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Enzymes' action on materials: Recent trends

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Abstract. In the era of increasing usage of materials, there is a growing concern of environmental aspects. This has motivated many scientists to research on greener materials and technologies. In this line, several biodegradable materials and biomolecules-based technologies have been developed in the recent years. This has triggered vivacious participation of various biomolecules such as DNA, proteins, enzymes, and cells in the field of 'materials'. Among the various biomolecules, enzymes have gained a significant position due to their applications in numerous fields including nanoparticles synthesis, polymer degradation, enzymatic lithography, biosensing and bio-fuel cells. This mini-review accounts on recent trends in selective fields such as nanoparticles synthesis, enzymatic polymer degradation and enzymatic lithography.

Keywords: Enzyme, polymer degradation, nanoparticle synthesis, enzymatic lithography

1. Introduction

Enzymes play a vital role in many systems due to their unique function. Enzymes are protein in nature along with a co-factor for maximum activity and are designed to carry out a specific task which is unique for each enzyme [1, 2]. Similar to "key-lock" fit, each enzyme fits uniquely with one substrate and catalyzes the reaction [3, 4]. Enzymes sourced from microorganisms or animal cell cultures bring about different substrate conversions in respective reactions. They have the ability to work as catalysts in moderate to wide range of pH, pressure and temperature. In addition, they possess substrate specificity under appropriate environment and thus assist in manufacturing industrially critical products without any contaminations. Because of these benefits, enzymes are exploited in a variety of applications such as paper, cosmetics, food, detergent, textile and pharmaceutical industries [5–9]. Currently enzymes are also exploited as drugs [10]. Nowadays rapid progression in the enzymology is primarily the result of contemporary biotechnology's accelerated evolution over the past decades. The bulk volume of industrially exploited enzymes is hydrolyzing in action, chiefly employed for the breakdown of several natural components [11]. Enzymes with proteolytic activity are dominantly exploited in the industry, because of their ample use in the dairy and surfactant industries. Different carbohydrate breaking enzymes stand for the second largest group, mainly cellulases and amylases are been exploited in the manufacturing industries such as the starch, textile, detergent and baking industries

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[12]. Enzyme catalysis of kinetic separation and asymmetric organic molecule synthesis to extract pure molecules has also been accelerated in the recent past. They are exploited to synthesize chiral molecules during the past decade due to the greater enantioselectivity [13, 14]. They are promising in the field of Green Chemistry mainly for the synthesis of chiral molecules, food additives or many drugs under the gentle eco-friendly conditions [15]. One more area where enzymes are widely exploited is bio-transformation of polymer materials. Polymer waste is extremely stable in the environment and their-biodegradability is limited. Recent reports include, the occurrence of polymer-degrading microorganisms in the environment, the isolation of new microorganisms for biodegradation, the discovery of brand-new degradation enzymes and the gene cloning for synthetic polymer-degrading enzymes [16]. One more enthralling and area of relevance for polymer degradation is "enzymatic lithography" and it is an eco-friendly approach to degrade polymer selectively and form desirable patterns that find numerous applications in biotechnology and electronics [17]. Current trends suggest the exploitation of enzymes in reduction processes for the synthesis of metal nanoparticles [18–21]. However enzyme catalysis is associated with a few problems respect to their activity and stability. These limitations can be surmounted by exploiting enzyme-reaction engineering [22]. This mini-review is aimed to provide a brief perspective on the recent trends in enzyme-based technology on materials. Representative examples from the literature are discussed to present an overview on the subject.

2. Enzymes sources and types

Enzymes are usually obtained from animals, plants and microbes. Nevertheless, the attributes of the enzymes' origin determines the convenience, cost, and recovery process. In broad, several enzymes analogous to those of plants and animals can be obtained from microorganisms as well. There is a disposition to employ the enzymes from microbial sources for commercial purposes, since they are made abundantly by this approach [23]. The microbial enzyme industry offers rapid and robust growth of microbial sources, and thereby enables one to produce enzymes in large quantities. Microbes can be manipulated at genetic level to obtain better strains – via recombinant DNA techniques, chiefly – to better the quality of the enzyme and to obtain higher yields [22].

2.1. Enzyme production

The enzyme industry rapidly progressed in the early 1980s and 1990s when it was established that enzymes can be obtained from the microbes. Traditionally, enzymes used to be extracted from animal and plant sources that resulted in reduced level of accessibility, swollen cost and inferior development of the enzyme industry. With the help of genetic engineering, desirable proteins are largely brought forth to satisfy the needs of enzyme industry. Therefore, to the highest degree biopharmaceuticals made nowadays are genetically modified products [46]. In recombinant synthesis, the initial step is to get the desirable DNA fragment; then the DNA is amplified and expressed in a suitable expression system. The yield, quality, production time and ease of extraction are crucial parameters for appropriate expression system for recombinant enzyme manufacturing [47]. Several enzyme expression systems have been established. These include cell cultures of bacteria, molds, mammals, yeasts, plants or insects, or via transgenic animals and plants. Various fermentation methods are employed to produce enzymes. Microbial cultures are grown on large scale fermenters for enzyme production under optimized growth conditions [48, 49]. Solid state fermentation and submerged fermentation methods are used commonly for enzyme production, but the former one is preferred due to better maintenance of aseptic conditions and process control.

3. Applications

3.1. Enzyme mediated nanoparticle synthesis

Synthesis of metal nanoparticles using biomolecules is attractive owing to their stability in colloidal solutions, different shapes and sizes. The broad range of nanoparticle utility is mainly due to the small size and greater surface area. A variety of synthetic approaches have been employed for metal nanoparticle synthesis [50]. However, these processes have some liabilities due to the use of harmful radiations and chemical processes. Therefore, a lot of attention has been given in the current scenario for green and sustainable synthetic approaches for nanoparticles of various sizes and shapes while preserving monodispersity. In this context, the reductive enzymes from microorganisms like bacteria and fungi have gained significant attention for the synthesis of metal nanoparticles.

Recent reports suggest the use of different reductases from *Fusarium oxysporum* for the synthesis of metal nanoparticles. In one such report, the extracellular Sulfate reductase from *Fusarium oxysporum* is employed to make cadmium sulfide nanoparticles of size 5–20 nm by the reaction of aqueous $CdSO_4$ solution with the enzyme. The enzyme reacts and converts sulfate ions to sulfide ions, which lead to the formation of CdS nanoparticles [51]. The same group, in another study, has exploited extracellular α-NADPH-dependent nitrate reductase for the synthesis of silver nanoparticles [52]. Brayner et al., has employed common cyanobacteria like *Anabaena, Leptolyngbya* and *Clathorix* to synthesize Au, Pd, Pt and Ag nanoparticles of regulated size in colloidal solution protected with capping protein. They identified the intracellular protein responsible for the synthesis of nanoparticles to be a nitrogenase enzyme [53].

Silver has been used in the form of silver salts for the treatment of several bacterial infections from time immemorial. But antibiotics discovery has reduced the use of silver significantly. However, due to rapid development of nanotechnology and silver in the form of nanoparticles, did a successful return as a potent germicidal agent in the form of nanolotions and nanogels that helped reducing the use of antibiotics [55]. A pure form of alpha-amylase was used to make silver nanoparticles. The alpha-amylase reduced the silver ions resulting in the fabrication of stabilized 22–44 nm silver nanoparticles [56]. Extracellular nitrate-dependent reductase from several strains of *Fusarium oxysporum* was extracted and used in the production of silver nanoparticles [57]. Similarly, nitrate-dependent reductases from *Aspergillus niger* was shown to be capable of synthesizing silver nanoparticles. These nanoparticles were bactericidal against gram-negative *Escherichia coli* and gram-positive *Staphylococcus aureus* bacteria [58, 59]. In other developments, supernatants of *Klebsiella pneumonia*, *Escherichia coli*, and *Enterobacter cloacae* (*Enterobacteriacae*) that contain nitroreductase enzymes were used to synthesize metallic nanoparticles of silver [60]. A dimeric hydrogenase enzyme extracted from *Fusarium oxysporum* was employed to synthesize platinum nanoparticles [61].

Gold nanoparticles (Au NPs) possess large potential as drug carriers and also for gene delivery in gene therapy. One more unique feature of Au NPs is thiol group interaction, creating highly controlled means of drug or gene release [62]. Due to the surface plasmon resonance in the visible light range, Au NPs are vastly used in optical biosensors. The extract from fungi *Sclerotium rolfsii* containing NADPH-dependent enzyme was used by Narayanan et al., to synthesize gold nanoparticles in less than 15 minutes. They demonstrated the controllability of size and shape of nanoparticles by altering the salt and cell extract ratios [63]. Atul kumar et al., has shown that the enzyme activity was retained while synthesizing gold nanoparticles. A pure form of alpha-amylase was used to synthesize gold nanoparticles by reduction of tetrachloroaurate. The enzyme readily stabilized nanoparticles by capping in colloidal solution [64].

In other reports, *in vitro* biosynthesis of gold nanoparticles capped with peptide was done with help of sulfite reductase α-NADPH-dependent and phytochelatin. The enzyme sulfite reductase reduced

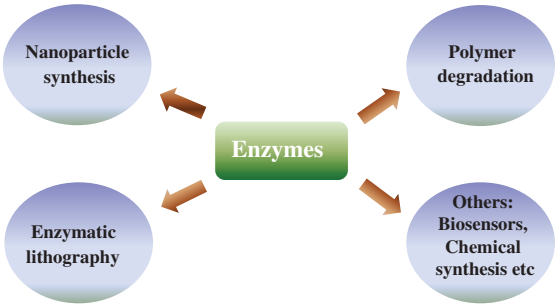

Fig. 1. Pictorial representation of various applications of enzymes.

gold ions resulting in the formation of a stable gold nanoparticle colloidal solution, with size ranging from 7–20 nm capped with the protein that stabilized the NPs in solution [65].

Rhodopseudomonas capsulata bacteria were employed for the synthesis of gold nanoparticles of different sizes and shapes. The key factor for controlling the size and shape of the gold nanoparticles was found to be the pH value of the reaction mixture. It was proposed that the possible mechanism liable for the reduction of Au (3+) to Au (0) that results in the fabrication of gold nanoparticles is by the secreted cofactor NADH- and NADH- dependent enzymes by the bacteria [66].

Morse et al., exploited hydrolase from marine sponge to catalyze the hydrolysis of gallium (III) nitrate that resulted in the polycondensation of gallium oxide to form nanocrystallites at low temperatures all along the length of the filaments [67].

Enzymatic synthesis of nanoparticles is highly dependent on the nature of the metal salt, enzyme and pH of the solution. The stability is brought about by the nature of capping proteins and interaction strength of proteins with metal nanoparticles. This may lead to varied morphologies and size control and monodispersity index.

3.2. Enzyme lithography

Lithography is a technique that allows one to create micro/nanostructures [70]. Traditionally photolithography is used in semiconductor industries for the fabrication of memory and circuit devices. Several advents of lithography such as dip-pen nanolithography, nanoimprint lithography, ion-beam lithography have been reported [71–73]. There is a huge interest in patterning of biomolecules as it could be beneficial for bio-sensing and tissue engineering [74–76]. However, the challenge is that biomolecules are irritable to the use of UltraVaccum, evaporation, UV irradiation, alkali or acid etching. In this backdrop, enzyme lithography has been identified as one of the suitable techniques to create micro/nanostructures at appropriate temperatures and pH values. This technique offers added advantages of high specificity for the substrates which is lacking in other lithographic techniques. In enzyme-lithography either the enzyme adds molecules to the substrate or etches away a part of the substrate.

One of the earliest reports on enzyme lithography was reported in 2003, in which a proteolytic enzyme was delivered onto selected locations of a thin film of bovine serum albumin (BSA). This has yielded selective etching of the BSA film on the locations where trypsin was delivered and thereby created structures at the nanoscale [77]. This was followed by further research in this area to develop techniques based on scanning probe microscope (SPM) and non-SPM based techniques [78].

Antonella Badia et al., demonstrated the enzyme lithography by employing the enzyme phospholipase A2. It catalyzes the hydrolysis of L-isomers stereoselectively, of the phospholipid bilayers made up of the l- and d- isomers of α-dipalmitoylphosphatidylcholine (DPPC) and

Table 1
Some of the industrially important enzymes

Enzyme	EC number	Source	Reference
Animal enzymes			
Rennet	3.4.23.4	Abomasum	24
Trypsin	3.4.21.4	Pancreas	25
Chymotrypsin	3.4.21.1	Pancreas	26
Lipase	3.1.1.3	Pancreas	27
Plant enzymes			
Papain	3.4.22.2	Papaya latex	28
α-Amylase	3.2.1.1	Malted barley	29
β-Amylase	3.2.1.2	Malted barley	30
Bromelain	3.4.22.4	Pineapple latex	31
endo-1,3-Glucanase	3.2.1.6	Malted barley	32
Bacterial enzymes			
α-Amylase	3.2.1.1	*Bacillus*	33
β-Amylase	3.2.1.2	*Bacillus*	34
Glucose isomerase	5.3.1.5	*Bacillus*	35
Penicillin amidase	3.5.1.11	*Bacillus*	36
Protease	3.4.21.14	*Bacillus*	37
Fungal enzymes			
α-Amylase	3.2.1.1	*Aspergillus*	38
Catalase	1.11.1.6	*Aspergillus*	39
Cellulase	3.2.1.4	*Trichoderma*	40
Dextranase	3.2.1.11	*Penicillium*	41
Lipase	3.1.1.3	*Rhizopus*	42
Rennet	3.4.23.6	*Mucor miehei*	43
Protease	3.4.23.6	*Aspergillus*	44
Lipase	3.1.1.3	*Candida*	45

α-dilauroylphosphatidylcholine (DLPC). The report demonstrates that the stereospecific action of enzyme can be exploited to modify polymer surface stereochemically [79].

Chow et al., has reported the use of deoxynucleotidyl transferase enzyme for enzymatic fabrication of DNA nanostructures. In their work, the capability of deoxynucleotidyl transferase to add mononucleotides at the 3' end of a short DNA (acts as an initiator) is exploited to extend a patterned self-assembled nanostructure of an oligonucleotide. Using this approach, oligonucleotide pattern having lateral dimensions in the range of 0.1 to 4 μm have been shown to grow to a height up to 121 nm in 2 h [80].

Jinho Hyun et al., demonstrated the negative enzyme dip-pen lithography. They employed endonuclease (DNase I) to selectively remove oligonucleotides from the gold substrate functionalized with DNA [81]. Another additive based example was demonstrated for patterned growth of polyaniline conducting polymer. To achieve this, horseradish peroxidase enzyme was inked onto the tip of an Atomic Force Microscope (AFM) [82]. Controlled movement and positioning of the tip on a surface containing aniline enabled oxidative polymerization to yield polyaniline catalyzed by hydrogen peroxide, which was produced by the enzyme immobilized on the AFM tip.

Soft-lithography based enzymatic lithography also has been reported in the literature. Typically, a stamp made up of poly(dimethlylsiloxane) (PDMS) containing the desired patterns is used to ink the enzyme that would be brought into contact with a polymer film so as to deliver the enzymes

Table 2
Representative enzymes used for the bio-synthesis of metal nanoparticles

Enzyme	Source	Type of particle	Size (in nm)	Reference
Sulfate reductase	*Fusarium oxysporum*	cadmium nanoparticles	5–20	51
Nitrate reductase	*Fusarium oxysporum*	silver nanoparticles	10–25	52
Nitrogenase	*Anabaena flos-aquae*	gold, palladium, platinum, silver	3.5–40	53
Nitrogenase	*Leptolyngbya foveolarum*	gold, palladium, platinum, silver	3.5–40	53
Nitrogenase	*Clathorix pulvinata*	gold, palladium, platinum, silver	3.5–40	53
Lysozyme	Hen egg white	Silver nanoparticles	10–62	54
α-amylase		silver nanoparticles	22–44	56
Nitrate-dependent reductase	*Fusarium oxysporum*	silver nanoparticles	20–50	57
Nitrate-dependent reductase	*Aspergillus niger*	silver nanoparticles	1–20	58,59
Nitroreductase	Klebsiella pneumonia	silver nanoparticles	28.2–122	60
Nitroreductase	Escherichia coli	silver nanoparticles	28.2–122	60
Nitroreductase	Enterobacter cloacae	silver nanoparticles	28.2–122	60
Hydrogenase	*Fusarium oxysporum*	platinum nanoparticles	100–180	61
NADPH-dependent reductases	Sclerotium rolfsii	gold nanoparticles	25	63
α-NADPH-dependent sulfite reductase	*Fusarium oxysporum*	gold nanoparticles	7–20	65
NADH- dependent enzymes	*Rhodopseudomonas capsulata*	gold nanoparticles	10–20	66
Hydrolase	*Tethya aurantia*	gallium		67
Sulfite reductase enzyme	*Thermomonospora sp*	gold nanoparticles	2–6	68
Laccase	*Pleurotus ostreatus*	gold nanoparticles	22–39	69

onto selective locations on the polymer film. The technique is also known as microcontact printing. Enzymatic degradation of the polymer film would then lead to the information of patterns as dictated by the features from the PDMS stamp. Guyomard-Lack et al., demonstrated this approach by delivering trypsin on a poly-L-lysine (PLL) polymer using a PDMS stamp [83].

Gross et al., utilized *Candida antartica* lipase B (CALB) to selectively etch the poly(ε–caprolactone) PCL thin film (100 to 300 nm thickness) by delivering the enzyme on designated locations achieved by using microcontact printing as well as polymer pen lithography [85]. In a later study, the same group optimized and extended the capability of the approach to fabricate high-throughput and high-resolution pattern fabrication by using the same enzyme and polymer system. In this case, the polymer crystallinity, enzyme concentration and moisture content were found to be the major parameters that define the etch rate as well as resolution of the patterns. With these optimal conditions, they have demonstrated the patterning on a thicker PCL film (0.1 to 2 μm) with a lateral resolution of <1 μm [84].

3.3. Polymer degradation

Polymer degradation research has attracted attention mainly due to the increased use of the polymeric materials in various domains like agricultural industry, packaging industry, and biomedical applications. Nevertheless, increased commercial utilization of polymers has led to the waste disposal problems. In this backdrop, polymer degradation research has attracted unspoken significant attention to address the environmental issues. Enzymatic degradation of few biopolymers is presented here from recent literature along with factors that can be decisive for degradation. Microorganisms in the environment degrade polymeric materials by the action of their secreted enzymes. In biomedical applications

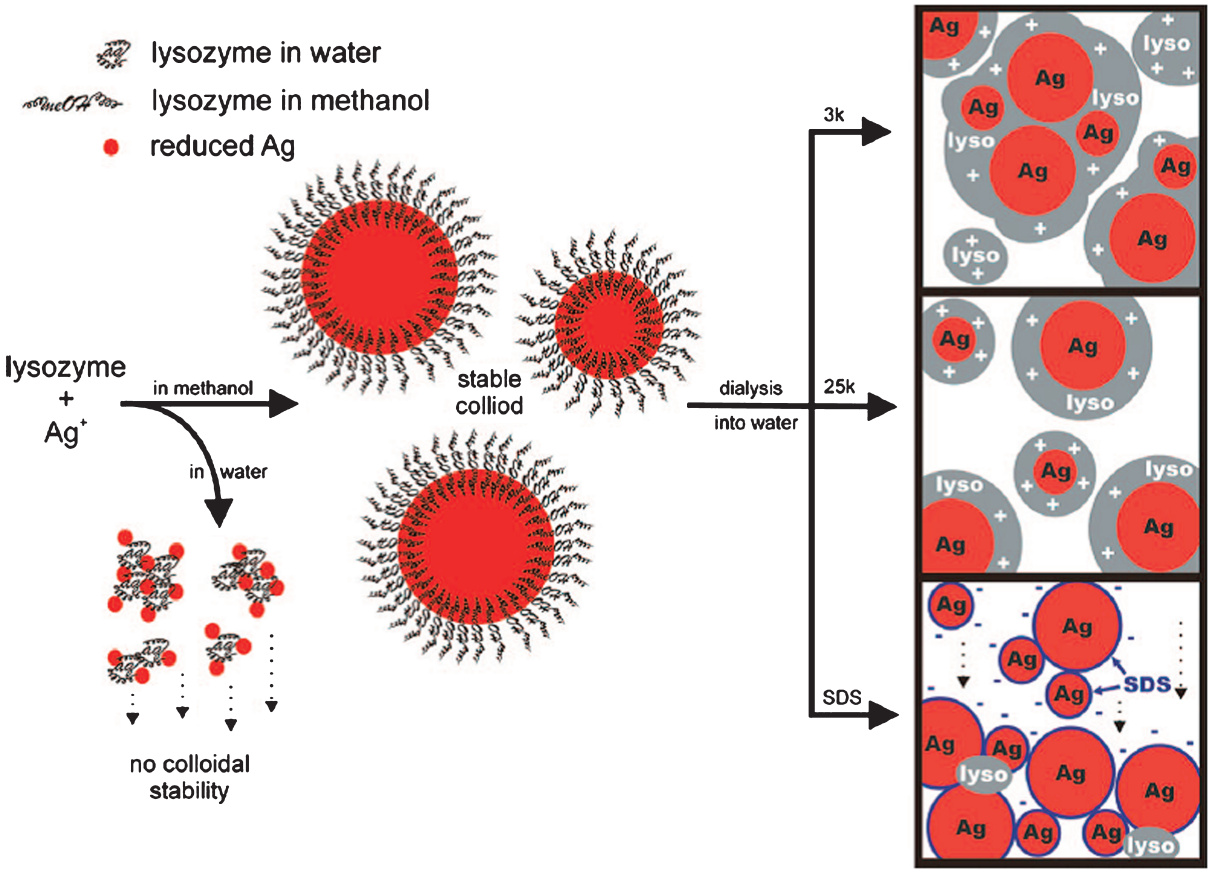

Fig. 2. Ag nanoparticle synthesis mediated by lysozyme (Reprinted with permission from [54]. Copyright (2009) American Chemical Society).

the used polymeric biomaterials may be degraded upon physical contact with body fluids and tissues by several enzymes via by oxidation or hydrolysis [86].

A study reported the radio-labeled polyester-urethanes (PU's) being degraded by cholesterol esterase, horseradish peroxidase and xanthine oxidase enzymes. It was found that the PU's were degraded faster by the action of cholesterol esterase; however the initiation of degradation was done by environmental oxidation. These *in vitro* studies suggested building of polymers with better degradation rates for *in vivo* implantation [87, 88]. The polymer degradation rate can be altered by changing the ratio of polymer blends. A study of Pseudomonas lipase mediated PCL degradation reports 100% weight loss for pure PCL polymer, while a blend of the same with 1% non-biodegradable polymer poly (styrene-co-acrylonitrile) (SAN) degradation has yielded only 50% weight loss. The difference in degradation pattern corresponds to the surface properties and crystal structure of the polymer blends. The enzymatic PCL degradation occurred at the amorphous surface of the polymer film. As the degradation proceeds, the content of the non-degradable component of the polymer blend increases at the surface and prevents the lipase from attacking the biodegradable PCL chains, there by stopping the polymer degradation abruptly [89].

In another similar study, degradation of poly (butylene adipate-co-butylene furandicarboxylate) (PBAFs) by porcine pancreas lipase reported that polymer degradation is influenced by the non-degradable crystalline part of the polymer. The amorphous component was degraded by the enzyme leaving the crystalline non-degradable part [90]. A study showed that the enzyme polyhydroxybu-

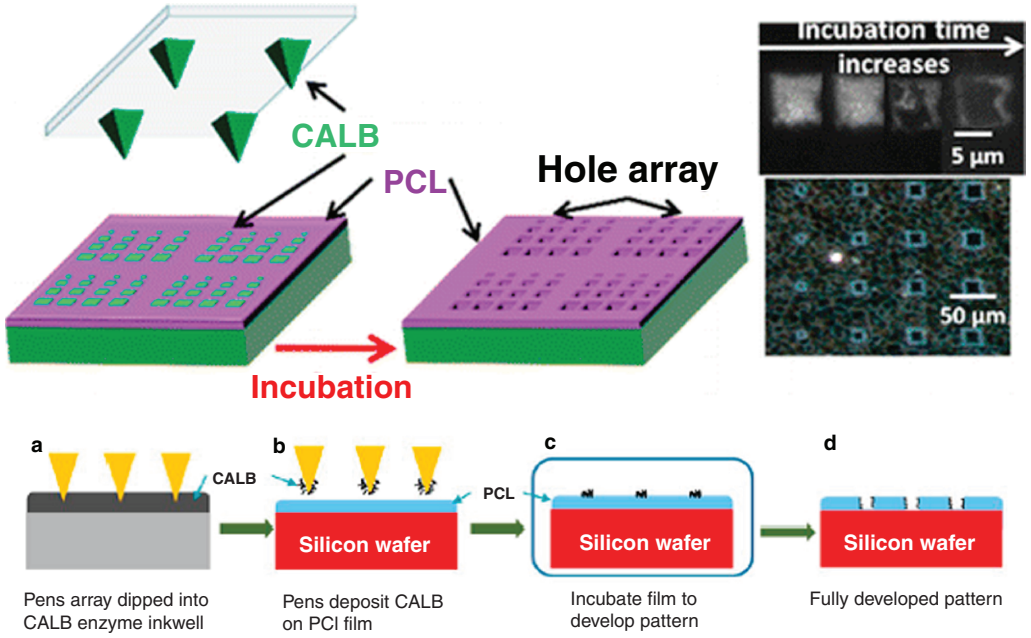

Fig. 3. Schematic representation of enzymatic lithography on PCL film (Reprinted with permission from [84]. Copyright (2009) American Chemical Society).

tyrate (PHB) depolymerase acts on polymer surface first, and then hydrolyze the amorphous part of the polymer and later the crystalline regions [91, 92]. The same was reported by Iwata et al. that enzymatic degradation is greatly influenced by the polymer subunit's crystalline nature and molecular conformation [93–95].

Reports suggest that polymer degradation by enzymes is highly dependent on reaction temperature. Degradation of cross-linked and non-crosslinked PCL polymer was done by the enzyme AK lipase in phosphate buffer saline (PBS) having pH 7.0. The enzymatic degradation gradually increased with increase in temperature till 55°C and 50°C for cross-linked and normal PCL, respectively. This suggests that the increased temperature increases the catalytic activity and reaches an optimum value. After attaining the optimum any further increase in temperature results in protein degradation and hydrolytic activity [96]. G. Madras et al. reported the effect of the alkyl group substitution on enzymatic and thermal degradation of poly n-(acrylates) [poly(methyl acrylate), poly(ethyl acrylate) and poly(butyl acrylate)]. Novozyme 435, lipolase and porcine pancrease were employed to degrade poly n-(acrylates) in toluene medium at various temperatures (40, 50 and 60°C). The enzymatic degradability order at 60°C precedes the identical order as of thermal degradation of the respective polymers [97].

Stereo conformation of polymer subunits also determines the degradation rate of polymer by enzymes' stereospecificity. Several reports suggested the degradation of poly (L- lactide) by different enzymes like pronase, Proteinase K or lipase [98–101]. Proteinase K breaks down the ester bonds joining the L-lactyl units preferentially compared to D-lactyl units [102–105]. Starch based co-polymers were also analyzed for their biodegradability by employing different enzymes. Pullulanase, glucoamylase and α-amylase mediated degradation of polymeric blends of corn starch with poly (ethylene-vinyl alcohol) copolymer and PCL (SEVA-C and SPCL, respectively) have been examined and reported that α-amylase was exhibiting superior activity to the glucoamylase [106]. A study has shown the enzymatic degradation of starch-PCL 3-D scaffold by lipase and α-amylase. It was also identified that both the enzymes were able to degrade the polymer and the polymer matrix was found to be porous in nature after the enzymatic process [107].

Table 3
Representative enzymes and their studies on polymer degradation

Enzyme	Polymer	Reference
Catalase, Horse radish peroxidase, Xanthine oxidase	polyester-urethane	87, 88
Lipase	poly (butylene adipate-co-butylene furandicarboxylate) (PBAFs)	89
Polyhydroxy-butyrate (PHB) depolymerase	PHB	91–95
Lipolase, Novozyme 435, Porcine pancreas	poly n-(acrylates)	97
Pronase, lipase, Proteinase K	poly(L- lactide)	98–105.
Pullulanase, G Glucoamylase, alpha-amylase	SEVA-C, SPCL	106,107
Lipase	PCL	88,95,107,108,109

In a relatively new approach, embedded enzymatic polymer degradation was reported by Gross et al. In this approach, an active enzyme is embedded in a polymeric matrix, which starts to degrade the whole matrix of the polymer as opposed to the conventional polymer degradation, where the degradation begins at the polymer film surface and then penetrate to the bulk. Candida antarctica Lipase B (CALB) blended with the surfactant sodium bis (2-ethylhexyl) sulfosuccinate was mixed with PCL in various proportions and casted into thin films. It was found that the PCL films containing 6.5 and 1.65% of CALB at pH 7.1 resulted in complete degradation of the polymer in 1 and 18 days, respectively [108]. This demonstrates the ability to tune the degradation kinetics of a polymer simply by altering the enzyme loading in the matrix. In a later study, the same group has optimized flow conditions and relative humidity for the PCL degradation [109].

Thus, it is clear that the polymer degradation is influenced by the surface and nature of chemical bonds existing in the polymer. Polymer blends with various components have a significant effect in the degradation capabilities of the final material. The presence of non-degradable polymer component in polymer blends will alter the polymer films' micro-structure and thereby making it more crystalline and less accessible to the enzyme leading to lesser extent of degradation. It is evident that large number of polymers will be degraded to a great extent if proper pH, temperature conditions along with an appropriate enzyme is provided. Further investigation and understanding of polymer degradation is required that are specific towards individual enzymes.

4. Perspective and conclusion

Extensive research is being carried out employing enzymes to improve the existing chemical technology and to adapt green technology. Still at large there is a considerable gap in this field which requires improvement. The use of enzymes for the synthesis of metal nanoparticles and functionalization of their surfaces is the current trend. Moreover, the metal nanoparticles synthesized were mostly spherical, irrespective of the enzyme or the capping agent used. Metal nanoparticle synthesis via enzymes is still in its infancy and vast applications are there to be explored, particularly in areas such as biosensors, targeted drug delivery, anti-microbial etc.

Enzymatic lithography is an eco-friendly approach that could yield desired biomolecular patterns without the need of toxic chemicals. This technique also offers benefits of specific recognition. This technique also is in its infancy and various enzymes could be studied for their efficiency is creating functional patterns over various substrates. Furthermore, new applications using this technique would be another area that has to be explored. Using the specific downward degradation of a polymer sheet in this technique, a polymer can be tuned either for 3-D tissue engineering scaffold or for developing nanofluidics and nano lab-on-a-chip fields.

The use of enzymes for biodegradation of polymers has resulted in extremely efficient and rapid hydrolysis process. Extensive scrutiny has to be done to establish polymer degradation pattern to overcome waste disposal troubles associated with the plastics. Embedded enzymatic polymer degradation is another area, where huge potential is there to develop polymers of controlled biodegradability that are suitable for implant applications. In addition to the above discussed applications, enzymes are going to dominate other research areas as well like biosensors [110], stereoselective chemical synthesis [111], drug delivery [112], bio-fuel cells [113] etc. The examples discussed herein are representative. The reader is encouraged to consult various review articles that are published in these areas for more specific information [114–120].

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