme are required to conduct dehydrogenation reaction. Picture 3 presents a mechanism of poly(vinyl alcohol) biodegradation by PVA dehydrogenase with the presence of Alcaligenes faecalis KK314.

Hatanaka et al. [22] isolated another PVA dehydrogenase depending on PQQ from Pseudomonas sp. 113P3 culture. The lack of enzyme activity in supernatant indicated that PVA had to be uptaken inside the cell, and then it was degraded by specific intracellular dehydrogenase. This enzyme catalysed the formation of β-diketone groups along polymer chains, which were hydrolysed to low molecular weight PVA by a specific hydrolase.

Shimao et al. [23] were the first who observed poly(vinyl alcohol) biodegradation in mixed cultures of some strains capable of PVA assimilation. All symbiotic cultures showed the presence of a strain producing an enzyme which degrades PVA and a strain which provides an essential cofactor [24]. The first cannot grow in axenic culture, but after adding supernatant of second strain culture into this culture, it acquires the ability to grow.

A pair of symbionts was made of, inter alia, strains of Pseudomonas putida VM15A and Pseudomonas sp. VM15C. P. putida VM15A is a strain which provides a cofactor, namely PQQ, whereas Pseudomonas sp. VM15C strain has two constitutive enzymes combined with cytoplasmic membrane which degrade polymer. PVA oxidase belongs to such enzymes. Its activity, contrary to second enzyme - PVA dehydrogenase, is not affected by Pyrroloquinoline Quinone provided by P. putida VM15A. Moreover, cytochrome reduction occurs during poly(vinyl alcohol) oxidation. This could indicate that polymer oxidation is related to a cell electron transport chain. It was suggested that PVA oxidase can be the subunit of oligomeric enzyme, to which PVA dehydrogenase belongs. PQQ can participate in the transfer of electrons from subunit oxidising PVA to other electron acceptor than oxygen as PVA dehydrogenase depending on PQQ does not consume oxygen during its activity. This can also explain the importance of PVA dehydrogenase as the primary electron acceptor during PVA metabolism under anaerobic conditions [25]. Picture 4 presents the proposed mechanism of PVA biodegradation in symbiotic culture of microorganisms.

From phylogenetic point of view, poly(vinyl alcohol) and other synthetic polymers appeared in the environment at relatively late stage of organisms’ evolution. Therefore, the majority of microorganisms are not able to consume PVA as the only source of carbon. However, it is possible that enzymes degrading PVA, such as PVA dehydrogenase Pseudomonas sp. VM15C, have appeared due to relatively recent evolutionary processes. PVA dehydrogenase seems to be functionally unique because its function considerably differs from functions of other quinoprotein dehydrogenases.
Mori et al. [26] were investigating the capacity for poly(vinyl alcohol) degradation in mixed culture. It was stated that Bacillus megaterium BXI bacteria demonstrated limited growth and PVA degradation in axenic culture, while significant growth in PVA assimilation was observed in the presence of other gram-positive bacteria. These bacteria did not degrade PVA which implied that poly(vinyl alcohol) degradation was the result of co-metabolism of both strains, however, it was not related to PQQ release.

There are few reports on PVA biodegradation by fungi. Basidium fungi Phanerochaete chrysosporium, commonly present in the environment, was the organism present during the majority of experiments. It is capable of degrading naturally occurring lignin, and also such xenobiotics as polycyclic hydrocarbons (PAHs) or synthetic polymers, such as polyethylene or nylon [27-29]. According to Mejia’s experiments [30], one of its lignolytic enzymes - lignin peroxidase (LiP) degrades PVA by forming carbonyl groups and double bonds along a polymer chain.

Biodegradation mechanism was proposed, in which epoxide is formed at the first stage. Then, double bonds were formed by elimination of water molecules. Formed free-radical intermediate product with produced benzaldehyde can be decomposed in next stages. Figure 5 presents the mechanism of PVA biodegradation by Phanerochaete chrysosporium.

Larking et al. [31] were investigating biodegradation with the part of Pycnoporus cinnabarinus which decays hard-degradable industrial dyes. They stated that chemical oxidation of PVA or the presence of co-metabolite – glucose, accelerates material degradation. Laccase - another lignolytic enzyme, was the only enzyme involved in this process.

Experiments conducted by Nishikawa and Hasegawa resulted in identification of Saccharomyces, Lipomyces and Rhodotorula yeasts which degrade and assimilate poly(vinyl alcohol). For PVA containing 12% of acetate groups, this process takes place regardless of acetic acid and its salts or esters presence in the solution. PVA is also biodegradable in the presence of Endomycetes, Zygosaccharomyces, Pichia and Nadsonia yeasts, but only if acetic acid derivatives appear as substrates. Besides these species, biodegradation of poly(vinyl alcohol) was not observed in axenic culture of yeasts [7].

**PVA application**

Nowadays, poly(vinyl alcohol) is the most commonly produced polymer soluble in water [32]. Poly(vinyl alcohol) foils are used in agriculture to deliver fertilizers, pesticides, herbicides and fungicides, as well as to coat seeds. Such prepared seeds are ready for germination only under adequate temperature and humidity conditions [33].

For environmental protection, poly(vinyl alcohol) can be used as a carrier of microorganisms. Gel manufactured by Kuraray company, composed mainly of PVA, has a porous, reticulate structure that can trap and carry microorganisms. A single gel bead with a diameter of 4mm can hold up to 1 billion microorganisms. PVA gel is used as an alternative to the activated sludge method in the treatment of wastewater [34].

For its biocompatibility, toxicity and lack of carcinogenic properties as well as due to excellent chemical properties, PVA is applied in modern technologies for use in medicine and pharmacy [35, 36]. The systems of controlled release of drugs are produced from this material. They can have the form of soft contact lenses, active dressing or tablets.

It is also used to manufacture surgery threads, implants, scaffolds for cells culture and artificial organs. Due to its hydrophilicity, PVA is most commonly used in combination with other polymers, such as poly(lactic acid) (PLA), poly(glycolic acid) (PGA) or poly(-caprolactone) (PCL) [36-38].

We can distinguish four types of systems of controlled release of active substance: controlled bulging, polymer dissolution, crystals dissolution and modulated. The system of controlled bulging is based on drug release caused by bulging of crosslinked polymers affected by water. The quantity of released active substance is controlled by the size of bulged material. Specified doses of drug are released in the system of polymer controlled dissolution due to gradual dissolution of PVA. Then, this material is excreted by an organism what eliminates the need to remove polymer implants. The system of controlled dissolution of crystals is based on the change of polymer phase from crystalline to amorphous phase. No necessity to use toxic cross-linkers is its advantage. In the modulated system, the action of external stimulus, such as put electric field, causes the release of a therapeutic substance. Also, self-modulated systems are possible. They are activated after the action of physiological or environmental stimulus in the surroundings of polymer [39].

For example, painkillers [36], antibiotics [40], hormones [41], anticancer drugs [38], vitamins [42] and other substances can be delivered through the systems of controlled release made from PVA.

In tissue engineering, PVA is applied as scaffolds for tissues regeneration, particularly for cartilaginous tissue regeneration. The regeneration process of these tissues is ceased at some moment because of more difficult access to oxygen and nutritive substances. This can be prevented by forming new blood vessels in a place of tissue regeneration. Matrices made from PVA are used for this purpose. They contain gelatin beads with basic Fibroblast Growth Factor (bFGF). After subcutaneous implantation to the target place, this factor is gradually released, leading to the growth of new blood vessels.

Also, the research into using poly(vinyl alcohol) for obtaining hybrid internal organs to replace damaged ones has been in progress. The prototype of hybrid pancreas has been constructed from live animal cells and a membrane protecting these cells against destruction by a receiver immune system. This membrane, made from PVA, is biocompatible with the ambient environment, not capsulated by tissues and is permeable for oxygen, glucose and insulin [39, 43].
PVA is also used in embolization procedure as the agent which blocks blood flow in blood vessels [44, 45].

Bibliography


2. Information from Vinyl Institute http://www.vinylinfo.org/UsesofVinyl.aspx


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SUNNY CHEMISTRY - the initiative of the Program Council of the monthly magazine CHEMIK and the Polish Association of Chemical Engineers, is supposed to change awareness and formulate a new way of thinking about chemistry in the society and science, technique and economy.

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