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Chemistry and Technology of Biocatalyzed Nanoengineering of Linen Textile Materials

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Abstract—The concept of nanoengineering of linen textile materials, realizing spatially localized effects of protein catalysts on polymeric cellulose companions and securing selective splitting of impurity compounds with preservation of technologically valuable nanoformations of binders in the fiber structure, is presented. The regulated degree of conversion of polysaccharides with the use of degradation products as secondary reagents for chemical transformations of lignin and polyphenolic compounds leads to the production of materials with new or improved functional and consumer properties.

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INTRODUCTION

In developed foreign countries textile manufacturers become more and more active in using the processes of biocatalysis at many technological stages of processing natural fiber materials, including cleaning of cotton fibers from natural impurities (8%), removing of starch sizing agents applied in the weaving process (14%), refining of color of the denim products (20%), obtainment of flax cotton from flax raw materials (5%), softening (3%), biopolishing, and antipilling treatment of materials (11%), intensification of bleaching (3%) and coloration effects (2%), biomineralization (4%), glazing (3%), special types of finish providing materials with new consumer properties (5%), wool biocarbonization (3%), degumming of natural silk (2%), and treatment of the finishing plant wastewaters (17%) [1].

At the same time, as demonstrated by the status analysis of the Russian market of textile products, domestic producers exhibit a rather low activity in abandoning out-of-date chemical methods for treatment of fiber materials. Apparently, one of the reasons for the observed stagnation is the textile industry withdrawal from state control and insufficient interest of the private sector in investing into knowledge-intensive biochemical technologies despite the optimistic forecasts of the Russian and foreign experts with regard to their high effectiveness, environmental safety, and social significance.

However, the inevitability for the Russian textile manufacturers to enter a new quality development level is evident, especially under current environmental conditions, a growing number of industrial and biological disasters, and allergic diseases of the population. Even now the leading global textile manufacturers are oriented at increasing a share of application of highly ecofriendly methods for treatment of fiber materials. In this case manufacturing of textile products with properties which were impossible to achieve using traditional chemical treatment methods is ensured not only due to a highly-selective character of biocatalysis processes but also due to implementation of fiber materials nanoengineering techniques, the essence of which is in the spatially limited impact of the protein catalyst nanosized molecules on the fiber matrix.

This principle lays the foundation for technologies of bioscouring of cotton textiles, involving regulated enzymatic influence on the nanolayer of the primary cell wall of the cotton fiber [2–4], where the majority of impurities are located. Due to higher orderliness of cellulose fibrils the secondary cell wall of the fiber is difficult to access for the globules of biocatalysts and, thus, it remains almost undamaged.

Nanoengineering methods also form the basis for wool treatment processes [5–7], involving the influence of keratinase enzymes on the layer of exocuticle (protein component of the wool fiber cuticle), which

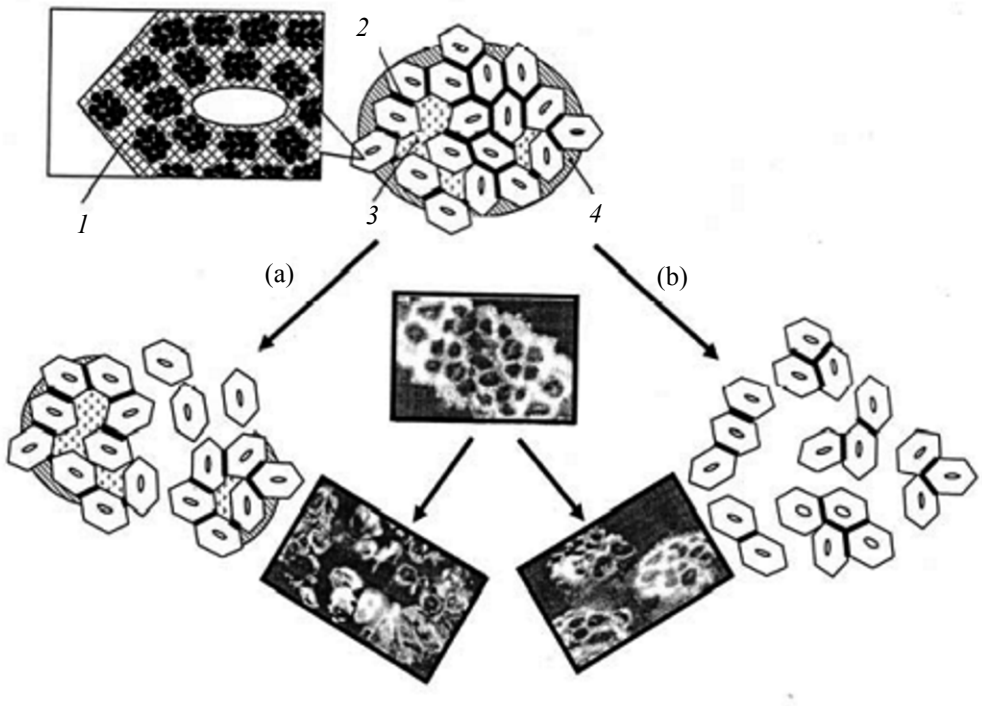


Fig. 1. Gradation of connective tissues in the structure of complex flax fiber and modification of their conditions as a result of (a) alkaline and (b) enzymatic treatment: (*1*) cell wall lignin-carbohydrate complex; (*2*) middle lamella; (*3*) intercellular formations; and (*4*) incrusts.

fixes the layer of epicuticle and is characterized by an increased content of cysteine protein in the molecules, ensuring the formation of multiple interchain and intrachain disulfide bridges. Biotreatment can replace chlorine treatment and cause softening and reduced peelability effects not due to shearing of the epicuticle but due to achieving its better adherence resulting from breaking of disulfide bonds in the layer of exocuticle and the formation of new bridges with the epicuticle proteins.

The widest opportunities for implementation of nanoengineering biochemical methods are found in linen production. This fact is related to the specific features of the morphological structure of flax fiber materials. Flax fiber is a multicomponent polymer system with multilayer organization, which makes it a perfect object for implementation of spatially localized biomodification techniques.

Principles of Biocatalyzed Nanoengineering of Linen Textile Materials

Within the framework of textile production flax fiber materials are processed in the form of complex fibers broken through the thickness, as elementary flax fiber has a smooth spindle-like surface and, unlike wool or cotton, possesses no clinging ability. At the spinning stage the processes of flax fiber preparation are aimed to ensure equal breaking of bast fiber bundles into groups of fibers of the optimal thickness and length, which is a rather difficult technological task solved through a number of multistage long-term actions [8–10]. For this purpose, mechanical methods of splitting and stretching of semifinished fiber materials (heckling) are used in combination with partial chemical splitting of cellulose companions, contained in industrial fiber of the flax roving, completed in the cycle of the woven fabric bleaching and finishing.

The required degree of fineness and uniformity of the formed yarn is ensured by means of rational splitting of substances binding elementary fibers together, at the same time, avoiding separation of the latter (yarn elementarization). For this purpose, if traditional chemical treatment processes are applied it is a common practice to regulate the amount of extractable non-cellulosic components. However, in order to prevent elementarization of the fiber material it is necessary to set limits not on the general total of

splittable impurities but on certain types of their structural formations.

Figure 1 contains a scheme illustrating the structure of industrial flax fiber taking into account the gradation of adhesive materials used in the plant biology depending on their massiveness in the structure of plant tissues. The following structural components of complex flax fiber are considered technologically valuable non-cellulosic polymer formations, maximal preservation of which is reasonable to ensure in the processes of flax roving preparation for spinning.

The lignin-carbohydrate complex of the cell wall is formed by sparsely crosslinked structures of lignin with hemicelluloses (mainly based on β-xylans) and pectin, binding cellulose macrofibrils of elementary fibers together; the complex ensures amorphousness and flexibility of elementary fibers [11] and quality of the yarn twisting.

Middle lamella are formed by layers of pectic substances and hemicelluloses in regions of firm adherence of the adjacent plant cells; their thickness does not exceed 100 nm. They are responsible for the strength of the adhesive contact of elementary fibers in the process of industrial fiber breaking.

Butt joints of elementary fibers are formed by lignin contained in sharp ends of plant cells. They ensure longitudinal adhesion of elementary fibers in case of splitting of complex fibers (not shown in the scheme).

Also there are greater structural formations of impurities, the size of which is comparable to the cross dimension of elementary fibers and can reach 15 μm. They hinder the spinning process, in connection with which it is necessary to perform as complete splitting of such formations as possible.

Incrusts are the remains of parenchymal tissues of the flax stem, located on the surface of the broken bast fiber bundles in a continuous layer or in fragments and binding large groups of elementary fibers together.

Intercellular formations are represented by deposits of adhesive substances between loosely joint plant cells, binding groups of elementary fibers together inside the bundle.

Incrusts and intercellular formations are hetero-polymeric systems containing pectic substances, neutral branched polysaccharides (mainly based on β-glucans), and glycoproteins, ensuring chemical cross-linking with the functional groups of pectins in the carbohydrate-protein complex. A part of intercellular formations contains lignin growing from butt joints in the form of rigid gridshell structures, which significantly deteriorates fiber processing in spinning, increases the breakage rate, and irregularity of the yarn [12], as well as hinders the achievement of the required degree of whiteness and softness of the woven fabric. The share of woody (lignified) intercellular formations is increased in fiber bundles from the root part of the flax stem and significantly increased in case of late flax pulling.

In the context of linen textile production coarse fiber types containing as much lignin as 5.5% and even 8 wt % account for more than 40% of the Russian raw material base [13]. Processing of such raw materials on the basis of traditional chemical treatment methods is difficult due to the fact that it is not possible to perform lignin destruction in lignified regions without breaking it in butt joints and the cell wall.

Another problem is searching for methods of rational splitting of adhesive materials in incrusts and intercellular formations of industrial fibers without damaging middle lamella. In particular, according to the data of microscopic studies of the surface of bast fiber bundles it is found [14] that it is not possible to destroy incrusts without losing spinning properties of the fiber if traditional chemical treatment methods are applied. These difficulties are related to the fact that chemical reagents cause degradation of polymers in all structural regions of the fiber, making it impossible to carry out selective splitting of impurity deposits. In this situation delicate middle lamella suffer significant damage, while solid deposits of incrusts and intercellular formations in the complex fiber structure remain insufficiently destroyed. As a consequence, the process is accompanied by a great number of elementary fibers splitting out of the bundle, which leads to a 10–25% loss of raw materials with downy waste of the spinning operation.

According to the data of the Lamberti Company, Italy [15], linen yarn from flax roving produced on the basis of a two-stage technology, involving alkaline boiling and peroxide oxidation, is characterized by high irregularity of structural, physical, and mechanical properties, which is reflected in high values of variability coefficients with respect to the yarn count – 2.75%, breaking length – 16.4%, and

breaking elongation – 12.2%. In this context, statistical data on the number of thickenings and thinnings with deviations exceeding half of the yarn average diameter (1911 and 1246 defects per 1000 m of the yarn, respectively) and the number of knots 3 and 5 times greater than the average diameter value (6152 and 836 defects per 1000 m of the yarn, respectively) also make a good illustration.

Biochemical methods for nanoengineering of linen textile materials make it possible to successfully solve the task of rational splitting of cellulose companions and ensure more even breaking of industrial flax fiber in the spinning process with minimization of the elementarization processes. Moreover, apart from selective splitting of large structural formations of adhesive substances of the complex fiber these methods provide conditions for intensification of lignin degradation in lignified intercellular regions and a possibility to establish a technology for bleaching of semifinished materials without using environmentally hazardous chlorine-containing oxidizing agents, at the same time, coping with high rigidity, which is a major drawback of Russian linen products.

The methods of nanoengineering of linen textile materials are based on the effects of spatially localized degradation of polymeric companions of flax cellulose in the presence of biocatalysts, ensuring selective action with preservation of nanosized formations of adhesive substances in the structure of complex and elementary fibers. The factor governing the course of chemical transformations at the structural level is the size of the enzyme globule reaching from 6–10 to 50–80 nm. Nanosized globules exhibit catalytic activity only in the regions of the fiber material that are not only accessible for penetration of protein molecules but also ensure spatial complementarity of the enzyme active site and the regions of the splittable material macromolecule [16].

The developed technological approaches to nano-engineering of semifinished linen textile materials using protein catalysts are based on the results of research works carried out in the following fields:

– selection of a multienzyme composition for making an impact on certain types of polymeric cellulose companions;

– dosed extraction of impurities from fiber materials at consecutive stages of textile production;

– determination of the degree of catalytic activity of the pectolytic enzyme complex taking into account their individual and cooperative effects, as well as the specific features of the chemical structure of polyuronide compounds contained in flax fiber;

– assessment of possibilities to use fermentation products as secondary reagents to control the course of redox transformations of lignin and other polyphenol compounds.

It is reasonable to perform selection of enzymes taking into account the required degree of removal of impurities from the fiber matrix. The following mathematical model was developed based on the results of works studying the influence of the residual content of polyuronides (P, wt %), hemicellulose (HC) compounds, and lignin (L) under conditions of their selective removal from the flax roving on linear density (T, tex), flexibility (F, mm), and specific breaking tenacity (BT_{SP}, cN/tex) of the resulting yarn. The composition–property mathematical model describing interdependence of the indicated parameters is given below:

$$T = 64.474 + 0.712P + 0.438P^2 - 4.733L + 1.281L^2 - 1.728HC + 0.093HC^2;\ r = 0.939;$$

$$F = 39.446 - 0.650P - 0.059P^2 + 10.201Л - 2.246L^2 + 3.908HC - 0.199HC^2;\ r = 0.974;$$

$$BT_{SP} = 12.705 + 0.559P - 0.370P^2 + 3.402L - 0.756L^2 + 0.751HC - 0.043HC^2;\ r = 0.959.$$

The identified patterns of influence of polymeric cellulose companions on the properties of the resulting yarn make it possible to find out that the increase in spinning properties of the fiber and quality of the resulting yarn is ensured in the first place by the removal of pectic substances, which act as an adhesive base of connective tissues in the structure of the bast fiber bundle, and lignin. The presence of extremum points on the curves of dependence of the textile material quality indicators from the content of lignin reflects the necessity of lignin degradation in lignified intercellular formations and undesirability of its splitting in butt joints binding elementary fibers together (accompanied by an increase in T and a decrease in BT_{SP}) and in the cell wall (a decrease in F). The extreme character of dependences of the yarn properties from the content of hemicelluloses confirms the technological necessity to preserve a part of neutral polysaccharides, as branching of their macromolecules ensures amorphization and increased flexibility of the fiber. After preparation of flax fiber for spinning the technological optimum of the residual content of

Table 1. Results of studies on chemical composition and patterns of enzymatic splitting of polyuronides in selective Varieties flax fiber and industrial raw materials of russian and foreign producers

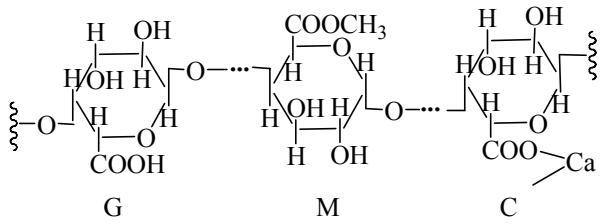
Type of fiber raw material	Share of structural units			Correlation coefficients for function (1)				
	G_G	G_M	G_C	*a*	*b*	*c*	*d*	*e*
Selective Variety A-93	0.26	0.51	0.23	3.19	10.62	0.68	0.38	4.57
Selective Variety Mogilevskii-2	0.25	0.63	0.12	1.79	12.68	1.17	0.99	9.88
Selective Variety Lenok	0.21	0.64	0.15	1.87	12.95	0.95	0.69	7.37
Selective Variety Aleksim	0.20	0.61	0.19	3.15	11.36	0.77	0.43	5.15
Selective Variety Zaryanka	0.20	0.51	0.29	3.56	10.64	0.55	0.35	4.52
Kostroma flax	0.31	0.59	0.10	1.74	11.43	1.01	0.47	8.37
Biisk flax	0.29	0.54	0.17	2.83	11.28	0.75	0.53	5.66
Tver flax	0.22	0.62	0.16	1.92	12.45	0.77	0.49	5.71
Vologda flax	0.19	0.59	0.22	2.29	11.31	0.71	0.41	4.24
Kaluga flax	0.19	0.61	0.20	3.12	11.35	0.78	0.42	4.32
Dutch flax	0.52	0.38	0.10	6.94	0.77	0.99	0.89	8.36
French flax	0.57	0.35	0.08	7.62	0.74	1.01	0.90	10.86

polyuronides in the fiber amounts to 0.35–0.45 wt %, of lignin 2–2.5 wt %, and of hemicelluloses 8–10 wt %.

Primary degradation of the polyuronidic adhesive base of binders in the course of pre-spinning fiber preparation is also observed by Lamberti specialists [15]. In particular, a significant improvement in the yarn quality indicators when using Bioprep, a pectin-degrading enzymatic agent produced by the Novozymes Company (Denmark), is shown. Despite the good results of application of this agent in foreign practice, its efficiency for processing of Russian flax fiber is insufficient; it is proposed to strengthen its effect by adding cellulolytic enzymes [17, 18]. However, on the basis of the authors' own experience and the opinion of the specialists from the Central Scientific Research Institute for Complex Automation in Light Industry [19] it is possible to speak about inadvisability to use cellulases at the stage of flax roving preparation. Their unfavorable impact is primarily related to intensification of β-glucans degradation in the structure of technologically valuable hemicellulosic compounds. The different effect of Bioprep on Russian flax fiber types is caused by the specific character of the chemical composition of pectic substances contained in this agent; and understanding of this interconnection makes it possible to optimize the composition of the applied multienzyme agent.

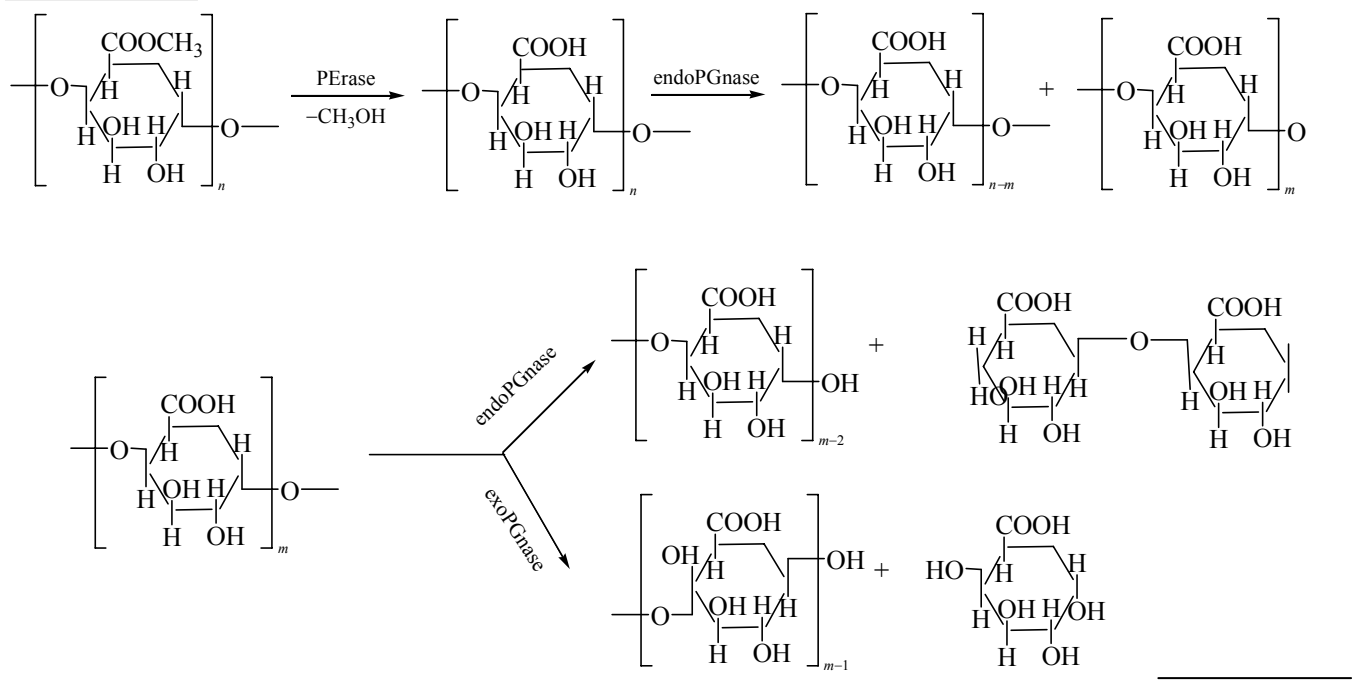
Patterns of Biocatalyzed Splitting of Pectic Substances in Preparation of Russian Flax Fiber

Pectic substances are represented by a complex of high-molecular compounds belonging to the class of polysaccharides, the main structural components of which are the units of galacturonic acid (G), its methoxylated derivatives (M), and calcium or magnesium salts (C), forming cross links between polymer chains:



The following four types of pectolytic enzymes take part in biocatalyzed hydrolysis of pectic substances: endopolygalacturonase (endoPGnase), exopolygalacturonase (exoPGnase), and exopolygalacturonosidase (exoPGdase), which affects α-1,4'-glycoside bonds in the polymer chain, and pectinesterase (PErase), catalyzing splitting of the ether bond in methoxylated units of polyuronide. In accordance with the present day ideas [20], degradation of polyuronides follows the general pattern of the serial-parallel effect of depolymerases.

According to the data [20, 21], the initial stage of hydrolysis takes place with dominant influence of either endoPGnase or PErase as the main depolymerizing enzyme depending on the ratio of unetherified and methoxylated forms of monomeric units in pectin. In particular, a high content of galacturonic acid units creates conditions for endoPGnase to display activity. Its effect discontinues in regions containing substituents (methoxylated or calcium-pectate forms) in the composition of elementary units, as the presence of these substituents breaks complementarity of the substrate molecule and the active site of polygalacturonase molecules. For efficient degradation of high-methoxylated pectin prior participation of PErase enzyme is necessary to convert the polymer into a free polygalacturonic form, which is affected by endo- and exodepolymerases resulting in the formation of oligomeric and low-molecular products as follows:



Therefore, optimization of conditions for biochemical removal of adhesive substances from the structure of bast fiber bundles in order to increase the quality of the produced yarn should be performed taking into account the structure of polyuronides.

According to the existing data, the degree of pectic substances methoxylation greatly varies in different plant types, as well as in the process of the plants maturation. Thus, sugar beet pectin is a low-methoxylated compound, containing ~35% of the methoxylated form [22, 23]. Apple and citrus pectin is 70–78% methoxylated [24], while cotton pectin is 85% methoxylated [22]. The results of studies on the structure of polyuronides of flax fiber materials are presented in works [25–27]. The applied methodological approach to assessment of conditions of carboxyl groups based on IR spectroscopy of polymer films is described in work [25].

Table 1 contains the results of the analysis of polyuronides in dew-retted flax fiber obtained from selective varieties of fiber flax cultivated under similar conditions at an experimental field of the Scientific and Research Institute of Agriculture (Kostroma obl., harvest of 2005), as well as from several types of industrial flax raw materials processed by textile manufacturers in 2006–2007.

It is found that pectins extracted from Russian flax types are high-methoxylated compounds, containing 1.4–1.6 times more etherified units (G_M) as compared to fibers from Dutch or French raw materials. It is registered that the studied pectins of Russian-produced flax also have a higher content of the calcium-pectate form (G_C) as compared to the imported fiber (up to 2.9-fold). As a consequence, a share of unsubstituted units of galacturonic acid (G_G), in relation to which endo- and exodepolymerases display activity, is 1.8–

2.7 times lower in Russian flax raw materials than in the samples of pectin contained in the structure of the imported fiber.

The results of studies dedicated to the kinetics of pectic substances degradation based on the use of agents with a varying ratio of the catalytic activity of the contained endo- and exogenous polygalacturonases (endoPGnase and exoPGnase, U ml^{-1}) and pectinesterase (PErase, U ml^{-1}) make it possible to find out correlation ratios for determination of the pectin splitting degree during 1 hour of biotreatment (ΔP, %/h) for all samples of flax materials [21, 27]. The obtained general pattern looks as follows:

$$\Delta P_i = a + b \cdot \text{PErase} + c\cdot\text{endoPGnase} + d\cdot\text{exoPGnase} + e \times \left(\frac{\text{endoPGnase}\cdot\text{exoPGnase}}{\text{endoPGnase} + \text{PGnase}}\right). \quad (1)$$

This pattern makes it possible to evaluate (according to the ratio of multipliers, see Table 1) the individual contribution of each type of enzymes, as well as manifestations of cooperativity in the effects of depolymerizing enzymes. The last summand formulated as a ratio of the product of the activity indicators of biocatalysts to their sum reflects the synergistic effect of the enzymes most adequately.

A generalized kinetic model of pectic substances degradation is developed on the basis of particular solutions. This model is applicable to all types of flax fibers and ensures adjustments for manifestations of activity of the pectinase complex components taking into account the level of the polymer accessibility for the corresponding types of enzymes. This model reflects the influence of the content of monomeric units in methoxylated and calcium-pectate forms in pectin on fiber processing efficiency with a sufficiently high correlation degree (r):

$$\Delta P_{\text{total}} = 0.21 + 12.37 \times \text{PErase}G_M + 1.39 \times \text{endoPGnase}^{1-G_C} + 0.71 \times \text{exoPGnase}^{1-G_C} + 1.40 \times \left(\frac{\text{endoPGnase}\cdot\text{exoPGnase}}{\text{endoPGnase} + \text{PGnase}}\right)^{1-G_K};$$

$$r = 0.9993. \quad (2)$$

Thus, a contribution of PErase enzyme grows in proportion to the increase in the content of monomeric units in pectin in the etherified form. A correcting factor ($1 - G_C$) of summands, reflecting the activity of depolymerizing enzymes, takes account a share of polymer chain units that are potentially susceptible to catalytic splitting, including both unetherified units and the methoxylated form, which is subjected to demethoxylation under the influence of PErase (i.e. exclusive of the calcium-pectate form). The model reflects a decrease in intensity of polyuronides degradation resulting from the high content of calcium-pectate units in the polymer chain observed for Russian flax raw materials. One of the methods to increase the efficiency of using bio-agents in processing of flax raw materials in solutions is the application of specially selected chelate compounds, capable of extracting Ca^{2+} ions from pectic substances with the formation of highly stable complexes, which makes it possible to reduce the content of the calcium form of galacturonic acid in the polymer by a factor of 2.5–3.

Verification of the proposed model [28] demonstrates that within a 95% confidence interval for determination of the degree of pectic substances splitting using multienzyme agents the difference between the experimental findings and the values calculated according to the above-given formula (2) does not exceed 3%. This fact confirms the efficiency of the proposed mathematical model with regard to the selection of composition of pectolytic enzymatic agents.

Application of the kinetic model (2) is not limited to the possibilities of forecasting or comparison of efficiency for various types of pectolytic agents according to the level of activity of the enzymes contained in the substances. First of all, the model is recommended for adjustment of formulations of industrial enzymatic solutions applied in textile production taking into account the characteristics of the structure of polyuronides of flax raw materials under conditions of different batches, suppliers, crop years, and varieties of the material.

Practice has shown that “universalization” of bio-agents based on obvious excess of enzymes or application of components unnecessary for some process or other is absolutely unacceptable for textile production; such a way significantly increases the costs of biotechnologies. Only well-timed adjustments of the applied formulations taking into account the properties of the modified fiber material make it possible to avoid inappropriate spending of enzymes and to increase profitability of biotechnologies, thus,

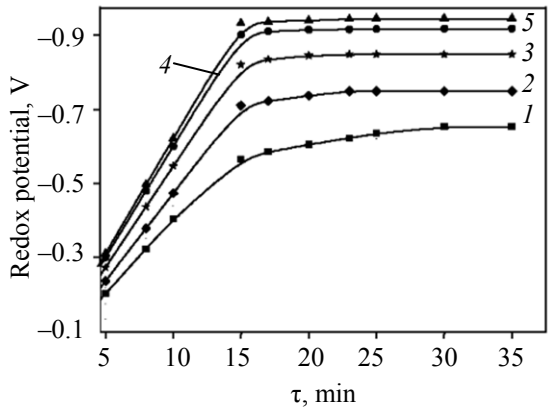


Fig. 2. Kinetic curves of redox potential dynamics in solutions of monosaccharides under anaerobic conditions: (*1*) glucose; (*2*) mannose; (*3*) galactose; (*4*) galacturonic acid; (*5*) xylose. Monosaccharide concentration 5.6 mM l^{-1}, pH = 11; temperature of 98 ºC.

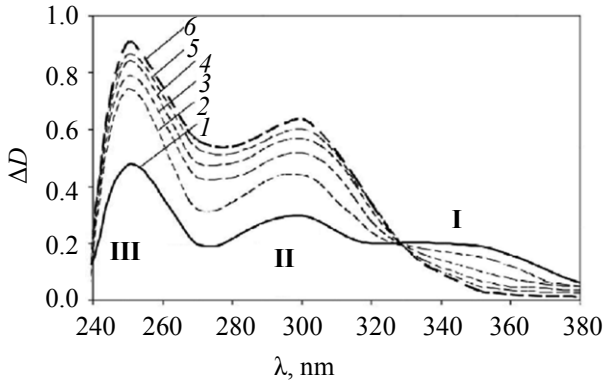


Fig. 3. Differential UV-spectra of alkaline lignin and its condition after treatment of polymer in solutions of reducing agents: (*1*) lignin; (*2*) galactose; (*3*) galacturonic acid; (*4*) xylose; (*5*) Na formaldehyde sulfoxylate; (*6*) Na borohydride.

creating innovative attraction for their application in textile production.

Method for Selective Degradation of Lignin in Flax Fiber

At present enzymatic degradation of lignin is performed only in the form of laboratory experiments. In particular, works [29–31] describe a mechanism of polymer transformation with participation of lignin peroxidase or Mn^{2+}-dependent peroxidase and accompanying groups of oxidoreductase enzymes and redox mediators. However, industrial production of biocatalysts of the required complex compositions has not been started yet in international practice and is not even expected in the short term.

We propose an original technique for complex splitting of polymer components in lignified intercellular formations based on redox transformations of lignin under the influence of enzymatic degradation products of polysaccharide impurities contained in them. While to extract components of the carbohydrate-protein complex of adhesive substances it is sufficient to perform their splitting to oligomers, a possibility to generate reactive monosaccharides in the system and use them as secondary reagents for implementation of desired chemical transformations is also taken into account.

The variety of compositions of polysaccharides in flax fiber (hemicelluloses and pectic substances) and the heteropolymeric character of their chemical structure predetermine a widest range of the obtained aldopyranose compounds. Work [32] demonstrates that reducing properties of aldose solutions depend on specific features of the chemical and stereoisomeric structure of the cyclic form of monosaccharides, which determine the stability of the pyranose cycle and the rate of tautomeric transition of molecules into the aldehyde form. The reducing capacity of solutions is increased when heated and alkalized, which is related to the reactions of retroaldol splitting of monosaccharides with breaking of the bond between carbon atoms and an increase in the content of reducing agents with shorter carbon chains [33, 34]. Dependence curves in Fig. 2 illustrate the dynamics of the growth in the reducing properties of solutions of certain polyoxyaldehydes formed, in particular, in the course of profound degradation of flax fiber polyuronide compounds. As can be seen, solutions of xylose, galacturonic acid, and galactose are the most reactive systems. Their influence on lignin is comparable to the effect of such strong reducing agents as sodium borohydride and sodium formaldehyde sulfoxylate (rongalite).

On the basis of the differential UV-spectroscopy method applied for aqueous and alkaline solutions of dioxane lignin [35] it is confirmed that the activity of reducing agents with respect to lignin is related to the presence of carbonyl groups in the composition of 20% of phenyl propane units of lignin [36]. Moreover, in 1/3 of them carbonyl is located in α-position of the propane segment, ensuring the auxochromic effect (strengthening of color as a result of the formation of the conjugated system of double bonds with aromatic fragments of the lignin molecule). Figure 3 demonstrates transformation of the spectrum of the

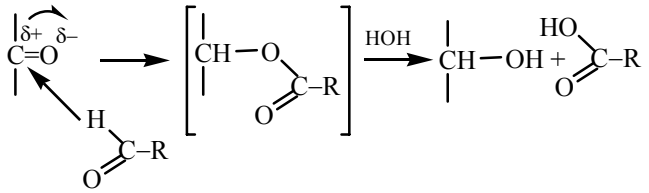
Table 2. Assessment of influence of flax roving reduction treatment on lignin reactivity

Reduction system	Redox potential (371 K), mV	Klason lignin content L_K,[a] mg g^{-1} of fiber	Degree of lignin removal after peroxide treatment of roving ΔL, %[b]
$NaBH_4$	–1205	29.6	84
Sodium formaldehyde sulfoxylate	–1030	30.8	79
Xylose, galacturonic acid, galactose	–910	36.3	70

[a] L_K parameter for fiber samples after enzymatic treatment or alkaline-reductive boiling (in coarse fiber and after alkaline boiling $L_{K(coarse)}$ = 60.5; $L_{K(alk/boil)}$ = 53.2 mg g^{-1} of fiber). [b] Value of ΔL for the process of alkaline boiling/peroxide treatment is 50%.

initial alkaline lignin as a result of its treatment with solutions of reducing agents. The spectra have peaks at wave lengths of 350, 300, and 250 nm, which mark the absorption of lignin by ionized phenolic units, correspondingly, in form (**I**), containing auxochromic carbonyl group [37], as well as in the form of coniferyl (**II**) and *n*-coumaric (**III**) alcohols.

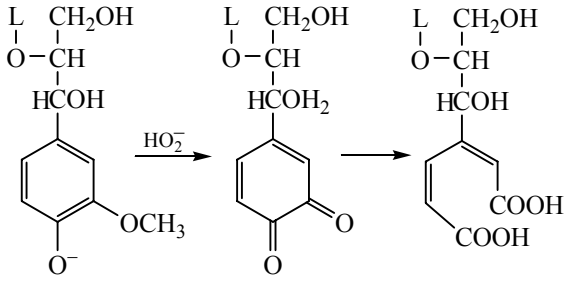
The presence of the isobestic point in the series of spectral curves testifies to mutually consistent transformations of form **I** into one of the reduced forms of structural units **II** or **III** depending on the presence or absence of the methoxyl group in the composition of chromophoric cycles. Similarities in the changes of the spectral curves make it possible to judge on uniformity of the processes in solutions of monosaccharides and borohydride, the effect of which on lignin is described in work [38]. It is evident that like in case of borohydride the transformation of carbonyl group in the presence of polyoxyaldehydes goes in accordance with the nucleophilic addition mechanism:



At the same time, it is found that auxochromic carbonyl reduction increases the number of reaction centers for sulfiting of lignin, which, as is known [36, 38], is based on nucleophilic substitution on α-atom of carbon. In the process only units in the free phenolic form undergo transformations, while structural fragments containing fixed phenolic hydroxyl are not exposed to sulfiting.

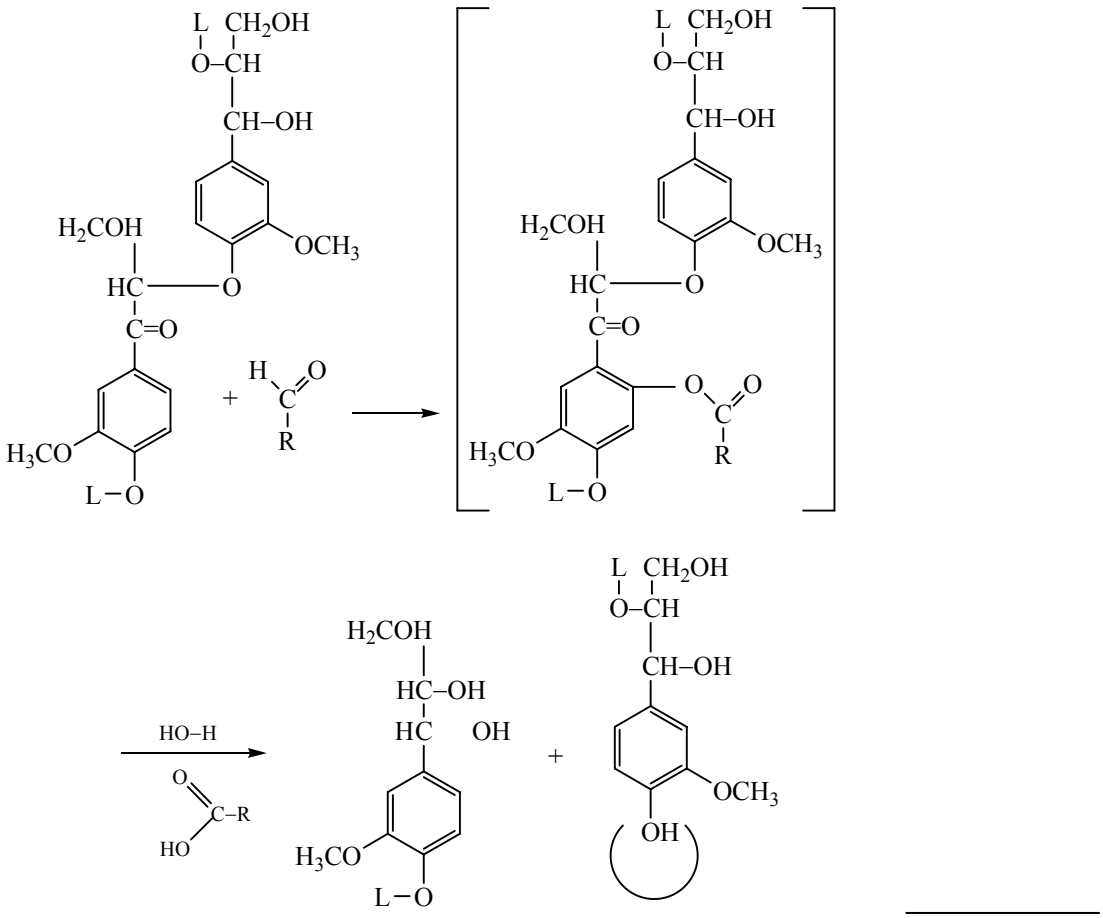
The results given in Table 2 provide objective evidence of an increase in reactivity of flax roving lignin after reduction treatment. The residual content of lignin insoluble in a 78% sulfuric acid (Klason lignin) in the fiber after the treatment with reducing agents decreases 1.96-fold and under the influence of polyoxyaldehyde solutions it is reduced by a factor of 1.67, while as a result of alkaline hydrolysis under conditions of traditional alkaline boiling this figure decreases not more than 1.14-fold.

Apparently, an additional number of units in the free phenolic form appear under the influence of polyoxyaldehydes as a result of destabilization and breaking of the ether bond between the structural units of lignin adjoining to the reducible carbonyl:



Difficulties in discoloration of lignin with hydrogen peroxide are related to the fact that chromophoric centers in units, phenolic hydroxyl of which is involved in the formation of ether bond between the structural elements of the polymer, do not undergo degradation. An increase in the content of units in the free phenolic form in lignin leads to significant intensification of the process of the following oxidation attack of the chromophoric centers by hydrogen peroxide (see scheme bellow).

The results of determination of lignin extraction degree ΔL indicate that the proposed alternative of enzymatic and peroxide treatment, which is a little inferior in efficiency to the alternative involving application of sodium borohydride or formaldehyde sulfoxylate before exposure to oxidizing agents, makes it possible to increase the degree of oxidative degradation of lignin by a factor of 1.4 as compared to alkaline-peroxide treatment of flax roving.



The production of polyoxyaldehydes is ensured by introduction of exogenous (saccharifying) enzymes into the composition of multienzyme agents for splitting of polyuronides in flax fiber. As indicated above, it is undesirable to break hemicelluloses in flax roving; therefore, it is impossible to use endogenous enzymes depolymerizing macromolecules of galactan and xylan. The required concentration of highly-active aldoses can be obtained as a result of complete conversion of degradable pectic substances, which, as is known, contain units of neutral saccharides in side branches. In this connection, it is recommended to add exogenous enzymes of xylosidase, galactosidase, and exopolygalacturonase in addition to main depolymerizing enzymes, ensuring maceration of complex flax fiber through degradation of the carbohydrate-protein complex of adhesive substances.

It is proved by experiments that exposure to these enzymes does not cause significant depolymerization of hemicelluloses; however, as a result of complete degradation of oligomeric polyuronides in intercellular formations it is possible to reach the required conditions for the reduction potential of the system to achieve the level exceeding –0.9 V in the course of activation treatment [13, 30]. The developed value of the potential is sufficient for redox transformations of lignin formations, located together with splittable polysaccharides, to take place. At the same time, nanosized formations of lignin in butt joints and in the cell wall of elementary fibers remain undamaged, which is principally different from the effects of traditional reductive boiling of the fiber.

The developed methods of maceration-delignification enzymatic treatment formed the basis for technologies of modification of flax fiber materials with application of bioagents. These technologies completely cover the cycle "roving–yarn–woven fabric–bleaching of coarse fabric–special finish."

Advantages of Biochemical Nanotechnologies for Production of Linen Textile Materials

A method proposed for efficient processing of Russian flax raw materials in the spinning industry is based on enzymatic-peroxide treatment of flax roving,

which is applicable to flax fibers of soft and medium grades [40]. The sequence and conditions of flax fiber processing were selected taking into account technological requirements to the process of preparation, characteristics of the polymer composition of flax fiber, and the specific character of the activity displayed by protein catalysts.

In particular, it is proved by experiments that the stage of deacidification directly before enzymatic treatment is necessary in order to remove acidic products of degradation of plant tissues of the flax stem, which are formed under conditions of primary treatment of the flax straw and extraction of flax fiber raw materials. This operation prevents enzymes from losing activity in the process of coarse roving treatment without introducing special buffer agents into the multienzyme solution. A bio-agent Polifan ML was created and approved for implementation of this method in industry. Polifan ML is an optimized complex of pectolytic enzymes and compatible proteases, which are necessary for degradation of glycoproteins in the carbohydrate-protein complex of connective tissues.

The cycle of flax roving preparation with the use of the enzymatic agent Polifan ML makes it possible to reduce the content of pectic impurities in the fiber by 80% and the content of proteins by 85%, at the same time, preserving 75% of hemicelluloses [39]. The enzymatic treatment increases lignin accessibility in lignified regions for alkaline and oxidizing agents in the following peroxide treatment, which contributes to a 1.35–1.4-fold decrease in the amount of lignin as compared to the coarse roving. As vividly illustrated by the data of Table 3, the optimal removal of impurities from fiber materials ensures increased uniformity of splitting of flax complexes in the processes of spinning without elementarization of the fiber and with no losses in the form of downy waste. This stage of treatment makes it possible to reach a 25% increase in the yield of high-quality linen yarn of grade 1 with a unique combination of properties: an increase in the yarn fineness and strength with a simultaneous decrease in variability coefficients with regard to linear density by a factor of 2 and to breaking strength –2.1.

For preparation of highly-lignified flax fiber varieties with increased rigidity which are difficult to process it is proposed to use a version of enzymatic-peroxide treatment ensuring maceration splitting of the adhesive base of incrusts and intercellular formations and achievement of reducing capacity by products of hydrolysis of polysaccharides, which is necessary for intensification of lignin degradation [41, 42]. Polifan MDL, a multienzyme maceration-delignifying agent, is developed to be used within the framework of this technology. Polifan MDL is an optimized composition of pectin-degrading enzymes with the globule size of 50–80 nm, proteases, and exogenous hemicelluloses. The enzymatic treatment within the framework of the technological process of the flax roving enzyme-peroxide treatment is performed in the following two stages:

stage I – localized splitting of adhesive substances of incrusts and intercellular formations at 50 °C for 110 min and profound degradation of polyuronide compounds with the production of monosaccharides;

stage II – activation of monosaccharides and reductive degradation of lignin at 95–100 °C; duration of the process selected taking into account the growth rate of the reducing capacity of polyoxyaldehydes (see Fig. 2) is 15–30 min.

Then the textile is subjected to oxidation at hydrogen peroxide concentration of 0.6–0.8 g l^{-1} (in terms of active oxygen) and total alkalinity of 6.5–7.0 g l^{-1} (in terms of NaOH) for 90–95 min.

In comparison with the traditional technology the biochemical method for preparation of highly-lignified fiber for spinning makes it possible to process highly rigid roving more efficiently and obtain fine yarn meeting the requirements of GOST 10078-85 State Standard for grade “1 highly linen.” As demonstrated by the data of Table 3, the resulting yarn is characterized by a significantly reduced number of thickening and thinning defects. It is an important indicator confirming the quality of semifinished material as at further technological stages fiber breakages are likely to take place at sections with such defects. After biochemical modification based on the proposed methodology the number of knots exceeding the average diameter of the fiber by a factor of 1.5 is reduced 3.8-fold and the number of thinnings less than 0.7 of the diameter decreases 2.4-fold. Moreover, the produced yarn has higher values of strength, deformation, and elastic properties, which ensures a 4-fold decrease in the semifinished material breakage rate at further stages of repeated winding and in the production of woven fabric.

Table 3. Yarn quality indicators for different product line groups using roving treated in accordance with traditional (control) and biochemical techniques with application of enzymatic agents

Property	Soft fiber		Coarse fiber		Dyed fiber	
	control	polifan ML	control	polifan MDL	control	polifan MDL
Linear density, tex	53.5	50.5	58.9	53.6	54.0	51.0
Yarn count	18.7	19.8	17.0	18.8	18.5	19.6
Thickness, average diameter d_{av}, mm	0.36	0.28	0.38	0.30	0.35	0.29
Variability coefficient for diameter, %	27.0	22.4	32.5	25.0	28.8	24.3
Number of defects per 100 m of yarn						
thickenings exceeding $1.5d_{av}$	250	165	412	108	280	201
thinnings below $0.7d_{av}$	392	73	433	183	400	123
Twisting of yarn, turns of twist per 1 m	560.3	541	536.8	550.0	540.2	556.8
Variability coefficient for twisting, %	9.3	7.1	8.5	7.4	9.4	7.3
Specific breaking tenacity, cN/tex	20.8	25.1	19.9	24.5	17.4	24.8
Variability coefficient for breaking tenacity, %	10	8.3	12.1	8.5	13.8	10.4
Breaking elongation, mm	8	11.8	8.7	11.7	6.1	9
Unit breaking toughness, kgs cm g^{-1}	44.3	48.6	41.7	55.2	39.3	47.9
Variability coefficient for breaking toughness, %	10	8.6	10.4	6.2	12.5	5.7
Resistance to repeated bending and abrasion, cycles	6897	10278	5595	8049	5586	9738
Share of relative elongation components in single-cycle tests, %:						
fast-reversible elongation	0.39	0.31	0.43	0.41	0.42	0.44
slow-reversible elongation	0.15	0.29	0.12	0.27	0.12	0.24
residual elongation	0.46	0.40	0.45	0.32	0.46	0.32
Yarn breakage rate, number of breakages per 100 spindle hours	50	32	80	44	66	32
Lightness of color, %	–	–	–	–	38.2	30.4
Range of lightness variability, %	–	–	–	–	7.2	2.3
Content of tetrabromindigo coloring agent, g kg^{-1}	–	–	–	–	7.7	11.2

In order to produce a range of colored yarn there is a combined method of preparation and vat coloration of roving from flax fibers of soft and medium grades [43]. The technological process is performed with application of Polifan MDL and suggests the use of products of enzymatic degradation of polysaccharides in flax fiber as secondary reagents for redox transformations of vat colors [44].

Combination of operations of the textile material preparation and vat coloration, for example, under conditions of reductive boiling of the roving [8], is problematic due to the low colorability of the material and high unevenness of the obtained colors. These drawbacks are related to low sensitivity of the untreated fiber material to sorption of the coloring agent, as well as to premature transformation of coloring agents into leucoform in the technological solution under the influence of traditionally applied reducing agents, which increases the affinity of the coloring agent to the fiber and hinders its even distribution on bobbins of the processed roving. In

case of separate implementation of the processes of roving preparation and coloration of the formed yarn [45] an additional impact of alkaline-reductive coloring solutions leads to excessive degradation of adhesive substances in flax complex fibers and, as a consequence, results in a reduction of strength and deformation properties of semifinished textile materials, which hinders processing of the materials at the weaving stage. The problem of complex treatment and coloration is relevant not only in relation to the production of colored and colored-woven fabrics but also in connection with the increasing volumes of consumption of colored yarn in knitting production.

Tests have demonstrated that the technology using enzymatic agents ensures higher spinning properties of the yarn and, correspondingly, better structural, physical, and mechanical characteristics of the flax yarn in combination with improved coloristic characteristics of the dyes. The properties of the obtained wet-spun yarn are in compliance with the requirements of grade "1 highly linen." Moreover, colorability of the material is increased: the content of the coloring agent in the fiber is 1.45 times higher than in case of the traditional technology application; lightness of the color is reduced by a factor of 1.25. Uniformity of the color of semifinished textile materials is also improved – variability of the lightness indicator in different regions of fiber material is reduced 3.1-fold.

The composition of the multienzyme agent is developed and the conditions of its application for complex splitting of starch size, pectic substances, and lignin in preparation of woven linen fabrics are elaborated [46–49]. The detected characteristics of the process of enzymatic desizing of linen fabrics [47, 48] are related to partial penetration of starch into the structure of complex fibers and the formation of hybrid polysaccharide fraction, the degradation of which is possible only in case of simultaneous exposure to amylolytic and pectolytic enzymes with the size of protein globules of 20–40 nm. Activation of polyoxyaldehydes produced as a result of such exposure ensures reductive transformations of natural coloring agents belonging to the class of flavanoids, contained in flax fiber, increases their solubility and intensifies the processes of lignin depolymerization described above. The method of enzymatic-peroxide bleaching of linen fabrics with the use of Polifan LT, which is developed and approved in industry [46, 49], makes it possible to reach the necessary level of capillarity and whiteness of textile materials without using the hypochlorination stage. The introduction of a multifunctional biomodification stage together with an increase in environmental indicators of the production process and the resulting products make it possible to reduce the total length of the technological cycle 2.5-fold and cut

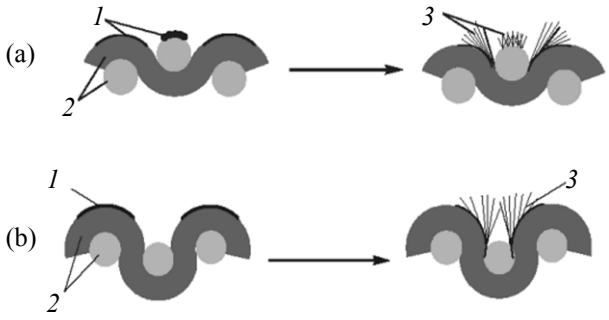


Fig. 4. Biomodified regions of (*1*) elementary fibers in (*2*) linen yarn and their mechanical splitting into (*3*)macrofibrillar bundles in creation of (a) nap-raising peach-skin effect and (b) suede-like effect.

Table 4. Effect of softening treatment on quality of textile materials and resistance of this effect to household use[a]

Agent	Rigidity of analyzed fabric samples, mN cm^{-2}		
	After softening treatment	After single wash	After five washes
Persoftal	31.4	47.7	60.2
Oteksin KS	36.2	42.7	67.5
Tratskan K	32.3	54.1	61.4
Velan	30.5	42.0	64.7
Neonol	47.8	62.1	75.9
Polifan PM	30.6	30.5	30.7

[a] Rigidity of the initial linen fabric is 153 mN cm^{-2}.

heat and electric energy consumption by a factor of 1.4.

Methods of biochemical nanoengineering are promising in the context of increasing consumer properties of the finished products. In particular, new variations of biomodification of linen fabrics at the final finishing stage are developed [50]. A distinctive feature of multienzyme agent Polifan PM is the presence of isoforms of enzymes with the globule size below 10 nm in its composition, which ensures biomodification of the textile material at the level of elementary fibers and, consequently, the achievement of efficient softening of linen fabrics. The influence of the multienzyme agent leads to the completion of degradation of lignin formations, including longitudinal butt joints between elementary fibers; thus, a 5-fold decrease in the rigidity index of the textile material is achieved. Moreover, as demonstrated in Table 4, unlike the results of application of chemical softeners, for the first time it becomes possible to achieve a wash-fast softening effect of textile articles.

A new type of finish, which is the first to allow achieving high-quality nap-raising effects on linen fabrics, is also of interest. Unlike cotton or wool textiles, under normal conditions linen fabrics are not liable to napping due to natural rigidity of elementary fibers. In case of using biochemical nanoengineering techniques it becomes possible to affect the lignin-carbohydrate complex of the cell wall of elementary fibers using enzymatic agents. In this process the interfibrillar matrix undergoes degradation only in a minor part of elementary fibers and not along their entire length but only in regions coming to the cloth bearing surface. In case of splitting of these regions into cellulose macrofibrils their cross breakages can take place in the course of the fiber processing using nap-raising equipment. In this case a uniform layer of undirected pile, the softness of which is attributable to the nanosized thickness of its fragments, is formed. The obtained pile layer possesses high resistance to abrasive effects as unsplitted ends of fibers are locked in the structure of the yarn. As a result of the yarn twisting new regions of elementary fibers coming to the bearing surface are subjected to biomodification and mechanical impact of the nap-raising equipment in every wave of weave of the woven fabric, due to which the mechanical strength of the fabric is reduced negligibly.

For different product line groups of linen fabrics with various structure and type of weave there are alternative textures of pile outlined in Fig. 4. A peach-skin alternative ensures an effect of delicate velvetiness. In particular, it is used for backing of suiting fabrics contacting with the human body and eliminates the feeling of cool surface, which is characteristic for linen textile materials. A suede-like alternative is applied on fabrics with three-dimensional weave and ensures masking of the structure. It can be recommended for the right side of the outerwear and for obtainment of conditioning materials with reduced heat transmission capacity.

Achievement of unique consumer properties of linen fabrics based on the new technology applied for their final finish makes it possible to broaden rather limited opportunities of linen fabrics design, as well as to increase consumer demand and competitive ability of the product.

CONCLUSIONS

Biochemical technologies for processing of linen textile materials are protected by a package of patents [40, 41, 43, 46, 50], prepared for implementation, and supported by a complex of services for analytical and organizational support both at the initial adjustment and commissioning stage and under conditions of continuous operation. This approach ensures maximum processing efficiency in combination with minimization of production costs.

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