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## Biotechnology in Textiles – an Opportunity of Saving Water

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## 1. Introduction

In the last few years biotechnology has been making its way into many areas of industry. Biotechnology is the application of life organisms and their components into industrial processes and products (Warke & Chandratre, 2003). The biological systems that have traditionally been used are organisms such as yeasts, fungi and bacteria. The progress of industrial biotechnology in the last twenty years, especially in molecular biology, protein engineering and fermentation technology, enhanced the development of new uses of enzymes in the food industry, the use spread into the areas of detergents, paper and leather industry, natural polymer modification, organic chemical synthesis, diagnostics ... The use of enzymes experienced an increase in the textile industry as well.

Amylases were the first enzymes applied in textile processing to remove starch-based sizes from fabrics after weaving. Later proteases were introduced into detergent formulations to remove organic protein-based stains from textile garments and cellulases to remove fibrillation in multiple washes. Further applications have been found for these enzymes to produce the aged look of denim and other garments (Gübitz & Cavaco-Paulo, 2001).

Today enzymes offer a wide variety of alternative, environment and fibre friendly procedures which are replacing or improving the existing classical technological procedures. Cellulases, proteases, amylases, catalases, pectinases, peroxidases and lactases are the enzymes that can replace aggressive chemicals (Cavaco-Paulo & Gübitz, 2003).

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 (Lu, 2005).

## 1.1 Cotton fibre

A mature cotton fibre is composed of several concentric layers and a central area called lumen. A cuticle, a primary cell wall, intermediary wall as well as secondary cell wall follow each other from the outer to the inner part of the fibre. The whole cotton fibre contains 88 to 96.5% of cellulose, the rest are uncellulosic substances, called incrusts (Karmakar, 1999). Pectins, waxes, proteins, minerals and other organic substances are classified as uncellulosic substances. The larger part of these substances is found in the cuticle and the primary cell

wall. During the growth of the fibres uncellulosic substances, especially waxes, protect them against the loss of water, insects and other outside influences that might damage the fibres. Furthermore, they also protect them against mechanical damage that can occur as a result of processing.

Row cotton fibres have to go through several chemical processes to obtain properties suitable for use. With scouring, non-cellulose substances (wax, pectin, proteins, hemicelluloses...) that surround the fibre cellulose core are removed, and as a result, fibres become hydrophilic and suitable for bleaching, dyeing and other processing.

Pectin, there is 0.4 to 1.2% of pectin in cotton fibres, acts as an adhesive, a glue between the cellulose and uncellulosic substances. By removing pectin, it is easier to remove all other uncellulosic substances. The processes of bioscouring that are in use today are based on the decomposition of pectin by the enzymes called pectinases.

#### **1.2 Pectin substances**

Pectin substances are generically called the complex polysaccharide macromolecules with high and varying molecular mass (Ridley et al., 2001). They are negatively charged and acidic. The primary chain is composed with  $\alpha$ -(1,4) linked molecules of  $\alpha$ -D-galacturonic acid. The side chains also contain molecules of L-rhamnose, arabinose, galactose and xylose that are connected to the main chain through their first and the second carbon atom. The structural formula of the primary chain of pectin- the polygalacturonic acid is shown in Figure 1.

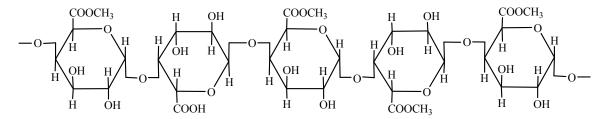


Fig. 1. Structural formula of the polygalacturonic acid.

The carboxyl groups of galacturonic acid are partially esterified by methyl groups and partially or completely neutralized by calcium, potassium, magnesium, iron, ammonium or other ions. Some of the hydroxyl groups on the second and the third carbon atom can be acetylated (Jayani et al., 2005; Kashyap et al., 2001; Gamble, 2003). With the help of electrostatic interactions unesterified or slightly esterified galacturonic groups with negative charge and calcium ions with positive charge form bonds. A calcium ion also bonds pectin with other polysaccharides. It forms a coordination bond between the hydroxyl group of the polysaccharide and an ionic bond with the carboxyl group of pectin. The removal of the calcium ion enhances the decomposition of the pectin substances rich in calcium (Losonczi et al., 2005).

#### 1.3 Enzymes

Enzymes are biological catalysts that accelerate the rate of chemical reactions (Cavaco-Paulo & Gübitz, 2003). The reaction happens with lower activation energy which is reached by forming an intermediate enzyme – substrate. In the reaction itself the enzymes are not used up, they do not become a part of the final product of the reaction, but only change the chemical bonds of other compounds. At the end of the reaction they are released and can participate again in the next biochemical reaction.

All known enzymes are proteins. They therefore consist of one ore more polypeptide chains and display properties that are typical of proteins. Some enzymes require small non-protein molecules, known as cofactors, in order to function as catalysts (Jenkins, 2003).

Generally they are active at mild temperatures. Above certain temperature the enzyme is denaturated. Enzymes have a characteristic pH at which their activity is maximal. Extreme pH values influence on the electrostatic interactions within the enzyme, leading to inactivation of enzyme. Other important factors that influence the effect of enzymatic processes are the concentration of enzyme, the time of treatment, additives like surfactants and chelators and mechanical stress.

Most enzymes are highly specific. They only catalyse a single reaction on a limited number of substrates. Enzymes are distinguished according to the form of the molecule and the charge distribution of the active side. The active side is an area where catalyses occurs and is just a small part of the enzyme. It must provide an environment where the substrate can bond and other molecules do not interfere with catalyses. The specific enzyme action has become known as the 'lock and key' model. The active side of the enzyme, the lock, with an accurately defined rigid structure can only suit a substrate, the key, which is adapted only to it.

Enzymes differ from chemical catalysts in several important characteristics (Cavaco-Paulo & Gübitz, 2003). Enzyme catalysed reactions are several times faster than chemically catalysed ones. Compared to the non-catalysed reaction the rates is from 10<sup>8</sup> to 10<sup>10</sup> higher (Faber, 1995 15). Enzymes have far greater reaction specificity than chemically catalysed reactions and rarely form byproducts. Enzymes catalyse a reaction under mild reaction conditions: the temperature is below 100°C, the atmospheric pressure and a pH of around 7 are needed.

#### **1.4 Pectinases**

The pectinolyic enzymes or pectinases are a heterogeneous group of related enzymes that hydrolise pectin substances present mainly in plants (Jayani et al., 2005). According to an international nomenclature they are classified into the third group (EC3) – hydrolases due to the specifics of the reactions (Holme, 2004).

The pectinases are produced by numerous microorganisms such as bacteria and fungi. Almost all of the commercial products of the pectinases are extracted from fungi, in industrial production of pectinolized enzymes fungi Aspergillus niger (Jayani et al., 2005) are most commonly used. This type of microorganism has a GRAS (Generally Regarded As Safe) status meaning that the produced metabolites are safe for use. Numerous types of from them including polymethylgalacturonases, pectinases can be produced polygalacturonases and pectinesterases. They can also be produced from other types of microorganisms, such as Penicillium frequentans, Mucur pusilus and others. Their stability is best in the pH range between 5 and 6. With the help of genetic changed microorganisms the alkaline pectinases were produced, they are active in the pH range between 8 and 9. The leading producers of the commercial products of pectinases are Novozymes (Netherlands), Novartis (Switzerland), Roche (Germany) and Biocon (India) (Gummadi & Panda, 2003 16). Products of pectinases are found under different names. Acid pectinases are marketed under the following commercial names: Forylase KL - Cognis, Viscozyme 120 L -Novozymes, Pectinase P9179, Pectinase p3026 - Sigma Chemical Co., Pectinase 62L -Biocatalysts, Multifect pectinase PL - Genencor International; and alkaline pectinases under the following commercial names: Bioprep 3000L, Pulpzyme HC, Scourzyme L -Novozymes, Baylase EVO - Bayer, Unizim PEC - Color-Center SA as well as numerous other names.

Pectinases are today among the enzymes with the best perspective for further application. They play an important role in the production of juices in food industry, in processing of the waste waters, the fermentation of coffee and tea, the preparation of animal forage and extraction of citric oil, in paper industry and have other biotechnological applications (Jayani et al., 2005). In the textile industry pectinases are used as agents in cotton scouring and in the biopreparation of bast fibers such as flax, ramie and jute (Holme, 2004).

## 1.5 Scouring

## 1.5.1 Alkaline scouring

The most commonly used procedure for removing noncellulosic material from cotton is the procedure of scouring with sodium hydroxide (classical or alkaline scouring). The procedure is performed at a high temperature in a bath that contains up to 4 % NaOH. Several auxiliary agents, such as wetting agents, emulsifiers and complexants, which improve the efficiency of scouring and reduce the damage of fibres, are also added to the scouring bath. Waxes, pectins, hemicellulose and proteins from the cuticle and the primary wall of the cotton fibres are efficiently removed in this procedure. Dust, different metal salts, chemical and processing impurities are also removed from the surface of the fibres, and partly also immature fibres and seed husks. Besides all the advantages mentioned, the procedure has some disadvantages. In the alkaline medium in contact with oxygen from the air oxycellulose can be formed on fibres. In cases when the concentration of the solution is irregular, the scouring is unequal since mercerisation of the cotton fibre can occur randomly. Having concluded the procedure, the fabric needs to be thoroughly rinsed and neutralised, with a considerable amount of water used in the process. Salts formed in neutralisation needs special procedures of cleaning. Due to high temperature a lot of energy is consumed in the process.

The efficiency of scouring is evaluated by determining residues of the different types of impurities, especially waxes and pectin that are found on the fibres (Preša & Tavčer, 2008a). Cracks are formed on the fibres, cuticle and the primary cell are removed. The fine structure of the fibre does not change, only the degree of crystallinity of cotton is slightly changed. The most noticeable change in the cotton fabric is the loss of mass. The lenght of the fabric shortens during boiling due to shrinking, causing the increasing of density and the tearing force. The most important change is the increased wettability which is a necessary property for a successful and even bleaching, dyeing and final treatment. The wettability needs to be good not only in spaces between the fibres but in the inner parts of the fibre as well (Lewin & Sello, 1983).

#### 1.5.2 Bioscouring

The disadvantages of scouring with sodium hydroxide have motivated textile industry to introduce more enhanced biologic agents which would be as effective in removing noncellulose substances as sodium hydroxide but would not have damaging effects on cellulose and would be less energy and water consuming. Favourable effects of scouring have been obtained with the enzymes pectinases (Etters, 1999; Hartzell & Hsieh, 1998; Li & Hardin, 1998; Csiszar e tal., 2001, Anis & Eren, 2001; Buchert et al., 2000), that catalyse the hydrolysis of pectin substances. Three main types of enzymes are used to break down pectin substances (Jayani, 2005): pectin esterases, polygalacturonases and pectin lyases. Considering the type of pectinases the bath may be slightly acidic or alkaline. It is recommendable to add the non-ionic surfactant into the bath and, depending on the type of

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the pectinases, a sequestering agent. The procedure is based on a fact that pectin acts as a type of cement or glue that stabilises the primary cell of the cotton fibres. When the pectinases are active a complex is formed between the pectinase and the pectin which causes the hydrolysis of the pectin substances. The result of this hydrolysis is a split of the bond between the cuticle and the cellulose body (Li & Hardin, 1998). The outer layers are destabilised and removed in the following procedures of rinsing. The enzymes are released and bond again with the pectin. The procedure is repeated until the enzyme is not destroyed chemically, with the change in pH or in the temperature (Etters et al., 1999). By removing pectin, other noncellulose substances are removed. The procedure of bioscouring gives softer fibres than conventional scouring, however the degree of whiteness is lower and the procedure is not appropriate for removing seed-coat fragments. The potencial advantages that make the enzyme scouring commercially appealing, are a higher quality of the fibres (softer to the touch and better strength), less waste waters, economy of energy and compatibility with other procedures, equipment and materials (Cavaco-Paulo & Gübitz, 2003).

#### 1.5.2.1 The development and conditions of bioscouring

The starting studies of enzyme treatment for scouring that is, cleaning of cotton fibres, were carried out by German researchers (Schacht et al., 1995; Rößner, 1995), and they included pectinases, proteases and lipases that act upon impurities and cellulases which hydrolyse the cellulose chain. Many other researchers followed in their path. They established that cellulases and pectinases are the most effective ones, lipases less with proteases being the least effective. On the basis of their studies they concluded that a simple procedure with pectinases in presence of non-ionic surfactant is sufficient to attain good absorbency (Li & Hardin, 1998; Hartzell & Hsieh, 1998; Buchert et al., 2000; Traore & Buschle-Diller, 2000; Galante & Formantici, 2003).

The first researches including pectinases as agents for scouring cotton were carried out to optimise the conditions of their activity. The concentration of the enzymes in the bath as well as time and temperature of treatment, pH of the bath, additives in the bath and the mechanical treatment all influence on the activity of the enzymes.

Due to a wide variety of enzymes, the added amount of pectinases strongly differs from research to research. The concentrations are usually low, from 0,05 to 2 % according to the weight of the fibres. The increase of concentration above the optimal value neither enhances nor improves the efficacy of the treatment.

The temperature of bioscouring is much lower compared to classic scouring, the optimal temperature is from 40 to 60 °C (Li & Hardin, 1998). Above the mentioned temperature the pectinases lose their activity since a higher temperature destroys the enzymes. However, a temperature that is too low does not suffice for removing the waxes, which have a melting point above 70 °C. A raise in temperature of the bath after completing the scouring is recommended for a better removal of the noncelullulosic material. A second reason for raising the temperature is also the deactivating of the enzymes. The pectinases alone are not harmful to the cellulose fibres, however, enzyme preparations often contain traces of cellulases which could be damaging to the fibres.

Beside the temperature, the pH of the environment is crucial for the activity and stability of the enzyme. The majority of enzymes are active in the pH range between 5 and 9. They are active in a wider pH range, however, at extreme values the three-dimensional form of the enzymes collapses and the enzymes lose their catalytic behaviour. Alkaline or acidic

environment depends on the type of pectinases. Acidic pectinases that function in a slightly acidic medium (pH between 4 and 6), as well as alkaline pectinases that function in a slightly alkaline medium (pH between 7 and 9) are known, both types have similar effects on cotton (Aly et al., 2004, Tzanov et al., 2001; Yachmenev at al., 2001). In acidic medium the pectin structure degrades without adding the pectinases which is often the reason for a better functioning of the acidic pectinases over the alkaline pectinases (Preša & Tavčer, 2008b).

In the starting researches, longer times of treatment were pointed out as the main disadvantage of the enzyme scouring (Sawada et al., 1998). By developing new pectinases, the times of treatment have shortened. Thus, the present forms of pectinases need 30 to 60 minutes for their functioning (Aly et al., 2004; Hartzell-Lawson & Durant, 2000).

Added surfactants also have a big influence on removing noncellulose impurities, however, caution is advised when adding surfactant. Anionic surfactants can form complexes with proteins and influence the structure. Cationic surfactants have a similar influence on proteins, however, with a lower affinity. Enzymes usually retain their catalytic activity in a solution with non-ionic surfactants, unless the concentration of the surfactants in the solution exceeds the critical micelle concentration (Li & Hardin, 1998). Non-ionic surfactants are compatible with enzymes and do not break their three-dimensional structure. They accelerate the effects of scouring due to lowering the surface tension of the fibres and an easier penetration of the enzyme into micropores and cracks of the fibres. Ultimately, the surfactants pull the enzyme back into the bath where it is available for further catalytic activity (). Surfactants take an active part in removing waxes and grease (Li & Hardin, 1998; Tzanov et al., 2001; Durden et al., 2001).

Enzyme inhibitors such as heavy metals and ionic detergents as well as product on the basis of formaldehyde need to be avoided since they deactivate the enzyme (Cavaco-Paulo & Gübitz, 2003; Li & Hardin, 1998).

One of the possibilities of improving the degradation of pectin is also the addition of the chelating agent. It is well known that calcium ions play an important part in the structure of the pectin, the  $Ca^{2+}$  ions bond the nonestrificated molecules of pectin. By removing the ion, the structure of the pectin is destabilised which enables the pectinases an easier access to the areas of attack.

Despite the good results in simultaneous activity of the chelating agent and the pectinases, caution is advisable in choosing the chelating agent. Chelating agents that are too strong also bond the metal ion, which is present in some types of enzymes, the so called metalo enzymes. The removal of this metal ion destroys the structure of the enzyme which causes a deactivation of the enzyme. Therefore, the use of weaker chelating agents, such as phosphate, silicate and carbon chelating agents, is recommended (Durden et al., 2001; Preša & Tavčer, 2008b).

The pectinases penetrate into the fibres through the cuticle in places where there are cracks and micropores, and catalyse the reaction of hydrolysis of the pectin molecules. The mixing loosens the bonds between the primary and the secondary wall of the cotton causing more micropores and cracks to appear on the surface of the fibres (Li & Hardin, 1998). This enables the enzyme to penetrate more easily into the inner part of the fibres. Introducing agitation into the procedure of scouring with the pectinases strongly enhances the absorption of the cotton fabric. The time of treatment is shortened and the amount of pectinases needed to attain good absorption of the fabric is lowered (Hartzel-Lawson & Durant, 2000).

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1.5.2.2 Influence of bioscouring on further finishing procedures

After the bioscouring the cotton fibres are darker than after alkaline scouring (Preša & Tavčer, 2009; Tavčer, 2008). In further bleaching with hydrogen peroxide, it was established that a better degree of whiteness can be attained on alkaline scoured sample than on the bioscoured one, however, it need to be taken into account that alkaline scoured fibred are very sensitive to oxidative damage during bleaching. More significant damage occures compared to the samples scoured with pectinases (Buschle-Diller et al., 1998).

Several researchers examined the possibilities of combining bioscouring with previous and following procedure. They achieved an adequate wettability by combining enzyme desizing and bioscouring (Lenting & Warmoeskerken, 2004; Yachmenev et al., 2001). Tzanov (Tzanov et al., 2001) used a desizing bath for scouring and it proved to be an important source of glucose in the following procedure with the glucose oxydases. The glucose oxidases produce hydrogen peroxide in water solutions in the presence of glucose from oxygen dissolved in water. The degree of whiteness attained in this procedure is lower than the degree of whiteness of the fibres bleached in a classic procedure with hydrogen peroxide.

Dyeing with direct and reactive dyes was efficient and equal on fabrics that were differently scoured (Canal et al., 2004; Preša & Tavčer, 2009). Etters (Etters et al., 2001) did not notice any statistically significant difference between the rate of uptake, equilibrium exhaustion, or colour depth on the cotton substrate between the two fabrics that were either alkaline scoured or bioscoured. On the contrary Losonczi and colleges (Loszonci et al., 2004) claim that classically scoured fabric compared to bioscoured fabric has a lighter colour. After previous bleaching of differently scoured fabric, no differences can be noticed in lighter dyeing. Treatments with or without the enzyme do not affect the evenness of the dyeing.

#### 1.6 Bleaching

Scouring is regularly followed by a bleaching process, which removes the natural pigments of cotton fibres. Cellulose fibres are most frequently bleached with hydrogen peroxide (HP) resulting in high and uniform degrees of whiteness. The water absorbency also increases, however, during the decomposition of hydrogen peroxide, radicals that can damage the fibres are formed. For this reason, organic and inorganic stabilizers and chelators are added to the treatment bath.

Hydrogen peroxide (redox potential is 1.78 eV) (1) is not ecologically disputable. The large amount of water used to rinse and neutralize the alkaline scoured and peroxide bleached textiles is ecologically disputable. Namely, the bleaching process is conducted in an alkaline bath at pH 10 to 12 and at temperatures up to 120°C. Due to high working temperature, a large amount of energy is consumed. Auxiliary chemicals added into the bath increase the TOC and COD values of effluents. Upon neutralization of highly alkaline waste baths, large amounts of salts are produced. Consequently, the textile industry is considered one of the biggest water, energy and chemical consumers (Alaton et al., 2006).

Bleaching with peracetic acid (PAA) is an alternative to bleaching with hydrogen peroxide (Gürsoy & Daioglu, 2000; Križman et al., 2005, Hickman, 2002; Prabaharan et al., 2000, Tavčer, 2008; Tavčer, 2010). It is a powerful oxidizing agent (redox potential: 1.81 eV) (Preša & Tavčer, 2009) with excellent antimicrobial and bleaching properties. It is efficient at low concentrations, temperatures and in neutral to slightly alkaline medium. Its products of decomposition are biologically degradable. In the past, it was prepared in situ from acetic acid anhydride and hydrogen peroxide (Rucker, 1989; Wurster, 1992). However, the risk of

explosion during the synthesis reaction prevented affirmation of PAA as a bleaching agent in industry. In recent years, PAA has become interesting (Hickman, 2002). Several commercial products are available as balanced mixtures of PAA, acetic acid and hydrogen peroxide (Equation 1). They are stabilized with a minimum amount of chelating agent. Today, PAA products available in the market are safe, simple to use, and price-effective. PAA induces epoxidation of coloured substances in fibres. Good bleaching results are obtained with low concentrations of PAA at the temperature 40 °C – 80 °C and the pH value 7 - 8. During bleaching, acetic acid is released from peracetic acid and the pH of the bleaching bath decreases. At the end of the process, the bleaching bath is slightly acidic and neutralisation is not necessary. Rinsing is needed only to remove wetting agents. PAA does not damage fibres. It decomposes to oxygen and acetic acid and is therefore environmentally safe (Križman et al., 2005; Križman et al., 2007, Preša & Tavčer, 2008b).

$$CH_{3} - C O + H - O - O - H \xrightarrow{\text{catalyst} \\ \text{stabilizer}} CH_{3} - C O + H_{2}O$$
(1)

Equation 2 shows the reaction that occurs when PAA is used for bleaching.

$$CH_3COOOH + impurities \longrightarrow CH_3COOH + oxidised impurities$$
 (2)

#### 1.7 Aim of research

Both processes, scouring with pectinases and bleaching with PAA, are conducted at temperatures of 50–60°C for 40–60 minutes and pH 5–8. If both processes could be combined into one process, huge amounts of water, energy, time, and auxiliary agents can be saved. In a previous study (Preša & Tavčer, 2008b), it was confirmed using a viscosimetric method that pectinases retain their activity in the presence of PAA and that combined processes are feasible.

The objective of our work was to compare the properties of enzymatically-scoured and PAA-bleached cotton fabrics treated by two-bath and one-bath scouring/bleaching methods, with respect to conventionally-treated fabrics (alkaline scoured and bleached with hydrogen peroxide). The degree of whiteness, water absorbency, fiber damage, and dyeability of woven fabrics were evaluated.

In addition, after all these treatments, pH, TOC, COD and BOD<sub>5</sub> values of the remaining baths were measured. The amount of water and heating energy used during the treatments and rinsing were measured as well.

## 2. Experimental

#### 2.1 Materials

Desized cotton fabric, 100 g/m<sup>2</sup>, was obtained from Tekstina, Slovenia. Acid pectinases Forylase KL (AP) was supplied from Cognis, Germany, and alkaline pectinases Bioprep 3000L (BP) from Novozymes, Denmark. Cotoblanc HTD-N (anionic wetting and dispersing agent, alkansulphonate with chelator) was supplied from CHT, Germany.  $H_2O_2$  35% (HP) and peracetic acid (PAA) as a 15% equilibrium solution in the commercial bleaching agent Persan S15 were obtained from Belinka, Slovenia. Foryl JA (nonionic wetting agent) and

Locanit S (ionic-nonionic dispersing agent) were obtained from Cognis, Germany and Lawotan RWS (nonionic wetting agent) was obtained from CHT, Germany. Sodium hydroxide was supplied from Šampionka, Slovenia, and acetic acid and sodium carbonate were supplied from Riedel-de Haen, Germany.

## 2.2 Treatment methods

The cotton fabric was scoured according to three different procedures using sodium hydroxide, acid pectinases or alkaline pectinases. The scoured fabrics were bleached with two bleaching agents: hydrogen peroxide and Persan S15. The abbreviation of processes and treatment conditions are displayed in Table 1. Enzymatic scouring and one-step treatments were performed 60 minutes at 55 °C, than the temperature of the bath was increased to 80 °C to for 10 minutes to deactivate the enzymes. To activate PAA in AP/PAA treatment, the pH was adjusted to 8 after 30 minutes. Demineralised water was used in all processes. The treatments were performed on the Jet JFL apparatus manufactured by Werner Mathis AG loaded with 50 g of fabric at a liquor ratio of 1:20. After all treatments, the bath was discharged and the jet was filled sequentially with fresh water heated to 80 °C, 60 °C and 25 °C to rinse the fabric. After alkaline scouring and peroxide bleaching, the fabrics were neutralised with a neutralizing bath containing acetic acid and rinsed with cold water.

Process	Conditions		
<b>AS</b> - Alkaline scouring	3 g/l NaOH, 2 g/l Cotoblanc HTD-N, 95°C,		
AS - Alkaline scouring	40 minutes		
<b>AP</b> - Scouring with acid pectinases	5 ml/l Forylase KL, 0,75 ml/l Foryl JA, 2		
AI - Scouring with actu pectiliases	ml/l Locanit S and CH <sub>3</sub> COOH to pH 5 - 5,5		
<b>PD</b> Courring with all caling postingers	0,05 % Bioprep, 0,5 g/l Lawotan RWS,		
<b>BP</b> - Scouring with alkaline pectinases	$Na_2CO_3$ to pH 8		
IID blog ship a with budge and generalide	7 g/l H <sub>2</sub> O <sub>2</sub> 35%, 1 g/l Cottoblanc HTD-N, 4		
HP - bleaching with hydrogen peroxide	g/l NaOH 100%, 95 °C, 45 min		
<b>DAA</b> blocking with noncetic still	15 ml/l Persan S15, 55 ml/l Na <sub>2</sub> CO <sub>3</sub> 0,5 M,		
<b>PAA</b> - bleaching with peracetic acid	0,1g/1 Lawotan RWS, pH 8, 55 °C, 40 min		
<b>AP+PAA</b> - one step scouring with acid	5 ml /l Forylase KL, 0.75 ml/l Foryl JA, 2		
pectinase and bleaching	ml/l LocanitS, 15 ml/l Persan S15		
<b>BP+PAA</b> - one step scouring with	0.05 % Bioprep 3000L, 0.1 mL/L Lawotan		
alkaline pectinase and bleaching	RWS, 15 mL/L Persan S15, pH 8 with NaOH		

Table 1. The abbreviation of processes and treatment conditions

## 2.3 Analytical methods

Prior to the measurements, fabrics were conditioned 24 hours at 20 °C and 65% relative humidity. The degree of whiteness and the colour values were measured on the Spectraflash SF600 Plus using the CIE method according to EN ISO 105-J02:1997(E) standard and EN ISO 105-J01:1997(E), respectively. Weight loss due to the pretreatments was determined by weighing the fabric samples before and after pretreatment and was expressed in percent. Water absorbency was measured according to DIN 53 924 (velocity of soaking water of textile fabrics, method for determining the rising height). Measurements of tenacity at maximum load were performed on Instron Tensile Tester Model 5567. The mean degree of polymerisation (DP) was determined with the viscosimetric method in cuoxam.

Samples of remaining bleaching and scouring baths were collected after all treatments. Their ecological parameters, such as pH, total organic carbon (TOC), chemical oxygen demand (COD) and biological oxygen demand (BOD<sub>5</sub>), were measured. TOC was measured on a Shimadzu TOC-5000A according to ISO 8245. COD was performed according to SIST ISO 6060, BOD<sub>5</sub> according to SIST ISO 5815, and biological degradation as a ratio of BOD<sub>5</sub> and COD.

The consumption of water for treatments was estimated by adding up all the sequential fillings of the jet apparatus with treatment and rinsing baths and the total consumption of 1 kg of fabrics was recalculated. The energy consumption was expressed with the amount of steam required to heat water to treat and rinse baths. The amount of steam required for heating one litter of water from certain starting temperatures to a defined final temperature was obtained from the technical documentation of the textile dyeing plant.

## 3. Results and discussion

## 3.1 Fabric properties

Table 2 represents the whiteness values, the loss of mass, rising height in warp direction, tenacity at maximum load and degree of polymerisation of differently pretreated cotton fabric samples.

	W	Mass loss (%)	Rising height (cm)	Tenacity (cN/tex)	DP
D	11.1		0	18.47	2482
AS	19.5	1.27	2.9	18.45	2432
AP	8.2	0.30	2.7	16.96	2451
BP	8.4	0.89	2.5	17.95	2385
AS+HP	84.1	1.52	3.0	16.65	1774
AP+HP	85.6	1.51	3.0	17.12	1947
BP+HP	85.1	1.62	2.8	16.83	2004
AS+PAA	72.7	1.30	2.8	16.94	2278
AP+PAA	57.7	0.65	2.9	18.12	2318
BP+PAA	57.3	0.95	2.9	13.75	2399
AP/PAA	68.7	0.40	2.7	16.94	2438
BP/PAA	69.6	0.60	2.8	18.84	2300

Table 2. Whiteness (W), the loss of mass, rising height in warp direction, tenacity at

maximum load, degree of differently pretreated and desized only (D) cotton fabric samples.

Alkaline scoured samples are whiter than enzymaticaly scoured ones. The degree of whiteness increased significantly after HP bleaching and the differences in whiteness from previous scouring disappeared. With PAA bleaching, a high degree of whiteness was not achieved and the differences in whiteness from the previous scouring remained visible. This occurs because bleaching with PAA proceeds at a low temperature and pH, where the impurities remaining after scouring could not be fully oxidised. Bioscoured fibres, which were not treated at high temperature and high pH, contained more waxes and other impurities that hindered the successful oxidation with PAA at mild conditions. Bleaching

the alkaline scoured fabrics with PAA is more effective since the impurities were removed from cotton fibers to a higher extent in the previous process and the pigments within fibers were more exposed to the oxidant's influence.

The degrees of whiteness after a one-bath treatment were higher than those after two-bath bioscouring and bleaching with PAA and close to the whiteness achieved after alkaline scouring and bleaching with PAA. The one-step process, namely ended with rising of temperature to 80 °C and this temperature activate the presented hydrogen peroxide, which improves the whiteness of fabric.

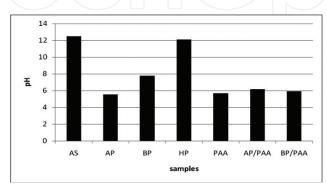
The loss of mass demonstrates that scouring with NaOH is more intensive and removes more incrusts than enzymatic scouring. In the following bleaching HP removed a large portion of compounds, which remained on fibers after scouring. The total mass loss after scouring and HP bleaching was similar for all samples.

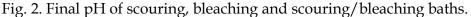
PAA bleaching also removed a certain part of the noncellulosic substances, which remained on fibers after scouring, but the quantity was lower relative to HP bleaching. Bleaching with PAA did not equalize the differences in the loss of mass, which is in agreement with the whiteness results. We can conclude that high temperature and high pH are conditions that contribute decisively to the removal of non-cellulosic impurities. Specifically, waxes cannot be removed completely when all processes are conducted at low temperatures and neutral pH, as is the case for bioscouring and PAA bleaching. The remained substances influence on the water absorbency and consequently alkaline scoured samples had the highest absorbency. Bleaching improved the absorbency of the scoured fabrics, particularly of enzymatically scoured ones. However, the difference in rising height was so small, that all the samples could be considered absorbent.

There were no higher differences in tenacity at maximum load between the de-sized and differently treated samples. On the other hand, the results of DP demonstrate, that bleaching with HP decreased the degree of polymerisation significantly, while other processes preserved the DP values close to the starting value. The bioscouring and bleaching with PAA in a one bath or two bath process causes no damage to fibers and this is one of the benefits of such processes.

#### **3.2 Ecological parameters**

Figures 2 to 5 present the final pH values of the remaining baths from different processes, total organic carbon (TOC), chemical oxygen demand (COD), biological oxygen demand (BOD<sub>5</sub>) and biological degradability of remaining baths (BOD<sub>5</sub>/COD ratio), respectively.





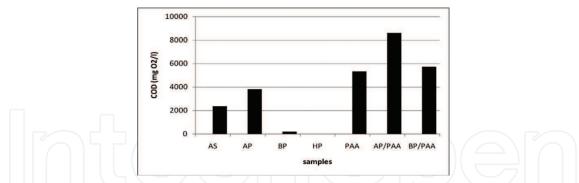


Fig. 3. COD values of scouring, bleaching and scouring/bleaching baths.

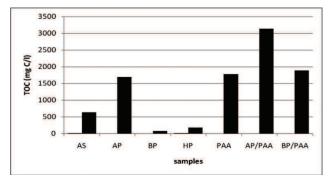


Fig. 4. TOC values of scouring, bleaching and scouring/bleaching baths.

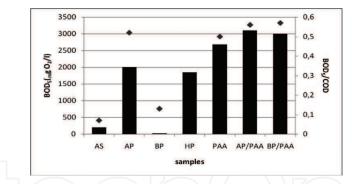


Fig. 5. BOD<sub>5</sub> values (column) and BOD5/COD (♦) of scouring, bleaching and scouring/bleaching baths.

Conventional treatment of cotton fibres was conducted in an alkaline environment: final pH at alkaline scouring and at bleaching with hydrogen peroxide was around 12.5. Such alkaline baths should be neutralized prior to drainage into the sewage system. At neutralization, salts that additionally load wastewaters are produced.

The processes of bioscouring and bleaching with PAA occurred between pH 5.5 and 8. The final pH value of the bath was 5.5 while scouring with acidic pectinases, and 7.5 while scouring with alkaline pectinases. While bleaching with PAA and at both combined processes, the final pH value of the bath was near 6. Since neither of these processes requires neutralization of fibres, the treatment process can be shorter and less expensive. Additionally, the remaining baths do not require the neutralization step prior to drainage into the sewage system, which also reduces the cost of processes.

TOC, COD and BOD<sub>5</sub> values show similar relations. The scouring bath with alkaline pectinases exhibited the lowest TOC and COD values. These values were so low that they did not exceed the limit values (TOC 60 mg C/L and COD 200 mg  $O_2/L$ ) for direct drainage into the sewage system (Decree, 1996). However, the scouring bath with acidic pectinases had high TOC and COD values that were even higher than alkaline scouring. The reason lies in the initial composition of the bath, which was prepared according to the producer's instructions and contained more auxiliary agents than the bath with alkaline pectinases, which contained only enzyme and wetting agent. We anticipate that the optimisation of the recipe of scouring with acid pectinases would improve its ecological parameters.

Among bleaching baths, the baths with PAA had higher TOC and COD values. PAA is an organic compound, which contributes to higher TOC and COD values, as well as acetic acid, which is present in the balanced mixture. Peracetic acid is decomposed in the waste bath to acetic acid and oxygen. Acetic acid, as such, is not ecologically disputable, and does not cause any problems in wastewaters in which it appears in the diluted state. Its biodegradability is 51 – 99% (Howard, 1990).

After bleaching with hydrogen peroxide, a certain amount of the non-used hydrogen peroxide remained in the bath. For that reason, we could not determine the real COD value, such that it is not presented in the diagram.

The BOD<sub>5</sub> values (columns of Figure 5) are high with the bioscouring baths with acidic pectinases (2000 mg  $O_2/l$ ) and the baths containing PAA (between 2680 and 3100 mg  $O_2/l$ ). The lowest BOD<sub>5</sub> value (25 mg  $O_2/l$ ) belongs to the bath with alkaline pectinases.

The baths with enzymes and PAA were biodegradable, while the bath was non-degradable after alkaline scouring. Biological degradability of the peroxide bleaching bath could not be determined in this manner.

#### 3.3 Consumption of water and energy

Figure 6 presents the amount of water and energy required for the treatment of 1 kg of material at a liquor ratio 1:20 for different processes.

The amount of water consumed for alkaline scouring and bleaching with HP was higher than the amount of water consumed for bioscouring and bleaching with PAA. After alkaline scouring and bleaching with hydrogen peroxide, the fabric must be neutralized. Neutralization is not required after bioscouring and bleaching with PAA because the pH value is only slightly acidic and is neutralized during the first rinsing. The process of bioscouring and bleaching with PAA consumed only 66.6% of water relative to alkaline scouring and bleaching with HP. During the one-bath treatment, the consumption of water was still lower, i.e. only 50% in comparison with two-bath process, and only 33% in comparison with conventional pre-treatment process.

While scouring with pectinases and bleaching with PAA, the consumption of energy required to heat the bath was also lower. Conventional processes of scouring and bleaching were performed at temperatures near the boiling point, whereas bioscouring and bleaching with PAA were conducted at a temperature of 55 °C. Due to the lower temperature, less energy was required, which is presented in Figure 4. Only 63.3% of the steam, which was required during alkaline scouring, was consumed at bioscouring, and only 30.5 % of the steam, which was required at bleaching with hydrogen peroxide, was consumed at bleaching with PAA. The lowest amount of energy was consumed by the one-bath process, i.e. 67.4% of steam was consumed by the two-bath process, while only 31.6% of the steam was required for alkaline scouring and bleaching with hydrogen peroxide.

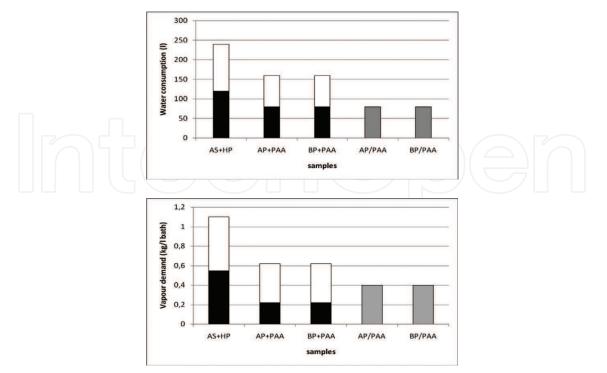


Fig. 6. Water (above) and energy (below) consumption for alkaline scouring and bleaching with hydrogen peroxide (AS+HP), for scouring with acid or alkaline pectinases and bleaching with PAA (AP+PAA, BP+PAA) and for combined bioscouring and bleaching with PAA (AP/PAA, BP/PAA) for treatment of 1 kg of fabric at liquor ratio of 1:20 (scouring  $\blacksquare$ , bleaching  $\Box$ , combined treatment  $\blacksquare$ ).

## 4. Conclusions

Bioscouring can be recommended as an adequate procedure for scouring of cotton. It is a simple, repeatable and safe procedure. The removal of pectin components from cotton adequately improves the water absorbencies of the fibres and facilitates the penetration of the dye and other substances into the fibre. Natural qualities of the cotton fibre are preserved, the fabric is softer to the touch than after classic scouring. Fibres are also less damaged. Biouscouring can also be used for mixtures of cotton and silk, wool and cashmere; in severe alkaline conditions of classic scouring, damage occurs on these fibres.

Sodium hydroxide is removed from the textile treatment procedures or its use is considerably lowered. Due to a lower pH of the bath, less rinsing is needed, what results in shorter times of treatment and lower use of water. Energy is economised as well, since the bioscouring occurs at a lower temperature. Direct dyeing without the intermediary bleaching is possible in the case of dyeing dark shades.

Waste waters are less polluted, the COD values of the scouring baths are thus lower due to the economised use of chemicals as well as BOD<sub>5</sub> values due to a smaller loss of the fibre weight.

However, bioscouring has a few disadvantages. Due to a relatively low treatment temperature, the waxes are not entirely removed. The attained degree of whiteness is lower compared to alkaline scoured or even desized fabric. Due to a lower pH the seed-coat fragments do not swell and are not so decolorized in bleaching.

Peracid bleaching can substitute the hydrogen peroxide bleaching when medium degree of whiteness is demanded. Cotton fibers, bleached with peracetic acid have appropriate water absorbancy and are not damaged. When bleaching with PAA, less water and energy is consumed, and the bleaching baths are biodegradable.

The consumption of water and energy is the lowest at one-bath processes of scouring/bleaching with pectinases and PAA. The degree of whiteness of fabrics is higher than at two-step scouring and bleaching with PAA, but lower than at bleaching with HP. The fabrics have good water absorbency, the fibres are not damaged and the remaining baths are biodegradable.

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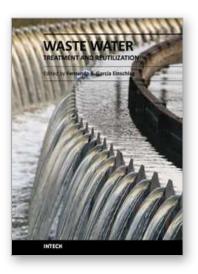
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