II International Conference Industrial Engineering and Environmental Protection 2012 (IIZS 2012)
October 31st, 2012, Zrenjanin, Serbia

BIOTECHNOLOGICAL APPLICATIONS OF PECTINASES IN TEXTILE PROCESSING AND BIOSCOURING OF COTTON FIBERS

Kiro Mojsov[1]
[1]University "Goce Delcev" Stip, Technological-technical Faculty, R. Macedonia
e-mail: kiro.mojsov@ugd.edu.mk

Abstract: This work represents a review of applications of alkaline pectinases in textile processing and bioscouring of cotton fibers, the nature of pectin and pectic supstances, and production of alkaline pectinases from various microorganisms. Over the years alkaline pectinases have been used in several industrial processes, such as textile and plant fiber processing, paper and pulp industry, oil extraction, coffee and tea fermentations, poultry feed and treatment of industrial wastewater containing pectinacious material. The use of enzymes in the textile chemical processing is rapidly gaining globally recognition because of their non-toxic and eco-friendly characteristic with the increasingly important requirements for textile manufactures to reduce pollution in textile production. Furthermore, the use of *pectinases* in conjunction with *amylases, lipases, cellulases* and other *hemicellulolytic* enzymes to remove sizing agents is attractive because enzymes are highly specific and efficient, and work under mild conditions, that results in reduced the use of harsh chemicals in the textile industry, process times, energy and water savings and improved product quality.
Key words: pectinases, application, textile processing, eco-friendly characteristics

INTRODUCTION

Textile processing is a growing industry that traditionally has used a lot of water, energy and harsh chemicals that result in waste streams causing high environmental burdens. With the increasingly important requirement for textile industries to reduce pollution in textile production, the use of enzymes in the chemical processing of fibres and textiles is rapidly gaining wider recognition because of their non-toxic and eco-friendly characteristics. Enzymes were discovered in the second half of the nineteenth centure, and since are routinely used in many environmentally friendly and economic industrial sectors. There is increasing demand to replace some traditional chemical processes with biotechnological processes involving microorganisms and enzymes such as *pectinases, xylanases, cellulases, laccases and ligninases* [3], [5], [10].
Today enzymes have become an integral part of the textile processing. There are two enzyme applications in the textile industry. Firstly, in the preparatory finishing area *amylases* are used for desizing process and secondly, in the finishing area *cellulases* are used for softening, bio-stoning and reducing of pillink propensity for cotton goods. Applications of *pectinases, lipases, proteases, catalases, xylanases* etc., included fading of denim and non-denim, bio-scouring, bio-polishing, wool finishing, peroxide removal, decolourization of dyestuff, etc.
Researchers have tried to apply enzymes into every step of textile wet processing, ranging from pretreatment, bleaching, dyeing to finishing, and even effluent treatment. Some applications have become well established and routine, while some have not yet been successfully industrialized due to technical or cost constraints. A famous example is bioscouring or biopreparation, a process that specifically targets noncellulosic impurities within the textile fabrics, with *pectinases* [27].
For fabrics made from cotton or blends, the warp threads are coated with an adhesive substance know as "size", to prevent the threads breaking during weaving. Although many different compounds have been used to size fabrics, starch and its derivatives have been the most common sizing agent. Starch is widely used as a sizing agent, being readily available, relatively cheap and based on natural, sustainable raw materials [22] . After weaving, the size must be removed again in order to prepare the fabric for dyeing and finishing. This process (desizing) must be carried out by treating the fabric with chemicals such as acids, alkali or oxidizing agents. The chemical treatment was not totally effective in removing the starch (which leads to imperfections in dyeing) and also results in a degradation of the cotton fiber resulting in destruction of the natural soft feel, or hand, of the cotton. However starchbreaking enzymes are preferred for desizing due to their high efficiency and specific action. Using amylase enzymes for the removal of starch is one of the oldest enzyme applications [14]. The use of enzymes such as pectinases in conjunction with amylases, lipases, cellulases and other

hemicellulolytic enzymes to remove sizing agents has decreased the use of harsh chemicals in the textile industry, resulting in a lower discharge of waste chemicals to the environment, improving both the safety of working conditions for textile workers and the quality of the fabric.
Before the fabric can be dyed, the applied sizing agent and the natural non-cellulosic materials present in the cotton must be removed (scouring). Conventionally the scouring process carried out by treating the fabric with caustic soda and sodium hydroxide at 70 °C to 90 °C. The use of traditional strongly alkaline process can have a detrimental effect on fabric weight (g/m^2) and on the environment. Enzymatic scouring makes it possible to effectively scour fabric without negatively affecting the fabric or the environment. Hydrolysis by enzymes such as pectinases promotes efficient interruption of the matrix to achieve good water absorbance without the negative side effect of cellulose destruction. This process is called bioscouring. It breaks down the pectin in the cotton and thus assists in the removal of waxes, oils and other impurities. The optimum temperature is 50-65 °C and pH between 7,5-9,0 [26], [32], [34], [38], [40]. The fabric gives better wetting and penetration properties, making subsequent bleach process easy and resultantly giving much better dye uptake.

Structure of cotton

Cotton is the most important of the raw materials for the textile industry. Cotton grows as unicellular fibre on seeds. The mature cotton fibre forms a highly convoluted flat ribbon, varying in width between 12 and 20 μm. Cotton fibres have a fibrillar structure. The primary wall in mature fibres is only 0.5-1 μm thick and contains about 50% of cellulose. Noncellulosic constituents consist of pectins, fats and waxes, proteins and natural colorants. The secondary wall, containing about 92-95% cellulose, is built of concentric layers with alternatic shaped twists. The layers consist of densely packed elementary fibrils, organized into microfibrils and macrofibrils. They are held together by strong hydrogen bonds. The lumen forms the centre of the fibres. Cotton is composed almost entirely of the polysaccharide cellulose. Chemical composition of cellulose is a linear (1→4)-linked polymer of β-D-glucopyranose. The degree of polymerization of cellulose varies with its source and the processing stage of the cellulosic material [23].
The primary wall is about 1 μm thick and comprises only about 1 % of the total thickness of cotton fibre. The major portion of the noncellulosic constituents of cotton fibre is present in or near the primary wall. Noncellulosic impurities, such as fats, waxes, proteins, pectins, natural colorants, minerals and water-soluble compounds found to a large extent in the cellulose matrix of the primary wall and to a lesser extent in the secondary wall strongly limit the water absorbency and whiteness of the cotton fiber [41]. Pectin is located mostly in the primary wall of the fibre. It is composed of a high proportion of D-galacturonic acid residues, joined together by α(1→4)-linkages. The carboxylic acid groups of some of the galacturonic acid residues are partly esterified with methanol. Pectic molecule can be called a block-copolymer with alternating the esterified and the non-esterified blocks. In the primary cell wall pectin is covalently linked to cellulose or in other plants to hemicellulose, or that is strongly hydrogen-bonded to other components. Pectin is like a powerful biological glue. The mostly water-insoluble pectin salts serve to bind the waxes and proteins together to form the fiber`s protective barrier.

Production of pectinases from microorganisms

Commercial sources of enzymes are obtained from three primary sources, i.e., animal tissue, plants and microbies. These naturally occurring enzymes are quite often not readily available in sufficient quantities for food applications or industrial use. However, by isolating microbial strains that produce the desired enzyme and optimizing the conditions for growth, commercial quantities can be obtained. This technique, well known for more than 3,000 years, is called fermentation. Most of the industrial enzymes are produced by a relatively few microbial hosts like *Aspergillus* and *Trichoderma* fungi, *Streptomyces* fungi imperfecti and *Bacillus* bacteria. Yeasts are not good produces of extracellular enzymes and are rarely used for this purpose. There is a large number of microorganisms which produce a variety of enzymes [7], [15]. Microorganisms producing enzymes of textile important are listed Table 1.

Table 1. Microorganisms producing enzymes of textile important

Microorganisms	Enzymes
1. Bacteria	
Bacillus subtilis	*Amylase*
B. coagulans	*α-amylase*
B. licheniformis	*α-amylase, protease*
2. Fungi	
A. niger	*Amylases, protease, pectinase, glucose oxidase*
A. oryzae	*Amylases, lipase, protease*
Candela lipolytica	*Lipase*
P. notatum	*Glucose oxidase*
Rhizopus sp.	*Lipase*
Trichoderma reesei	*Cellulase*
T. viride	*Cellulase*
Ascomycetes	*α-amylase*
Basidomycetes	*α-amylase*
Aspergillus sp.	*Pectinase, lipase*

The enzymes are inducible, i.e., produced only when needed, and they contribute to the natural carbon cycle. Pectolytic enzymes or pectinases are classified according to their activity on the main polygalacturonan backbone chain [35]. Pectinases comprise a group of enzymes that catalyze the breakdown of substrates containing pectin. Pectinases are classified into three classes: pectin esterases, depolymerizing enzymes (hydrolases, lyases), and protopectinases. Some of the alkaline pectinases from microbial sources are listed in Table 2.

Table 2. Microbial sources of alkaline pectinases, properties and applications

Microorganisms	pH (opt.)	T (opt.), ^{o}C	Application	Reference
1. Bacteria				
Bacillus sp. DT-7	8	60	Degumming	Kashyap et al. 2001
Bacillus sp. MG-cp-2	10	60	Degumming	Kapoor et al. 2000
Bacillus sp. NT-33	10.5	75	Degumming	Cao et al. 1992
2. Fungi				
Penicillium italicum	8	50	Food industry	Alana et al. 1991
Aspergillus fumigatus	3-9	65	Degumming	Baracat et al. 1993
Amycolata sp.	10.25	70	Degumming	Bruhlmann et al.1994
Streptomyces sp.QG-11-3	3-9	60	Biobleaching	Beg et al. 2000

Several methos, such as submerged fermentation (SmF), solid-state fermentation (SSF) and whole cell immobilization have been successfully used for alkaline pectinase production from various microorganisms [12], [18]. The production of alkaline pectinase in SmF cultures is reported to be induced by supplementing the production medium with different nitrogen and carbon sources containing pectinaceous substances such as pectin polymer, cheap agricultural residues such as ramie fiber or leaves, citrus pectin, orange peel, wheat bran rice husk, etc. [5], [9],[18], [36].
SSF is the growth of organisms on solid substrates in systems with continuous gas phase and no free-flowing water. Agro-industrial residues such as wheat bran, rice bran, sugarcane bagasse, corncobs, and apple pomace are generally considered the best substrates for processes [6], [28], [30].
For practical applications, immobilization of microorganisms on solid materials offers several advantages, including repeated usage of enzyme, ease of product separation and improvement of enzyme stability [18].

Properties of enzymes used in textiles

1. **Enzyme accelerates the reaction**
   - An enzyme accelerates the rate of particular reaction by lowering the activation energy of reaction
   - The enzyme remains intact at the end of reaction by acting as catalyst
2. **Enzymes operate under milder condition**
   - Each enzyme have optimum temperature and optimum pH i.e. activity of enzyme at that pH and temperature is on the peak
   - For most of the enzyme activity degrades on the both sides of optimum condition
3. **Alternative for polluting chemicals**
   - Enzymes can be used as best alternative to toxic, hazardous, pollution making chemicals
   - Also some pollutant chemicals are even carcinogenic. When we use enzymes there is no pollution
4. **Enzyme acts only on specific substrate**
   - Most enzymes have high degree of specificity and will catalyse the reaction with one or few substrates
   - One particular enzyme will only catalyse a specific type of reaction. Enzymes used in desizing do not affect cellulose hence there is no loss of strength of cotton
5. **Enzyme are easy to control**
   - Enzymes are easy to control because their activity depends upon optimum condition
6. **Enzymes are biodegradable**
   - At the end of reaction in which enzymes used we can simly drain the remaining solution because enzymes are biodegradable and do not produce toxic waste on degradation hence there is no pollution

APPLICATIONS OF PECTINASES IN TEXTILE PROCESSING

Textile industry uses various chemical agents in the different wet processes. These chemicals, after their use, cause pollution in the efluents, some of them are corrosive that could damage equipment and the substrate. However, by introducing enzymatic processes an environment friendly production can be ensured. The serious wastewater pollution caused by conventional textile finishing has oriented the research towards application of enzymes in textile wet proceses. One of the oldest technology being used today is based on *amylase*-catalysed hydrolysis of the starch size. The advantage of these enzymes is that they are specific for starch, removing it without damaging to the support fabric. An amylase enzyme can be used for desizing processes at low-temperature (30-60 ºC) and optimum pH is 5,5-6,5 [13]. In the last two decades several other enzymatic processes have also been developed for the different wet processing of textile goods in wide-ranging operations from cleaning preparations to finishing processes. *Cellulases, hemicellulases* and *pectinases* (hydrolyses) acting on native cellulosic fibres (cotton, flax, hemp, jute, etc.) became the target enzymes in textile bioprocessing.
Recent results indicate that certain enzymes may be used effectively in the cleaning procedure of cotton. The scientific interest in this process is reflected in the number of papers published during recent years describing biopreparation results obtained, using various enzymes from different sources. But enzymatic biopreparation of cotton represents a fairly new approach and is still mostly in the development stage.
Pectinases catalyse the degradation of pectin. The total degradation is resulted by the harmonized work of several enzymes with different activities. These enzyme are in synergism with each other. There is a nondepolymerase in pectin degrading enzyme system: *pectin esterase*. This enzyme catalyses the cleavage of ester bond of poligalacturonan, as a conseqvence the degree of esterification decreases. Free carboxyl group and methyl alcohol are produced in the reaction [2]. *Polygalacturonase,* which is a depolymerase enzyme, catalyses the cleavage of $\alpha(1\rightarrow4)$ bonds in pectic polymer chain, releasing water and reducing groups at the chain ends. *Exopolygalacturonase* works at the end of the chain, while *endopolygalacturonase* works randomly within the chain [2].

Transeliminase or shortly *lyase* (depolymerase) catalyses the cleavage of α(1→4) bonds in polygalacturonan chain without releasing water and creating a double bond between the C4 and C5 athoms. Endo and exo enzymes work within or at the end of the chain, respectively, similar to the *polygalacturonases* [37].
Cellulases catalyse the degradation of cellulose. All *cellulases* have an identical chemical specificity towards the β(1→4) glycosidic bonds, but they differ in terms of the site of attack on the solid substrates (*exoglucanase* and *endoglucanase*). These enzymes catalyse the hydrolyses of the glycosidic bonds by general acid catalysis [20]. *β-glucosidases* cleave cellobiose and other soluble oligosaccharides to glucose, which is an importan step since cellobiose is an end-product inhibitor of many *cellulases* [20].
Xylanases catalyse the hydrolysis of xylan, the major constituent of hemicellulose. Xylans are heteropolysaccharides with a homopolymeric backbone chain of (1→4)-linked β-D-xylopyranose units. Two types of *xylanases* are distinguished, one is a non-branching, which does not liberate arabinose, while the other is a debranching, which liberates arabinose from the side chain substituents in addition to cleaving main chain linkages. *Endo-* and *exoxylanase* work within or at the end of the chain, respectively [21].

APPLICATIONS OF PECTINASES FOR BIOSCOURING OF COTTON FIBRES

Biopreparation of cotton

Scouring is removal of non-cellulosic material present on the surface of the cotton. Raw cotton contains about 90 % of cellulose and various noncellulosics such as waxes, pectins, proteins, fats, lignin-containinig impurities and colouring matter. The goal of the cotton preparatory process is the remove the hydrophobic and noncellulosic components and produce highly absorbent fibres that can be dyed and finished uniformly. In the conventional energy and chemical intensive process concentrated sodium hydroxid solution and additional hydrogen peroxide and sodium hypochlorite solutions are applied for removing the impurities from greige cotton. The mild reaction conditions offered by enzymatic treatment provide an environmentally friendly alternative. Pectinases, cellulases, proteases and lipases have been investigated most commonly and compared to alkaline scouring.
In generally cellulase and pectinase are combined and used for Bioscouring. In this pectinase destroy the cotton cuticle structure by digesting the pectin and removing the connection between the cuticle and the body of cotton fibre whereas cellulase can destroy cuticle structure by digesting the primary wall cellulose immediately under the cuticle of cotton. But at present, the only commercial bioscouring enzyme products are based on pectinases. Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD) of enzymatic scouring process are 20-45 % as compared to alkaline scouring (100 %). Total Dissolved Solid (TDS) of enzymatic scouring process is 20-50% as compared to alkaline scouring (100%). Handle is very soft in enzymatic scouring compared to harsh feel in alkaline scouring process. Enzymatic scouring makes it possible to effectively scour fabric without negatively affecting the fabric or the environment. It also minimises health risks ence operators are not exposed to aggressive chemicals. Bioscouring process provides many advantages, such as reduced water and wastewater costs, reduced treatment time and lower energy consumption because of lower treatment temperature. Moreover, the weight loss in fabric is reduced, and fabric quality is improved with a superior hand and reduced strength loss. [31].

Bioscouring with pectinases

Pectinase, as the name suggests, hydrolysis pectins that are present in cotton as a non-cellulosic impurity. The best kinds of pectinase are those, which can function under slightly alkaline conditions even in the presence of chelating agents. Such enzymes are called "alkaline pectinases". Most conventional pectinases are usually inactive under these commercially useful conditions, their optimum activity lying in the slightly acidic region.
Bioscouring is a process by which alkaline stable pectinase is used to remove pectin and waxes selectively from the cotton fibre. Unlike the traditional alkaline scouring, this process is substrate-specific and does not alter the cellulose component. The treatment here is also rather lower than that of

the high-temperature alkaline scouring. The bioscouring however does not swell or remove the seed coat fragments called motes. This can be beneficial when scouring for the "natural look", because of the mote and colour retention in the cotton fabrics scoured with this process, pastel or light shades need to be bleached prior to dyeing, but medium to dark shades can be dyed directly after bioscouring. Some of the researchers have reported that pectinase treatment alone results in adequate wettability [11], [17], [25], [38], [39], however, others have found only a little improvement in water absorbency [16]. Yachmenev and his co-workers obtained better absorbency and whiteness after the treatment with alkaline pectinase than with acidic pectinase [40]. During the pectinase treatment the pectin content of cotton fibre can be decreased by about 30 %. Removal of pectin results in lower amounts of waxes on the cotton surface and subsequently in improved water absorbency of the fabric, which supports the hypothesis of chemical linkage between pectin and waxes. The enzymatic treatment has no effect on the tensile strength [11]. Pectin acts as a sort of cement or matrix that stabilizes the primary cell wall of the cotton fibres. The enzymes will degrade pectin during incubation, thereby destabilizing the structure in the outher layers.The weakened outher layers can be removed in a subsequent wash process [29]. Pectinase treatment modifies the morphology of cotton fibres. After pectinase treatment the fibre surface becomes perforated at first, an further treatment results in cellulose fibrils protruding from the surface of the fibre [24].

Waxes have a melting point about 70 ^{o}C, therefore during pretreatment they melt and disperse into the treatment bath or they are redistributed on the fibre surface and the thickness of the fabrics increases. The bioscouring of cotton using pectinase enzyme with multiple mixed surfactant and organic solvent has also been investigated. Three mixed surfactants used were: C12-14 syntetic alcohol with ethylene oxide 5 mol; C12-14 syntetic alcohol with ethylene oxide 9 mol; Coconut alcohol with ethylene oxide 18 mol; The natural product D-limonene has been used as an organic solvent to improve the extraction of wax. It has been found that the addition of small amounts of non-ionic surfactants in the bioscouring solution greatly enhances the effectiveness of the removal of cotton wax without inhibiting the activity of pectinase [33].

Novozymes, Bayer and Dexter Chemical Corporation have introduced an enzymatic alternative for scouring woven and knitted cotton fabrics in the textile industry on the basis of an alkaline pectinase (Dextrol Bioscour 3000) produces by a genetically modified *Bacillus* strain. For cotton knits, enzyme is added in this step. The temperature is brought to 57 °C and held for 10 minutes. This is the actual bioscour part of the procedure. The bath is then heated to 95 °C to melt and emulsify waxes and held for 5 minutes. The scour is followed by at least one 80 °C rince before proceeding for dyeing. The later modifications include reducing the time used for rinsing by skipping the drain step and going directly to another flow wash. Among other modifications, at 50 °C rinse prior to the bioscour process has proved to be effective in helping remove knitting oils and reducing foam levels. For cotton woven fabrics, batch process, pad-batch bioscour as well as continuous bioscouring have also been suggested. The bioscouring process results in textiles being softer than those scoured in the conventional sodium hydroxide process [29].

Recipe for conventional alkaline scouring process

Sodium hydroxide, 20 %

Non-ionic surfactant, 2 g/l

Wetting agent, 1-2 g/l

Temperature, 100 °C

Time, 60-90 mins

Liquor ratio, 20 : 1

Recipe for enzymatic scouring using pectinase

Pectinase, 7-8 g/l (in acetate buffer solution)

pH, 4.0

Temperature, 40 °C

Time, 45 mins

Liquor ratio, 50 : 1

CONCLUSION

Pollution free processes are gaining ground all over the world. Enzymes emerging as the best alternative to the polluting textile processing methods. Enzymes also saving lot of money by reducing water and energy consumption which ultimately reduce the cost of production.
Biotechnology offers a wide range of alternative environmentally-friendly processes for the textile industry to complement or improve the conventional technologies. The use of various enzyme is in the early stages of development but their innovative applications are increasing and spreading rapidly into all areas of textile processing. These enzymatic processes are gives the similar results as that of conventional methods The textile industry can greatly benefit from the expanded use of these enzymes as highly specific and efficient, non-toxic, environmenatally friendly compounds, work under mild conditions (pH, temperature) with low water consumption that results in reduced the use of harsh chemicals in the textile industry, process times, energy and water savings and improved product quality. The conventional alkaline scouring carried out with hot caustic soda is unquestionable an energy, water and chemical-intensive process.
Biopreparation of the cellulosic fibres is an enzyme-aided process by which the noncellulosic "impurities" (waxes, pectic substances, proteins, lignin-containing and colouring materials, etc.) are removed mainly by pectinase rich enzymes.
Advances in enzymology, molecular biology and screening techniques provide possibilities for the development of new enzyme-based processes for a more environmentally friendly approach in textile industry. It seems that in the future it will be possible to do every process using enzymes.

REFERENCES

[1] Alana, A., Liama, M., Serra, J.L., Purification and some properties of the pectin lyase from *Penicillium*. FEBS Lett, Vol. 280, pp.335-350, 1991.
[2] Bailey, M.J., Pessa, E., Strain and Process for Production of Polygalacturonase, Enzyme and Microbial Technology, Vol.12, pp. 266-271, 1990.
[3] Bajpai, P., Application of enzymes in the pulp and paper industry, Biotechnology Progress, Vol.15, pp.147-157, 1999.
[4] Baracat, M.C., Vanetti, M.C.D., Araujo, E.F.D., Silva, D.O., Partial characterization of *Aspergillus fumigatus* polygalacturonases for the degumming of natural fibres, J Ind Microbiol, Vol.11, pp.139-142, 1993.
[5] Beg, Q.K., Bhushan, B., Kapoor, M., Hoondal, G.S., Production and characterization of thermostable xylanase and pectinase from a *Streptomyces sp*. QG-11-3, J Ind Microbiol Biotechnol, Vol.24, pp.396-402, 2000.
[6] Blandino, A., Iqbalsyah, T., Pandiella, S.S., Cantero, D., Webb, C., Polygalacturonase production by *Aspergillus awamori* on wheat in solid-state fermentation, Applied Microbiology and Biotechnology, Vol.58, pp.164-169, 2002.
[7] Boyer, P.D., The enzymes,3rd ed., Academic Press, Inc., New York, Vol.5, 1971.
[8] Bruhlmann, F., Kim, K.S., Zimmerman, W., Fiechter, A., Pectinolytic enzymes from actinomycetes for the degumming of ramie bast fibers, Appl Environ Microbiol, Vol.60, pp.2107-2112, 1994.
[9] Bruhlmann, F., Purification and characterization of an extracellular pectate lyase from an *Amylocota sp.,* Applied and Environmental Microbiology, Vol.61, pp.3580-3585, 1995.
[10] Bruhlmann, F., Leupin, M., Erismann, K.H., Fiechter, A., Enzymatic degumming of ramie bast fibers, Journal of Biotechnology, Vol.76, pp.43-50, 2000.
[11] Buchert, J., Pere, J., Scouring of cotton with Pectinases, Proteases and Lipases, Textile Chemist and Colorist&American Dyestuff Reporter, Vol.32(5), pp.48-52, 2000.

[12] Cao, J., Zheng, L., Chen, S., Screening of pectinase producer from alkalophilic bacteria and study on its potential application in degumming of ramie, Enzyme and Microbial Technology, Vol.14, pp. 1013-1016, 1992.
[13] Cavaco-Paulo A. & Gübitz G. M., Cambridge:, Textile processing with enzyme, Woodhead Publishing Ltd, ISBN 18557366101, 2003.
[14] Ciechańska D., Kazimierczak J., Fibres & Textiles in Eastern Europe, Vol.14, No 1(55), pp. 92-95, 2006.
[15] Fersht, A., Enzyme structure and mechanism, San Francisco: Brenda, W.H., The comprehensive enzyme information system, 50–2. ISBN 0-7167-1615-1 , 2007.
[16] Hartzell, M.M., Hsieh, Y-L., Enzymatic Scouring to Improve Cotton Fabric Wettability, Textile Research Journal, Vol.68(4), pp.233-241, 1998a.
[17] Hartzell, M.M., Hsieh, Y-L., Pectin-Degrading Enzymes for Scouring Cotton, In ACS Symposium Series 687 (eds. Eriksson K. and Cavaco-Paulo A.) Washington, D.C.,pp.212-227, 1998b.
[18] Kapoor, M., Beg, QK., Bhushan B., Singh, K., Dadhich, K.S., Hoondal, G.S., (2001). Production and partial purification and characterization of a thermo-alkali stable polygalacturonase from *Bacillus sp.* MG-cp-2, Process Biochemistry, Vol.36, pp.467-473.
[19] Kashyap, D.R., Vohra, P., Soni, S.K., Tewari, R., Degumming of buel (*Grewia optiva*) bast fibers by pectinolytic enzyme from Bacillus sp. DT7, Biotechnol Lett, Vol.23, pp.1297-1301, 2001.
[20] Koivula, A., Linder, M., Teeri, T.T., Structure-function Relationships in *Trichoderma* Cellulolytic enzymes, In *Trichoderma* and *Cliocladium* Vol.2 Enzymes,Biological Control and Commercial Applications (eds. Harman G.E. and Kubicek C.P.), Taylor&Francis, pp.3-23, 1998.
[21] Kulkarni, N., Shendye, A. and Rao, M, Molecular and Biotechnological Aspects of Xylanases. FEMS Microbiology Review, Vol.23, pp.411-456, 1999.
[22] Lange, N.K., Textile Chemist and Colorist, Vol.29, pp.23-26, 1997.
[23] Lewin, M., Pearce, E.M., Handbook of Fibre Chemistry, Marcell Dekker Inc., New York, 1998.
[24] Li, Y., Hardin, I.R., Treating Cotton with Cellulases and Pectinases: Effects on Cuticle and Fibre Properties, Textile Research Journal, Vol. 68(9), pp.671-679, 1998a.
[25] Li, Y., Hardin, I.R., Enzymatic Scouring of Cotton-Surfactants, Agitation, and Selection of Enzymes Textile Chemist and Colorist, Vol. 30(9), pp.23-29, 1998b.
[26] Losonczi, A., Csiszar, E. Szakacs, G. and Kaarela, D., Bleachability and Dyeing Properties of Biopretreated and Conventionally Scoured Cotton Fabrics, Textile Res. J., Vol.74(6), pp.501-508, 2004.
[27] Lu, H., Insights into Cotton Enzymatic Pretreatment. International Dyer, No.4 10-13, ISSN 0020 – 658X, 2005.
[28] Maldonado, M.C., Saad, A.M.S., Production of pectin esterase and polygalacturonase by *Aspergillus niger* in submerged and solid state systems, Journal of Industrial Microbiology and Biotechnology, Vol.20, pp.34-38, 1998.
[29] Nierstrasz, V.A., Warmoeskerken, M.M.C.G., Process Engineering and Industrial Enzyme Applications. In Textile Processsing with Enzymes (eds. Cavaco-Paulo, A., Gűbitz), Woodhead Publishing Ltd, Cambridge, England, pp.129-131, 2003.
[30] Pandey, A., Selvakumar, P., Soocol, C.R., Nigam, P., Solid state fermentation for production of industrial enzymes, Current Science, Vol.77, pp.149-162, 1999.
[31] Pawar, S.B., Shah, H.D., Andhorika, G.R., Man-Made Textiles in India, 45(4), pp.133, 2002.
[32] Rajendran R, Sundaram SK, Radhai R, Rajapriya P., Bioscouring of cotton fabrics using pectinase enzyme its optimization and comparison with conventional scouring process, Pak J Biol Sci., Vol.14(9) pp.519-25, 2011.
[33] Rossner U, Melliand Textilberichte, Vol.74, pp.144, 1993.; Li Y H and Hardin I R, Textile Chemist and Colourist, Vol.29, pp.71, 1997.
[34] Sawada, K., Tokino, S., Ueda, M. Wang, X.Y., Bioscouring of cotton with pectnase enzyme, Journal of the Society of Dyers and Colourists, Vol.114 (11), pp.333-336, 1998.

[35] Sakai, T., Sakamoto, T., Hallaert, E., Vandamme, E.J., Pectin, pectinase and protopectinase: production, properties, and applications, Advances in Applied Microbiology, Vol.39, pp.213-294, 1993.

[36] Said, S., Fonseca, M.J.V., Siessere, V., Pectinase production by *Penicillium frequentans*, World J Microbiol Biotechnol, Vol.7, pp.607-608, 1991.

[37] Sakamoto, T., Hours, R.A., Sakai, T., Purification, Characterisation and Production of Pectic Transeliminases with Protopectinase Activity from *Bacillus subtilus,*Biosci. Biotech. Biochem., Vol. 58(2), pp.353-358, 1994.

[38] Tzanov, T., Calafell, M., Guebitz, G.M. and Cavaco-Paulo, A., Bio-preparation of cotton fabrics, Enzyme and Microbial Technology, Vol.29, pp.357-362, 2001.

[39] Traore, M.K., Buschle-Diller, G., Environmentally Friendly Scouring Processes, Textile Chemist and Colorist&American Dyestuff Reporter, Vol.32(12), pp.40-43, 2000.

[40] Yachmenev, V.G., Bertoniere, N.R. and Blanchard, E.J., Effect of Sonication on cotton Preparation with Alkaline Pectinase. Textile Res. J., Vol.71(6), pp.527-533, 2001.

[41] Yamamoto, R., Buschle-Diller, G., Takagishi, T., Eco-friendly processing of cotton: application to industrial manufacturing. In: Proceedings of the American Chemical Society National Meeting, April 2001, San Diego, Calif. ACS, Washington D.C., 2001.