Microbial Enzymes from In Vitro to Market

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Ashish Vyas and Abdulhadi Yakubu

Abstract

A number of chemical transformations sustaining life are mediated by enzymes. All metabolic processes in the system are accelerated by these macromolecules. Microorganisms producing complex enzyme systems are pivotally found in all environments, where they exist in consortia with other microorganisms. In laboratory, optimum synergism amongst enzymatic systems and condition needs to be explored for complete harnessing of its application. Last four to five decades have shown exploration of research with promising insight into the working mechanism of microbial enzymes. The requirement for particular enzymatic formulations is growing more rapidly than ever before, and this demand has become the driving force for research on enzymatic systems. This chapter focuses on the applications, commercialization and market outreach of microbial enzymes from in vitro to market.

Keywords

Industrial utilization · Microbial enzymes · Sustainable environment

A. Vyas (✉)
Department of Microbiology and Biochemistry, School of Bioengineering and Biosciences, Lovely Professional University, Phagwara, Punjab, India
e-mail: ashish.vyas@lpu.co.in

A. Yakubu
Department of Microbiology and Biochemistry, School of Bioengineering and Biosciences, Lovely Professional University, Phagwara, Punjab, India

Department of Science Laboratory Technology, College of Science and Technology, Jigawa State Polytechnic, Dutse, Jigawa State, Nigeria

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12.1 Introduction

Microbial enzymes are used in various eco-friendly purposes. These microbial enzymes can expedite and fasten up reactions by forming transitional complexes states and reduce the activation energy of the reactions. Microbial enzymes are widely used in large number of fields like medicinal products, cosmetics, manufacturing of food and feedstuffs. These microbial catalysts are applied in cleansers in detergents, for pulp and paper applications, in textile applications, for fuel generation and for the creation of pharmaceuticals and chiral substances. Microbial feed enzymes are used in degrading specific components of feed which otherwise would have been harmful or of no value to mulching animals. Microbial enzymes is also helpful in medical field by preventing the spreading of septicaemia such as in healing of wound, lysis of vein thrombosis and acute therapy for myocardial infarction, in diagnosis and curing of different types of leukaemia. This chapter is centred on three pivotal points. Primarily, the focus is given to the latest scenario about the applications of enzyme in the field of food and feed industry, cosmetics and food processing followed by characteristics of enzymes, and sources are discussed. Lastly, enzyme companies involved in large-scale production with highlights, features and commercial brand names have been discussed. In spite of the fact that enzyme formulations have been utilized by humankind over a long history, leaps forward are expected to expand their uses in more extensive applications with progressive unrivalled execution. Moreover, endeavours are made to draw a clear situation about the industrial structure of worldwide enzyme market regarding India.

12.2 History of Microbial Enzymes

Dr. Christian Hansen, a Danish chemist, in 1874 extracted the enzyme rennet from saline solution from dried calf stomach. Since then a lot of exploration and elucidation related to microbial enzymes have occurred. French scientist Louis Pasteur in the eighteenth century reported the fermentative activity of microorganisms. The main utilization of cell-free proteins was the utilization of rennin aspartic protease characterized from calf or sheep stomach in cheddar making. The principal commercial enzyme (trypsin) was set up by Rohn in Germany in 1914, isolated and separated from animals and utilized in detergents to degrade proteins. This enzyme trypsin has shown extraordinary results that original small packet size made the German housewives suspicious, so the product had to be reformulated and sold in larger packages. The innovation of microbial proteases has revolutionized the production of detergent industries. Novozymes is a global biotechnology located in Denmark, and its application to major detergents started around 1965. The company has shown remarkable performance of laundry detergents with improved stain removal, garment care, wash performance and replacing ingredients like surfactants and builders by more environment-friendly product. A 2.9 billion people's clothes were estimated to be washed per week with a detergent containing enzyme in 2015.

As world current population increased to about 7.4, the need for laundry detergent is now around 40%.

Alpha-amylases and glucoamylases have completely replaced the traditional acid hydrolysis, converting 95% of starch to yield simple monosaccharide sugar. Apart from detergent companies, starch processing industries are the second highest enzyme users. Many different extracellular enzymes such as pectinases, esterase, cellulases and many others were reported to replace conventional (chemical) method of ink removal from waste paper. These enzymatic proteins are accounted for eco-friendly to be earth well-disposed when contrasted with regular strategy (Table 12.1).

12.3 Enzyme Application Based on Fields

Microbial enzymes can be the effective mitigator for solution against set goal no 6, 11 and 12 of UN Sustainable Development Goals (SDGs) and the 2030 Agenda for sustainable development. Many industrial products and manufacturing processes were improved by microbiological enzymatic solutions, thereby saving energy, water and raw materials as well as decreasing waste and emissions. The United Nation goal no. 6 has been planned to ensure the presence and availability along with sustainable utilization and proper management of water and hygienic sanitation for all. Target 6.3 further explains that by 2030, there will be improved water quality by decreasing contamination, taking out dumping and limiting arrival of perilous synthetic concoctions and materials, dividing the extent of untreated waste water and significantly expanding reusing and safe reuse all around. A large number of Novozymes enzymatic arrangements help clients and shoppers spare water during application contrasted with regular strategies. For instance, compounds can be utilized in the material business to consolidate procedures and spare critical measures of water. Different Novozymes arrangements help clients in the mash and paper industry to address lignin lethality in effluents created during the generation procedure. It additionally offers answers for waste water treatment and slop decrease for city and mechanical applications. In China, Novozymes is attempting to understand water difficulties in the south-eastern industrialized region just as in the less grown north-west. Their microbial waste water treatment arrangements have been applied in processing plants in Ningxia, Shanxi, Xinjiang and Inner Mongolia, guaranteeing the consistent release of waste water and improved water accessibility. As a feature of the objectives in China's Water 10 Plan, the organization is as of now cooperating with accomplices to extend utilizations of bio-arrangements into recuperation and protection from dark smelly water bodies. Table 12.2 summarizes the aspects of this hydrolytic inducible enzyme and its usage in industry.

Table 12.1 Biochemical characterization of cellulases enzymes with sources and location

Sample	Location	Organism	Enzyme	pH	Temp (°C)	References
Soil	Pulp and paper industries, India	*Bacillus subtilis*	Cellulase	4.0	60	Pala et al. (2004)
Soil	Macuya rainforest, Pucallpa, Peru	*Aspergillus sp.* LM-HP32, *Penicillium* sp. LM-HP33 and 37	Cellulase	4.8–9.4	28	Vega et al. (2012)
Soil	Iguazu rainfalls, Argentina	*Penicillium sp.* CR-313 and *Penicillium sp.* CR-316	Cellulase	4.5	65	Picart et al. (2007)
Waste paper	USM campus, Penang, Malaysia	*Aspergillus niger*	Cellulase, hemicellulose	6.0	50	Lee et al. (2013)
Agricultural waste	Cairo, Egypt	*Bacillus thuringiensis* MAM-29, MAM-38	Cellulase, xylanase	3–7.6	60–80	Abo-State et al. (2013)
Waste photocopy paper	Medellin, Colombia	NA	Cellulase, amylase	7.0	40	Gil et al. (2013)
Wild herbivore, rain deer	Wayanad, Kerala, India	*Escherichia coli* SD5	Cellulase, xylanase	NA	37–39	Vinod Kumar et al. (2018)
Soil, compost, animal waste slurry	Jeju Island, South Korea	*Bacillus subtilis* C5–16 and S52–2	CMCase, avicelase, xylanase	5.0	50	Kim et al. (2012)
Waste paper	NA	NA	Cutinase, amylase	9–11	50	Wang et al. (2018)
Water	Lonar Lake, Buldhana, Maharashtra, India	Many haloalkaliphilic bacteria	Lipase, amylase, caseinase, cellulose	10.5	23	Kanekar et al. (2008)
Soil	Vellore, Tamil Nadu, India	*Streptomyces sp.*	Xylanase	7.5	37	Kalpana and Rajeswari (2015)

Old newsprint, magazine, inkjet, xerox	Chandigarh, Punjab, India	*Bacillus halodurans* FNP135	Xylanase	8–9.5	65	Virk et al. (2013)
Soil	Ambala Cantt., Haryana, India	*Bacillus pumilus*	Xylanase	6–11	60	Nagar et al. (2012)
Soil	Tianshan Xinjiang, China	*Streptomyces rameus* L2001	Xylanase	5–8	70	Li et al. (2010)
Industrial effluents	Shreyan paper industry, Ahmedgarh, Punjab India	*Aspergillus nidulans* KK-99	Xylanase	8–8.5	55	Taneja et al. (2002)
Compost pit	BREC Sadra, Gujarat, India	*Bacillus altitudinis* DHN8	Xylanase	8.0	45–55	Adhyaru et al. (2017)
Waste paper	Chandigarh, Punjab, India	*Bacillus halodurans*	Xylanase and Laccase	8–9.5	65	Virk et al. (2013)
Soil	Effluents of paper industries, India	*Bacillus pumilus* AJK10414	Xylanase, Pectinase	8.5	55	Singh et al. (2012)

Table 12.2 Industrial use of enzymes

Application fields	Enzyme	Technical benefits
Pulp and paper industry	Amylases	Cleaving starch molecules to reduce the viscosity for surface sizing in coating but not used for dry strength agent additive
	Lipases	Deinking to control pitch in pulping processes
	Cellulases	Improving softness by hydrolysing cellulose fibres, breaking weak points in fibres, increasing the flexibility of fibres
	Mannanases	Degrading the residual glucomannan to increase brightness
	Laccases	Bleaching to increase brightness
	Beta-xylanases	Pulp bleaching process efficiency
Laundry detergents	Proteases	Hydrolysis of protein-based stains in fabrics into soluble amino acids
	Lipases	Collar and cuff cleaning by removing the thick spots of oils
	Cellulases	Separation of the microfibres and fuzz to give glossier appearance
	Amylases	Removing resistant starch residues
Cosmetics industry	Oxidases, peroxidases, polyphenyl oxidases	Hair dyeing
	Protein disulphide isomerases, glutathione sulfhydryl oxidases, transglutaminases	Hair waving
	Papain, bromelain, subtilisin	Gentle peeling effect in skin care
	Amyloglucosidase, glucose oxidases	Toothpastes and mouthwashes
Dairy industry	Chymosin, lipases, lysozymes	Cheese manufacturing
	Beta-galactosidases, lactases	Conversion of lactose to glucose and galactose for lactose intolerance
Juice industry	Amylases and glucoamylases	Breaking down starch into glucose
	Pectinases	Acting on soluble pectin hydrolysis, increase in overall juice production
	Cellulases and hemicellulases	Acting on soluble pectin hydrolysis and on cell wall components of pectinases
	Laccases	Increase the susceptibility of browning during storage
	Naringinases and limoninase	Hydrolysis of narginin, resulting in decreases in bitterness of compounds
Starch processing	Beta-amylases	Cleaving alpha 1, 4 glyosidic bond from non-reducing ends amylose, amylopectin and glycogen

(continued)

Table 12.2 (continued)

Application fields	Enzyme	Technical benefits
	Pullulanases	Attacking alpha 1,6 linkages, liberating straight chain residues of oligosaccharides of glucose residues linked by alpha 1,4 bonds
	Neopullulanases and amylopullulanases	Attacking on both alpha 1,6 and 1,4 linkages
	Beta-amylases	Cleaving alpha 1, 4 glyosidic bond from non-reducing ends amylose, amylopectin and glycogen

12.4 Enzyme-Producing Microbes

12.4.1 Cellulase and Its Industrial Applications

Cellulases of microbial origin are found to be engaged as the significant enzymes based on its multi-isoenzymatic as well as broad applications. Bacteria and fungi mainly reported for this enzyme are *Trichoderma, Thermomonospora, Fusarium* and *Aspergillus*. Research on cellulase enzymes complex and related hydrolases is mainly confined to animal feed, food application, brewery and wine industry. The last 50 years of research witnessed progress in isolation, purifying, characterizing and elucidating the mechanism of action of cellulases from microbes. Recent use of these enzymes in textile, laundry, pulp and paper industries has given a new spurt for screening of new microbes having extremophilic, alkaliphilic and thermotolerent properties for novel industrial applications from lab to market utilization.

12.4.1.1 Industrial Application of Xylanase

This enzyme is a hydrolytic compound which breaks the β-1, 4 spine of the multiplex plant cell wall. Apart from cellulose, xylan is the second largest polysaccharide. Among different clusters of microorganisms that are engaged with breaking down of xylanase include fungi, bacteria, yeast and actinomycetes. In this process, bark removal, grinding and screening of wood usually take place. At that point a chip experiences boiling procedure with the goal that the amount of microbes will reduce drastically. This will be followed by cooling the chips and then inoculated with biopulping organism. The biopulping procedure is savvy and mechanically possible. The fundamental bit of leeway is the abatement in the utilization of vitality just as the expansion in factory utilization. These procedures additionally allow an upgrade of paper and decreased ecological problems. Based on previous research, it was assumed that pre-dyeing strategy of this enzyme is less expensive and eco-accommodating than chemicals. It additionally diminishes the critical measure of synthetic compounds that enjoyed request to get splendour in synthetically

blanching procedure. In a customary strategy for paper-making process, the makers utilize unsafe synthetic compounds which confer negative effect to the earth.

A large quantity of xylanase was reported from *Bacillus pumilus* SV-205 under an optimized fermentation conditions. The bacterium secretes maximum amount of cellulase-free xylanase in combination with yeast and peptone which also enhanced highest xylanase production that differ from other combinations. The enzyme maintained a thermal stability of 65% activity after incubation at 60 °C for 2 h (Nagar et al. 2012). Biobleaching capability was also reported from xylanase produced by *Streptomyces* L2001 at a maximum temperature of 70 °C and 5.3 pH (Li et al. 2010) as well as *Bacillus altitudinis* DHN8 (Adhyaru et al. 2017). Using response surface technology, enzyme yield was improved by optimizing submerged fermentation conditions which include incubation time, temperature, agitation speed, sorghum straw, inoculum size and gelatin. Improvement of enzyme production was expressed using response surface methodology (RSM) which gives twofold increase in activity compared to conventional method used for biodeinking and biobleaching.

12.4.1.2 Industrial Application of Laccase

In 1883, Yoshida discovered laccase from Japanese lacquer tree called *Rhus vernicifera* and reported as one of the oldest and most widely reported industrial enzymes. A high quantity of this enzyme was present in fungi and plants including basidiomycetes, white rot and ascomycetes. Among these, white rot and basidiomycetes are found to be involved in the breaking of lignin using enzymes like manganese-dependent peroxidases, peroxidases and laccase. In different ways, this laccase usually participate in cellular process such as plant pathogenesis and sporulation. Increase in pulp strength can be achieved by woody chips pretreated with lignolytic fungi.

Some of the industrial uses of this enzyme include kraft pulp biobleaching. It was reported that 25% decrease of chloride application and 1.8 unit increase in brightness of kraft pulp were observed when SL4 lignocellulotic fungi was used (Kaur and Nigam 2014). Decrease in lignin content in the wood of eucalyptus with its application in biobleaching was found in *C. albidus.* Another vital application of this enzyme is the removal of toxic waste from pulp industries that contain high amount of phenolic compounds. Virk et al. (2013) reported an increase in ink removal of this enzyme in combination with xylanase when some physico-chemical and nutritional parameters were optimized using response surface methodology which is the first time when mediator supplements were not added in this enzyme treatment. A synthetic dye was reported to be removed by alkaline laccase produced from *Myrothecium verrucaria* 24G-4 (Sulistyaningdyah et al. 2004).

12.4.1.3 Industrial Application of Amylase

This enzyme is capable of breaking down starch into different types of products such as dextrin and glucose units. It is found in plant, animals as well as microorganisms.

Moreover, microbial amylases are now used instead of conventional chemical methods in pulp and paper industries because of its low cost. Microbial amylase is the first commercially available enzyme of fungal origin produced in 1894 with therapeutic application in digestive disorder. By the application of ethoxylated fatty acid as surfactant, amylase and cellulase were used for the deinking of waste paper. In this process, temperature of 40 C was applied with floatation consistency of 0.8% in 6 min. The result indicated an increase in brightness as well as reduction of residual ink as compared to control which contained a denatured enzyme (Gil et al. 2013).

The enzyme is basically divided in to endo- and exoamylase. The former hydrolyses oligosaccharides different lengths in a random way, while the latter hydrolyses in a non-reducing end by forming short end products. Some fungi responsible for the production of these industrial enzymes include *Aspergillus* species of *niger, flavus* and *oryzae.* These species have the ability of generating high amounts of this enzyme for commercial purposes.

12.4.1.4 Industrial Application of Lipase

In 1834, J. Eberle was the first person to discover the presence of lipase enzyme in the pancreas, which was also isolated in 1856 by C.I. Bernard. This enzyme has the ability to yield glycerol and fatty acids when working with carboxyl ester found in triacylglycerol under aqueous conditions (Gupta 2004). In the area of biotechnology, microbial lipase plays a vital role because of its versatility, excellent production of large quantity and highly use in different industries. Microorganisms responsible for the production of lipase include fungi such as *Acinotobacter radioresistens, Aeromonas hydrophila* as well as *Aspergillus oryzae* (Andualema and Gessesse 2012). Pitch described hydrophobic contents of wood which happen to be one of the cheap sources of paper. This usually brings more problems in paper processing industries. Moreover, this enzyme can remove pitch from the pulpy deposits at the point of processing paper. Almost 90% of triglycerides found in the pitch can be converted into monoglycerides, glycerol and fatty acids by this enzyme that has low stickiness and high hydrophilic activity (Jaeger and Reetz 1998). This was confirmed by Nippon, a paper industry in Japan, where 90% of woody triglycerides were hydrolysed by this enzyme produced from *Candida rugosa.* Some of the major applications of lipase are the ability to increase the rate of pulping, improve brightness and decrease the level of pollutant from waste water, time and cost of composite. Lipases isolated from *Pseudomonas* sp. (KWI-56) have been found to increase pulp brightness and reduce residual ink concentration, while a thermoalkalophilic *Bacillus coagulans* BTS-3 found to produce lipases was used to remove ink from waste paper when grown at an optimized culture conditions.

12.5 International Market Scenario

In recent year, global industrial enzyme markets are expected to create new opportunities to market players. The market has an estimate of $4.2 billion in 2014, and it was expected to reach $6.2 billion between 2015 and 2020 with almost 7% increase. The major factor for driving growth of global industrial market is obtained from beverage, paper and personal care industry. Approximately, 4000 different enzymes are known currently, and at least 200 of these enzymes are of microbial origin. Moreover, the large-scale industrial production of these microbial enzymes is just 20. However, based on the current investigations of enzyme production biochemistry, recovery methods and process fermentation, a large number of industrial enzymes can be achieved. Novozymes from Denmark, DuPont from the United States and Roche from Switzerland are three major global producers of enzymes with about 75% of the total enzyme production. In addition to these three major producers, a total of 12 major and 400 small industries supplied global enzyme needs presently. Some of the problems encountered by these companies are the market competitiveness, less profit gap as well as technology intensive (Table 12.3).

12.6 Indian Market Scenario

Some of the leading players in the India Enzymes Market are BASF SE, Associated British Foods PLC, Novozymes A/S, Dyadic International Inc., Advance Enzyme Technology Ltd., etc. As per latest survey, India Enzymes Market is anticipated to post robust growth by 2023, owing to the growing application of enzymes for fermentation of milk to produce dairy products like curd, yogurt, cheese, etc. The flourishing dairy industry in India is anticipated to have a positive impact on the demand for industrial enzyme in the coming years. Moreover, the increasing popularity of recombinant enzymes across various industries like detergents, pharmaceuticals, etc. as a result of constant innovations in R&D to increase the yield and improve enzyme specificity as well as stability will propel the growth of the market further. Additionally, the rising use of proteases in detergents coupled with the growing use in animal feed is projected to drive the growth of the market during the forecast period. Based on the type, the protease segment is expected to grow at the highest CAGR by 2023, due to widespread use in food, beverage, detergents and pharmaceuticals. Based on the application, the pharmaceutical segment is expected to lead the market during the forecast period, owing to the rising R&D in recombinant techniques that have enabled in improving the yields of enzymes through fermentation, increased stability and altered specificity. Biotech industry in India is just 2% of the global market, but as the investment opportunity is expanding, it turns more into global visibility. The Indian modern chemical showcase is taking advantage of the interest for simpler plans, helping expanded utilitarian advantages and multi-application profiles. The influx of international companies coupled with expansion of research and production in Indian companies has led to customer's choice with wider inventive products to choose.

Table 12.3 Microbial enzymes – in vitro to market outreach

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
Mixed enzymes	Rossari Biotech, Mumbai	Rexsize LHT New Liquid	Mixing of hydrolytic enzymes at high temperature desizing of fabric	A unique mix of several enzymes with efficient fabric desizing	The main type of basic reactions involved in enzymatic desizing: absorption of water, pH buffering, anti-catalytic action – fibre sizes are destabilized by swelling, entry and breaking.
				Biodegrades the starch-based sizes without redeposition	
				Highly concentrated product	
				Size removal is fast	
				Operates over wide temperature range (70–110 °C) and at pH range of 6.5–8.0	Gelatinization: enzyme acts like the scissors working at molecular level – it washes off and disperses the degraded products. Rexsize LHT New Liquid degrades starch into smaller soluble fragments called disaccharides (maltose) by hydrolysis and hence can be easily removed from the fabric.
				Operational by exhaust as well as semi-continuous process	
				Economical in use	
				Eco-friendly operation and non-corrosive to the equipment	
Amylases	Maps Enzymes Limited (Formerly Maps (India) Limited)	Palkozyme	Alpha-amylase for low-medium temperature conventional desizing	–	For fabrics produced using cotton or mixes, the twist strings are covered with a sticky substance known as 'size'; to forestall the strings breaking during weaving. Albeit a wide range of mixes have been utilized to estimate textures; starch and its

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Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
					subsidiaries have been the most well-known measuring operator. In the wake of weaving, the size must be expelled again so as to set up the texture for colouring and finishing.
					This procedure (desizing) must be done by treating the texture with synthetic substances, for example, acids, soluble base or oxidizing operators. Anyway starch-breaking compounds (amylases) are favoured for desizing because of their high effectiveness and explicit activity. Amylases achieve total evacuation of the size with no destructive impacts on the texture. Another advantage of enzymes contrasted with solid synthetic concoctions referenced above is that chemicals are conditioned neighbourly.

Cellulases	Maps Enzymes Limited (Formerly Maps (India) Limited)	Palkofeel Palkosoft	Biological polishing with the help of cellulases with mixing of fabric and garment		Cotton and other normal filaments dependent on cellulose can be improved by an enzymatic treatment known as biopolishing. This treatment gives the texture a smoother and glossier appearance. The treatment is utilized to expel 'fluff' – the small strands of fibre that distend from the outside of yarn. A chunk of fluff is known as a 'pill' in the material exchange. After biopolishing, the fluff and pilling are decreased. Different advantages moving fluff are a milder and smoother handle, and prevalent shading brilliance.
Cellulases	Maps Enzymes Limited (Formerly Maps (India) Limited)	Palkowash	In processing of garments with process of biological stonewashing with cellulase	–	Denim finishing Numerous garments of clothing are exposed to a wash treatment to give them a somewhat worn look; model is the stonewashing of denim pants. In the conventional stonewashing process, the blue denim was blurred by the grating activity of pumice stones on the article of clothing surface. These days, denim finishers are utilizing an exceptional cellulase. Cellulase

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Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
					works by relaxing the indigo colour on the denim in a procedure known as 'bio-stonewashing'. A little portion of chemical can supplant a few kilograms of pumice stones. The utilization of less pumice stones brings about less harm to article of clothing, machine and less pumice dust in the clothing condition.
					Bio-stonewashing has opened up new potential outcomes in wrapping up denim by expanding the assortment of completions accessible. For instance, it is presently conceivable to blur denim to a more noteworthy degree without risking harming the article of clothing. Efficiency can likewise be expanded in light of the fact that clothing machines contain less stones or no stones and more garments. Maps scope of cellulases offers a for denim finishing, each

					with its own extraordinary properties. These can be utilized either alone or in mix with pumice stones so as to acquire a particular look.
Catalase	Maps Enzymes Limited (Formerly Maps (India) Limited)	Palkoperox	Enzymatically removing the residual hydrogen peroxide after bleaching of cotton with the help of catalase		Bleach clean-up regular textures, for example, cotton, are typically blanched with hydrogen peroxide before colouring. Fades are exceptionally receptive synthetics, and any peroxide left on the texture can meddle with the colouring procedure. A careful 'fade clean-up' is fundamental. The conventional technique is to kill the sanitizer with a lessening specialist, yet the portion must be controlled absolutely. Catalysts present an increasingly advantageous option since they are simpler and speedier to utilize. A little portion of catalase is equipped for separating hydrogen peroxide into water and oxygen. Contrasted and the customary clean-up techniques, the enzymatic procedure brings about cleaner squander water or diminished water utilization.

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Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
					Maps offers catalase for expelling remaining hydrogen peroxide after the fading of cotton. It diminishes the flushing important to expel dye, or it very well may be utilized to supplant synthetic medicines.
Multicomponent enzyme	Maps Enzymes Limited (Formerly Maps (India) Limited)	Palkoscour	In the case of native cellulose, bio-scouring needs to be done by partial and complete removal of non-cellulosic fraction with the help of isoenzymic form of cellulases		Bio-scouring. Cotton yarn or texture, preceding colouring or printing, experiences various procedures in a material handling unit. A significant procedure is scouring. Right now, cellulosic segments from local cotton are totally or incompletely removed. Scouring gives a texture with a high and even wet capacity, so it very well may be blanched and coloured effectively. Today, profoundly basic synthetic compounds scathing soft drink are utilized for scouring. These synthetic concoctions not just expel the non-cellulosic polluting

					influences from the cotton but in addition assault the cellulose prompting overwhelming quality misfortune and weight reduction in the texture. Besides, utilizing these dangerous synthetic compounds brings about high COD (substance oxygen request), BOD (organic oxygen request) and TDS, in the waste water. Recently another enzymatic scouring process known as 'bio-scouring' is utilized in material wet-preparing in which all non-cellulosic segments from local cotton are totally or halfway evacuated. After this bio-scouring process, the cotton has an unblemished cellulose structure, with lower weight reduction and quality misfortune. The texture gives better wetting and infiltration properties, making consequent blanch process simple and resultantly giving much better colour take-up.

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Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
Amylases	Rossari Biotech, Mumbai	Rexsize MHT-A Liquid	Exceptionally viable mix of amylase-based compound for cotton and its mixes with peak activity at 60–65 °C	Good compatibility with chemicals in desizing bath	Starch comprises of two unique polysaccharides (amylose and amylopectin) and is insoluble is water; henceforth, it must be decayed into pieces which break up more effectively. The conditions of enzymatic desizing is as per the following: wetting, pH buffering, hostile to synergist activity; swelling, infiltration, breaking and de-adjustment of size layers; gelatinization; enzyme attack – the compound plays the job of atomic scissors; wash off, scattering of the debasement items. Rexsize MHT-A Liquid corrupts starch into more little solvent pieces called disaccharides (maltose) by hydrolysis and thus can be effectively expelled from the texture.
				Applicable by exhaust as well as pad batch method	
				Highly effective for removal of size	
				Apply at 600C with pH 6–7.5	
				Biodegradable and non-corrosive in nature	
				Economical operation	
Amylases	Rossari Biotech, Mumbai	Rexsize LHT Conc Liquid	Highly effective blend of amylase-based enzyme for cotton and its blend. It has peak activity at 85C	A unique blend of several enzymes for efficient fabric desizing	
				Biodegrades the starch-based sizes without redeposition	
				Highly concentrated product	
				Operates at an optimum temperature of 60–70 ° C and at pH range of 6.5–8.0	
				Operational by exhaust as well as semi-continuous process	
				Imparts soft hand to the fabric	
				Very economical in use	
				Eco-friendly operation	

	Rossari Biotech, Mumbai	BioD −15 Plus Powder	Highly effective blend of amylase-based enzyme for cotton and its blends. It has peak activity at 98 °C	Good compatibility with chemicals in desizing bath	
				Applicable by exhaust as well as pad batch method	
				Highly effective for removal of size with partial scouring property	
				Apply at 60–98 °C with pH 6.5–7.5	
				Biodegradable and non-corrosive in nature	
				Economical operation	
Cellulases	Rossari Biotech, Mumbai	Biofast 50 Liquid	Acid cellulase enzyme for biopolishing with minimum colour loss	Anamorphic stage of *Trichoderma* cellulase having high FPAse activity	Due to the ring dyeing of warp yarn of denim, a faded fashionable look is obtained. These types of washdown effects are created due to the removal of dyes by abrasion on the garments. Washdown effects are achieved due to bio-abrasion where the enzymes act on cellulose, and hydrolysing it gives the desired look. Biofast 50 Liquid attacks the shapeless areas, creating better cutting, great puckering impacts yet with higher back recolouring look at impartial or designed cellulases.
				Hydrolytically cracks the cellulose on the surface of the garment	
				Acts only on crystalline regions ensuring minimum colour loss	
				Imparts soft feel to the fabric	
				Gives clean surface to the fabric	
				Improves drapability of the garments	
				Operates at 55 °C temperature and pH range of 4.5–5.0	
				Eco-friendly operation	

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Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
Cellulases	Rossari Biotech, Mumbai	Biofast FNB Conc Liquid	A unique easy-to-use cellulase that allows biopolishing to occur under neutral conditions	Hydrolyses the cellulose on the surface of the fabric	Biofast FNB Conc. Fluid is utilized for bio-cleaning or depilling of cellulosics which improve the texture quality, frequently done after overwhelming handling where pills are raised. Cellulase catalysts debilitate the strands distending from the surface by debasement, ideally of the formless structure of the fibre. The enzyme-weakens the filaments which are touchy to shear powers endless supply of adequate shear the fibre will separate from the surface. This results in improved pilling opposition, more brilliant hues, cleaner surface, improved drapability and expanded delicate quality, decrease in the measure of dead and youthful cotton.
				Imparts soft feel to the garment	
				Its high crystalline type activity cuts good amount of cotton fuzz, giving good lustre and clean surface to the fabric	
				Improves drapability of the garments	
				Operates at 55–60 C temperature and pH 6.5–7.0	
				Eco-friendly operation	

Cellulases	Rossari Biotech, Mumbai	G-Zyme BCS Conc Liquid	A cellulase enzyme preparation to produce a very fast biopolishing effect	Highly concentrated product	Cellulase chemicals are utilized for bio-cleaning or depilling of cellulosics which improve the texture quality, frequently done after substantial preparing where pills are raised. Cellulase proteins debilitate the strands distending from the surface by debasement, ideally of the formless structure of the fibre. The enzymatic action improves pilling opposition, more splendid hues, cleaner surface, improved drapability and expanded non-abrasiveness, decrease in the measure of dead and juvenile cotton. G-Zyme BCS Conc Liquid is biopolishing cellulase yet in addition reasonable for blurring where high aggression is required.
				Fast biopolishing effect	
				Prevents redeposition of fuzz by keeping it in suspension due to additional dispersing property	
				Softens the fabric, with excellent fuzz cutting	
				Imparts clean and fresh look with brightening effect	
				Gives natural and permanent finish on garments	
				Operates at a temperature of 55 °C and pH of 4.5–5.0	
				Eco-friendly operation	

(continued)

Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
Cellulases	Rossari Biotech, Mumbai	G-Zyme BSL Liquid	New generation, fully formulated engineered enzyme for effective biopolishing as well as fading	High aggression	Due to the ring dyeing of warp yarn of denim, a faded fashionable look is obtained. These types of washdown effects are created due to the removal of dyes by abrasion on the garments. Washdown effects are achieved due to bio-abrasion where the enzymes act on cellulose and hydrolysing it gives the desired look. G-Zyme BSL Liquid attacks the amorphous regions producing better cutting, good puckering effects and as it engineered with its CBD'S knocked off produces far less back staining compared with acid cellulases. Can call this product as high-speed enzyme with neutral looks.
				Very low back staining	
				Good pH and temperature tolerance	
				High economy as fully formulated	
				Good indigo retention with grey cast	
				Excellent fuzz cutting	
				Operates over wider range of temperature 55 to 60 °C and pH 4.5–5.5. Highest activity at 600 C	
				Eco-friendly operation and non-corrosive to the equipment	

Cellulases	Rossari Biotech, Mumbai	Genecel HEBPL CONC Liquid	High activity acid cellulase for biopolishing	Aggressive acid cellulase-based enzymes for biopolishing Hydrolyses the cellulose on the surface of the fabric High cutting giving clean look to the fabric Excellent inner softness and smooth handle Exhibits good drapability to the fabric Specially recommended for biopolishing of hosiery and defibrillation of Tencel Not recommended for use after dyeing as it gives colour loss Operates at a temperature of 55 °C and pH range of 4.5–5.0	Genecel HEBPL CONC Liquid is used for biopolishing or depilling of cellulosics which improve the fabric quality, often done after heavy processing where pills are raised. Cellulase enzymes weaken the fibres protruding from the surface by degradation, preferably, of the amorphous structure of the fibre. The enzyme-weakened fibres are sensitive to shear forces, and upon application of sufficient shear, the fibre will break from the surface. This results in improved pilling resistance, brighter colours, cleaner surface, improved drapability and increased softness, reduction in the amount of dead and immature cotton.
Cellulases	Rossari Biotech, Mumbai	Genecel JNI Liquid	An acid cellulase enzyme for washdown effect on denims	Aggressive acid cellulase-based enzymes for biopolishing Hydrolyses the cellulose on the surface of the fabric High cutting giving clean look to the fabric Excellent inner softness and smooth handle	Genecel JNI Liquid is utilized for bio-cleaning or depilling of cellulosics which improve the texture quality, regularly done after overwhelming handling where pills are raised. Cellulase chemicals debilitate the strands jutting from the surface by debasement, ideally of the nebulous structure of the fibre.

(continued)

Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
				Exhibits good drapability to the fabric	This enzymatic action results in improved pilling obstruction, more splendid hues, cleaner surface, improved drapability and expanded delicate quality, decrease in the measure of dead and youthful cotton.
				Specially recommended for biopolishing of hosiery	
				Operates at a temperature of 55 C and pH range of 4.5–5.0	
Cellulases	Rossari Biotech, Mumbai	Koldenz CONC Liquid	An acid cellulase for biopolishing at room temperature	Acid cellulase-based enzymes for biopolishing	Koldenz CONC Liquid is used for biopolishing or depilling of cellulosics which improve the texture quality, regularly done after overwhelming handling where pills are raised. Cellulase compounds debilitate the strands projecting from the surface by corruption, ideally of the indistinct structure of the fibre. This enzymatic action results in improved pilling obstruction, more brilliant hues, cleaner surface, improved drapability and expanded delicate quality, decrease in the measure of dead and juvenile cotton.
				Hydrolyses the cellulose on the surface of the fabric	
				High cutting giving clean look to the fabric	
				Excellent inner softness and smooth handle	
				Exhibits good drapability to the fabric	
				Specially recommended for biopolishing of hosiery and defibrillation of Tencel	
				Operates at room temperature and pH range of 4.5–5.0	

Cellulases	Rossari Biotech, Mumbai	Neutrox BDN 100 New Powde	True neutral cellulase enzyme for washdown effect on garments	Neutral cellulase enzyme for washdown effect on garments Minimum back staining Large grain size Excellent colour contrast Applicable at low MLR Operates at 50–60 °C temperature and pH 6.5–7.5	Due to the ring dyeing of warp yarn of denim, a faded fashionable look is obtained. These types of washdown effects are created due to the removal of dyes by abrasion on the garments. Washdown effects are achieved due to bio-abrasion where the enzymes act on cellulose, and hydrolysing it gives the desired look. Bio-washing improves the texture quality, regularly done after overwhelming handling where pills are raised. Cellulase compounds debilitate the strands projecting from the surface by corruption, ideally of the indistinct structure of the fibre. This enzymatic action results in improved pilling obstruction, more brilliant hues, cleaner surface, improved drapability and expanded delicate quality, decrease in the measure of dead and juvenile cotton.
Cellulases	Rossari Biotech, Mumbai	Neutrox Cool Powder	Special cold enzyme of true neutral cellulytic type for fading and biopolishing	True neutral cellulase enzyme designed for washdown effect on garments Neutral biopolish so shade change and colour loss will be less Brighter shades due to combination with surface	Due to the ring dyeing of warp yarn of denim, a faded fashionable look is obtained. These types of washdown effects are created due to the removal of dyes by abrasion on the garments. Washdown effects are achieved due to

(continued)

Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
				active agents for better dispersion and cleaning of fabric surface	bio-abrasion where the enzymes act on cellulose, and hydrolysing it gives the desired look. Neutrox Cool Powder attacks the amorphous regions producing better cutting, good puckering effects, and being true neutral engineered enzyme, it works at low temperature and gives less back staining compared to acid cellulases and conserves energy. Neutrox Cool Powder is used for biopolishing or depilling of cellulosics which improve the texture quality, frequently done after overwhelming handling where pills are raised. Cellulase catalysts debilitate the strands jutting from the surface by debasement, ideally of the formless structure of the fibre. This enzymatic action results in improved pilling opposition, more splendid hues, cleaner surface, improved drapability and expanded delicate quality, decrease in the measure of dead and youthful cotton.
				For the first time a biopolishing in powder form to be used at room temperature	
				Operates at 30–20 ° C temperature and pH range of 6.0–8.0, peak activity at 30 °C and 6.5pH	
				Easy to handle	
				Saves energy	
				Lower dusting	
				The enzyme bath water is harmless to operators and non-corrosive to equipment	
				Recommended mainly for higher-end garment finish	
				Eco-friendly operation	

Cellulases	Rossari Biotech, Mumbai	Neutrox MKL Liquid	Neutral cellulase enzyme for biopolishing with minimum colour loss	Hydrolyses the cellulose on the surface of the fabric	Neutrox MKL Liquid is used for biopolishing or depilling of cellulosics which improve the texture quality, regularly done after overwhelming preparing where pills are raised. Cellulase catalysts debilitate the filaments projecting from the surface by debasement, ideally of the shapeless structure of the fibre. The compound-debilitated filaments are delicate to shear powers endless supply of adequate shear the fibre will part from the surface. This results in improved pilling opposition, more brilliant hues, cleaner surface, improved drapability and expanded non-abrasiveness, decrease in the measure of dead and juvenile cotton.
				Imparts soft feel to the garment	
				Its high crystalline type activity cuts good amount of cotton fuzz giving good lustre and clean surface to the fabric	
				Improves drapability of the garments	
				Operates at 55–60+ C temperature and pH 6.5–7.0	
				Eco-friendly operation	
Cellulases	Rossari Biotech, Mumbai	Neutrox MKL Super Liquid	Neutral cellulase enzyme for biopolishing with minimum colour loss	First time telescoping of four processes in one: dyeing, scourboosting, peroxide neutralization and biopolishing	Biofast SB-NP Super Liquid has unique feature of boosting scouring efficiency, thereby enhancing penetration of dyestuff and other auxiliaries, resulting into even dyeing in a single step. It also carries out residual hydrogen peroxide removal very effectively along with excellent
				Saving in processing time, energy, water	
				Improved absorbency	
				Uniformity in dyeing with increased dye uptake	

(continued)

Table 12.3 (continued)

Enzymes	Industries involved in enzyme production	Commercial brand product	Features	Highlights	Application
				Effectively carries out residual peroxide killing and biopolishing	biopolishing or depilling of cellulosics which improve the fabric quality, often done after heavy processing where pills are raised.
				Hydrolyses the cellulose on the surface of the fabric	
				Softens the fabric, with excellent fuzz cutting	
				Gives clean surface to the fabric	
				Improves drapability	
				Operates at a temperature of 40–60 °C and pH of 4.5–7.0, peak activity at temperature 55 °C and pH 5.5	
				Eco-friendly operation	
Cellulases	Rossari Biotech, Mumbai	Rexsize Cool Liquid	A liquid desizer for efficient removal of size from fabric	Biodegrades the starch sizes very effectively	Starch consists of two different polysaccharides (amylose and amylopectin) and is insoluble in water; hence, it must be decomposed into fragments which dissolve more easily. The mechanism of enzymatic desizing is as follows: wetting, pH buffering, anti-catalytic action; swelling, penetration, cracking and destabilization of size layers; gelatinization; enzyme attack – the enzyme
				Operates in the temperature range of 60–110 ° C/steaming and at neutral pH	
				Broad temperature-sensitive amylase blend, maintains the desizing efficiency even at lower temperatures	
				Operates by exhaust, semi-continuous and steaming method	

				Eco-friendly operation and non-corrosive to the equipment	takes the role of molecular scissors; wash off, dispersion of the degradation products. Rexsize ECE Liquid degrades starch into smaller soluble fragments called disaccharides (maltose) by hydrolysis and hence can be easily removed from the fabric.
Cellulases	Rossari Biotech, Mumbai	Rexsize LHT 100 Liquid	Blend of enzymes for desizing having peak activity at 80–85 °C	A unique blend of several enzymes for efficient fabric desizing	Starch consists of two different polysaccharides (amylose and amylopectin) and is insoluble in water; hence, it must be decomposed into fragments which dissolve more easily. The mechanism of enzymatic desizing is as follows: wetting, pH buffering, anti-catalytic action; swelling, penetration, cracking and destabilization of size layers; gelatinization; enzyme attack – the enzyme takes the role of molecular scissors; wash off, dispersion of the degradation products. Rexsize LHT 100 Liquid degrades starch into smaller soluble fragments called disaccharides (maltose) by hydrolysis and hence can be easily removed from the fabric.
				Biodegrades the starch-based sizes without redeposition	
				Highly concentrated product	
				Operates at an optimum temperature of 70–85 C and at pH range of 6.5–8.0	
				Operational by exhaust as well as semi-continuous process	
				Very economical in use	
				Eco-friendly operation	

12.7 Conclusion and Future Prospects

Microbial enzyme field needs more research and development expenditures. Research should also focus in and around isolating novel organisms from unexplored virgin sites across the globe. Establishment of data bank having information on enzyme production will also provide impetus to microbial enzymologist. Discovery of more enzymes from microbial technology will lead enhanced and improved enzyme products with different physiological conditions. Industrial enzymes are pivotal in current commercial status of biotechnology and may lead to more new discoveries of industrial applications in India.

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