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Effects of Various Techniques of Untanning Of Leather Shavings on The Properties of The Protein Hydrolysate.

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ABSTRACT

There are many recycling techniques of primary waste of chrome-tanned leather, i.e. leather immediately after tanning. We explore possible ways of recycling of secondary tanning waste – low-grade final products (wool skin). The first step in our research was to determine the optimal untanning procedure. The criteria included the duration of untanning, physical and chemical properties of protein hydrolysate. We conducted alkaline heat treatment to obtain protein hydrolysate. We examined its amino acid composition and analyzed correlations between dynamic viscosity of the hydrolysate and the processing parameters. In order to improve the technology of hydrolysate production, we developed a mathematical model of alkaline treatment of collagen-containing waste. This model gives opportunity to choose the processing parameters basing on the desired properties of the resulting hydrolysate.

Keywords: Chrome-tanned leather shavings, untanning, protein hydrolysate, mathematical modelling, wool skin

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INTRODUCTION

The issue of recycling and efficient use of leather industry waste is a real challenge for scientists and manufacturers of the world. Chrome-tanned protein waste can be divided into two types: chrome-containing leather waste immediately after tanning and leather waste after the whole set of currying processes – for instance, low-grade final product, leather scraps. The most hazardous leather waste is produced in case of noncompliance with storage and use regulations due to forming compounds of hexavalent chromium, which are toxic [1]. Considering large amounts of low-grade final products and shavings that remain after currying, recycling secondary tanned waste is an urgent issue.

It is hard to remove chromium compounds from tanned leather waste due to specific features of dermis, high tolerance to hydrothermal treatment. At the same time, resulting waste contains quite valuable organic substances that can be used for production of glue, gelatin, biofilm, products of collagen hydrolysis, protein hydrolysate, etc.

There are various research methods aimed at efficient recycling of chromium-containing leather shavings. For instance, this type of waste, having high sorption capacity, is used to remove oil from industrial discharge waters and polluted coastal territories [2-3], as well as to remove dyes from waste waters of consumer goods industry [4].

Protein extraction from leather waste is another recycling approach, used to obtain products of collagen hydrolysis, collagen matrices, films, protein hydrolysates. Apart from one-stage acid hydrolysis, protein hydrolysate can be obtained from collagen-containing chromium-tanned leather shavings in a two-step process:

1) initial untanning by aqueous solution of different salts;
2) further treatment with enzymes, acids, alkali under an elevated temperature [5-6].

Protein hydrolysates are highly soluble and undergo high-degree hydrolytical degradation (Klompong *et al.,* 2007) [7]. Hydrolysate characteristics directly influence its functional properties (Kristinsson, & Rasco, 2000) [8].

Our research was aimed at studying the effects of various techniques of untanning on physical and chemical properties of protein hydrolysate, as well as developing mathematical model of processing of collagen-containing waste with alkaline solutions. The subject of our study included secondary collagen-containing materials – sheepskin with defects, such as holes, bald patches, marking brands, and cuts.

EXPERIMENTAL

Substances and chemicals

Low-grade products (wool skin) were selected on the premises of OOO “MIP “ECOM”, Ulan-Ude, Russian Federation, according to GOST 1821-75 [9].

All chemicals conformed to Pro Analysi (p. a.) quality.

Experimental techniques

In order to examine physical and chemical properties of sheepskin samples, we used the methods as described in GOST (determination of gravimetric water content [10], ash [11], chromium oxide [12], free fatty substances [13], shrinkage temperature [14], pH of aqueous extraction [15]).

The current research included the following methods of untanning of chromium-containing waste: magnesia, liming, acidic, salt, and alkali-soda. Prior to tanning, hairs were removed, sheepskins were soaked for 12 hours, cut into stripes 1.5-2.0 cm wide, minced to obtain 0.5×0.5 mm pieces. Wet fine-cut samples were weighed. Table 1 summarizes untanning conditions.

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Table 1 – Technological conditions of untanning of chrome-tanned samples of low-grade wool skin

Untanning method	Technological parameters	Type and volume of chemicals	Equipment
Magnesia	Volume ratio=0.5; Pressure 1atm Duration – 6 hours	Magnesium hydroxide – 4% by mass of wet fine-cut samples	VK-75 PT autoclave
Liming	Volume ratio=5; Temperature 18-22 °C; Duration 3-10 days	Slaked lime – 3-4% by mass of fine-cut sheepskin, Ammonium sulfate – 1.5% by mass of fine-cut sheepskin	Enameled vessel V=2000 cm^3
Acidic	Volume ratio=2; Duration 24 hours; Temperature 22 °C	Oxalic acid – 40 g/dm^3 Magnesium hydroxide – 4% by mass of fine-cut sheepskin	Glass vessel V=2000 cm^3
Salt	Volume ratio=2; Temperature 22 °C; Duration 2-3 days	Ammonium oxalate – 40 g/dm^3	Glass vessel V=2000 cm^3
Alkali-soda	Volume ratio=3; Duration 8 hours; Temperature 22 °C	Sodium hydroxide – 100 g/dm^3, Sodium carbonate – 150 g/dm^3	Glass vessel V=2000 cm^3

After untanning of low-grade wool skin, we assessed the residual content of chromium oxide in fine-cut samples according to GOST [12].

Protein hydrolysate was obtained with a previously developed technique [16] with the addition of soda in the amount of 10-40 g/dm^3.

Protein content in the hydrolysate was determined with LK-100 automatic unit (LOIP) for Kjeldahl digestion; LOIP LK-500 Kjeldahl nitrogen and protein analyzer (LOIP).

Amino acid composition was determined by capillary electrophoresis, using “Kapel’105-M” capillary electrophoresis system (Lyumeks) [17].

Dynamic viscosity was measured with DV-II+ Pro viscometer (Brookfield) [18], using ReocalcT Software (Brookfield).

We investigated the effect of alkali treatment on the properties of protein hydrolysate as the result of alkali treatment compared with heat treatment by the method of mathematical modeling [19].

RESULTS

All physical and chemical properties of wool skin samples were compliant with GOST [14]. Visual assessment of wool skin samples after untanning showed that treatment had not led to any changes in the appearance of the samples, except for the exposure to alkali-soda, which led to dissolution of the samples (99.46%).

The diagram (Figure 1) shows the results of the analysis of chromium oxide content in the samples after untanning.

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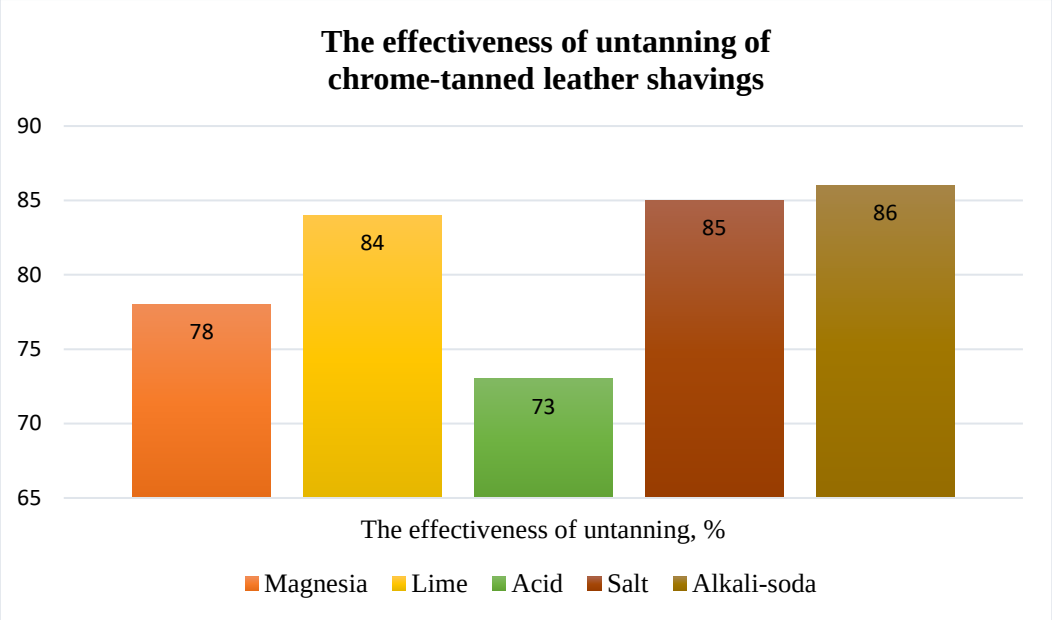

Figure 1 – Effectiveness of untanning of collagen-containing waste

As one can see from the data shown in Figure 1, initial content of chromium oxide in leather was 3.4% on average. The maximal untanning effect was achieved with alkali-soda treatment (86%), whereas acidic method of untanning provided minimal removal of chromium oxide from wool skin leather (73%). The major disadvantage of alkali-soda treatment is significant loss of sample mass, including valuable proteins. High level of efficiency of liming was attained in case of long-term (up to 10 days) treatment of samples, which is unacceptable from the perspective of the industrial application of this method.

Salt and magnesia methods were selected for further production of the protein hydrolysate according to the registered technology [16], as they provide sufficient removal of chromium oxide from leather (85% and 78%, respectively) over a short period of time (6-72 hours). We determined physical and chemical properties of the resulting hydrolysate. The data are shown in Table 2.

Table 2 – Physical and chemical properties of the protein hydrolysate

Parameters	Untanning technique by	
	Magnesia	Salt
Mass fraction of protein, %	18.12	17.97
Residue on evaporation, %	4.72	4.53
pH	10.16	9.38
Density, kg/m³	1025	1030

Figure 2 shows IR-spectra of the protein hydrolysates of samples after different untanning processing by: a) magnesia; b) salt

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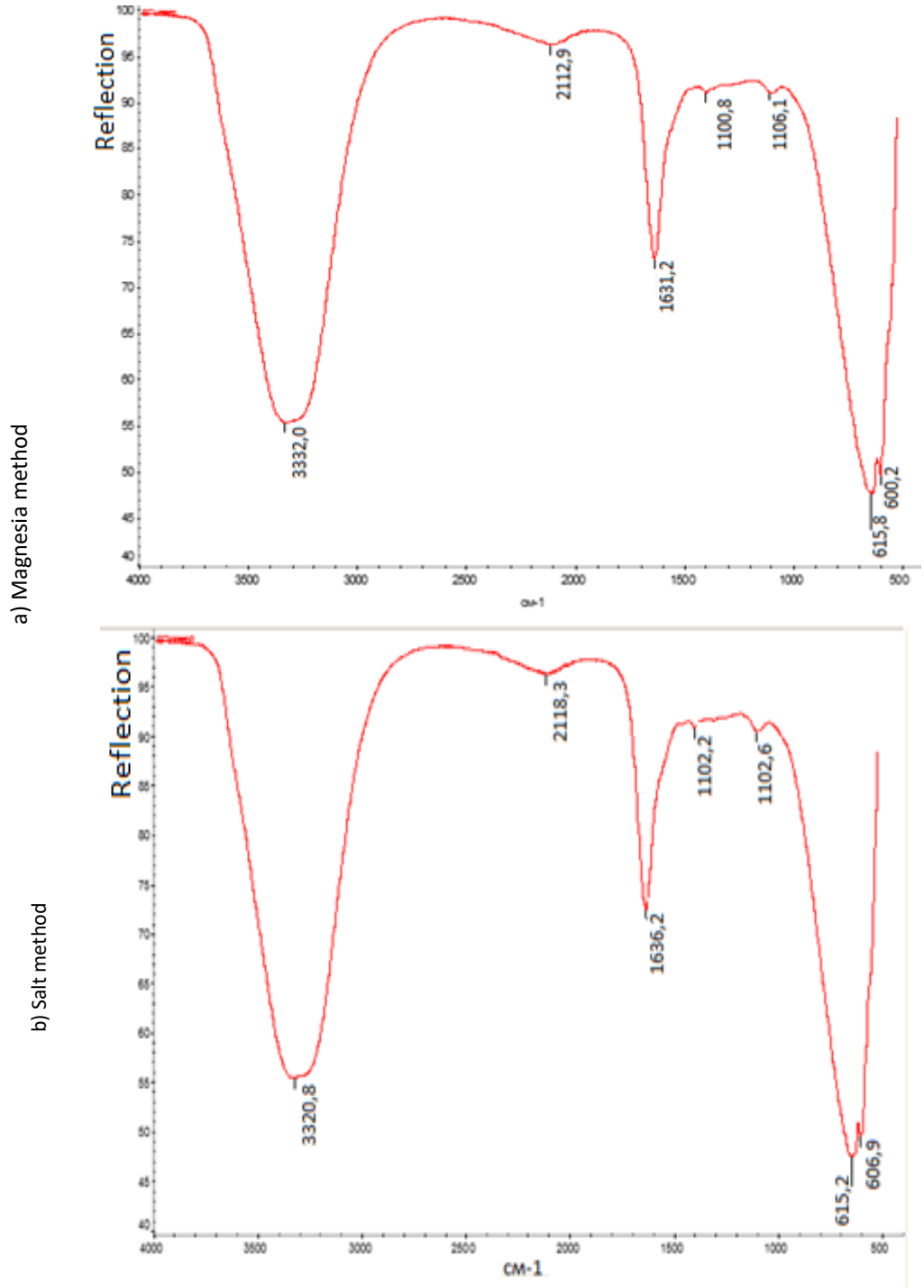

Figure 2 – IR-spectra of the protein hydrolysate

Figure 2 shows that IR-spectra of the protein hydrolysates, obtained with different untanning methods, are identical.

Figure 3 represents the results of the analysis of amino acid composition of the protein hydrolysate after magnesia untanning processing.

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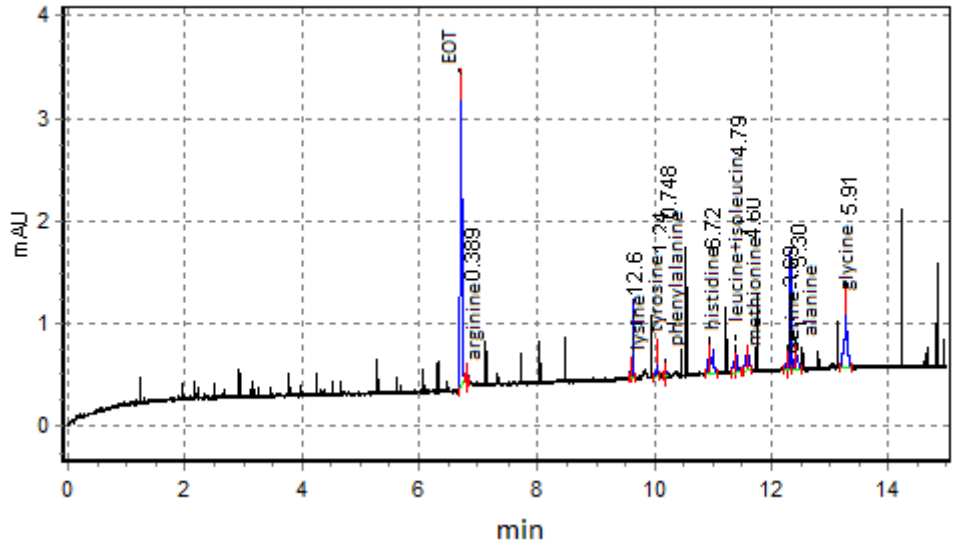

Figure 3 – Electrophoregram showing amino acid composition of the protein hydrolysate after magnesia untanning treatment

Figure 3 shows that the protein hydrolysate contains amino acids, typical for proteins. Lysine has the highest concentration in the protein hydrolysate – 12.6 mg/dm^3, the next amino acid is histidine – 6.72 mg/dm^3, glycine – 5.91 mg/dm^3, alanine – 5.30 mg/dm^3. The lowest observed concentrations belonged to arginine – 0.389 mg/dm^3 and phenylalanine – 0.748 mg/dm^3.

Figure 4 represents the results of the analysis of amino acid composition of the protein hydrolysate after salt untanning processing.

The figure shows that the protein hydrolysate contains amino acids, typical for proteins. The protein hydrolysate has the highest content of leucine+isoleucine – 13.1 mg/dm^3, lysine – 10.8 mg/dm^3, the next amino acid is histidine – 6.71 mg/dm^3, glycine – 12.2 mg/dm^3, alanine – 12.5 mg/dm^3. We observed the lowest concentrations of arginine – 0.346 mg/dm^3, tyrosine – 0.671 mg/dm^3, phenylalanine – 0.699 mg/dm^3, serine – 0.757 mg/dm^3.

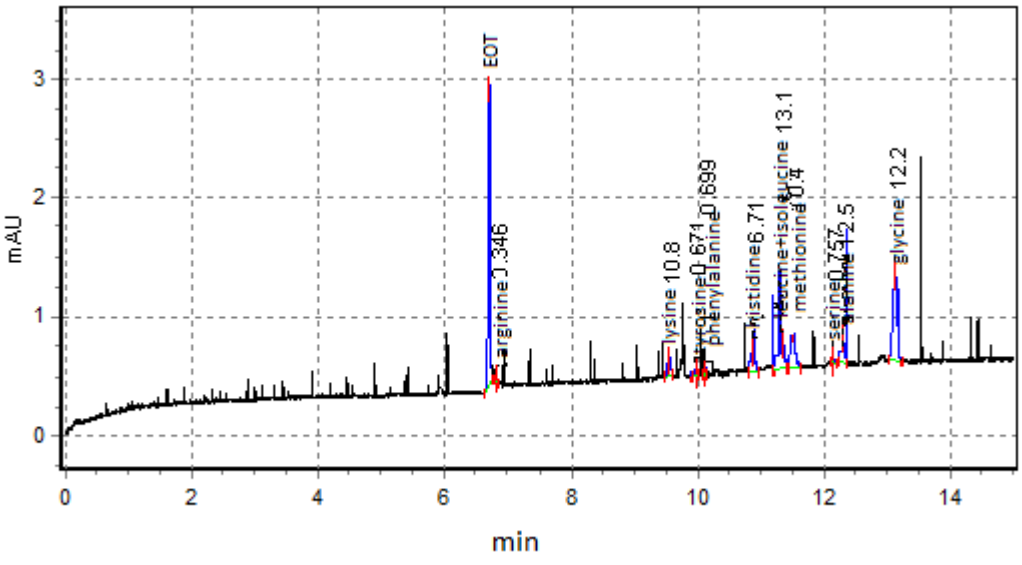

Figure 4 – Electrophoregram showing amino acid composition of the protein hydrolysate after salt untanning treatment

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DISCUSSIONS

Explored untanning techniques allow to markedly decrease the content of chromium oxide down to 0.5-1.0%. Basing on the results of the comparative analysis of the untanning methods, we have come to the following conclusions. Liming technique is unsuitable due to long duration and high water demand, as the treated mass should undergo multiple washing to remove undissolved lime. Alkali-soda method was rejected, as the samples were almost completely dissolved.

IR-spectra of the hydrolysates after magnesia and salt untanning are marked by peaks at 3320 – NH_2 groups, 3000-3500 – CO-NH; 21,187 – NH groups, 1636 – NH, NH_2, CO-NH groups. All these groups are typical for proteins. Identical spectra indicate that proteins are not destructed during untanning or preparation of the protein hydrolysate.

Protein hydrolysate is different from the products of collagenolysis, as it is marked by low molecular weight, indicating massive break-down of molecular bonds within the collagen structure. The analysis of the amino acid composition confirms this idea. The comparative analysis of the amino acid composition of the hydrolysates shows that the set of amino acids is the same in the protein hydrolysates after different untanning treatment. They contain equal number of amino acids – 11. However, the concentrations of individual amino acids are different. For instance, the protein hydrolysate, produced with salt untanning method, contains twice as much glycine and alanine. The concentration of tyrosine, on the contrary, is higher in the protein hydrolysate after magnesia untanning treatment. The discrepancies in the amino acid concentrations can be explained by different impact rate of the untanning agents on the proteins.

We obtained a second order polynomial regression equation. Extended matrices of full factor experiment were set up.

Regression equation for heat treatment in alkaline solutions:

$$y_{t.i} = 1.81 - 0.78x_{1i} - 0.26x_{2i} - 0.07x_{3i} + 0.03x_{1i}x_{2i} + 0.005x_{1i}x_{3i} + 0.0002x_{2i}x_{3i}$$

According to the results of comparing theoretical and experimental data in the course of development of mathematical model, we traced the identical error with the maximum deviation of no more than 0.13%. These parameters suggest that the calculated regression equations provide robust models of alkaline treatment processes, fully describing the influence of individual factors and their joint impact.

We analyzed the regression equations to determine, which factors have the most pronounced influence on solution viscosity. The maximal values of coefficients are observed at x_{1i} and x_{2i}, while the magnitude of their joint influence is comparable to the linear impact of x_{3i} – it means that the linear connections with treatment duration and temperature, as well as their joint influence, have the highest effect on the output. The concentration of sodium bicarbonate has almost no impact on the viscosity of the solution, but the presence of minimal amounts of soda in the solution allows one to reach the maximum value of 5.942; therefore, there is no reason to neglect this parameter. However, one should consider the fact that the regression equation provides mathematical description of three factors within certain ranges; therefore, it is possible to create the surface of influence of the process temperature and duration on dynamic viscosity of the fluid, which will provide approximate demonstration of the process (Figure 5).

We developed a mathematical model of correlation between treatment conditions and dynamic viscosity of the protein hydrolysate and calculated response surface to provide graphical representation (Figure 5)

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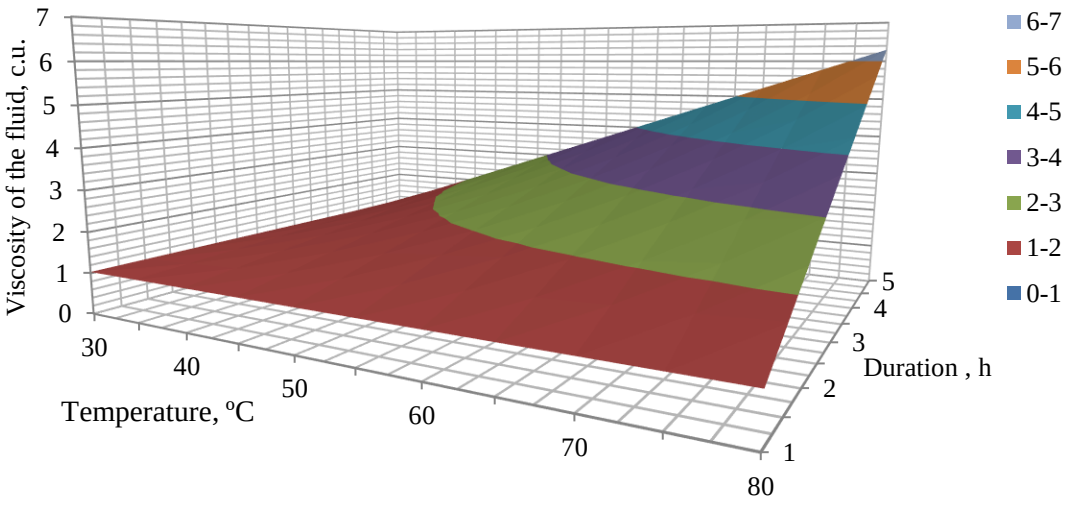

Figure 5 – Influence of processing temperature and duration of the process on viscosity of the protein hydrolysate solution

As one can see in Figure 5, peak values of solution viscosity fall into the marginal area of the response surface. It means that high viscosity can only be achieved under the maximum temperature values. The following distribution over layers is marked by lower viscosity, yet wider range of possible parameters.

Therefore, possible implementation of the developed regression equations will allow one to control the processes of the alkaline treatment, basing on the required characteristics of the product. Translational model also provides clear mathematical description of changes in viscosity of the solution, influenced by three major parameters of the alkaline treatment.

CONCLUSION

Our research has revealed an optimal technique of untanning of chromium-containing waste, particularly low-grade semi-products of woolskin. The studied methods – salt and magnesia treatment – are highly efficient in the context of residual content of chromium compounds in leather.

The analysis of amino acid compositions has confirmed the hypothesis of correlation between the amino acid concentrations and the impact rate of the untanning agent on the proteins.

We developed the mathematical model of producing the protein hydrolysate that gives one opportunity to choose the parameters of processing the collagen-containing materials basing on the desired viscosity of the protein hydrolysate. This model is particularly suitable for the protein hydrolysate, obtained from secondary materials – low-grade wool skin.

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