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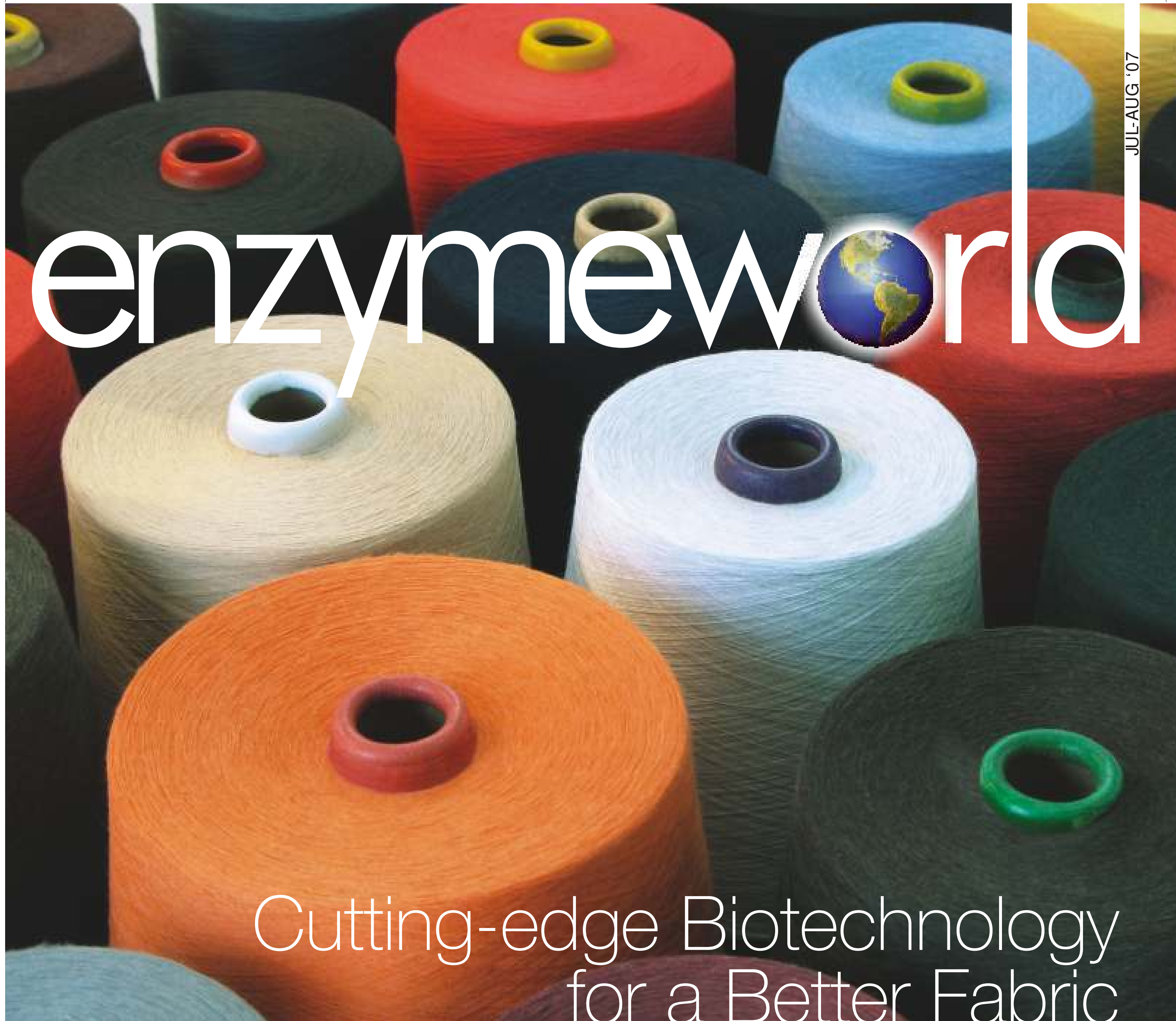
enzymeworld



Cutting-edge Biotechnology for a Better Fabric



contents

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advanced enzymes

Where ENZYME is Life

Dear Friends,

The Textile Industry has strategic importance to India as it is the 2nd largest employer in the country, next only to agriculture. India being the world's 2nd largest nation with over 1 billion people, after food clothing is most important.

The Textile Industry has been modernising in India and composite mills are becoming the norm once again. The whole industry is quite old and has witnessed many changes. At the same time, Indian entrepreneurship can truly take advantage of a good technical pool and emerge as the Number One supplier for high quality fabrics and garments, with China being the 'mass producer' for textile goods. There is a very big opportunity for the Indian Textile Industry in this segment.

In the last few decades we have witnessed the perils of pollution caused by irresponsible use of carcinogenic chemicals, dyes and non-ecofriendly auxiliaries! In several places, where textile processing is a major industry, water is highly polluted. We today cannot drink even a drop of water any where near Tirupur in South India or near Pali and Balotra in Rajasthan! When I met various textile and garment processors in Ludhiyana, I warned them that this should not happen in a state like Punjab!

Gurudev Poojya Sri Sri Ravi Shankarji has always told us to take responsibility and solve problems. There are two major challenges going forward - Quality and Environment Protection! We at ADVANCED ENZYMES have accepted this challenge and responded with several ECO-SAFE solutions. In order to help the textile and garment industry, we have expanded our service team as well as our Research & Development Labs to solve this problem right from the 'molecular' level. Over and above this, we have set up a separate blending facility at Shahpur near Mumbai to help us quickly service customer needs.

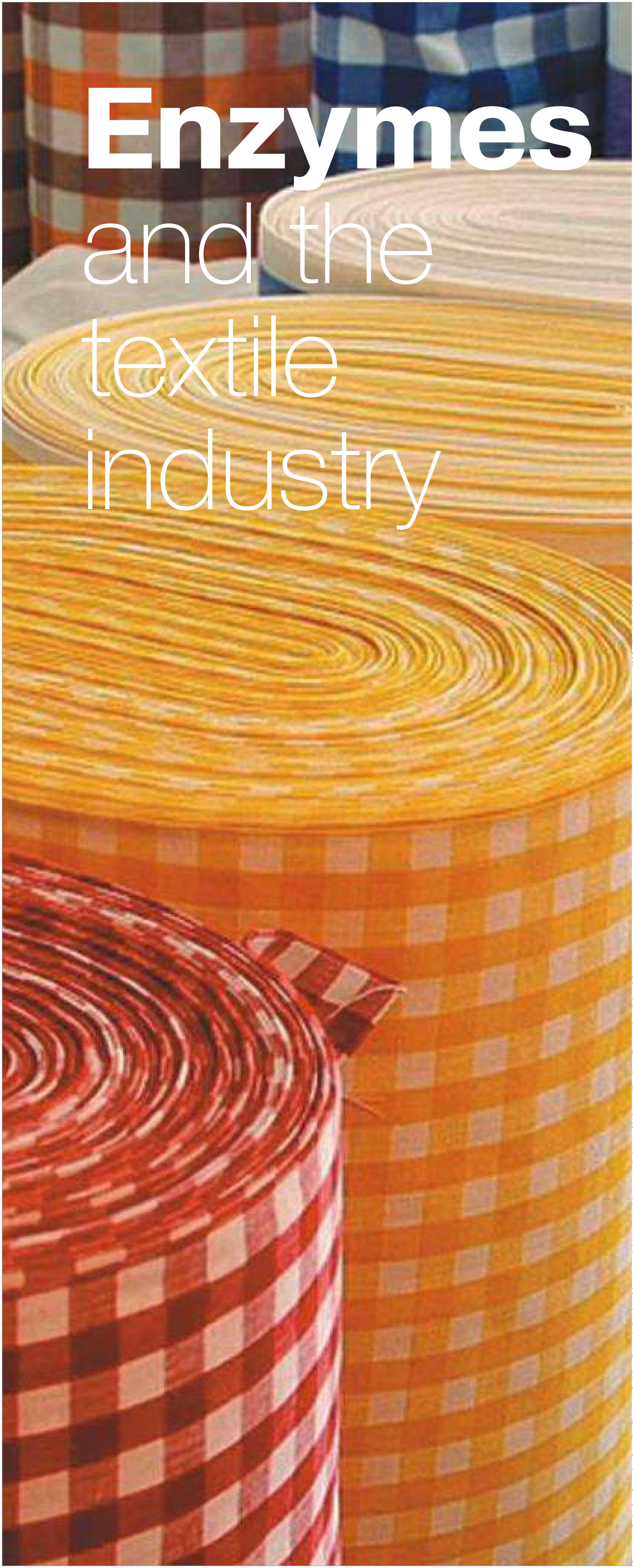
This issue of Enzyme World is a showcase of our commitment. This is the beginning. In the near future, my team is committed to offering better products that will not only improve the Quality of the textile goods by several notches, but also improve Cost Efficiencies significantly!

I welcome each and every technocrat to this mission to help enlarge the scope and area of the challenge.

Jai Gurudev.

C. L. Rathi,
Managing Director

Enzymes and the textile industry



Enzymes have found wide application in the textile industry replacing harsh and hazardous chemicals in various processes. They go a long way in improving production methods and fabric finishing by reducing effluent processing cost.

One of the oldest applications in this industry is the use of Amylases to remove starch size. The warp (longitudinal) threads of fabric are often coated in starch in order to prevent them breaking during weaving. Scouring is the process of purifying fabric of the native cellulosic fibres from impurities such as waxes, pectins, hemicelluloses and mineral salts. Research has shown that pectin acts like glue between the fibre core and the waxes, but can be destroyed by an Alkaline Pectinase. An increase in 'wettability' can thus be obtained.

Cellulases have become the tool for fabric finishing. Their success started in denim finishing when it was discovered that cellulases could provide the fashionable stonewashed look, traditionally achieved through the abrasive action of pumice stones. Cellulases are also used to prevent pilling and improve the smoothness and colour brightness of cotton fabric in a process that is called Biopolishing. In addition, a softer handle is obtained.

Creating an aged look on denim was the fashion of the time, while there was an increased emphasis on moving to environment friendly processes. Laccase and different types of peroxidase helped meet this need, being very effective for giving a novel finish to denim while replacing bleaching chemicals. Catalases

are used for degrading residual hydrogen peroxide after the bleaching of cotton. Hydrogen peroxide has to be removed before dyeing. Proteases are used for wool treatment and the degumming of raw silk.

Furthermore, Advanced Enzymes is developing various enzymes and enzyme based solutions, replacing conventional age-old enzymes and hazardous chemicals. Thus, reducing processing time, minimising processing cost and the use of water. Enzymes for applications like wetting, dye wash off, bleaching any sort of dye etc., are in the advanced stage of development. If enzymes can do such wonders, let us first understand WHAT ARE ENZYMES? AND WHY WE NEED THEM?

WHAT ARE ENZYMES?

Enzymes are proteins. Like other proteins, enzymes consist of long chains of amino acids held together by peptide bonds. They are present in all living cells, where they perform a vital function by controlling the metabolic processes whereby nutrients are converted into energy and fresh cell material. Furthermore, enzymes take part in the breakdown of food material into simpler compounds. Some of the best known enzymes are those found in the digestive tract where pepsin, trypsin and peptidases breakdown proteins into amino acids, lipases split fats into glycerol and fatty acids, and amylases breakdown starch into simple sugars.

Enzymes are basically biocatalysts. Enzymes are capable of performing these tasks because, unlike food proteins such as egg albumin, gelatine or soya protein, they help to catalyze reactions. This means that by their mere presence, and without being consumed in the process, enzymes can speed up chemical processes that would otherwise run very slowly, if at all.

ENZYME PROPERTIES

1. Enzymes are specific

Contrary to inorganic catalysts such as acids, bases, metals and metal oxides, enzymes are very specific. In other words, each enzyme can breakdown or synthesize one particular compound. In some cases, they limit their action to specific bonds in the compound with which they react. Most proteases, for instance, can breakdown several types of protein, but in each protein molecule only certain bonds will be cleaved depending on which enzyme is used.

2. Enzymes are very efficient catalysts

For example, the enzyme catalase, which is found abundantly in the liver and in the red blood cells, is so efficient that in one minute one enzyme molecule can catalyze the breakdown of five million molecules of hydrogen peroxide to water and oxygen.

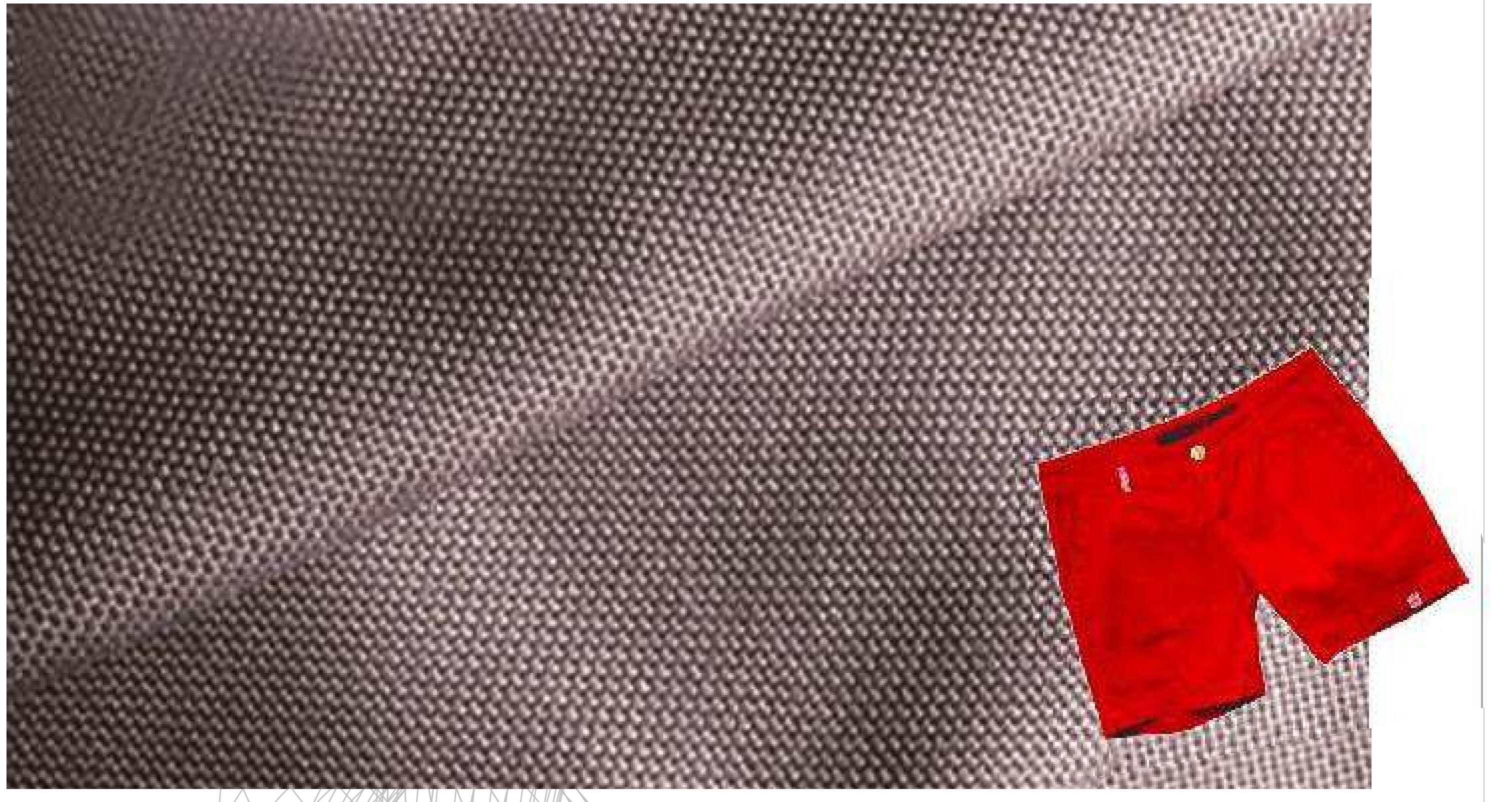
3. Origin - natural source

Enzymes are present in all biological systems. They come from natural systems and when they are degraded, the amino acids of which they are made of can be readily absorbed back into nature.

4. Enzymes work only on renewable raw material

Fruits, cereals, milk, fats, meat, cotton, leather and wood are some typical candidates for enzymatic conversion in industry. Both the usable products and the waste of most enzymatic reactions are non-toxic and readily broken down.

Need of **enzymes** in the textile industry



The textile processing industry is classified into Textile Fabric Processing and Wet Garment Processing. To give various finishes to fabric different auxiliaries, polymers, dyes, specialty chemicals are being used abundantly. These chemicals are hazardous in nature. Many of them are carcinogenic and very dangerous to handle, putting life at risk. Most of them are not eco-friendly. Due to such hazards, the government and regulatory bodies are banning textile processing using such chemicals or the release of waste liquor into nature. The use of enzymes is the ONLY feasible solution to counteract such problems faced by industrialists today.

Enzymes can be used to replace hazardous chemicals, thus saving energy and preventing pollution. They are also highly specific, which means fewer unwanted side effects and creation of by-products in the production process. Enzymes themselves are biodegradable, so they are readily absorbed back into nature.

CONVENTIONAL PROCESSES AND ENZYMES IN THE TEXTILE INDUSTRY

a. Enzymatic desizing of cotton fabric

Although many different compounds have been used to size fabric over the years, starch has been the most common sizing agent for more than a century and this is still the case today, though use of PVA and gums is on the rise. After weaving, the size must be removed to prepare the fabric for the finishing steps of bleaching or dyeing. Enzymes are used for desizing woven fabrics because of their highly efficient and specific way of desizing without harming the yarn.

As an example, desizing on a jigger is a simple method where the fabric from one roll is processed in a bath and re-wound on another roll. First, the sized fabric is washed in hot water (80-95°C) to gelatinise the starch. The desizing liquor is then adjusted to pH 5.5-7.5 and a temperature of 60-80°C depending on the enzyme. The fabric then goes through an impregnation stage before the amylase is added. Degraded starch in the form of dextrins is then removed by washing at 90-95°C for two minutes. The jigger process is a batch process.

By contrast, in modern continuous high-speed processes, the reaction time for the enzyme may be as short as 15 seconds. Desizing on pad rolls is continuous in terms of the passage of the fabric. However, a holding time of 2-16 hours at 20-60°C is required using low-temperature alpha-amylases before the size is removed in washing chambers. With high-temperature amylases, desizing reactions can be performed in steam chambers at 95-100°C or even higher temperatures to allow a fully continuous process.

b. Scouring with enzymes

Before cotton yarn or fabric can be dyed, it goes through a number of processes in a textile mill. One important step is scouring - the complete or partial removal of the non-cellulosic components of native cotton such as waxes, pectins, hemicelluloses and mineral salts as well as impurities such as machinery and size lubricants. Scouring gives a fabric high and even 'wettability' that can be bleached and dyed successfully. Today, highly alkaline chemicals such as sodium hydroxide are used for scouring. These chemicals not only remove the impurities but also attack the cellulose, leading to a reduction in strength and loss of weight of the fabric. Furthermore, the resulting wastewater has a high COD (chemical oxygen demand), BOD (biological oxygen demand) and salt content.

The use of an alkaline pectinase reduces the environmental impact, and in most cases, works out to be more economical. The new process is called Biopreparation. It has been welcomed because rinsing water can be reduced by more than half compared to the traditional processes. This is especially advantageous since hot rinsing water is used, in effect considerable amounts of energy can be saved. Quality improves too. The enzymatic treatment leaves the cellulose structure almost intact, so it reduces weight loss and strength loss. Bio-scouring has a number of potential advantages over traditionally prepared textiles. It reduces total water consumption by around 25 per cent; the treated yarn/fabrics retain their strength properties; and weight loss is much less as compared to the traditional way of processing. Bioscouring also gives softer cotton textiles.

c. Enzymes for denim finishing

Most denim jeans or other denim garments are subjected to a wash treatment to give them a slightly worn look. In the traditional stonewashing process, the blue denim is faded by use of pumice stones. This causes high wear and tear of machines.

However, too much abrasion can damage the fabric, particularly hems and waistbands. This is why denim finishers today use cellulases to accelerate the abrasion by loosening the indigo dye on the denim. Since a small dose of enzyme can replace several kilograms of stones, the use of fewer stones results in less damage to garments, less wear on the machines, and less pumice dust in the working environment. Productivity can also be increased through laundry machines containing fewer stones but more garments. With a stone-free process, the need for the removal of dust and small stones from the finished garment is reduced. There is also no sediment in the wastewater, which can otherwise block drains.

Denim garments are dyed with indigo, which adheres to the surface of the yarn. The cellulase molecule binds to an exposed fibril (bundles of fibrils make up a fibre) on the surface of the yarn and hydrolyses it, but leaving the interior part of the cotton fibre intact. When the cellulases partly hydrolyse the surface of the fibre, the indigo is partly removed and light areas are created. This is also known as the SALT AND PEPPER EFFECT.

Both neutral cellulases acting at pH 6-8 and acid cellulases acting at pH 4-6 are used for the abrasion of denim. There are a number of cellulases available, each with its own special

properties. These can be used either alone or in combination in order to obtain a specific look. Practical, ready-to-use formulations containing enzymes are available.

Application research in this area is focussed on preventing or enhancing backstaining depending on the style required. Backstaining is defined as the redeposition of released indigo onto the garments. This effect is very important in denim finishing. Backstaining at low pH values (pH 4-6) is relatively high, whereas it is significantly lower in the neutral pH range. Neutral cellulases are therefore often used when the objective is minimal backstaining.

Enzymes have opened up new possibilities in denim finishing by increasing the variety of finishes available. For example, it is now possible to fade denim to a greater degree without running the risk of damaging the garment. The denim industry is driven by fashion trends. The various cellulases available for modifying the surface of denim give fashion designers a pallet of possibilities for creating new shades and finishes. The combination of new looks, lower costs, shorter treatment times and less solid waste has made abrasion with enzymes the most widely used fading process today. Incidentally, since denim fabric is always sized, the complete process also includes desizing of the denim garment.

d. Cellulases for the Biopolishing of cotton fabric and lyocell

Cotton and other natural and man-made cellulosic fibres can be improved by an enzymatic treatment called Biopolishing. The main advantage of Bio-polishing is the prevention of pilling. Cellulases hydrolyse the microfibrils (hair or fuzz) protruding from the surface of yarn because they are most susceptible to enzymatic attack. This weakens the microfibrils, which tend to break off from the main body of the fibre and leave a smoother yarn surface. A ball of fuzz is called a 'pill' in the textile trade. These pills can present a serious quality problem since they result in an unattractive, knotty appearance. After biopolishing, the fabric shows a much lower pilling tendency. Other benefits of removing fuzz are a softer, smoother feel and superior colour brightness. Unlike conventional softeners, which tend to be washed out and often result in a greasy feel, the softness-enhancing effects of biopolishing are washproof and non-greasy.

For cotton fabrics, the use of biopolishing is optional for upgrading the fabric. However, biopolishing is almost essential for the new polynosic fibre lyocell (the leading make is known by the trade name Tencel®). Lyocell is made from wood pulp and is characterised by a tendency to fibrillate easily when wet. In simple terms, fibrils on the surface of the fibre peel up. If they are not removed, finished garments made with lyocell will end up covered in pills. This is the reason why lyocell fabric is treated with cellulases during finishing. Cellulases also enhance the attractive, silky appearance of lyocell. Lyocell was invented in 1991 by Courtaulds Fibres (now Acordis, part of Akzo Nobel) and at the time was the first new man-made fibre created in 30 years.

e. Enzymes for wool and silk finishing

The biopolishing of cotton and other fibres based on cellulose came first, but in 1995 enzymes were also introduced for the biopolishing of wool. Wool is made of protein and so this treatment features a protease that modifies the wool 18 fibres. 'Facing up' is the trade term for the ruffling up of the surface of wool garments by abrasive action during dyeing. Enzymatic treatment reduces facing up, which significantly improves the pilling performance of garments and increases softness.

Proteases are also used to treat silk. Threads of raw silk must be degummed to remove sericin, a proteinaceous substance that covers the silk fibre. Traditionally, degumming is performed in an alkaline solution containing soap. This is a harsh treatment because the fibre (the fibrin) is also attacked. However, the use of proteolytic enzymes is a better method because they remove the sericin without attacking the fibrin. Tests with high concentrations of enzymes show that there is no fibre damage and the silk threads are stronger than with traditional treatments.

Specialty Products by **Advanced Enzymes**

a. Enzymatic desizing of cotton fabric

Although many different products are available in the market place for desizing, however desizing efficiency is the key parameter. Desizing is the heart of all textile processes and inefficient desizing leads to poor finishing subsequently. Advanced Enzymes introduces RAPIDENZ series of enzymes offering desizing efficiency as high as 8.5 tegava.

Desizing in continuous flash steam process for denim is also offered by Advanced Enzymes.

b. Scouring with enzymes

Conventional scouring enzymes offered in the market place are alkaline pectinases. Advanced Enzymes with its application research has come out with two enzyme solutions i.e. ADDSCOUR L and ADDSCOUR P, which are capable of even removing other impurities like wax, gums, glues, ash etc. from cotton thereby increasing absorbency further while also offering the advantage of a zero caustic process. With process parameter alteration and different combinations, ADDSCOUR does give a whiteness index equivalent to the chemical process.

c. Enzymes for denim finishing

The key strength of Advanced Enzymes is CELLULASE and its applications. Cellulase does find significant application in textile processing.

For denim garment processing, Advanced Enzymes offers a completely cool process. This is achieved with COOL TYPE AMYLASES and COOL TYPE CELLULASES. Options are available for acid and neutral pH processing. Products like RAPIDENZ COOL and ADDCOOL are being used for this process.

The cool process not only saves energy but also imparts excellent finish to the garments.

d. Cellulases for the Biopolishing of cotton fabric and lyocell

Innovation in the fashion industry and hybridization of cottonseeds made cotton or lyocell too weak, and made it more susceptible to pilling tendency, colour loss with greater impurities. The demand for high colour retention with the bald effect after processing is on the rise. Unless a specific cellulase is designed to do this, such effects cannot be achieved.

Advanced Enzymes offers ADDCOOL, SEBRITE and DENIBRADE series of products to impart such finishes. Products are available in acid and neutral pH processing at ambient or high temperatures.

Dipak Roda
G.M. - Marketing

Advanced Enzymes Product List for the Textile Industry

Sr. No.	Product Name	Application
1.	RAPIDENZ COOL	Low temperature fungal amylase for desizing of cotton and denim
2.	SUPERSIZE XS	Medium temperature amylase for desizing of cotton and denim
3.	RAPIDENZ HT 40L	Thermostable bacterial amylase for desizing of cotton and denim
4.	ADDSCOUR	Alkaline pectinase for bioscouring of woven cotton and knit
5.	DENICELL MD	Economy liquid acid cellulase for biowashing of denim
6.	DENICELL XP	Liquid acid cellulase for biowashing of denim
7.	DENICELL	Concentrated liquid acid cellulase for bio-washing of denim
8.	ADDZYME FDX	Economy acid cellulase for biowashing of denim
9.	FADEX 500	Aggressive acid cellulase for biowashing of denim
10.	DENIBRADE 10 C	Powder acid cellulase for biopolishing of knits to impart the bald effect and to develop high contrast finish on denim
11.	COLDFADE 3535	Room temperature acid cellulase for biowashing of denim
12.	SEBRITE PRIME	Economy acid cellulase for biopolishing of cotton and denim
13a.	SEBRITE BP	Aggressive acid cellulase for biopolishing of cotton and denim with increased colour retention
13b.	SEBRITE BP +	Concentrated acid cellulase for biopolishing of woven garments
14.	NEUTRASTONE NC PLUS	Aggressive neutral cellulase for biowashing of denim to impart a blackish tone and prevent backstaining
15.	NEUTRASTONE NC ULTRA	Economy neutral cellulase for biowashing of denim to impart a greyish black tone
16.	NEUTRASTONE NC	Neutral cellulase for stonewashing of denim
17.	ENZYSTONE N160	Neutral cellulase for stonewashing of denim for a high contrast effect
19.	FADEX HB 2M / 5M	One step desizing and stonewashing enzyme with negligible backstaining
20.	ADDCOOL NC 501L	Low temperature hybrid cellulase for biowashing and biopolishing of cotton and denim
21.	ECOWASH BB	Indigo bleaching enzyme without mediator at neutral pH
22.	INDIGO WASH	Laccasse for bleaching of indigo denim
23.	ADDOX	Catalase for hydrogen peroxide decomposition
24.	ECOWASH DWO	Enzyme based dye washing off agent

Advanced Enzymes New Product Release

Solution from Advanced Enzymes: **FADEX HB 5M**

During the weaving of cotton textiles, the threads are exposed to considerable mechanical strain. Prior to weaving on mechanical looms / machines, warp yarns are often coated with size starch or starch derivatives or CMC / PVA / PAA in order to increase their tensile strength and to prevent breaking. Cotton wax and other lubricants can be applied to yarn in order to increase the speed of cotton weaving. Also waxes of higher melting points are being introduced. Wax lubricants are predominantly triglyceride ester based lubricants.

The Conventional Process

In general, after the textile has been woven, the fabric proceeds to a desizing stage, followed by one or more additional fabric processing steps. Desizing is the act of removing size from textiles and is the heart of textile processing. After weaving, the size coating must be removed before further processing the fabric in order to ensure a homogeneous and wash-proof result. The preferred method of desizing is enzymatic hydrolysis of the size by the action of enzymes.

For the manufacture of denim cloth, the fabric is cut and sown into garments and finished afterwards. In particular, for the manufacture of denim garment, different enzymatic finishing methods have been developed. GOOD finishing of denim garment normally is initiated with an enzymatic desizing step, during which garments are subjected to the action of enzymes in order to remove sizing components, it also provides softness to the fabric and makes the cotton more accessible to the subsequent enzymatic finishing steps. In general, the processing conditions are pH 6 - 8 and temperature 45 - 90 °C depending on the type of desizing enzyme used, with time varying from 20 minutes to one hour. Most of the times, after desizing, the wax either remains or redeposits on the fabric and as a result, the fabric gets darker in shade, gets glossy spots, and becomes stiffer. Good and complete enzyme based desizing products should remove these wax components fully.

Post desizing, the denim garment is subjected to various finishes using different methods. For many years denim jeans manufacturers have washed their garments in a finishing laundry with pumice stones to achieve a soft-hand as well as the fashionable 'stonewashed' look. This abrasion effect is obtained by locally removing the surface bound dyestuff. Cellulase enzymes have been introduced into the finishing process, turning the stonewashing process into a 'biostoning process'. The goal of a biostoning process is to obtain a distinct, but homogeneous abrasion of the garment (stonewash appearance). Biostoning is done at pH 5 - 7 and temperature 35 - 60 °C, with time varying from 30 minutes to two hours depending on process suitability and effects desired. However, uneven stonewashing ('streaks' and 'creases') is very often the requirement. As a consequence repair work ('after-painting') is needed on a major part (up to about 80%) of the stonewashed jeans that have been processed in the laundries. Doing finishing in split stages leads to high water consumption, increased processing time, less production and at times uneven results with streak marks.

The FADEX HB 5M Process

Advanced Enzymes took on the task to design and offer a product and a process to overcome the current problems. And FADEX HB 5M **turned out to be on excellent solution.** FADEX HB 5M provides a one-step process for enzymatically desizing and stonewashing dyed denim. The process comprises of washing denim garments in ONE BATH, saving water, time and energy and giving a finish equal or better than that of the current methods available. FADEX HB 5M has amylase and a SPECIAL streak-reducing cellulase.

FADEX HB 5M process can be carried out in the presence of conventional textile finishing agents, including wetting agents, polymeric agents, dispersing agents etc. A conventional wetting agent may be used to improve the contact between the substrate and the enzymes used in the process. The dispersing agent may suitably be selected from nonionic, anionic, cationic, ampholytic or zwitterionic surfactants. Conventional finishing agents that may be present in a process include (but are not limited to) pumice stones and perlite. Perlite is a naturally occurring volcanic rock. Preferably, heat expanded perlite may be used. Heat

expanded perlite may, for example, be present in an amount of 20-95 w/w % based on the total weight of the composition.

Example:

The following example illustrates the effect of adding FADEX HB 5M (with a streak-reducing cellulase for one step desizing abrasion process) in order to reduce the number of streaks on denim jeans or other garments and to produce denim garment, especially jeans, with a uniformly localized colour variation with reduction in process time cycle and reduced consumption of water.

Lab scale wash trials were carried out with FADEX HB 5M and the conventional process of two stage desizing and cellulase treatment. FADEX HB 5M was treated under the following conditions:

Denim: 14.5 Oz Arvind Denim, 10 legs of approx 100 gms each

Product: FADEX HB 5M

Trial A:

MLR: 1:10 Dose: 1.5 gpl **Time:** 60 - 90 Min

Washing was carried out in rotary drum washer of 5 kg capacity.

Drying: The samples were dried in IFB tumble-dryer.

Evaluation:

Five persons skilled in the art of evaluating denim were asked to grade the denim legs (two legs from each trial, leg '1' and '3' from Trial B of split process, leg '2' and '4' from Trial A of FADEX HB 5M). It was found that the results of FADEX HB 5M were comparable or better over the split process. No issues / problems of streak marks were observed. Moreover, FADEX HB 5M process exhibited considerable water and time saving.

The use of FADEX HB 5M led to a more economical process development with the advantages of lower consumption of water and power, both in short supply in today's scenario. There are more aggressive and novel products under development at Advanced Enzymes for making processes more COST EFFECTIVE and ECO-FRIENDLY.

Enzymes and backstaining

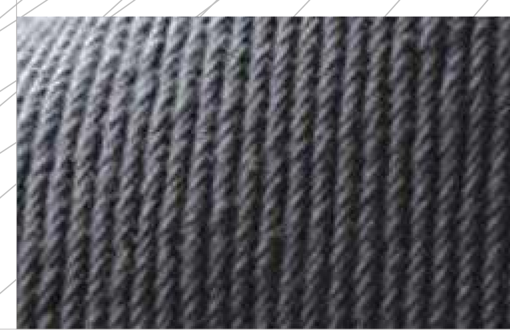
STAINCLEAR is the answer from Advanced Enzymes

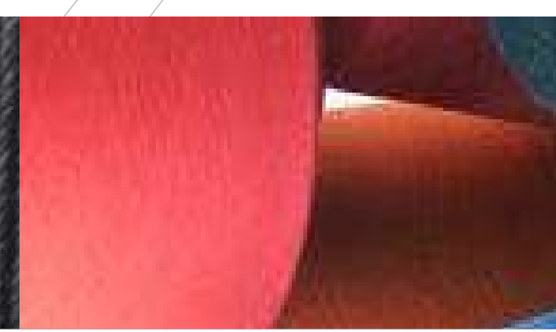
Denim is cotton cloth that has been dyed, usually blue, with the dye indigo. One desirable characteristic of indigo-dyed denim cloth is the alteration of dyed threads with white threads, which upon normal wear and tear gives denim a white on blue appearance. A popular look for denim is the stonewashed look. Traditionally stonewashing has been performed by laundering the denim material in the presence of pumice stone, which results in fabric with a faded or worn appearance with the desired white on blue contrast appearance. Now it is being done using enzymes. This stonewashed look primarily consists of removal of dye to yield a material with areas that are lighter in colour, while maintaining the desirable white on blue contrast, and a material that is softer in texture.

Enzymes, particularly cellulases, are currently used in processing denim. In particular, cellulases have been used as a replacement for or in combination with pumice stones for the traditional 'stonewashing' process used to give denim a faded look. Use of enzymes to stonewash has become increasingly popular because use of stones alone has several disadvantages. For example, stones used in the process cause wear and tear on the machinery, they cause environmental waste problems due to the grit produced and result in high labour costs associated with the manual removal of the stones from pockets of garments. Consequently, reduction or elimination of stones in the wash may be desirable.

However, even though the use of enzymes such as cellulase may be beneficial as compared to stones alone, there are some problems associated with the use of enzymes for this purpose. For example, one problem with some ACID CELLULASES is what could be described as an incomplete removal of dye caused by 'redeposition' or 'backstaining' of some of the dye back onto the fabric during the enzymatic stonewashing process. Such backstaining causes blue coloration of the surface, resulting in less contrast between the blue and white threads and abrasion points (i.e. a blue on blue look rather than the preferred white on blue).

The problem of redeposition of dye during stonewashing has been a concern of denim processors. Previous attempts to address the problem include addition of extra anti-redeposition chemicals, such as surfactants or other agents into the cellulase wash to help disperse the loosened indigo dye and reduce redeposition. In addition, denim processors have tried using cellulases with less specific activity on denim along with extra rinsing. This results in additional chemical costs and longer





processing times. Another method to address the redeposition problem includes adding a mild bleaching agent or stain removing agent in the process. This method affects the final shade of the garment and increases processing time.

While these methods aid to some limited degree in the reduction of redeposition, the methods are not entirely satisfactory and some objectionable backstaining remains. Use of enzymes and stones together may be advantageous in overcoming this redeposition problem, however, it leaves the processor with some of the problems associated with the use of stones. Based on the shortcomings of previously attempted methods, there was a need for a more environmentally friendly and more cost-effective method to address the issue of backstaining of dye during stonewash treatment.

Accordingly, Advanced Enzymes directed its research team to find an enzymatic composition or method that would enhance the removal of the dye during stonewashing where Backstaining Celluloses or Acid Cellulases are use. To overcome the problems, the Advanced Enzymes' team came up with STAINCLEAR, an eco-friendly backstaining removing (not preventing) enzyme-based solution.

Users have found that treatment of cotton indigo-dyed denim with an effective amount of a Backstaining Cellulase and added STAINCLEAR improved backstaining over only Acid Cellulases or Backstaining Cellulases. The result of this treatment with such a composition is an improvement in the contrast between white and blue threads, achieving more complete stained dye removal (more like that achieved with pumice stones). The improvement in the contrast is due to specific bond breaking by STAINCLEAR between cotton and indigo developed during cellulose treatment of denims and also the deterioration of cellulose specific proteins responsible for backstaining.

One of the Advanced Enzymes lab demonstration details are as follows:

A lab scale 5 kg garment washing machine was used for the trials. Approximately 1 kg of desized denim garment was placed in the machine. The machine was filled with 20 L hot water and brought it to 55 °C. pH was adjusted to 4.5 using acetic acid. Once pH was established, Acid Cellulase DENICELL (AETL BRAND) was added at a rate of 1 ml of product/L of wash liquor. STAINCLEAR was used in two ways in two different sets of trials. In the first trial, STAINCLEAR was used along with DENICELL (treatment time 60 min) and in the second trial, STAINCLEAR was used in the rinsing bath after DENICELL treatment (15 min). Dose used was 1 - 1.5 gpL. After this, the bath was dropped, rinsed and finished.

It was observed that the denims treated with STAINCLEAR had better glaze, abrasion and whiteness compared to denims treated ONLY with DENICELL. STAINCLEAR used in the same bath with cellulase yielded better backstaining and lesser abrasion. Whereas STAINCLEAR used in the rinsing bath gave higher abrasion and marginally lesser backstain removal. But both the cases were better than chemical based backstaining removing chemicals. The documented data and treated swatches shall be available on request.

STAINCLEAR was an effective solution to problems occurring during the cellulase treatment, primarily for backstaining issues without affecting too much the base indigo shades.

Hence, the ADVANTAGES of using STAINCLEAR are clear:

1. Removal of backstaining rather than preventing it. Hence one can use cellulases freely for denim washing

2. Eco-friendly product

3. Increased abrasion may lead to better cost to performance ratio

4. Lesser wastage

STAINCLEAR is certainly an answer to hazardous, chemical based backstaining preventing agents.

Bleaching of Indigo Denim

Inspired by modern processing technology and driven by the demand for stylish textile designs produced by means of washing, bleaching and dyeing, efforts are ongoing to vary denim garments and produce, for example, a worn look. Denim is a warp and weft weaving technique wherein the warp consists of a cotton yarn dyed with a blue dye and the weft consists of an undyed, substantially white cotton yarn. The weft may be pretreated, for example, by extraction with a caustic solution to remove hemicelluloses and seeds. Blue denim, a fabric often used for producing blue jeans, is a three-leaf warp body (K 2s/1), for example, which has a warp which is dyed blue by means of indigo

dye or a combination of indigo dye and sulfur black or sulfur blue dyes mainly on the fabric surface. As examples of sulfur black dyes typically employed mention is made of Ultra Black and Indigo Black. As an example of sulfur blue dyes typically employed mention is made of Indigo Blue. When the combination of indigo and sulfur dyes is employed to produce the blue dyed warp, the sequence of dying the yarn is spoken of as sulfur bottom dyed yarn (sulfur dye being applied first) or sulfur top dyed yarn (sulfur dye being applied after the indigo dye). The substantially white weft is visible on the underside of the fabric in contrast to the blue dyed fabric topside. Industrial laundries have until now attempted to produce stylish textile designs by employing various techniques including mechanical methods, such as stonewashing, and/or chemical methods, such as chemical and enzyme-washing.

A. Chemical Method

Finished ready-to-wear garments are turned inside out and pre-washed and/or desized. The garments are then removed from the washing machine, turned right side out and washed in a suitable machine with calcareous sandstone (pumice stone) in a weight ratio of 1:3, i.e. 1 kg of garment to 3 kg of stone. The garment is then removed

from the machine, the stones are removed, and the garments are bleached with sodium hypochlorite to produce a desired shade of colour. This bleaching process is done where the fabric is treated with sodium hypochlorite at 60 $^{\circ}$C and pH 11 - 12 for up to 20 minutes, followed by a neutralisation step and a rinsing. Use of hypochlorite is undesirable, both because chlorite itself is undesirable and because the neutralisation subsequently generates high amounts of salts leading to disposal and pollution problems like increase in BOD and COD levels in effluent and subsequent effluent processing cost.

Chlorine Free Bleaching methods / products are also developed and are commercially available. The steps followed for bleaching by this method is:

a. Placing denim textile material in water and heating to 75 $^{\circ}$C.
b. Adding to the water a dispersing agent which is effective to retard deposition of dyestuff stripped from the warp yarn during bleaching onto the weft yarn and which is comprised of polymers.
c. Bleaching the denim textile material by adding to the water a bleaching solution, typically hydrogen peroxide which is aqueous and alkaline. Alternatively, bleaching agent which is selective for the indigo dye or the indigo derivative dye of the warp yarn and which is selected from the group consisting of formamidine sulfinic acid, at least one reducing carbohydrate, and mixtures thereof is used for bleaching.

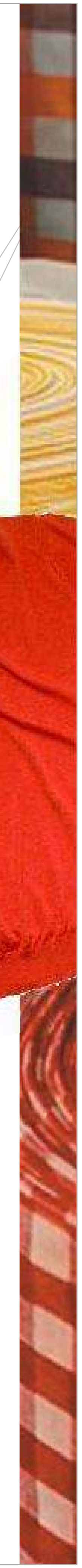
The typical process here is to treat denim similar to hypochlorite based products with only difference in the product composition.

All chemical based bleaching processes lead to high processing time, lot of hazardous waste coming out adding to effluent cost.

B. Enzyme Method - BIOTECHNOLOGY

Modern society expects biotechnology to be the answer for many worldwide problems like depletion of energy sources, incurable illnesses and pollution, among other problems. Industrial use of biotechnology, known as white biotechnology, is bringing about new products and processes aimed at the use of renewable resources, as well as the application of green technologies with low energy consumption and environmentally healthy practices.

Textile processing is a growing industry that traditionally has used a lot of water, energy and harsh chemicals, starting from pesticides for cotton growing to high amounts of wash waters that result in waste streams



causing high environmental burdens. As textile fibres are polymers, the majority being of natural origin, it is reasonable to expect there would be a lot of opportunities for the application of white biotechnology for textile processing. Enzymes - nature's catalysts - are the logical tools for development of new biotechnology-based solutions for textile wet processing.

Enzyme based products are the biotechnological alternative to chemical based bleaching of indigo denim. Here also, two categories are available:

I) Laccase based systems / products

II) ECOWASH BB - A product first developed for commercially use in the world by Advanced Enzymes.

I) Laccase Systems

Laccases (EC. 1.10.3.2 p-benzenediol: oxygen oxidoreductase) belong to a family of multi-copper oxidases. Laccases are widely occurring enzymes in higher plants, fungi, some insects and bacteria. They are characterised by low substrate specificity, oxidising various substrates, including diphenols, polyphenols, different substituted phenols, diamines, aromatic amines, and even inorganic compounds like iodine. Laccases oxidize their substrates by a one-electron oxidation mechanism, and they use molecular oxygen as an electron acceptor. Among Laccases the primary sequence, induction mechanism, physico-chemical (e.g. isoelectric point and carbohydrate content) and biochemical characteristics are variable. The copper binding sites of Laccases are, however, strictly conserved.

In order to create a successful bio-based pretreatment for denims, Laccase was introduced first for enzymatic bleaching process. The group of enzymes called Laccases or Phenol Oxidases possesses the ability to catalyze the oxidation of a wide range of phenolic substances, including indigo. Laccases alone are not effective as bleaching agents, but need to be applied with a mediator molecule, which is the actual substrate of the Laccase and which mediates electron transfer from, for example, indigo to molecular oxygen. Laccase mediator systems have been used to reduce backstaining, enhance abrasion levels and bleach indigo in denim processing. However, it has not been possible to show bleaching effects with a Laccase mediator system on greige cotton.

Commercially available Laccase are applied on denims at pH 4.5 - 5.0 and temperature 50 - 60 °C at dose level of 1 - 4% OWG and MLR of 1: 5 - 1:8. Treatment time depends on the amount of fading required. But it may vary from anything between 15 to 45 minutes. Laccase has limitations to use. It has to be used after cellulase treatment or pumice stone treatment for effective bleaching effect. Direct use after desizing will lead to non-effective fading. Cost to performance ratio is another limiting factor as of now. So there is need for further improvement in Laccase for Indigo Denim bleaching.

II) ECOWASH BB

Driven by more and more demand from our clients for eco-friendly processes, limitations in the available tools to bleach indigo and with the mission to provide ecosafe solutions, Advanced Enzymes has developed an enzyme and non-mediator based enzyme solution: ECOWASH BB to bleach indigo denim.

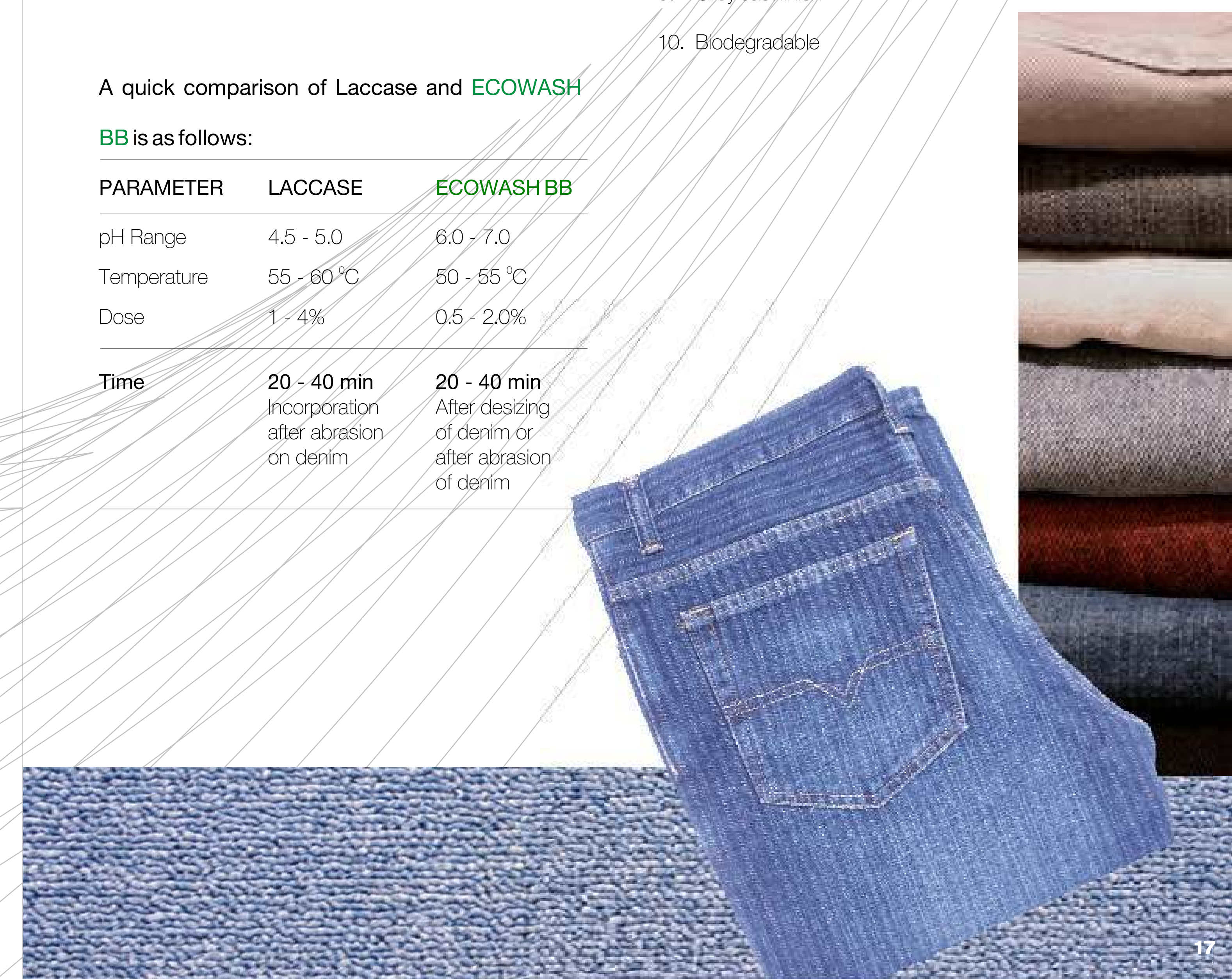
ECOWASH BB is a unique blend of several enzymes from the class of oxidoreductase, works in neutral pH range of 6.0 - 7.0 and temperature of 50 - 55 °C.

A quick comparison of Laccase and ECOWASH BB is as follows:

PARAMETER	LACCASE	ECOWASH BB
pH Range	4.5 - 5.0	6.0 - 7.0
Temperature	55 - 60 °C	50 - 55 °C
Dose	1 - 4%	0.5 - 2.0%
Time	20 - 40 min Incorporation after abrasion on denim	20 - 40 min After desizing of denim or after abrasion of denim

Advantages of using Ecowash BB:

1. Faster action time over Laccase
2. Effective for longer durations in aqueous media over Laccase mediator system
3. Cleaner look and raised grainy effect on denims
4. Reduction in the use of acetic acid
5. Low energy cost
6. Increase in production
7. Can be used directly after desizing, eliminating the need of cellulase use in selective washes
8. Less weight loss of denim
9. Grey cast finish
10. Biodegradable



Technology & **the People**

Advanced Enzymes is driven by its people and technology. Watch for details of new product developments - neutral cellulase for biopolishing and ambient temperature neutral cellulase for high contrast denim finishing in the next issue.

Advanced Enzymes products soon to be launched

a. Microbiopolishing
b. Wetting Enzymes
c. Dye Wash Off Enzymes
d. Softening Enzymes

A better life. A better world. With Enzymes.

It is proven that enzymes have immense potential to replace hazardous chemicals in the processing industry. Advanced Enzymes has a dedicated team of application specialists and enzymologists who are working day in and out to design innovative, eco-safe products to replace hazardous chemicals in textile processing. At Advanced Enzymes, enzyme is indeed life, and we are committed to making life better for all with enzymes.

In the **News**

Enzyme **Quiz**

Choose the answer which BEST completes the following statements or answer the following questions. Circle the number of the answer which is correct. (3 pts.)

1. An enzymatic reaction that occurs at 20°C would be expected to double its rate at a minimum temperature of...
   (1.) 21°C
   (2.) 30°C
   (3.) 40°C
   (4.) 10°C
2. A substrate cannot bind to an enzyme if the enzyme has...
   (1.) an attached cofactor
   (2.) a competitive inhibitor in its active site
   (3.) an activating modulator in its allosteric site
   (4.) an attached coenzyme
   (5.) all of the previous
3. Noncompetitive inhibitors render the enzyme helpless by...
   (1.) altering their shape, making their active site inoperable
   (2.) binding to their proposed substrate
   (3.) Filling their active site
   (4.) denaturing their proposed substrate
   (5.) destroying their allosteric sites
4. Under what conditions will an allosteric enzyme bind to a substrate?
   (1.) when an inhibitor binds to its active site
   (2.) when an activating modulator binds to its allosteric site
   (3.) when an activating modulator binds to its active site
   (4.) whenever their is sufficient substrate
   (5.) when any modulator joins its active site
5. In negative feedback mechanisms, when excess metabolic end products fill allosteric enzyme sites...
   (1.) more enzymes are created
   (2.) the metabolic reaction is halted
   (3.) they behave as activating modulators
   (4.) metabolic activity is increased
   (5.) the enzymes are denatured
6. According to the induced-fit hypothesis, the active site of an enzyme...
   (1.) is rigid and inflexible
   (2.) is precisely tailored to fit a substrate perfectly
   (3.) may change its shape to fit any substrate
   (4.) binds to any substrate
   (5.) fits a specific substrate imperfectly, creating a stressed situation
7. Enzymes accelerate biochemical reactions by...
   (1.) raising the energy of activation
   (2.) altering the direction of the reaction
   (3.) raising cellular temperatures
   (4.) changing equilibrium concentrations
   (5.) lowering activation energy
8. Cyanide bonds easily to the metallic portion of cytochrome molecules (respiratory enzymes). Which concept does this most directly illustrate?
   (1.) Steric hindrance
   (2.) Competitive inhibition
   (3.) Induced fit
   (4.) Noncompetitive inhibition
   (5.) Entropy

Base your answers to questions 9 and 10 below on reading the passage that follows and on your knowledge of biology.

A student ground 1 gram of fresh liver in a mortar, placed the ground liver in a test tube, and added 1 ml of peroxide. The gas that was generated was collected. A glowing splint burst into flames when placed in the gas. The student then repeated the procedure, using one gram of boiled liver and one gram of liver treated with a strong acid. When peroxide was added to each sample of liver, no gas was generated.

9. The gas that was generated was most likely...
   (1.) Oxygen
   (2.) Nitrogen
   (3.) Carbon Dioxide
   (4.) Hydrogen
   (5.) Ammonia
   (6.) Water Vapor
10. If the substance in the liver that acted on the peroxide was an enzyme, it could...
   (1.) be recovered from the living tissue that had not been boiled or treated with acid after the reaction ceased
   (2.) not be recovered because it was consumed while engaging in its catalytic reaction activity
   (3.) not be recovered because there is no enzyme in liver that catalyzes the breakdown of peroxide
   (4.) not be recovered because grinding would break up the molecule
   (5.) be recovered only before the peroxide was added