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Biodegradation of Natural Rubber by Fungi and Bacteria

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ABSTRACT

Environmental pollution is currently one of the major problems that are threatening biodiversity, ecosystems, and human health around the world. Natural rubber, which is one of the most significant polymers due to its variety of uses, has now become a serious environmental concern. Rubber waste management poses one of the greatest problems because it is extremely resilient and persists in the environment despite several mitigation efforts. Biodegradation is an eco-friendly alternative to conventional disposal methods and has gained tremendous interest in recent years. Several studies on rubber biodegradation utilizing fungi and bacteria have been reported. However, except for a few studies on technical applications, the majority of research on these microbes has focused on the fundamentals of rubber biodegradation. The challenge with biodegradation as a potential solution for rubber waste management is that we have limited mechanistic insight into rubber biodegradation, and the complicated composition of rubber products inhibits cell growth and activity of microbes. Thus it becomes important to fully comprehend the mechanism of rubber biodegradation and continue the search for new microbial strains so that the acquired knowledge can be utilized to develop a biodegradation process suitable for scale-up. In this short review, rubber degradation using fungi and bacteria is highlighted.

INTRODUCTION

Rubber, due to its exceptional qualities like flexibility, longevity, and a wide range of uses, has become one of the most essential commodities in today's world. As per the Malaysian Rubber Council, world production of rubber increased by 3.3% to 7.0 million tonnes in the first quarter of 2021, compared to 6.8 million tonnes in the same period of 2020. Similarly, world consumption of rubber grew by 14.8% to 7.4 million tonnes in the first quarter of 2021, compared to 6.5 million tonnes in the same period of 2020 (Malaysian Rubber Council 2021). With the increase in consumption, wastage of rubber in the form of used rubber products especially scrap tires has increased. Rubber waste management is an extremely challenging task for Municipal Corporation. The biggest challenge comes in the form of recycling. Rubber is highly durable and inherently non-biodegradable, leaving them stagnant in landfills for hundreds of years, occupying valuable space. Many cities have scrap tire stockpiles, which cause public health, environmental, and aesthetic issues (Yehia 2004).

Many plants, primarily from the Euphorbiaceae, Compositae, Moraceae, Eucommiaceae, Celastraceae and Apocynaceae families, produce rubber by enzymatic activities. Chemically NR is a polyisoprene polymer. There are mainly two types of polyisoprenoids based on isomerism, the cis isomer natural rubber (NR) [poly(*cis*-1,4-isoprene)] and the trans isomer gutta-percha (GP) [poly(*trans*-1,4-isoprene)] (Fig. 1).

Natural Rubber can be obtained from plants such as *Hevea brasiliensis* (rubber tree), *Parthenium argentatum* (guayule), *Taraxacum kok-saghyz* (Russian dandelion), *Dyera costulata* (jelutong). Gutta-percha on the other hand can be obtained from *Palaquium gutta* (gutta-percha), *Manikara zapota* (chico), *Eucommia ulmoides* (Tochu), *Euonymus europaeus* (spindle tree), *Mimusops balata* (balata) (Yikmis & Steinbüchel 2012). For commercial uses, NR is produced from the latex of *Hevea brasiliensis*, a South American plant endemic to the Amazon Valley. The first scientific or commercial interest in rubber was demonstrated by Frenchman Charles Marie de Condamine, who submitted a report to the

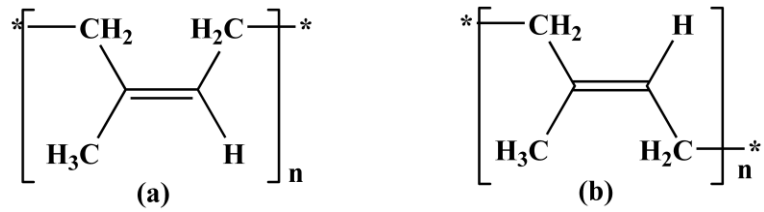

Fig. 1: Chemical structures of (a) Natural rubber (NR) [poly(cis-1,4-isoprene)] (b) Gutta-percha (GP) rubber [poly(trans-1,4-isoprene)].

Paris Academy of Sciences in 1745 after visiting Ecuador and observing the local use of *Hevea* latex. Priestly, an English scientist, named the raw material, 'Rubber' in 1770 after noticing that it can remove pencil markings. In 1839, Goodyear discovered vulcanization, ushering in a golden age of the rubber industry (Hurley 1981). The discovery of synthetic polyisoprene by German scientist Fritz Hofmann in 1909 paved the way for large-scale production of synthetic poly (cis-1,4-isoprene) with a molecular structure similar to NR (Yikmis & Steinbüchel 2012).

The latex (colloid liquid in the aqueous phase) of these plants is converted to rubber by coagulation (chemically and electrically) and drying. Rubber at this stage is a soft, sticky, thermoplastic material with low tensile strength and elasticity. These properties have a straightforward molecular structural basis. A variety of polymeric chains of varying lengths make up the material. Most notably no crosslinking is present. As a result, while being known for millennia, rubber in this form did not find any significant application until the discovery of vulcanization (Kumar & Nijasure 1997). During vulcanization (Fig. 2) rubber is heated in the presence of sulfur, resulting in the three-dimensional cross-linking of chain rubber molecules (polyisoprene) bonded to each other by sulfur atoms. Other compounds such as hydrogen sulfide, sulfur monochloride, benzoyl chloride, etc. can also be applied for vulcanization. This process improves the elasticity, tensile strength, resilience, and water-absorbing capacity of rubber. Moreover, vulcanized rubber is resistant to oxidation, abrasion, wear, and tear. It also has a wide useful range of temperatures.

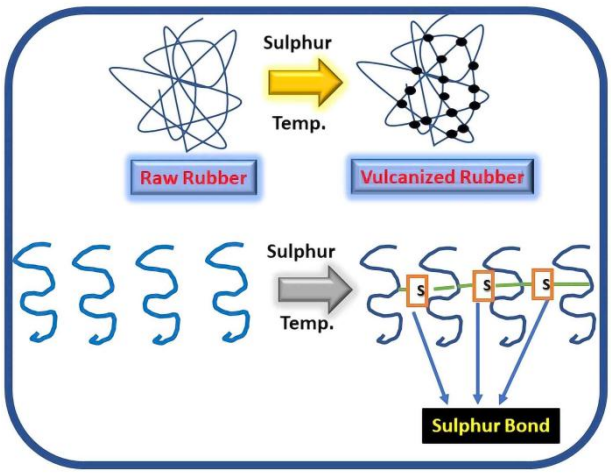

Fig. 2: Mechanism of vulcanization.

As discussed earlier, the major problem with rubber products is their disposal after use. One way to counter this problem is recycling. However, unlike polythene, it cannot be simply melted and reshaped again into the product due to cross-linking (formed during vulcanization) (Nayanashree & Thippeswamy 2013). The rubber wastes such as tires are conventionally buried in landfills or are held in stockpiles. However, it does nothing to help with the disposal issues that come with rubber waste as it is not biodegradable in landfills and remains immobile in stockpiles leading to several environmental problems. Rubber waste especially tires can also be thermally degraded at around 800°C to produce Tar Pyrolysis Oil (TPO), which has diesel-like properties. This process, in addition to being complex, costly, and labor-intensive, has the potential to pollute the air and water due to poor process management. Tyre abrasion has been identified as one of the primary sources of microplastics which subsequently enter the food chain and cause biological contamination. Left-over tire crumbs can be utilized to generate asphalt for roads, playground rubber flooring, sports or bicycle tracks, or to alter the structural qualities of concrete. However, in all of these mitigation methods, the rubber remains in the environment and is degraded very slowly (Basik et al. 2021). In recent times, microbial bioremediation of wastes has gained tremendous interest. Bioremediation using bacteria and fungi has found its way into many diverse applications such as treatment of antibiotics present in water (Singh et al. 2017), textile azo dye decolorization and detoxification (Karnwal 2019), oil cleaning from water bodies (Rahul et al. 2018), removal of pesticide (Sidhu et al. 2019), herbicides (Digvijaya et al. 2017, Mukherjee et al. 2018), explosive materials (Gorontzy et al. 1994), toxic heavy metals (Gehlot & Singh 2018, Karnwal 2018, Kaur et al. 2018 & Mishra et al. 2016) from soil and water, etc. Biodegradation is an eco-friendly

alternative to traditional disposal methods in which microorganisms break down complex organic compounds in waste products into simpler compounds and eventually into the water and either carbon dioxide (aerobic) or methane (anaerobic). While microorganisms can break down the majority of natural substances, they frequently lack the enzymes required to degrade most manmade compounds, including synthetic rubbers. Compounds with a molecular structure that microorganisms have not been exposed to (for example, synthetic rubbers and polymers) are typically resistant to biodegradation. They ultimately endanger the ecosystems by contaminating and accumulating in the environment.

During microbial degradation, rubber polymers are mineralized and redistributed through the Elemental cycles (Enoki et al. 2003, Cui et al. 2005). The biodegradation process progresses through four stages: bio-deterioration, bio-fragmentation, assimilation, and mineralization. The chemical and physical properties of the polymer are altered during the first stage, while enzymatic cleavage permits the polymer to be broken down during the second. The assimilation is the uptake of molecules by microbes; and finally, the mineralization phase, which is characterized by the emission of CO_2 and H_2O in aerobic settings and CO_2, CH_4, and H_2O in anaerobic conditions (Pathak & Navneet 2017).

Microbial rubber degradation has been the subject of a number of studies in recent years. Both fungi and bacteria have been shown to degrade rubber, however, the process is slow (Onyeagoro et al. 2012). NR is primarily composed of hydrocarbons, with minor amounts of lipids, sugar, resins, proteins, and minerals. The growth of microorganisms is aided by these organic contaminants. Microbial processes have advantages over chemical and physical processes as they are non-toxic and do not produce any hazardous substance. However, several challenges remain, the most significant of which being microorganism sensitivity to numerous chemicals, including rubber additives, which are used to improve tire durability and operation across a wide temperature range (Yikmis & Steinbüchel 2012). This brief review outlines the microbial degradation of rubber by fungi and bacteria.

DEGRADATION OF NATURAL RUBBER BY FUNGI

De Vries was the first to explore the biodegradation of rubber by fungi. The biodegradation of rubber was studied using several *Penicillium* and *Aspergillus* strains in a 10% (w/v) aq. NaCl liquid medium with natural rubber as the substrate. After a 19-month to 5-year incubation period, there was a 6% rise in biomass and a 15.5–30.9 percent drop in the weight of the rubber material (Shah 2020). Schade reported the growth of fungi *Monascus purpureus* and *Monascus rubber* on purified natural rubber substrate (Schade 1937). After a decade Kalinenko (1938) identified fungal strains from *Aspergillus* and *Penicillium* as rubber degraders (Kalinenko 1938). In soil burial tests conducted on NR vulcanized sheets of specific composition, Kwiatkowska et al. (1980) discovered considerable weight losses after 91 days, equivalent to 40% of the initial weight. They identified *Fusarium solani* fungal strain on the rubber's surface and held it responsible for the observed weight loss by degradation (Kwiatkowska et al. 1980). Borel et al. (1982) found that *Fusarium solani* degrades rubber faster than other fungi utilized in his studies, such as *Paecilomyces lilacinus*, *Phoma eupyrena*, and *Cladosporium cladosporioides* (Borel et al. 1982). A fungal strain, *Penicillium variable* was isolated by Williams from a damaged NR sample following soil burial. Using solution viscosity measurements, Williams discovered a 15% decrease in the molecular weight of polyisoprene after 70 days due to breakdown by the *Penicillium variable* (Williams 1982). Atagana et al. (1999) in their study on fungal degradation of waste from the rubber processing industry, demonstrate that *Mucor* species have the potential to metabolize the aqueous fraction obtained during coagulation of latex thereby lowering BOD in a reasonable manner (Atagana et al. 1999). Stevenson et al. (2008) proposed a multistage tire rubber recycling process that included using the fungus *Recinicium bicolor* in the first stage of detoxification to remove pollutants that inhibit microbial growth (Stevenson et al. 2008). Nayanashree et al. isolated two fungal strains of *Aspergillus niger* and *Penicillium* from rubber pieces that had previously been dumped in the soil. Both these strains were found to be effective in rubber degradation with *Aspergillus niger* showing 28.3% degradation, while *Pencillium* sp. showing a 25.9% degradation in two months (Nayanashree & Thippeswamy 2013). Mohamed et al. studied the ability of *Penicillium chrysogenum* and *Aspergillus niger* to metabolize and degrade rubber latex obtained from *Calotropis procera* by analyzing the rise in fungal protein content, reduction in molecular weight and intrinsic viscosity of latex and growth of these stains on rubber surface (Mohamed et al. 2017). Singh et al. (2017) in their study found that fungal species *Aspergillus niger* and *Phlebia radiate* can degrade NR, with *Aspergillus niger* having the highest degrading potential, accounting for 27.27% on the scale of NR weight loss (Singh et al. 2017). In his study on the biodegradation of unvulcanized natural rubber by microorganisms, Bosco et al. (2018) discovered that filamentous fungus (*Alternaria alternata*) isolated from an NR surface and yeast (*Rhodotorula mucilaginosa*) isolated from NR liquid culture were both effective in promoting NR biodegradation (Bosco et al. 2018). Recently genome sequencing of a fungal species *Rigidoporus microporous*

was carried out by Oghenekaro et al. (2020) This fungus is known to cause white root rot disease in the rubber tree and can grow on latex. In the genome sequencing, however, no homologs of bacterial proteins involved in latex degradation were found thus indicating that not all latex-tolerant strains have rubber-degrading genes (Oghenekaro et al. 2020, Basik et al. 2021). The role of fungus in rubber deterioration is mostly descriptive, indicating solely its potential to degrade NR. Table 1 summarizes the list of fungi mentioned in this review.

DEGRADATION OF NATURAL RUBBER BY BACTERIA

Many studies have been carried out in recent years to identify and characterize the efficient rubber-degrading bacteria, as well as to understand the metabolic basis for natural rubber breakdown. Until recently, many bacterial strains have been discovered that can consume rubber as their only source of carbon and energy (Shah et al. 2013). These bacteria can be categorized into two groups based on their differing methods of rubber degradation. The Members of the first group (Group B) produce translucent halos when grown on solid media containing latex particles, indicating the excretion of a polyisoprene-cleaving enzyme (Fig. 3a), while members of the second group (Group A) do not form translucent halos or develop on latex plates, instead require direct contact with the rubber and grow adhesively on its surface in liquid cultures using it as the source of carbon and energy (Fig. 3b) (Linos et al. 2000).

The most effective Group B members include *Streptomyces*, and *Micromonospora*, whereas CNM (*Corynebacterium, Nocardia, Mycobacterium*) are the most potent rubber degraders from Group A (Shah 2020). The first publication on microbial degradation of NR was done by Akio et al. where they used *Nocardia* sp. strain 835A to degrade NR vulcanizates (Tsuchii et al. 1985). The majority of known NR degraders are Gram-positive bacteria, which have been widely reported, whereas, only a few Gram-negative NR-degrading bacteria have been discovered and described in the scientific literature. *Xanthomonas sp*. strain 35Y (Tsuchii & Takeda 1990) (now reclassified as *Steroidobacter cummioxidans* strain 35Y (Sharma et al. 2018)) is the first Gram-negative bacteria known to degrade rubber. Table 2 summarizes the list of NR degrading bacteria reported in the literature until now.

Research has confirmed that there are three enzymes responsible for the degradation of natural rubber; Latex clearing protein (Lcp) which was first identified and characterized in *Streptomyces* sp. strain K30 (Rose et al. 2005) and Rubber oxygenase (RoxA and RoxB) first found in *Xanthomonas* sp.

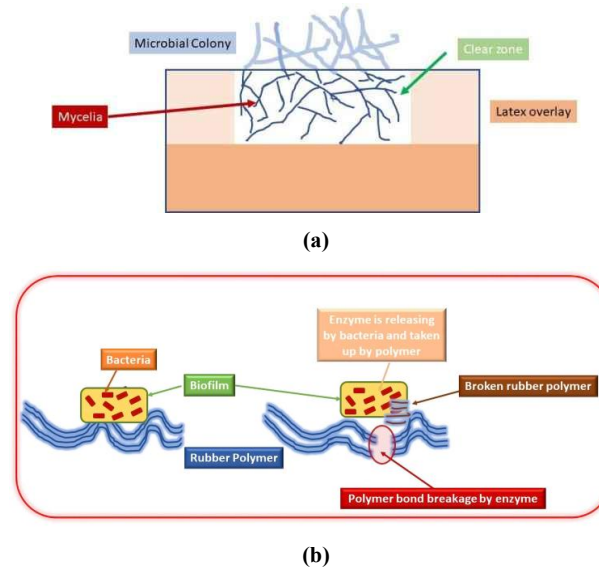

Fig. 3: NR degradation strategy of (a) Group B bacteria and (b) Group A bacteria. (Adapted from Basik et al. 2021).

Table 1: List of NR degrading fungal strains mentioned in this review.

Fungal Strain	References
Monascus rubber, Monascus purpureus	(Schade 1937)
Fusarium solani	(Kwiatkowska 1980)
Paecilomyces lilacinus, *Phoma eupyrena*, *Cladosporium cladosporioides*	(Borel et al. 1982)
Penicillium variable	(Williams 1982)
Mucor species	(Atagana et al. 1999)
Recinicium bicolor	(Stevenson et al. 2008)
Aspergillus niger	(Nayanashree & Thippeswamy 2013, Singh et al. 2017)
Penicillium chrysogenum	(Mohamed et al. 2017)
Phlebia radiate	(Singh et al. 2017)
Alternaria alternata, Rhodotorula mucilaginosa	(Bosco et al. 2018)

Table 2: List of NR degrading bacteria reported in the literature.

Bacteria	Group	Enzyme Involved	References
Gram-positive			
Streptomyces sp. strain K30	B	Lcp	Birke et al. 2015, Rose & Steinbüchel 2005, Röther et al. 2016, Yikmis et al. 2008
Streptomyces sp. strain CFMR 7	B	Lcp	Nanthini et al. 2017, Nanthini & Sudesh 2017
Streptomyces griseus 1D	B	Lcp	Bode et al. 2001, Jendrossek et al. 1997
Streptomyces coelicolor 1A	B	Lcp	Jendrossek et al. 1997, Bode et al. 2000
Micromonospora aurantiaca W2b	B	Unknown	Linos et al. 2000
Rhodococcus rhodochrous RPK1	A	Lcp	Watcharakul et al. 2016
Gordonia westfalica Kb2	A	Lcp	Berekaa et al. 2000
Gordonia polyisoprenivorans VH2	A	Lcp	Hiessl et al. 2012, Oetermann et al. 2018
Gordonia polyisoprenivorans Kd2	A	Lcp	Berekaa et al. 2000, Linos et al. 1999
Nocardia nova SH22a	A	Lcp	Luo et al. 2014
Nocardia farcinica E3	A	Lcp	Ibrahim et al. 2006
Nocardia farcinica NVL3	A	Lcp	Linh et al. 2017
Paenibacillus lautus	A	Unknown	Hapuarachchi et al. 2016
Achromobacter sp.	A	Unknown	Berekaa et al. 2005
Mycobacterium fortuitum NF4	A	Unknown	Linos et al. 2000
Gram-negative			
Steroidobacter cummioxidans strain 35Y	B	RoxA, RoxB	Sharma et al. 2018
Rhizobacter gummiphilus NS21	B	RoxA, RoxB	Imai et al. 2013
Pseudomonas aeruginosa AL98	A	Unknown	Linos et al. 2000
Pseudomonas citronellolis	A	Unknown	Bode et al. 2000
Acinetobacter calcoaceticus	A	Unknown	Bode et al. 2001

strain 35Y (Jendrossek & Reinhardt 2003). To date, almost all gram-positive rubber-degrading bacteria have been found to release the Lcp protein, whereas gram-negative bacteria have been shown to carry the RoxA and RoxB genes (Shah et al. 2020).

RUBBER DEGRADING ENZYMES AND MECHANISM

Rubber is a high molecular weight polymer that cannot be absorbed directly by cells; instead, it must first be broken down extracellularly into low molecular components that may then be transported over the cell membrane and used for metabolism. Previous research works on rubber degradation has therefore largely focused on extracellular enzyme attack on the polyisoprene molecule (Birke et al. 2017). For this, both Gram-positive and Gram-negative bacteria use two unrelated types of enzymes i.e., Latex clearing protein (Lcp) and Rubber oxygenase (RoxA and RoxB). Lcp is a mono-heme cytochrome-b protein while Rubber oxygenase Rox A and Rox B are both Diheme cytochrome-c dioxygenase proteins (Shah 2020). Several studies have been reported on RoxAs and Lcps (Birke et al. 2015, IIcu et al. 2017, Schmitt et al. 2010, Seidel et al. 2013 & Yikmis et al. 2012) whereas RoxB has been discovered only recently (Birke et al. 2017). The amino-acid sequences of RoxAs and RoxBs have no notable similarities to those of Lcps. Regardless, all three enzymes attack the polyisoprene molecule's cis double bond oxidatively, resulting in cleavage products with aldehyde and keto end groups, as well as some isoprene units in between. Rubber is broken down by Lcp into a variety of compounds, ranging from C20 tetra-isoprenoids to higher oligo-isoprenoids. RoxA, on the other hand, only makes one polyisoprene cleavage product, ODTD, a C15 oligoisoprenoid. The active sites of Lcp and RoxA are distinct, as evidenced by their diverse products. The active site of Lcp is thought to be more surface accessible and should be closer to the substrate-binding site, whereas the active site of RoxA is buried deep within the enzyme structure and has no direct open access to the protein surface. An exo-type cleavage mechanism is proposed for RoxA to explain the regular spacing between

two adjacent cleavage sites, however, an endo-type cleavage mechanism is proposed for Lcp to explain the wide range of cleavage products (Birke & Jendrossek 2014, Jendrossek & Reinhardt 2003). Although RoxB and RoxA share the same fundamental amino acid sequence and other features, the cleavage products for RoxB were discovered to be identical to those observed for Lcp. RoxB is related to Lcp and, unlike RoxA, cleaves polyisoprene in an endo-type manner, as indicated by the detection of a variety of oligo-isoprenoids of varying lengths (Birke et al. 2017). Table 3 summarizes the characteristics of NR degrading enzymes.

The Lcp, RoxA, and RoxB enzymes are responsible for the extracellular cleavage of polyisoprene. These degraded isoprene derivatives are transported into the bacterial cell which is responsible for rubber degradation. In the intracellular space of the bacteria, there are a variety of enzymes that are found to be important for rubber metabolism. These enzymes are Acyl CoA Synthase, Acyl CoA Dehydrogenase, Dienoyl CoA Reductase, Enoyl CoA Isomerase, Enoyl CoA Reductase, 3-Hydroxylacyl CoA Dehydrogenase, Acyl CoA Acetyltransferase, α-Methylacyl Racemase, Acyl CoA Dehydrogenase, Acyl CoA Hydratase, 3-Hydroxyacyl CoA dehydrogenase, and Acyl CoA Acetyltransferase. As a result of degradation by these enzymes, further degradation is done by the beta-oxidation process. During these processes, the degradation product of the rubber finally is converted into the propionyl-CoA and acetyl-CoA which are easily taken up by the bacteria for their metabolic processes like glycolysis and TCA cycle (Methylcitrate cycle and Methylmalonil pathway). The time required for the degradation depends on the amount of propionyl-CoA and acetyl-CoA formed and taken by bacteria during the number of cycles for degradation. Fig. 4 explains the role of different enzymes in the rubber degradation by different bacterial enzymes.

FUTURE PERSPECTIVES

Although NR biodegradation is a more environmentally acceptable alternative to traditional disposal methods, it is a slow and low-yielding process. This is because living microbes catalyze solid and impure substrates, resulting in slow

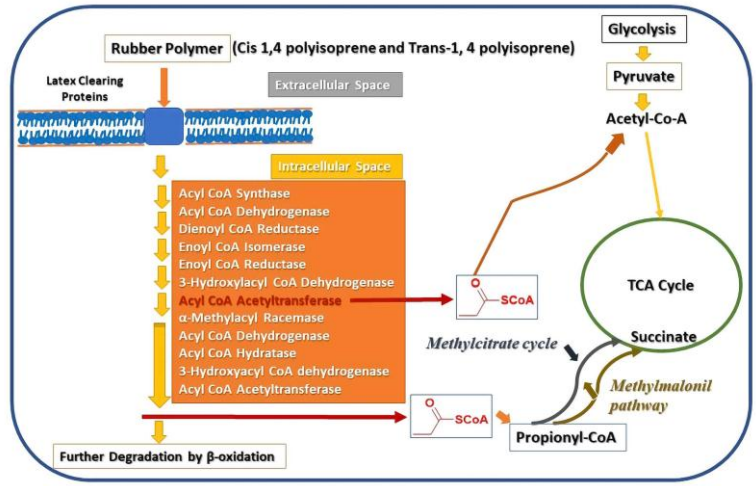

Fig. 4: Proposed metabolic pathway of poly(cis-1,4-isoprene) in *Gordonia polyisoprenivorans* VH2 (Adapted from Andler 2020).

Table 3: Characteristics of NR degrading enzymes (Shah et al. 2020, Basik et al. 1989, Birke et al. 2017, Birke & Jendrossek 2014)

	RoxA	RoxB	Lcp
Identified and Characterized from	*Xanthomonas sp.* strain 35Y	*Xanthomonas sp*. strain 35Y	*Streptomyces sp*. strain K30
Bacteria	Gram-negative	Gram-negative	Gram-positive
Co-factor	Diheme cytochrome-c dioxygenase protein	Diheme cytochrome-c dioxygenase protein	Mono-heme cytochrome-b protein
Molecular Mass	≈73 kDa	≈70 kDa	≈40 kDa
Mechanism of Cleavage	Exo	Endo	Endo
Rubber Degradation Product	12-oxo-4,8-dimethyltrideca-4,8-diene-1-al (ODTD) a C15 oligo-isoprenoid	Mixture of C20, C25, C30 and higher oligo-isoprenoids	Mixture of C20, C25, C30 and higher oligo-isoprenoids

reactions (long incubation periods), and they are sensitive to chemical substances such as rubber additives in most cases. A cost-effective strategy for treating massive amounts of rubber waste has been proposed using enzymes with a high-efficiency expression system and a low-cost recovery methodology. It is critical to screen all of the enzymes involved in total rubber degradation to achieve this (Andler 2020).

For an effective rubber waste recycling approach, Stevenson et al. proposed a multistage process involving detoxification, desulfurization-devulcanization, and total or partial biodegradation. The detoxification process involves the use of certain fungal and bacterial species to remove toxic additives from the rubber. This is followed by desulfurization-devulcanization which involves removing the sulfur cross-links in the vulcanized rubber by the sulfur utilizing microorganisms. Detoxification boosts the biodegradability of rubber while also lowering the environmental risks connected with its disposal. It has also been shown to help in the growth of desulfurizing bacteria for devulcanization (Stevenson et al. 2008). Table 4 lists some fungi and bacteria which can be utilized for Detoxification and Devulcanization in multistage NR degradation.

Another approach toward sustainable NR degradation would be to combine green chemistry with biological processes. Catalytic agents for the oxidation of specific rubber additives obtained from the vulcanization process, in particular, can be exceedingly effective and time-saving when compared to biological procedures. However, in doing so green chemistry principles, such as the use of cleaner solvents, the reduction of by-products, and the reduction of energy requirements, should be considered (Andler 2020).

The resistance of synthetic rubber towards microbial biodegradation is mostly because they have not been available for long enough in natural evolution for microorganisms to create degradative enzymes to use the compound. To degrade novel synthetic compounds, microorganisms will need to acquire new genes and genetic functions that encode catabolic enzymes. Gene transfers between microorganisms can result in the emergence of a specific degradative pathway. In response to synthetic compounds, microbes have occasionally shown response by producing degrading enzymes, however, there may be no optimal control on the pathway. Thus, to sum up, microbes need a long period to acclimatize to synthetic material, and to achieve effective biodegradation of synthetic rubber, this natural process of biodegradation should be accelerated. Recently a novel material ENSO RESTORE^{TM} RL a rubber additive was proposed to attract the specific naturally occurring microbes and rapidly acclimatize them to synthetic material. This additive has a unique property that it is inert to rubber resin and does not contribute directly to rubber degradation thus preserving the rubber's shelf life. The test results showed the effectiveness of ENSO RESTORE^{TM} RL to acclimatize the flora within the test inoculum such that synthetic rubber can be used as the only carbon source and effectively biodegraded. Unexpectedly, this material was shown to work for synthetic rubber but not for natural rubber. It was found that the anaerobic environment such as those found in landfills only promotes the biotic degradation process through extra-cellular and intra-cellular enzymes and not the abiotic oxidation through free oxygen which is the first step in the natural rubber degradation. Furthermore, most of the earlier studies on NR materials involved isolated microbes and enhanced environmental conditions which do not correspond to the natural habitat involving multiple different species (2013).

Table 4: Some useful microbes screened for the detoxification and devulcanization (Stevenson et al. 2008)

Fungi	Bacteria
Detoxification	
Pleurotus. sajor-caju	*Rhodococcus* sp.
Trametes versicolor	*Corynebateria*
Recinicium bicolor	*Pseudomonas*
	Escherichia coli
Desulfurization-Devulcanization	
	Thiobacillus. ferrooxidans
	Thiobacillus. thioparus
	Thiobacillus. thiooxidans.
	Rhodococcus
	Sulfolobus acidocaldarius

By using the microbial consortia and imitating the microbial activity naturally present in tire dump soil, Bosco et al. (2018) investigated the biodegradation of rubber. This naturally chosen microbial biomass was found to be capable of utilizing NR as the only source of carbon and breaking down NR efficiently, as evidenced by a 15.6 percent dry weight loss. The predominant bio degraders in this investigation were found to be aerobic biomass, primarily filamentous fungi (Bosco & Mollea 2021)

CONCLUSION

Natural rubber (NR) is one of society's most significant polymers. It is a valuable raw material that is utilized to produce over 40,000 distinct products. Medical equipment, surgical gloves, plane, and automobile tires, pacifiers, apparel, and toys are just a few of the products made from it. Today, synthetic polyisoprene with a purity of 98 to 99% may be produced with physical qualities that are identical

to natural rubber. However, they lack the stress stability, processability, and other characteristics of natural rubber (Rose et al. 2005). The major problem associated with the rubber products is their disposal after use, as there is currently a lack of knowledge on the fate of rubber materials in nature. The rate at which rubber decomposes is determined by the type of rubber, its composition, and the surrounding environment. Rubber can be reused, recycled, or disposed of using conventional methods, however, the end product is still rubber mostly microparticles that disintegrate slowly in the environment (Basik et al. 2021). Scientists have been exploring several ways to efficiently break down rubber and rubber-generated wastes since the discovery of distinct rubber-degrading microbes and their genes responsible for the enzymes that digest different types of rubber. Microbial degradation is eco-friendly, which is why it is preferred over chemical and physical degradation. Fungi have been tested for their ability to degrade NR since 1928. However, later publications on rubber-degrading fungi were essentially descriptive, merely stating that it could degrade NR. Many bacterial strains that can use rubber as their sole source of carbon and energy have been discovered to date. However, with the exception of a few studies on technical applications, most research on these bacteria has concentrated on the fundamentals of rubber biodegradation. Despite our growing knowledge of enzyme activity, we still have a limited understanding of enzyme action on rubber substrates and the bacteria, molecular, and environmental factors that influence it. As a result, it's critical to keep looking for new strains and completely comprehending the mechanism of rubber biodegradation to apply the wealth of knowledge gathered to build NR biodegradation processes and systems that can be scaled up.

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