UDK 677+687

YU ISSN 0040-2389

SAVEZ INŽENJERA I TEHNIČARA TEKSTILACA SRBIJE

Naučni i stručni časopis tekstilne i odevne industrije

tekstilna industrija

1868 - 2015

SCIENTIFIC AND PROFESSIONAL JOURNAL OF THE UNION OF TEXTILE ENGINEERS AND TECHNICANS OF SERBIA

Godina LXII • Broj 2 (april - jun) • 2015. godina • Beograd

UDK 677 + 687

YU ISSN 0040-2389

SAVEZ INŽENJERA I TEHNIČARA TEKSTILACA SRBIJE
Naučni i stručni časopis tekstilne i odevne industrije

tekstilna industrija

1868 – 2015

SCIENTIFIC AND PROFESSIONAL JOURNAL OF THE UNION OF TEXTILE ENGINEERS AND TECHNICANS OF SERBIA

Volume LXII • Number 2 • Beograd 2015 • Page 87

Publisher: Textile Engineers and Technicians Union of the Republic Serbia

Editoral offices: Serbia, 11000 Beograd, Kneza Miloša 7a/II, tel/fax: 011/3230-065

e-mail: saveztekstilacasrbije@gmail.com; casopistekstilnaindustrija@gmail.com

CONTENTS

Mojsov Kiro[1]

Naučni rad

UDC: 677.21.027.254.1

ENZYMATIC DESIZING OF COTTON FABRIC AND GLUCOSE GENERATION IN DESIZING LIQUOR

Abstract: *Woven fabrics of cotton, on the warp threads are coated with an adhesive sizing agent to prevent the threads from breaking during weaving process. Starc and its derivatives are the most common sizing agents. After weaving, starch must be removed in order to prepare the fabric for dyeing and finishing. This process is called as desizing and it include the use of chemicals such as acids, alkalis and oxidising agents with traditional process. Because of that in the world are carried out investigations to replace conventional chemical textile processes by eco-friendly and economically attractive bioprocesses using enzymes. This paper will show the experimental results relating to the selection of the enzyme and process optimization made in order to increase the content of glucose in the desizing liquor of starch sized cotton fabric. The results showed that the commercial desizing enzyme preparations of α-amylase enzymes were not appropriate to produce a large quantity of glucose, and whilst used commercial food amyloglucosidase preparation, produced a large quantity of glucose after the optimization process.*

Keywords: bio-process, eco-friendly characteristics, α-amylase, amyloglucosidase.

ENZIMNO ODSKROBLJAVANJE PAMUČNE TKANINE I GENERISANJE GLUKOZE U RASTVOR ODSKROBLJAVANJA

Apstrakt: *Pamučne tkanine, na osnovi niti su obložene lepljivim sredstvom za učvršćivanje da spreči niti od lomljenja prilikom procesa tkanja. Skrob i njegovi derivati su najčešći sredstva za skrobljenje. Posle tkanja, skrob mora biti uklonjen u cilju pripreme tkanina za bojenje i dorade. Ovaj proces se zove odskrobljavanje i uključuje korišćenje hemikalija kao što su kiseline, alkalije i oksidaciona sredstva u tradicionalnom procesu. Zbog toga u svetu se sprovode istraživanja da zameni konvencionalne hemijske tekstilne procese sa ekološki i ekonomski atraktivnim bioprocesima pomoću enzima. Ovaj rad će pokazati eksperimentalne rezultate koji se odnose na izbor enzima i proces optimizacije napravljen u cilju povećanja sadržaja glukoze u rastvor odskrobljavanja skrobljene pamučne tkanine. Rezultati su pokazali da komercijalne enzimne preparate za odskrobljavanja od α - amilaza enzima nisu bili odgovarajuće da proizvede veliku količinu glukoze, a dok upotrebom komercijalnog prehranbenog preparata amiloglukozidaze, proizveo je veliku količinu glukoze posle optimizacije procesa.*

Ključne reči: *bio-proces, ekološke karakteristike, α-amilaza, amiloglukozidaza.*

1. INTRODUCTION

Biotechnology is a field of applying living organisms and their components in the industrial processes and products. During the last years, intensive research of biotechnological processes involving enzymes and microorganisms has been made in the field of textile technology. Biotechnology as an ecological advantageous and moreover economical beneficial technology plays an increasingly important role in the industrial wet textile pretreatment and the finishing processes. Therefore, in an enzymatic pretreatment, the textile substrate is less damaged when compared to a classical pretreatment. In many

[1] Faculty of Technology, University of Stip, Republic of Macedonia, e-mail: kiro.mojsov@ugd.edu.mk

industries, enzymes are used as biological catalysts to replace harsh chemicals or perform reactions under milder conditions.

Enzymes are specific and fast in action and small amounts of enzyme often save large amounts of raw materials, chemicals, energy and water. Especially in textile manufacturing the use of enzymes has a long tradition. The first microbial amylases were used in the 1950s for the removal of starch sizes, which today is routinely used by the industry [1, 2]. Amylases are enzymes which hydrolyse starch molecules to give diverse products, including dextrins and smaller polymers composed of glucose units [3]. Sizing is carried out in the weaving to protect the warp yarn during the weaving process from damage or interruption. The size forms a protective film on the warp yarn, protruding fiber ends causing loom stops are minimized. About 75% of sizing agents used worldwide are starch and its derivatives [4].

Desizing is a typical process in pretreatment of cotton woven fabrics and cotton blends but also necessary for all grey synthetic materials containing sizes. The sizing agents on the warp yarns, have to be removed before further processing in textile finishing [4, 5]. In the past, hydrogen peroxide and sodium hydroxide were generally used as desizing agents. But this method was not economically and environmental-friendly. In conventional method of starch removing from a cotton article large amount of water, chemicals and energy have to be consumed. 50-80% of the Chemical Oxygen Demand (COD) in the effluents of textile finishing industries is caused by sizing agents [6]. To decrease water and the chemicals consumption and in response to the environmental concerns, enzymatic desizing of cotton seems a practical alternative. Alternatively, the peroxide could be produced in situ by enzymatic system glucose oxidase/glucose [7].

The commercially available enzymes are manufactured from the micro-organisms by fermentation. Further purification steps involve precipitation, extraction, centrifugation and filtration. 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 [8].

Starch comprises a mixture of a linear polymer and a branched polymer. Amylose is a flexible linear polymer of glucose residues joined by alpha-1,4 glycosidic bond, which may be formed from several thousand glucose molecules. Amylopectin, however, is a branched molecule of glucose residues joined by either the α-1,4 or α-1,6 glycosidic bond. There are four groups of starch-converting enzymes: endoamylases, exoamylases, debranching enzymes and transferases. Endoamylases (α-amylase) are able to cleave α-1,4 glycosidic bonds present in the inner part of the amylose or amylopectin chain. Exoamylases (β-amylase, amyloglucosidase or glucoamylase) act on the external glucose residues of amylose or amylopectin and produce glucose, maltose or dextrin. Debranching enzymes (isoamylase and pullanase) that exclusively hydrolyse α-1,6 glycosidic bonds. Transferases are able to cleave α-1,4 glycosidic bonds of the donor molecule and transfer part of the donor to a glycosidic acceptor with the formation of a new glycosidic bond [9].

The commercial desizing enzymes (α-amylases) are used to produce of glucose in the desizing bath, but these enzymes do not seem appropriate for this purpose [1, 10], whereas amyloglucosidases are suitable enzymes to degrade starch until it becomes glucose [6, 11, 12]. The applications of amyloglucosidase extend to many beverage and other food industries [13]. Amyloglucosidase is also widely used in brewing, paper, food, pharmaceutical and textile industries [14].

Glucose oxidase enzymes (GO_x) are representative of the oxidoreductase group. They catalyse the oxidation of glucose to gluconic acid and simultaneous production of hydrogen peroxide for bleaching. In this study was examined the desizing effect of amyloglucosidase/pullanase enzyme mixture and α-amylases in desizing liquor.

2. EXPERIMENTAL PART

2.1. Materials

Commercial desizing enzymes (α-amylase) from several suppliers and one commercial amyloglucosidase/pullanase enzyme mixture for food industry.

Raw cotton fabric with a mass per square meter of 160 g/m^2. The cotton fabric was sized with starch sizing agent and 5% starch was present in the sized fabric.

2.2. Treatment methods

Desizing trials (enzyme type and desizing recipe) were performed according to recommendations by manufacturers listed in Table 1 with fabric specimens of 15 grams (30 x 30 cm^2) at a liquor ratio of 1:10 with distilled water. The process time was 30 minutes. Optimization trials were performed for the amyloglucosidase/pullanase enzyme mixture.

2.3. Glucose assay

Glucose amounts of desizing liquor were measured by use of a GO_x solution (glucose oxidase > 15.000 U/l, peroxidase > 100 U/l, 4-aminoantiprin-0.5 mmol/l, 4-hydroxybenzoic acid-10 mmol/l, phosphate-119 mmol/l and stabiliser). Temperature: 37 °C; Primary wavelength: 500 nm; Desizing liquor: GO_x solution ratio (1 : 150); Incubation time: 5 minutes; Sensibility: 0-6300 mg/l.

Reactions can be written as:

Glucose + O_2 + H_2O → Gluconoic acid + H_2O_2
H_2O_2 + Hydrobenzonoic acid + 4-aminoantiprin → Quinoneimine dye + H_2O

The absorbance of the solution was measured using a spectrophotometer of 500 nm. The absorbance of the desizing liquor was compared to the absorbance of standard glucose solution (5.55 mmol/l). The glucose content of desizing liquor (G_d) was calculated by with formula: $\mathbf{G_d = (A_d/A_s)\cdot G_s}$, where G_d and A_d are the glucose amount (mg/l) and absorbance of the desizing liquor, and G_s and A_s are of standard glucose solution [15,16].

3. RESULTS AND DISCUSSION

3.1. Enzyme desizing and glucose generation in desizing liquor

Amylases are used in textile industry for desizing process. Desizing involves the removal of starch from the fabric which serves as the strengthening agent to prevent breaking of the warp thread during the weaving process. The α-amylases remove selectively the size and do not attack the fibres [3, 17].

The glucose generation in desizing liquor during the desizing process were compared with several commercial desizing enzymes (α-amylases) and one commercial amyloglucosidase/pullanase enzyme mixture for food industry is shown in *Table 2*. Results indicate an acceptable desizing effect, but very low glucose generation for α-amylases. The amount of glucose in desizing bath of approximately 4.000 mg/l or 400-600 mg/l hydrogen peroxide generation of glucoseoxidase is required to obtain a satisfactory whiteness [11]. The results reported in *Table 2* for the amounts of glucose from commercial desizing enzymes (α-amylases) (average of 201 mg/l) were not enough, and despite their well-known and satisfactory desizing effect [1, 3, 18]. These enzymes do not seem appropriate for this purpose. The low amount of glucose generation can be due to the reaction mechanism of α-amylases, which are endoamylase enzymes.

Amyloglucosidases and pullanases are debranching enzymes that can to degrade starch in the desizing bath, until it becomes glucose. The results reported in Table 2 for the amounts of glucose from amyloglucosidase/pullanase enzyme mixture (average of 3.659 mg/l) representing the great increase in the amount of glucose generation compared to that of α-amylases. Despite the grate increase in the amount of glucose, it were still under required level of approximately 4.000 mg/l to obtain a satisfactory whiteness [11]. Therefore to investigate any further increase in the glucose content of the de-sizing liquor of a starch-sized cotton fabric were performed optimization trials for the amyloglucosidase/pullanase enzyme mixture.

3.2. Optimisation of enzyme dosage by amyloglucosidase/pullanase depending on the glucose generation

A set of trials were performed to find an optimum enzyme dosage by amyloglucosidase/pullanase mixture to generate maximum glucose during the

Table 1.- *Enzyme types and desizing parameters recommended by manufacturers*

Enzyme type	Dosage, g/l	Temp., °C	pH	Time, min	Supplier
α-amylase-1	0.2-1.0	60-105	5.5-7.5	30	AB Enzymes GmbH, Darmstadt, Germany
α-amylase-2	0.25-1.3	70-80	6-7	30	Novozymes A/S, Bagsvaerd, Denmark
α-amylase-3	0.5-1.5	80-105	5.8-7	30	Shanghai S.B.Inc., Shanghai, China
α-amylase-4	0.5-2.0	60-100	5.4-8	30	CHT, Istanbul, Turkey
Amyloglucosidase/pullanase for food industry	no data for desizing process				Novozymes A/S, Bagsvaerd, Denmark

Table 2.- *Glucose generated in desizing liquor during enzyme desizing process*

Enzyme type	Dosage	Temperature, °C	pH	Time, min	Glucose, mg/l
α-amylase-1	1.0 g/l	90	6.5	30	200
α-amylase-2	1.3 g/l	70	6.5	30	215
α-amylase-3	1.5 g/l	90	6.4	30	185
α-amylase-4	2.0 g/l	80	6.7	30	205
amyloglucosidase/pullanase for food industry	0.4 % (owf)	60	4.2	30	3659

desizing process. *Figure 1* illustrates enzyme dosage by amyloglucosidase/pullanase mixture of 0.4 to 2.0% (owf) (on weight of material) depending on the glucose generation in desizing process. The minimum necessary enzyme dosage is 0.4 % (owf). However, increasing the enzyme dosage to 1.0% (owf) resulted in higher glucose amounts than 0.4% (owf). From these results was chosen an optimum enzyme dosage of amyloglucosidase/pullanase mixture of 1.0% (owf).

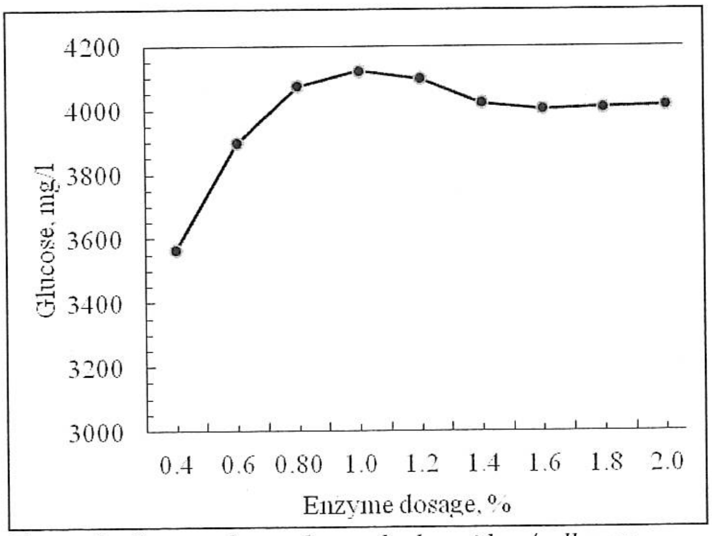

Figure 1.- *Enzyme dosage by amyloglucosidase/pullanase depending on the glucose generation*

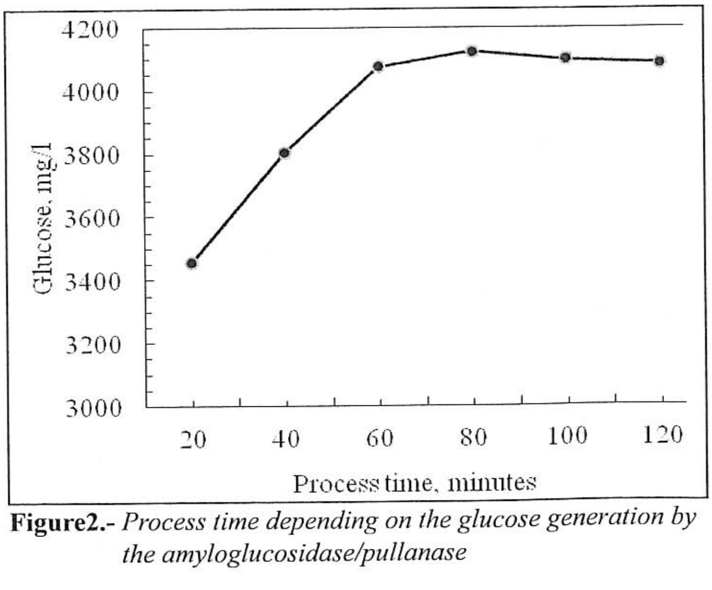

Figure2.- *Process time depending on the glucose generation by the amyloglucosidase/pullanase*

3.3. Optimisation of process time depending on the glucose generation by the amyloglucosidase/pullanase

Recommended process parameters for amyloglucosidase/pullanase mixture were not available for desizing process, but pH 4.1-4.3 and temperature of 60-63 °C have been applied to produce glucose from starch in food industry.

Since there were no data available concerning the use of amylopectinase/pullanase mixture in desizing process, the effect of the process time was also examined in order to find optimum circumstances. Trials were performed for 120 minutes with 20 minute intervals. *Figure 2* illustrates the process time by amyloglucosidase/pullanase mixture of 20 to 120 minutes depending on the glucose generation in desizing process. Increasing the process time to 80 minutes resulted in higher glucose amounts, while the process time of 100 to 120 minutes in smaller glucose amounts. From these results was chosen an optimum process time of 80 minutes.

3.4. Optimisation of pH depending on the glucose generation by the amyloglucosidase/pullanase

pH of 4.1-4.3 was recommended by the manufacturer of the amylo-glucosidase/pullanase mixture for glucose production from starch in food industry, but however it lacked appropriate pH for desizing process.

Since there were no data available concerning the use of amylopectinase/pullanase mixture in desizing process, the effect of pH was also examined in order to find optimum circumstances. Trials were performed for pH of 3.0-4.6 with 0.2 intervals. *Figure 3* illustrates pH depending on the glucose generation in desizing process by amyloglucosidase/pullanase mixture. Increasing

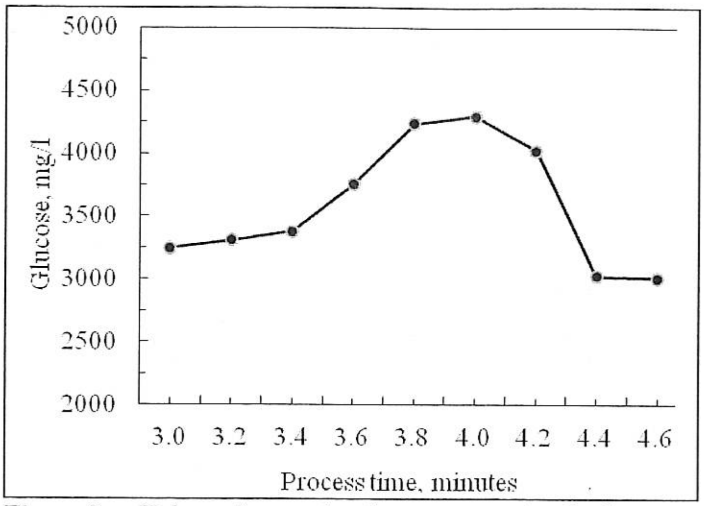

Figure 3.- *pH depending on the glucose generation by the amyloglucosidase/pullanase*

pH to 4.0 resulted in higher glucose amounts, while pH of 4.2-4.6 in smaller glucose amounts. From these results was chosen an optimum pH of 4.0.

4. CONCLUSIONS

Textile processing is a growing industry that traditionally has used a lot of water, energy and harsh chemicals. As textile fibers 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 to textile processing. Biotechnology offers the potential for new industrial processes that require less energy and are based on renewable raw materials. It is important to note that biotechnology is not just concerned with biology, but it is a truly interdisciplinary subject involving the integration of natural and engineering sciences.

When all the benefits of using enzymes are taken into consideration, it`s not surprising that the number of commercial applications for enzymes is increasing every year. Enzymes can be used in order to develop environmentally friendly alternatives to chemical processes in almost all steps of textile fibre processing. New enzymes with high specific activity, increased reaction speed, and tolerance to more extreme temperatures and pH could result in development of continuous processes. The textile industry can greatly benefit from the expanded use of these enzymes as non-toxic, environmentally friendly compounds.

A commercial enzyme that used in the food industry was utilized in this study.The performance of this commercial enzyme amyloglucosidase/pullanase mixture was tested and were performed optimization trials to investigate any further increase in the glucose content of the de-sizing liquor of a starch-sized cotton fabric. Optimum circumstances obtained were: enzyme dosage – 1.0% (owf); process time – 80 minutes at 60 °C; pH – 4.0.

There is still considerable potential for new and improved enzyme applications in future textile processing. It seems that in the future it will be possible to do every process using enzymes.

5. REFERENCES

[1] Ciechańska, D., Kazimierczak, J., *Enzymatic Treatment of Fibres from Regenerated Cellulose.* Fibres & Textiles in Eastern Europe, 14, No 1(55), 92-95 (2006).

[2] Marcher, D., Hagen, H. A., Castelli, S., *Entschlichten mit Enzymen.* ITB Veredlung, 39(3), 20-32 (1993).

[3] Gupta, R., Gigras, P., Mohapatra, H., Goswami, V. K., Chauhan, B., *Microbial α-amylases: a biotechnological perspective.* Process Biochemistry, 38, 1599-1616 (2003).

[4] Cavaco-Paulo, A., Gübitz, G. M., *Textile processing with enzyme*, Woodhead Publishing, Cambridge, 2003, p. 240.

[5] Hashem, M. M., *An approach to a single pretreatment recipe for different types of cotton.* Fibers Text. East. Eur., 15(2), 85 (2007).

[6] Opwis K, Knittel D, Kele A, Schollmeyer E., *Enzymatic recycling of starch-containing desizing liquors.* Starch/Starke 51, 348–353 (1999).

[7] Schacht, H., Kesting, W., Schollmeyer, E., *Perspectiven Enzymatischer Prozesse in der Textilveredlung.* Textilveredlung, 30, 237–243 (1995).

[8] Fersht, A., *Enzyme structure and mechanism* (second edition), de W.H. Freeman & Company, New York, 1985, p. 475.

[9] Maarel, M. J. E. C., Veen, B., Ultdehaag, J. C. M., Leemhuis, H., Dijkuisen, L., *Properties and applications of starch-converting enzymes of the α-amylase family.* Journal of Biotechnology, 94, 137–155 (2002).

[10] Kirk, O., Borchert, V., Fuglsang, C. C., *Industrial enzyme applications*. Current Opinion in Biotechnology,13, 345-351 (2002).

[11] Buschle-Diller, G., Yang, X. D., *Enzymatic Bleaching of cotton Fabric with Glucose Oxidase*. Textile Research Journal, 71(5), 388-394 (2001).

[12] Tzanov, T., Calafell, M., Guebitz, M., Cavaco-Paulo, A., *Bio-preparation of cotton fabrics*. Journal of Biotechnology, 93, 87-94 (2002).

[13] Crabb, W.D. and Shetty. J.K., *Commodity Scale Production of Sugars from Starches*. Current opinion in Microbiology, 2, 252-256 (1999).

[14] Norouzian, D., Akbarzadeh, A., Scharer, J. M. & Moo Young, M., *Fungal glucoamylases*. Biotechnol. Adv., 24, 80–85 (2006).

[15] Aniş, P., Davulcu, A., Eren, H. A., *Enzymatic Pre-Treatment of Cotton. Part 1. Desizing and Glucose Generation in Desizing Liquor*. Fibres & Textiles in Eastern Europe, 16(4), 100-103 (2008).

[16] Sarita, S., Shirmir, D. B. and Mehnaaz, F. A., *Supporting Information GOx signaling triggered by aptamer-based ATP detection*. Electronic Supplementary Material (ESI) for Chemical Communications, 1-9 (2012).

[17] Feitkenhauer, H., *Anaerobic digestion of desizing wastewater: influence of pretreatment and anionic surfactant on degradation and intermediate accumulation*. Enzyme Microb. Technol. 33, 250–258 (2003).

[18] Lange, N.K., *Lipase Assisted Desizing of Woven Cotton Fabrics*. AATCC Review, 29 (6), 23–26 (1997).