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COMPREHENSIVE FORMATION OF LEATHER SEMI-FINISHED PRODUCTS USING ENZYMES

The object of research is the process of complex formation of elastic leather using enzymes of proteolytic and hydrolytic action.

One of the most problematic areas is the decrease in the elasticity of the leather semi-finished product at the stage of its dehydration. Enzyme treatment increases the mobility of the microfibrillar structure of the semi-finished product due to the destruction of physical intermolecular bonds.

During the study, a proteolytic enzyme was used at the stage of bating the pelt and enzymes of hydrolytic action were used to treat the tanned chrome semi-finished product.

A semi-finished product was obtained, which is characterized by an increase in porosity compared to the original semi-finished product. The porosity of the semi-finished product increases by 22% in the case of using enzyme treatment at the bating stage and by 67% with repeated treatment of the tanned semi-finished product with enzymes. This is due to the fact that the proposed enzyme treatment promotes the removal of glycosaminoglycans from the dermis at the bating stage. Further use of enzymes after tanning of the semi-finished product contributes to the destruction of carbohydrate bonds with collagen macromolecules, which ensures an increase in its physicochemical properties. The peculiarity of this effect can be explained by the presence of an active center in enzymes, which forms enzyme-hydrocarbon-collagen complexes with carbohydrates and collagen macromolecules. Inside the formed complexes, the destruction of existing bonds occurs and the separation of carbohydrates from the collagen of the dermis.

This provides the possibility of obtaining a leather semi-finished product, which is characterized by an increase in the tensile strength and elongation at 9.8 MPa by 8.4 and 23.0%, respectively, and these indicators reach 20.7 MPa and 48.0% compared to the indicators of the tanned semi-finished product.

Keywords: *enzymatic plasticization, enzymes of proteolytic and hydrolytic action, leather semi-finished product, physicochemical properties.*

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1. Introduction

The technology of leather production is characterized by multi-stage, the use of a large number of chemical reagents of different composition. To a large extent, this is due to the specific composition of natural raw materials and the need to obtain leather material with a set of specified properties. Taking into account the properties of leather raw materials and component composition, depending on its type, already at the first stage of formation of elastic material, its dee p structural changes occur. At the same time, after alkaline treatment, it is necessary to remove calcium, interfibrillar proteins from the obtained semi-finished product as much as possible and increase the mobility of the fibrillar structure of dermis collagen. This is accompanied by peptization of non-collagenous proteins, a significant decrease in the density of the dermis structure and an increase in the activity of collagen macromolecules. In the future, during acid-salt treatment of the semi-finished product, the structural distances between macromolecules increase, which contributes to the effective volumetric interaction of the main chromium compounds with dermis collagen. To increase the mobility of the fibrillar structure of tanned collagen after neutralization of the semi-finished product, it is necessary to stabilize the mobility of the dermis elements for further liquid finishing processes.

In this regard, there is a need to develop new and improve existing technologies with the economy of natural raw materials when using chemical reagents of special action at different stages of manufacturing a semi-finished product of increased elasticity. At the same time, taking into account the technological necessity of using a significant number of environmentally hazardous reagents, including chromium compounds, reducing their costs is of significant importance. To ensure the high quality of the obtained leather materials, scientifically substantiated conditions for carrying out physicochemical processes and operations provided for by the technological regulations are necessary.

At different stages of the technology of production of natural leather, environmentally safe biologically active reagents of catalytic action are used – enzymes of various chemical composition [1]. In leather production, hydrolases, enzymes of the maltavamorin series [2] or proteases [3] are traditionally used. In the production of elastic leather, the soaking- liming and tanning processes are of great importance, especially with the use of enzymes. The use of enzymes in the soaking-liming processes of leather production is due to the acceleration of the soaking process by using alkaline proteases without damaging the dermis structure [4]. At the same time, the fiberization of the structure is enhanced to increase its accessibility at the following stages of technological processing [5]. At the soaking stage, the use of amylolytic, lipolytic and proteolytic enzymes is effective [6], which contribute to its rapid and complete hydration, removal of contaminants [7]. It has been shown that the use of lipase contributes to the acceleration of water absorption by raw materials, the destruction of soluble dermis proteins, and the dispersion of fats [8]. At the same time, enzymes of proteolytic action play an active role in the structural transformations of dermis collagen. They enhance the reactivity of chemical reagents and contribute to the energetic destruction and removal of ballast protein-hydrocarbon complexes from the dermis [9]. At the stage of degreasing the leather semi-finished product, the use of alkaline lipases and their combination with proteases is envisaged [10]. The use of enzymes for degreasing the semi-finished product allows replacing part of the surfactants [11].

The use of enzymes at the dehairing stage improves the quality and physical and mechanical properties of the leather [12] and reduces the burden on the environment [13]. When liming, special enzymes are used to improve the quality of the pelt and during the bating process, acidic and neutral proteases are used [14]. The quality of the leather can also be improved by using the elastase enzyme, which increases its elastic-plastic properties. The dehairing of the leather material and the effective removal of interfiber substances can be achieved by using neutral proteases in combination with hydrogen phosphate and magnesium ions [15]. In [16], the authors recommend using the enzymes Lithudac L and Novo Bate WB in an acidic environment for dehairing. When preserving hair, the combined use of alkaline protease with thiourea (II) oxide is known. To soften the pelt, proteolytic enzymes are used [17], which contribute to the loosening of dermal collagen and peptization of non-collagenous components [18].

Reducing the duration of soaking fur raw materials for fresh-dry preservation and reducing the defect rate of hide during mechanical processing is achieved by increasing their plasticity [19]. For this purpose, a number of enzyme reagents are used, such as amylosubtilin G3x, maltavamorin G10x, pectofoetidin P10x. At the same time, when processing sheepskin with dense skin tissue and dimensions of more than 90 sq. dm, the enzymes maltavamorin G10x, protosubtilin G3x, pectofoetidin P10x are used in combination with surfactants. This accelerates the removal of lipids, contaminants, protein and hydrocarbon substances from hair and skin tissue. It should be noted that surfactants perform a dispersing function in relation to the products of destruction of the dermis components of skin tissue during mechanical action on it. The activity of such enzymes significantly depends on the temperature and pH of the environment.

It is also important to ensure the accessibility of the structure at the tanning-finishing stage, which contributes to the acceleration of the diffusion of chemical materials into the dermis structure, their uniform distribution and fixation without excessive shrinkage of the structure [20]. The treatment of the semi-finished product with enzymes [21] contributes to increasing its hydrothermal stability and vapor permeability. At the liquid finishing stage, enzymes ensure more efficient selection of synthetic dyes from the working solution, their uniform distribution and binding to the dermis structure [22]. The use of enzymes in combination with dyes accelerates their diffusion into the dermis structure [23].

Thus, the analysis of publications on the use of enzymes at different stages of the technology of elastic leather production indicates the specificity of their catalytic action on the raw material and the semi-finished product obtained from it. In this regard, there is a need to conduct scientific research to find and establish optimal conditions for the use of enzymes. In particular, this concerns the use of a complex of enzymes for the gradual formation of the properties of an elastic leather semi-finished product in pre-tanning and post-tanning treatments.

Thus, *the object of research* is the process of complex formation of elastic leather using enzymes of proteolytic and hydrolytic action.

The aim of research is to determine the influence of enzymes of proteolytic and hydrolytic action on the physicochemical properties of a leather semi-finished product.

To achieve the set aim, it is necessary to perform the following tasks:

1. To investigate changes in the physicochemical properties of a leather semi-finished product under the influence of enzymatic treatment.
2. To determine the sorption-diffusion properties of the semi-finished product at different stages of its formation.
3. To determine the gradual changes in the physicomechanical properties of the semi-finished product during the formation of an elastic leather material.

2. Materials and Methods

The work used raw materials from cattle – beef hides with an average paired weight of 17 kg, a thickness of 2.4–3.7 mm. To study the process of tanning-bating of unpaired pelt, the hide samples were treated in an alkaline medium using sodium sulfide and calcium hydroxide, respectively, in concentrations of 0.7 and 12 g/l. In this case, the water/mass ratio of the paired raw materials was 5/1 at a temperature of 18–22°C and duration of 5 and 12 days. After that, the obtained pelt was delimed with ammonium sulfate with subsequent bating. To soften the pelt, the enzyme pancreatin was used at a temperature of 35–37°C and a pH of the technological medium of 7.8–8.5 and a duration of the process of 1, 2 and 3 hours.

To soften the obtained pelt – a dehaired and delimed semi-finished product, the enzyme pancreatin technical with an activity of 600 units/g was used, which contains a number of proteolytic enzymes – proteinases and peptidases, the main of which is the proteinase trypsin, as well as lipase, amylase and elastase. At the beginning of liquid finishing, the tanned semi-finished product was treated with a solution of high-temperature enzymes of hydrolytic action of bacterial origin. Amylosubtilin and glucavamorin were used – respectively, alpha-amylase and glucoamylase preparations with activities of 1500 ± 150 and 3000 ± 300 units/g. The choice of different enzymes for the specified stages of processing is associated with their different effects on the structure of the dermis. It should be noted that enzymes that effectively remove glycosaminoglycans from the interfibrillar structure are used to soften the pelt. In post-tanning treatments, enzymes are used that destroy hydrogen bonds during dehydration of the semi-finished product, which ensures its elasticity.

To reduce the duration of the tanning processes and post-tanning treatment with hydrolytic enzymes, a semi-finished product obtained using resource-saving technology was used. This technology involves carrying out the tanning process with reagents, % of the raw material mass: water – 130, sodium hydrosulfide – 1.5; calcium hydroxide – 4.0; sodium sulfide – 1.5 for 12 hours in the spent soaking solution at a temperature of 27–29°C.

To obtain a structured leather semi-finished product, the pelt was pickled at a temperature of 23–25°C for 3.0–3.5 hours. Before tanning, the pelt was treated with an electrolyte-resistant emulsion of dipropylene glycol sulfonated ether at a consumption of 0.5% for 10 minutes. Chromium (III) sulfate with a basicity of 36% was added to the spent technological solution with a consumption of 1.2% Cr_2O_3. After the cut of the hooves was completely stained with a tanning solution, the basicity of chromium (III) sulfate was increased with magnesium oxide. The tanning of the hooves was completed at a pH of the technological solution of 3.9–4.2. At the same time, after tanning, the semi-finished product acquired a heat resistance of 104–105°C. After the samples were left to stand for one day, they were cut into 1.2–1.4 mm thick. Further processing of the tanned semi-finished product was carried out with high-temperature enzymes amylosubtilin and glucavamorin in a ratio of 1/1 with a consumption of 0.2% of the mass of the semi-finished product for 10–15 min. At the same time, the consumption of water with a temperature of 75–70°C was 100%. Samples of leather semi-finished product were dried at a temperature of 20–23°C to residual moisture of 16%.

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To study the structural transformations of dermis collagen in the processes of tanning and bating under the action of the enzyme pancreatin, a method was used to determine the amount of melted gelatin before the beginning of the destruction of the main polypeptide chains of collagen from the treated pelt. In this case, the amount of melted gelatin is determined in a solution at a temperature of 65 ± 1°C for 1.5 hours. To assess the effectiveness of the processes of processing the semi-finished product using enzymes, tests of the sorption-diffusion and physico-mechanical properties of the obtained semi-finished product were carried out according to the methods [24]. At the same time, before determining the complex of physico-chemical properties of the delimed and bated semi-finished product, their alcohol-ether dehydration was carried out. The physical and mechanical properties of the leather semi-finished product were determined on a RT-250M tensile machine (Ukraine) by deforming the samples at a speed of 90 mm/min. All test samples were standardized under normal conditions before testing.

3. Results and Discussions

The research results of liming-bating process of the pelt obtained by resource-saving technology are given in Table 1. As the data obtained show, with an increase in the liming duration by 2.4 times, the melting of gelatin from the delimed pelt increases from the dense area – the rump by 2.8 times, and from the bottom areas – by 3.4 times. At the same time, the efficiency of gelatin melting is higher in the bottom areas – by 21.4%. The melting of gelatin from the bated pelt within one hour with liming duration of 5 days with an increase in the enzyme concentration by 3.7 times increases by 1.7 times for the rump and by 2.0 times for the bottom areas. At the same time, in the case of liming for 12 days, such an increase in gelatin melting is manifested for the rump and bottom, respectively, by 2.0 and 1.2 times. The results obtained indicate that gelatin melting is facilitated by an increase in the enzyme concentration. Therefore, the duration of the bating process for 1 h at a pancreatin concentration of 0.6 g/l is sufficient for the pelt ash for 5 days to obtain elastic leather. This may indicate the pelt production, in the structure of which amide intermacromolecular bonds are destroyed and carboxyl and amine groups are released. An increase in the duration of the ash can also is accompanied by the destruction of polypeptide chains of collagen macromolecules.

Table 1

Gelatin melting from different topographical areas of delimed and bated pelt

Semi-finished product area	Liming duration, days	Gelatin melting, % of dry mass			
		De-limed	Bated, hours		
			1	2	3
Rump	5	3.0	8.7/14.9	7.1/14.5	5.7/14.2
Rump	12	8.3	12.4/27.4	12.7/28.4	12.4/28.8
Bottom area	5	4.0	12.1/23.8	11.7/23.1	11.4/18.7
Bottom area	12	13.5	34.0/41.2	29.9/37.5	29.8/36.7

Note: pancreatin consumption during bating in the numerator and denominator, respectively, 0.6 and 2.2 g/l

To assess the influence of the layered density of the pelt on the features of the dermis structure, a study of the kinetics of gelatin melting from the papillary and reticular layers from the rump area of the pelt was conducted. The results of this study during scalding at a pancreatin concentration of 2.2 g/l and a temperature of 18–22°C for 5 days are given in Table 2. The data obtained show that at the maximum concentration of pancreatin under the specified technological conditions after deliming of the pelt, gelatin melting from the papillary layer is 3.6 times greater than that from the reticular layer. However, after bating the pelt for 1 hour, gelatin melting from the papillary layer increases. At the same time, the duration of bating has a slight effect on gelatin melting. When comparing the melting of gelatin from the papillary and reticular layers after bating for 1 hour in relation to the delimed pelt, an increase of 5.5 and 7.4 times is observed, respectively. Thus, the efficiency of melting gelatin from the bated pelt increases significantly after 5 days of ashing. At the same time, the absolute values of gelatin melting indicate almost 100% destruction of collagen of the papillary layer of the dermis, with the exception of the basal membranes.

Table 2

Layer-by-layer melting of gelatin from the bated pelt

Pelt	Gelatin melting, % dry weight, from the layer	
	Papillary	Reticular
Delimed	16.9	4.7
Bated 1 hour	93.6	34.8
Bated 3 hours	99.5	36.1

The research results of the pelt bating obtained by implementing the resource-saving technology with pancreatin are given in Table 3. With an increase in the enzyme concentration in the working solution from 0.125 to 0.5 g/l, the melting of gelatin from the rump area of the pelt increases and reaches a maximum value at a consumption of 0.05% of the pelt mass. At the same time, the condition of the obtained pelt corresponds to the organoleptic assessment as good. At lower concentrations of pancreatin, the pelt quality decreases. At the same time, the melting of gelatin from the rump area increases significantly at water/pelt ratio of 1.5/1.0–1.0/1.0.

Table 3

Properties of bated pelt when using pancreatin

Consumption, % of the mass of the raw material		Gelatin melting, % of dry mass	Organoleptic evaluation of the bated pelt
Pancreatin	Water		
0.0001	400	7.4	unsatisfactory
0.0015	300	7.7	insufficient
0.001	200	8.9	satisfactory
0.05	100	12.1	good

To assess the structural changes and operational properties of the leather semi-finished product obtained by the developed technology, a set of physicochemical properties was determined at different technological stages of its formation (Tables 4, 5 and Fig. 1). It should be noted that the traditional technology [25] does not provide for enzymatic treatment of the tanned semi-finished product. From the data presented in Table 4, it is clear that after the process of pelt bating, all the sorption-diffusion indicators of the semi-finished product increase. At the same time, this applies to the greatest extent to the air permeability from the tanned side and the specific surface area and to a lesser extent to the porosity of the semi-finished product.

After tanning, all the studied indicators decrease and this is most evident in the air-vapor permeability and specific surface area with minimal decrease in porosity. After enzymatic treatment, all the indicators increase, and the air-vapor permeability and specific surface area increase to a greater extent with smaller changes in porosity. The obtained data indicate that at successive stages of formation of the structure of the leather semi-finished product, deep structural changes in the dermis occur. At the bating stage, the pelt is effectively freed from glycosaminoglycans and other non-collagenous components with the destruction of their bonds with the polypeptide chains of collagen.

Table 4

Sorption-diffusion properties of leather semi-finished products at different stages of their formation

Indicator	Semi-finished products*			
	a	*b*	*c*	*d*
Porosity, %	57.0	66.0	62.0	69.0
Maximum water vapor sorption, %	48.0	57.0	53.0	64.0
Hygroscopicity, %	54.0	63.0	52.0	66.0
Specific surface area, m^2/g	112.0	137.0	84.0	141.0
Capillary moisture during watering, %	89.0	106.0	89.0	112.0
Vapor permeability, ml/($cm^2 \cdot h$), from the tanned side	12.0	15.0	11.0	16.0
Vapor permeability, ml/($cm^2 \cdot h$), from the front side	3.5	6.0	3.0	7.0
Air permeability, $cm^3/(cm^2 \cdot h)$, from the tanned side	690.0	910.0	530.0	975.0
Air permeability, $cm^3/(cm^2 \cdot h)$, from the front side	570.0	690.0	380.0	710.0

Notes: * – semi-finished product: *a* –delimed; *b* – bated; *c* – tanned; *d* – enzyme-treated

At the tanning stage, during the structuring of the dermis collagen, new chemical and physical bonds of chromium complexes with the released carboxyl groups of the collagen side chains in its hydrophilic areas are formed. Such a densification of the structure is accompanied by a significant decrease in sorption-diffusion indicators. Further, during enzymatic plasticization, a decrease in the density of the dermis collagen structure and an increase in the mobility of structural elements are observed due to the destruction of hydrocarbon bonds with collagen macromolecules. The specific nature of the change in the structural properties of dermis collagen at different stages of the formation of the semi-finished product is due to the fact that the nature of its porous structure is determined not only by a change in the absolute size of the pores, but also by their size distribution [18–26]. In this case, the features of the interaction of dermis collagen with water when using it as an element in determining a particular indicator are of significant importance. In particular, when determining vapor permeability, not only water is sorbed by dermis collagen, but also its desorption in the process of diffusion through the capillaries of the dermis of the semi-finished product. Thus, the nature of the stage-by-stage change in the properties of the leather semi-finished product indicates that the most structurally sensitive indicators are vapor permeability, air permeability and its specific surface area, and technologically important ones are vapor permeability, air permeability and porosity.

All physical and mechanical properties after bating of the pelt, tanning and enzymatic treatment of the tanned semi-finished product increase, with the exception of the residual elongation after tanning (Table 5, Fig. 1 and 2).

The maximum effect is observed for the residual elongation and stress at crack of the face layer after bating. After tanning, the greatest effect is manifested for the strength of the semi-finished product at break and stress at crack of the face layer. In the case of enzyme-treated semi-finished samples, the maximum effect is on the elongation at break and residual elongation. The remaining parameters have a relatively minimal value.

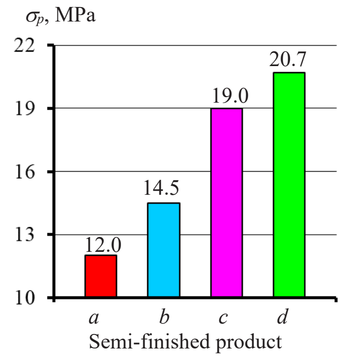

Fig. 1. Dependence of the tensile strength σ_p on the stage of formation of the semi-finished product: *a* – delimed; *b* – bated; *c* – tanned; *d* – enzyme-treated

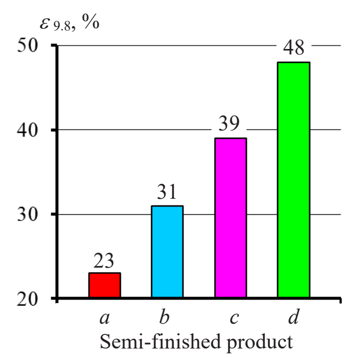

Fig. 2. Dependence of elongation $\varepsilon_{9.8}$ on the stage of formation of the semi-finished product: *a* – delimed; *b* – bated; *c* – tanned; *d* – enzyme-treated

Thus, the nature of the stage-by-stage dependence of the change in the indicators of the leather semi-finished product on the stage of its formation indicates a gradual strengthening of its structure. It should be noted that at the stage of enzymatic treatment of the tanned semi-finished product, the plasticization of the structure of the semi-finished product also occurs due to the use of hydrolytic enzymes. According to the complex of properties, the semi-finished product obtained after enzymatic treatment can be used for the manufacture of a wide range of elastic leather materials [19–27] for footwear and clothing and haberdashery purposes.

Table 5

Physical and mechanical properties of the leather semi-finished product at diff erent stages of its formation

Indicator	Semi-finished product			
	Delimed	Bated	Tanned	Enzyme-treated
Stress at crack occurrence of the face layer, MPa	10.2	14.5	18.8	20.7
Elongation at break, %	36.0	47.0	56.0	77.0
Residual elongation, %	6.0	11.0	8.0	10.5

Research limitations. The studies conducted concern a semi-finished product obtained from large cattle raw materials. In the future, when using another type of leather raw materials, additional studies of the influence of technological regulations on the physicochemical properties of elastic leather are necessary.

Prospects for further research. The use of enzymes in technology allows to obtain leather materials that meet the requirements of international regulatory documents, provided that environmentally friendly and resource-saving technologies are used. In particular, the range of elastic leather materials for various purposes can be expanded.

4. Conclusions

1. The complex of properties of the leather semi-finished product during its staged formation using enzymes of proteolytic and hydrolytic action was studied. At the same time, changes in the physicochemical properties of the pelt were established, including the gelatin melting index. It was found that with an increase in the duration of tanning by 2.4 times, the gelatin melting index from the delimed pelt increases from the dense area – the rump by 2.8 times, and from the bottom areas – by 3.4 times.

2. The complex of sorption-diffusion indices of the delimed, bated, tanned and enzyme-treated semi-finished product was established. In particular, it was found that the semi-finished product after bating and enzyme treatment is characterized by an increase in porosity by 22.0 and 67.0%, respectively, due to an increase in its vapor and air permeability. These processes are due to the removal of glycosaminoglycans from the pelt under the action of the enzyme pancreatin at the stage of bating the semi-finished product. The interaction of basic chromium compounds with collagen of the dermis and the subsequent destruction of carbohydrate bonds with collagen macromolecules under the action of the enzymes amylosubtilin and glucavamorin contributes to the increase in the physicochemical parameters of the semi-finished product.

3. Changes in the physicomechanical properties of the semi-finished product at different stages of the technological cycle were revealed. Thus, the semi-finished product at the stage of enzymatic treatment is characterized by an increase in the ultimate strength at break and elongation at a stress of 9.8 MPa by 8.4 and 23.0%, respectively, and these indicators reach 20.7 MPa and 48.0%. At the same time, enzymatic treatment of the semi-finished product does not lead to a decrease in the strength of the front layer, which brings the stress index at the appearance of cracks in the front layer to 20.7 MPa. The resulting semi-finished product can be used for the production of elastic leather for a wide range of purposes, when it is formed in stages using enzymes of proteolytic and hydrolytic action.

Conflict of interest

The authors declare that they have no conflict of interest related to this research, including financial, personal, authorship, or other aspects that could have influenced the research or its outcomes as presented in this article.

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Manuscript has no associated data.

Use of artificial intelligence

The authors confirm that no artificial intelligence technologies were used in the preparation of this work.

Authors' contributions

Anatolii Danylkovych: Terminology, Conceptualization, Formal analysis, Writing – original draft, Writing – review and editing, Supervision; ***Olena Okhmat***: Terminology, Conceptualization, Investigation, Formal analysis, Writing – original draft, Writing – review and editing.

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