



STUDY OF THE EFFECT OF ONION HUSK ETHANOL EXTRACT ON THE CHEMICAL COMPOSITION AND MICROSTRUCTURE OF MEAT PATES

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Abstract

The wide use of antioxidants is due to their involvement in free radical processes in foods and human body. Interest in the use of low-value raw materials providing products with functional properties and increasing their shelf life is rapidly increasing. However, any changes in the formulation and technology may affect the properties and composition of the finished product. During the work, the effect of replacing 34% (sample 1) or 17% (sample 2) beef broth with 70% water-ethanol extract of yellow onion peels in the formulation of the experimental meat pates was investigated. The control product contained only beef broth as liquid. The total antioxidant capacity by the DPPH radical method (TAC_{DPPH}), fatty acid composition and amino acid composition were determined; microelement content analysis, proteomic and microstructural studies of meat pate samples with and without the addition of extract were also carried out. For 14 days, TAC_{DPPH} values of experimental pates were higher than in control by at least 2.32 times ($P < 0.10$). Samples 1 and 2 were characterized by a decrease in the concentrations of zinc, manganese and magnesium by no more than 14% ($P < 0.10$), with a simultaneous increase in selenium, copper, potassium and calcium of 8% to 17.35% ($P < 0.10$) depending on the microelement. The mass fraction of protein in experimental pates 1 and 2 was higher by 6.76% and 2.73% ($P < 0.10$), respectively, which was due to a decrease in moisture because of ethanol evaporation. Replacing the broth in the formulation affected the decrease in the protein biological value, as evidenced by a decrease in amino acid scores (AASs). However, a decrease in the AAS difference coefficient in experimental pates 1 and 2 by 7.71% and 3.07%, respectively, led to an increase in the biological value of the pates by 7.7% and 3.06%, respectively. Based on the results of proteomic and histological analysis, it was revealed that the addition of ethanol extract did not lead to significant changes in the protein composition and microstructural characteristics of the test samples.

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Introduction

Meat and meat products play an important role in the diet, as they are a good source of energy and nutrients such as essential amino acids, proteins of high biological value, minerals (iron, zinc, selenium, manganese) and B vitamins, especially vitamin B12 [1]. On the other hand, some nutritionists associate increased consumption of meat products with an increased risk of developing a number of diseases, for example, coronary heart disease and certain types of cancer [2]. In addition, lipids and proteins contained in meat products are prone to oxidation with the formation of toxic substances, which leads to a deterioration in organoleptic characteristics and a decrease in nutritional properties [3]. To reduce these processes, synthetic antioxidants are widely used, which may negatively affect the human body [4,5].

Consumer awareness of the value of healthy foods [6] has contributed to changes in dietary habits and traditions, which has increased interest and demand for fortified foods and the replacement of synthetic preservatives with natural ingredients [7,8]. This trend stimulates the search for and study of new sources of plant-based antioxidants to develop food products with improved nutritional value and beneficial properties. Numerous studies show the effectiveness of using plant-based antioxidants instead of artificial antioxidants in extending the shelf life of meat products [9,10,11,12,13]. Thus, Kim et al. [9] demonstrated that the addition of 70% ethanol extracts of *Pimpinella brachycarpa* and *Aralia elata* to raw beef burgers helped to reduce the concentration of lipid peroxidation products and microorganisms, as well as to improve the color stability of meat when stored at 4°C for 12 days. In another

work, Das et al. [11] showed that the use of mature *Moringa oleifera* leaf extract in cooked goat meat burgers at the level of 100 mg/100 g reduced oxidative spoilage over a longer period of time compared to the common synthetic antioxidant, butylated hydroxytoluene. In addition, the positive physiological effects of compounds with antioxidant properties have been proven [14,15]. The widely known properties of plant-based antioxidants are due to their ability to neutralize free radicals (FR), as well as interrupt oxidative chain reactions and stimulate the activity of the body's antioxidant system.

At the moment, there are numerous technologies for creating products of animal origin in combination with plant raw materials. The authors of the publication [16] characterize such products as foods with an increased biological value and a balanced amino acid composition. In articles [17,18], the authors consider methods for producing meat products with a high content of polyunsaturated fatty acids, probiotics, biologically active plant compounds (antioxidants, dietary fiber) while maintaining their sensory characteristics.

In the meat products market, pates are in great demand, which is due to the unique original taste because of the plant components they contain (pistachios, prunes, etc.) [19].

Furthermore, the technology for the production of meat pates is one of the most convenient for adding additional components, such as plant extracts, emulsions, powders, dietary fiber, etc., since a high degree of homogenization contributes to their uniform distribution in the product. However, due to high homogenization, humidity, the use of by-products, and the presence of a significant amount of fatty raw materials, pates in coating are perishable food products, so the use of antioxidants in their production is an important component. Scientific research has also demonstrated the effectiveness of using extracts of spices, medicinal and food plants as natural antioxidants, and some scientific works have shown their antimicrobial potential [20,21]. A particularly interesting area is the use of low-value plant raw materials to provide products with functional properties and increase their shelf life, which makes it possible to convert by-products into goods with high commercial value. There is an increase in research of antioxidants obtained from processing wastes from the fruit and vegetable industry, which reduces the cost of functional food products and also complies with the principles of a circular economy [22,23]. Previously, we demonstrated that the addition of yellow onion peel ethanol extract to the formulation of meat pate contributed to an increase in the total antioxidant capacity in relation to transition metal ions and the content of phenolic compounds simultaneously with a decrease in oxidative spoilage [24]. However, any changes in the formulation and technology of meat products may affect the properties and composition of the finished product. Therefore, the purpose of this work was to study the effect of 70% ethanol extract of yellow onion

peels, rich in plant-based antioxidants, namely quercetin and its glycosides, on chemical composition and microstructure of meat pates.

Objects and methods

The objects of the study were yellow onion peels (*Allium cepa*) purchased at the VkusVill store, manufactured by Agroleto LLC, Krasnodar, Russia, and developed meat pates with and without the addition of 70% water-ethanol extract of yellow onion peels.

To prepare onion husk extract (OHE), crushed onion husk (particle size 5 mm or less) were mixed with 70% ethanol in a ratio of 1:15 (g: ml), infused for 24 hours at $22 \pm 2^\circ\text{C}$, and then filtered through a folded paper filter. The extract was stored in a sealed dark vial at 4°C . The total antioxidant capacity according to the DPPH method (TAC_{DPPH}) was 2.919 ± 0.058 mmol equiv. quercetin / l.

Three samples of the experimental meat pates were produced in the Department of Applied Scientific and Technological Development of the V. M. Gorbатов Federal Research Center for Food Systems in accordance with the formulation presented in [24] and in Table 1.

Table 1. Formulations of the experimental meat pates

Ingredients	Pate samples		
	Control	Sample 1 (max)	Sample 2 (1/2 max)
Main raw materials, kg per 100 kg of blanched products			
Beef flank, blanched	35	35	35
Beef liver, trimmed, blanched	23	23	23
Lean pork, blanched	20	20	20
Pork heart, blanched	10	10	10
Wheat flour	5	5	5
Whole cow's milk powder	2	2	2
Fried onions	5	5	5
Spices and other materials, g per 100 kg of unsalted raw materials			
Table salt	1400	1400	1400
Granulated sugar	300	300	300
Ground black pepper	100	100	100
Ground allspice	50	50	50
Ground mustard	500	500	500
Ground nutmeg	50	50	50
Liquid, liters per 100 kg of main raw materials			
Beef broth	20	13.2	16.6
OPE	0	6.8	3.4

Control product and two experimental products with the addition of the maximum tolerable concentration (100 mg rutin/day) of antioxidants (Sample 1) and 50% of the maximum concentration (Sample 2) in equivalent quercetin based on the adequate intake (AI) of flavanols and their glycosides in accordance with the Appendix 5 "Values of daily consumption of food and biologically active substances for adults as part of specialized food products and dietary supplements" of the Uniform sanitary-epidemiological and hygienic requirements for products (goods) subject to sanitary-epidemiological supervision

(control) (as amended as of November 14, 2023)¹. OHE was added instead of broth at the levels of 6.8 l/100 kg of the main raw materials for test sample 1 and 3.4 l/100 kg of the main raw materials for test sample 2, which corresponded to TAC_{DPPH} values of 19.85 and 9.94 mmol equiv. quercetin/100 kg of the main raw materials, respectively (or 6.0 and 3.0 g of quercetin/100 kg of the main raw materials).

The technology for preparing experimental meat pates corresponded to the technology presented in [24]. Briefly, meat raw materials were prepared, cut into pieces of 200 to 300 g and blanched separately in water at 95 ± 5 °C for 15 to 20 minutes (liver), 120 minutes (pork heart) or 40 minutes (beef flank). At the same time, the onions were peeled, chopped in Bosch MCM3501M food processor (Bosch, Škofja Loka, Slovenia) and blanched with sunflower oil in a frying pan until golden brown. Heat-treated meat raw materials and onions were separately ground in Hurakan HKN-12SC meat grinder (Hurakan, Guangzhou, China) to a particle size of 2 to 3 mm. The pate was mince using a cutter blender (Robot-Coupe, Montceau-Les Mines, France). Fried onions, dry ingredients, broth and OHE were added to the finished emulsion and processed until a homogeneous minced mass was obtained. Chopping was carried out at 3000 rpm for 5 minutes. This product was used as a control sample. The technological difference that changed the composition of the final product was the replacement of 34% (Sample 1) and 17% (Sample 2) of beef broth with OHE. The final product temperature at the end of homogenization was higher than 40 °C. The product was packaged in 100 ± 1 g vacuum packaging (VakumPak-M, Webomatic, Bochum, Germany), PA/PE, 150 × 200 mm, 70 μm thickness; cooked in a water bath (EKROS4310, St. Petersburg, Russia) for 20 to 30 minutes at a temperature of 72 °C in the geometric center of the bar; then cooled to 4 °C and stored at 4 °C. Temperature was monitored using WT-1 digital thermometer (Xuzhou Sanhe Automatic Control Equipment Co., Ltd., Xuzhou, China).

To study the effect of OHE on the chemical composition and microstructure of meat pate, water-soluble and fat-soluble vitamins, minerals, microelements, amino acids, fatty acids (FA) were determined in freshly prepared samples; atherogenic index was calculated; proteomic analysis was carried out using electrophoresis; and histological analysis was carried out. TAC was determined by the DPPH method on the day of preparation (day 0) and on days 3, 5, 7 and 14 of storage at 4 °C.

To determine the TAC of pates, ethanol extracts were prepared, for which an average sample of the experimental product was mixed with 96% ethanol in a ratio of 1:5 (g: ml), homogenized using hand-held S10 homogenizer (Stegler, China) for 2 minutes at 9000 rpm. Ethanol extracts were infused for 60 minutes at 22 ± 2 °C, followed

by filtering through a folded paper filter, and then stored at minus 40 °C. TAC values of onion peel and meat pate extracts were determined by the DPPH radical method on SF-2000 spectrophotometer (OKB Spektr, Russia) according to the published method [25]. A stock 1 mM ethanol solution of the DPPH radical (Santa Cruz Biotechnology, USA) was prepared in a dark glass container, which was infused in the dark at a temperature of 22 ± 2 °C for 12 hours. Before measurements, DPPH working solution with a concentration of 100 μM was prepared, the absorbance of which was at least 1.00 ± 0.05 optical units. To determine TAC values of extracts, 1.52 ml of DPPH working solution and 80 μl of a sample or 96% ethyl alcohol as a control sample or quercetin at concentrations of 100 to 275 μM were added to glass tubes to plot a calibration curve. The reaction mixture was shaken vigorously and incubated in the dark at 22 ± 2 °C for 30 min. Measurement of the absorbance of solutions was carried out in cuvettes with a distance between the working faces of 1 cm at a wavelength of 517 nm. Measurements for each sample were carried out in quadruplicate. TAC values were calculated from the calibration plot (R² > 0.99) using equation (1) and expressed in mmol equiv. quercetin/l of OHE or in μmol equiv. quercetin/100 g of product.

$$TAC = \frac{\left(\frac{D_k - D_o}{D_k}\right) \times 100\% - 4.6904}{0.3081} \quad (1)$$

where D_k is the absorbance of the control sample; D_o is the absorbance of the sample.

To study the physicochemical composition of meat pates, the mass fractions of protein were determined according to GOST 25011–2017², the mass fractions of carbohydrates according to MU 1–40–3805³ and the mass fractions of fat according to GOST 23042–2015⁴.

Concentrations of water-soluble and fat-soluble vitamins in experimental meat pates were measured according to GOST R 55482⁵ and GOST 32307⁶, respectively.

To study the variability of the microelement composition of the control and experimental meat pates, the content of iron, zinc, copper, lead, cadmium was determined according to GOST 30178–96⁷, the content of

² GOST 25011–2017. “Meat and meat products. Protein determination methods: Retrieved from <https://docs.cntd.ru/document/1200146783> Accessed February 7, 2024 (In Russian)

³ MU1–40–3805. “Methodological instructions on laboratory quality control of public catering products” Retrieved from <https://docs.cntd.ru/document/1200049293> Accessed February 7, 2024 (In Russian)

⁴ GOST 23042–2015. “Meat and meat products. Fat determination methods” Retrieved from <https://docs.cntd.ru/document/1200133107> Accessed February 7, 2024 (In Russian)

⁵ GOST R 55482. “Meat and meat products. Method for determination of water-soluble vitamins” Retrieved from <https://docs.cntd.ru/document/1200104685> Accessed February 7, 2024 (In Russian)

⁶ GOST 32307. “Meat and meat products. Determination of fat-soluble vitamins by high performance liquid chromatography” Retrieved from <https://docs.cntd.ru/document/1200107182> Accessed February 7, 2024 (In Russian)

⁷ GOST 30178–96. “Raw material and food-stuffs. Atomic absorption method for determination of toxic elements” Retrieved from <https://docs.cntd.ru/document/1200021152> Accessed February 7, 2024 (In Russian)

¹ “Uniform sanitary-epidemiological and hygienic requirements for products (goods) subject to sanitary-epidemiological supervision (control) (as amended as of February 22, 2022)” Retrieved from <https://docs.cntd.ru/document/902249109> Accessed January 20, 2024

magnesium, sodium, potassium, manganese according to GOST R 55484–13⁸, the content of calcium according to GOST R 55573–13⁹, the content of selenium according to GOST 31707–12¹⁰, the content of arsenic according to GOST R 51766–01¹¹, the content of mercury according to GOST R 53183–08¹².

The amino acid composition of pates was determined according to GOST 34132–2017¹³, the content of oxyproline according to GOST 23041–2015¹⁴, the content of tryptophan according to GOST R 70149–2022¹⁵.

The biological value of pates was assessed by calculating amino acid scores (AASs), AAS difference coefficient (AASDC, %), rationality coefficient of amino acid composition (R_c , fractions of units), coefficient of utility and indicator of comparable excess content of essential amino acids (EAAs) (σ , mg/g of reference protein) according to [26].

The biological value of pate protein was determined by calculating amino acid scores for essential amino acids according to the formula (2).

$$C_i = \frac{A_i}{A_{i, \text{ref}}} \cdot 100 \quad (2)$$

where C_i is the amino acid score of the i -th EAA, %; A_i is the content of i -th EAA in 1 g of the protein under study, mg/g; $A_{i, \text{ref}}$ is the content of the i -th EAA in 1 g of “reference” protein according to FAO/WHO, mg/g.

The excess amount of EAAs not used for plastic needs was determined by calculating the AAS difference coefficient (AASDC, %) according to the formula 3.

$$\text{AASDC} = \frac{\sum_{i=1}^n |(C_i - 100)|}{n} \quad (3)$$

where C_i is the AAS of the i -th EAA, %; n is the number of EAAs.

⁸ GOST R55484–13. “Meat and meat products. Determination of sodium, potassium, magnesium and manganese by flame atomic absorption” Retrieved from <https://docs.cntd.ru/document/1200103312> Accessed February 7, 2024 (In Russian)

⁹ GOST R55573–13. “Meat and meat products. Determination of calcium by atomic absorption and titrimetric methods” Retrieved from <https://docs.cntd.ru/document/1200105941> Accessed February 7, 2024 (In Russian)

¹⁰ GOST 31707–12. “Foodstuffs. Determination of trace elements. Determination of total arsenic and selenium by hydride generation atomic absorption spectrometry (HGAAS) after pressure digestion” Retrieved from <https://docs.cntd.ru/document/1200098581> Accessed February 7, 2024 (In Russian)

¹¹ GOST R51766–01. “Raw material and food-stuffs. Atomic absorption method for determination of arsenic” Retrieved from <https://docs.cntd.ru/document/1200025461> Accessed February 7, 2024 (In Russian)

¹² GOST R53183–08. “Foodstuffs. Determination of trace elements. Determination of mercury by cold-vapour atomic absorption spectrometry (CVAAS) method after pressure digestion” Retrieved from <https://docs.cntd.ru/document/1200076584> Accessed February 7, 2024 (In Russian)

¹³ GOST 34132–2017. “Meat and meat products. Determination of amino acids composition of animal protein” Retrieved from <https://docs.cntd.ru/document/1200146930> Accessed February 7, 2024 (In Russian)

¹⁴ GOST 23041–2015. “Meat and meat products. Method for determination of oxyproline” Retrieved from <https://docs.cntd.ru/document/1200123926> Accessed February 7, 2024 (In Russian)

¹⁵ GOST R70149–2022. “Meat and meat products. Determination of the mass fraction of tryptophan by spectrophotometric method” Retrieved from <https://docs.cntd.ru/document/1200184801> Accessed February 7, 2024 (In Russian)

The balance of EAAs in relation to the physiologically necessary norm (reference) was characterized by the rationality coefficient of amino acid composition (R_c , fractions of units), which was calculated according to the formula 4.

$$R_c = \frac{\sum_{i=1}^k (A_i \cdot k_i)}{\sum_{i=1}^n A_i} \quad (4)$$

where A_i is the content of the i -th EAA in 1 g of the protein under study, mg/g; k_i is the coefficient of utility of the i -th EAA to the limiting amino acid, fractions of units.

The coefficient of utility (utilization) reflects the balance of EAAs in relation to the reference and was calculated according to the formula 5.

$$k_i = \frac{C_{\text{min}}}{C_i} \quad (5)$$

where C_{min} is the minimum EAA score of the protein under study in relation to the reference protein, fractions of units; C_i is the score for the i -th EAA in relation to the reference protein, fractions of units.

Calculation of the indicator for comparable excess content of EAAs (σ , mg/g of reference protein) showing the total mass of EAAs not used for anabolic needs, was calculated according to the formula 6.

$$\sigma = \frac{\sum_{i=1}^n (A_i - C_{\text{min}} \cdot A_{i,3})}{C_{\text{min}}} \quad (6)$$

Fatty acid composition (36 acids) of the test samples was determined according to GOST R 55483–2013¹⁶. The calculation of the atherogenic index (AIP) of pates was carried out according to the formula (7) given by Ulbricht T. L. V. and Southgate D. A. T., 1991 [27].

$$\text{AIP} = \frac{(C_{12} + C_{14} + C_{16} + \text{Trans FA})}{(\text{PUFA} + C_{18:1} + \text{other MUFA})} \quad (7)$$

The determination of the tolerable level of toxic elements in meat pates was carried out according to TR TS 021/2011 “On food safety”¹⁷, SanPiN2.3.2.560–96 “Hygienic requirements for the quality and safety of food raw materials and food products”¹⁸.

One-dimensional electrophoresis was carried out in a 10% polyacrylamide gel in the presence of sodium dodecyl sulfate (SDS-PAGE) in VE-10 chamber (Helicon, USA) at a constant current and voltage of 60 V and 120 V, for 2 hours, until the front stain (bromophenol blue) reached the bottom edge of the gel sheets. A marker consisting of molecular weight standards (Thermo, Latvia) was used as a standard solution. Staining was carried out with Coomassie brilliant blue G-250 (PanReac, Spain). To remove the

¹⁶ GOST R55483–2013. “Meat and meat products. Determination of fatty acids composition by gas chromatography” Retrieved from <https://docs.cntd.ru/document/1200103852> Accessed February 7, 2024 (In Russian)

¹⁷ TR TS 021/2011. “Technical Regulations of the Customs Union On food safety (as amended as of July 14, 2021)” Retrieved from <https://docs.cntd.ru/document/902320560#8Q20M0>. Accessed April 4, 2022

¹⁸ SanPiN2.3.2.560–96. “Hygienic requirements for the quality and safety of food raw materials and food products”. Retrieved from <https://docs.cntd.ru/document/9052436>. Accessed April 03, 2022

unbound stain, 10% acetic acid (Reagent Component, Russia) was used. To increase resolution, staining with silver nitrate (PanReac, Germany) was additionally performed according to Blum's method [28]. Computer densitometry of one-dimensional electropherograms in a wet state was carried out using Bio-5000 Plus scanner (Serva, Germany) in 600 ppi 2D-RGB mode in triplicate. Visualized protein fractions were interpreted using UniProt database [29].

To study the microstructure, three pieces of $1.5 \times 1.5 \times 0.5$ cm were selected from each sample, which were placed in the chamber of MIKROM-HM525 cryostat (Thermo Scientific, USA). Slices 14 μm thick were made (three sections from each piece), mounted on Menzel-Glaser glasses (Thermo Scientific, USA) and stained with Ehrlich hematoxylin and 1% aqueous-alcoholic eosin solution (BioVitrum, Russia) according to the generally accepted method [30]. The study of histological preparations and their photography were carried out on AxioImaiger A1 light microscope (Carl Zeiss, Germany) using AxioVision 4.7.1.0 image analysis software (Carl Zeiss, Germany).

Statistical analysis was carried out using Microsoft Excel and Statistica 10.0 software packages. The results were presented as means and standard deviations ($M \pm SD$). Statistical significance was calculated using nonparametric Mann-Whitney U-tests (for two independent groups). A probability of $P < 0.1$ was selected as the level of significance.

Results and discussion

The results of determining TAC_{DPPH} in ethanol extracts of meat pates stored at 4°C and the difference between the control and test samples are presented in Table 2.

Table 2. TAC_{DPPH} values of meat pates

Days	TAC_{DPPH} , $\mu\text{mol equiv. quercetin}/100 \text{ g of product}$				
	Control	Sample 1 (max)	Sample 2 (1/2 max)	$\Delta(S1-C)$	$\Delta(S2-C)$
0	15.45 ± 1.05	$49.59 \pm 2.66^*$	$35.84 \pm 1.91^{*,\#}$	34.14	20.39
3	4.36 ± 0.63	$44.33 \pm 3.20^*$	$25.45 \pm 5.75^{*,\#}$	39.96	21.08
5	7.22 ± 0.67	$41.18 \pm 3.22^*$	$21.13 \pm 1.89^{*,\#}$	33.97	13.92
7	6.51 ± 0.59	$38.09 \pm 2.09^*$	$25.54 \pm 0.84^{*,\#}$	31.58	19.03
14	8.24 ± 0.51	$41.79 \pm 1.31^*$	$24.34 \pm 0.25^{*,\#}$	33.55	16.10

^{*}, significant difference between the control and test samples, Mann-Whitney U-test, P -value < 0.10 ; [#], significant difference between test samples, Mann-Whitney U-test, P -value < 0.10

During 14 days of storage, a decrease in total antioxidant capacity was observed in all samples. TAC values in the test samples during all days of storage were significantly higher than that in the control pate. Thus, TAC_{DPPH} values of the experimental product with the maximum concentration of OHE (Sample 1) exceeded those of the control sample by $34.64 \pm 3.15 \mu\text{mol equiv. quercetin}/100 \text{ g of product}$, and the corresponding values of the experimental pate with 50% OHE concentration (Sample 2) exceeded those of the control sample by $18.10 \pm 3.02 \mu\text{mol equiv. quercetin}/100 \text{ g of product}$. It was determined that reducing the

concentration of OHE introduced into the meat product by 50% significantly reduced the contribution of the plant extract to TAC_{DPPH} , as evidenced by the excess of the difference between Sample 1 and the control pate ($\Delta(S1-C)$) and the differences between Sample 2 and the control at day 0 of approximately 1.7 times. Based on the results of the experiment, it was determined that within 14 days of storage, the addition of OHE to the formulation of meat pate provides a significantly increased TAC in relation to nitrogen-containing free radicals in comparison with the control sample, with previously proven reduction in oxidative spoilage presented in [24]. Other researchers have shown the effectiveness of using peanut skin extract to inhibit oxidative spoilage in meat burgers, i. e. the reduction of lipid and protein oxidation products [31]. Based on the results of work [32], it was determined that the introduction of an antioxidant complex into the formulation of meat burgers contributed to an increase in the antioxidant capacity of the test sample by 9 times in comparison with the control, while the increased antioxidant capacity values were maintained when the products were stored for 28 days.

The results of determining the concentrations of water-soluble and fat-soluble vitamins in meat pates and the calculated percentages of AI of these vitamins according to the values of daily consumption of nutrients and biologically active substances for adults in the composition of specialized food products (SFP) and biologically active supplements¹⁹ are presented in Table 3.

In all pate samples, the content of vitamins D3, B7 and C was below the detection limit. The concentrations of vitamins A, E, B1 and B12 significantly decreased in the order of Sample 2 > Control > Sample 1, and vitamins B2, B3, B5, and B9 significantly decreased in the order of Sample 1 > Sample 2 > Control. According to the results of determining vitamins in all experimental meat pates, a high content of B vitamins was noted, while not exceeding the tolerable upper intake level of all vitamins except B12. It is known that products of animal origin, in particular liver, which was the main component of the experimental meat pate, are characterized by a high content of B vitamins, especially B12, which is an important nutrient for the cognitive and neurological functions of the body [33]. In addition, beef liver is rich in components such as protein, heme iron, and zinc [33]. Liver contains more than $10 \mu\text{g}$ of vitamin B12 per 100 g, but its absorption is markedly reduced when its concentration in the product exceeds $2 \mu\text{g}$, with only about 20% of the vitamin being effectively absorbed [34]. Thus, a significant excess of vitamin B12 over AI in all the resulting pates will contribute to the absorption of about $2.6 \mu\text{g}/100$ of product, which is about 85% of the daily requirement. During the work, it was shown that the addition of an ethanol extract of plant-based antioxidants

¹⁹ "Uniform sanitary-epidemiological and hygienic requirements for products (goods) subject to sanitary-epidemiological supervision (control) (as amended as of February 22, 2022)" Retrieved from <https://docs.cntd.ru/document/902249109> Accessed January 20, 2024

Table 3. Vitamin content and their% of AI in meat pates

Vitamin	Intake, per day		Control		Sample 1 (max)		Sample 2 (1/2 max)	
	AI	UL	C, in 100 g	% of AI	C, in 100 g	% of AI	C, in 100 g	% of AI
A, µg	900	3000	156.90 ± 2.23	17.43	147.95 ± 2.79*	16.44	188.35 ± 2.97* [#]	20.93
D3, µg	10	15	< 1.0	0	< 1.0	0	< 1.0	0
E, mg	15	150	1.34 ± 0.06	8.93	1.21 ± 0.03*	8.07	1.67 ± 0.03* [#]	11.13
B1, mg	1.5	5.0	1.59 ± 0.08	106.0	2.25 ± 0.06*	150.0	2.52 ± 0.05* [#]	168.0
B2, mg	1.8	6.0	2.10 ± 0.05	116.67	2.54 ± 0.06*	141.11	2.37 ± 0.06* [#]	131.67
B3, mg	20	60	17.25 ± 0.39	86.25	19.36 ± 0.19*	96.80	18.27 ± 0.24* [#]	91.35
B5, mg	5	15	5.46 ± 0.07	109.20	6.27 ± 0.11*	125.40	5.64 ± 0.46 [#]	112.80
B6, mg	2.0	6.0	0.36 ± 0.04	18.0	0.41 ± 0.01	20.50	0.48 ± 0.04* [#]	24.0
B7, µg	50	150	< 1.0	0	< 1.0	0	< 1.0	0
B9, µg	400	600	77.08 ± 0.21	19.27	79.09 ± 2.27*	19.77	78.71 ± 0.39*	19.68
B12, µg	3.0	9.0	13.23 ± 0.99	441.0	10.85 ± 0.25*	361.67	14.86 ± 0.19* [#]	495.33
C, mg	90	900	< 1.0	0	< 1.0	0	< 1.0	0

*, significant difference between the control and test samples, Mann-Whitney U-test, P-value < 0.10; #, significant difference between test samples, Mann-Whitney U-test, P-value < 0.10; C, concentration of vitamins, mg or µg depending on the vitamin; UL, tolerable upper intake level.

Table 4. Microelement composition of meat pates

Microelement	Intake, per day		Control		Sample 1 (max)		Sample 2 (1/2 max)	
	AI	UL	C, in 100 g	% of AI	C, in 100 g	% of AI	C, in 100 g	% of AI
Iron, mg	18 / 10 [^]	40 / 20 [^]	3.50 ± 0.01	19.44 / 35.0	3.33 ± 0.03 *	18.5 / 33.3	3.56 ± 0.04 * [#]	19.78 / 35.6
Magnesium, mg	400	800	23.58 ± 0.20	5.90	23.37 ± 0.25	5.84	20.49 ± 0.11 * [#]	5.12
Sodium, mg	1300 ^{**}	—	598.96 ± 12.05	—	661.45 ± 6.11 *	—	687.27 ± 6.49 * [#]	—
Potassium, mg	2500	3500	419.54 ± 2.57	16.78	462.60 ± 2.74 *	18.50	479.18 ± 5.47 * [#]	19.17
Zinc, mg	12	25	3.73 ± 0.03	31.08	3.45 ± 0.04 *	28.75	3.07 ± 0.04 * [#]	25.58
Copper, mg	1	3	3.44 ± 0.02	344	3.91 ± 0.03 *	391	3.76 ± 0.01 * [#]	376
Calcium, mg	1000	2500	15.61 ± 0.02	1.56	17.19 ± 0.07 *	1.72	18.31 ± 0.01 * [#]	1.83
Selenium, µg	55 / 75 [^]	150	35.0 ± 1.0	50.0	38.0 ± 1.0 *	54.29	38.0 ± 1.0 *	54.29
Manganese, mg	2	5	0.08 ± 0.0	4.0	0.08 ± 0.0	4.0	0.09 ± 0.01 * [#]	4.0
Lead, mg	—	—	< 0.01	—	< 0.01	—	< 0.01	—
Cadmium, mg	—	—	< 0.01	—	< 0.01	—	< 0.01	—
Arsenic, mg	—	—	< 0.01	—	< 0.01	—	< 0.01	—
Mercury, mg	—	—	< 0.002	—	< 0.002	—	< 0.002	—

*, significant difference between the control and test samples, Mann-Whitney U-test, P-value < 0.10; #, significant difference between test samples, Mann-Whitney U-test, P-value < 0.10; ^, norm for women/men; C, concentration of microelements, mg or µg; UL, tolerable upper intake level; **, only in SFP for athletes' nutrition.

obtained from onion peels to the formulation of meat pate did not have a negative effect on the composition of vitamins, and in the case of such compounds as B1, B2, B3, B5, B6, B9, the increase in concentrations was proportional to the addition of OHE.

The results of determining the concentrations of microelements and toxic elements in pates, as well as the calculated percentage of AI according to the values of daily intake of nutrients and biologically active substances for adults in the composition of specialized food products (SFP) and biologically active supplements²⁰ are presented in Table 4.

According to TR TS 021/2011 “On food safety”²¹, the concentrations of toxic elements such as lead, cadmium, arsenic and mercury in all samples were significantly be-

²⁰ “Uniform sanitary-epidemiological and hygienic requirements for products (goods) subject to sanitary-epidemiological supervision (control) (as amended as of February 22, 2022)” Retrieved from <https://docs.cntd.ru/document/902249109> Accessed January 20, 2024

²¹ TR TS021/2011. “Technical Regulations of the Customs Union On food safety (as amended as of July 14, 2021)” Retrieved from <https://docs.cntd.ru/document/902320560#8Q20M0>. Accessed April 4, 2022

low the tolerable level, which indicates the safety of meat pates. It was determined that the concentrations of microelements were significantly different between samples. The concentrations of iron and manganese decreased in the order of Sample 2 > Control > Sample 1, and the content of sodium, potassium and calcium decreased in the order of Sample 2 > Sample 1 > Control. It was noted that the highest amount of zinc and magnesium was contained in the control sample and exceeded the similar values of Sample 1 and Sample 2 by 0.28 and 0.66 mg of zinc/100 g of product (P < 0.10), respectively, and by 0.21 and 3.09 mg magnesium/100 g of product (P < 0.10), respectively. Copper concentration was highest in the pate with the maximum amount of OHE (Sample 1) and amounted to 3.91 ± 0.03 mg/100 g, which significantly exceeded the amount of the microelement in the Control and Sample 2 by 0.47 and 0.15 mg/100 g (P < 0.10), respectively. An increase in selenium content in the test samples was noted, which amounted to 38.0 ± 1.0 µg/100 g of product. Analysis of the microelement composition demonstrated that the addition of OHE to meat pate led to a decrease in the con-

centrations of zinc, manganese and magnesium by no more than 14% ($P < 0.10$), simultaneously with an increase in the amount of selenium, copper, potassium and calcium from 8% to 17.35% ($P < 0.10$) depending on the microelement. This corresponds to the data of [35], in which the authors demonstrated that onion peels are a rich source of calcium (1.8 to 16.5 mg/g), potassium (11.1 to 15.9 mg/g), selenium (0.00003 to 0.00093 $\mu\text{g/g}$) and other microelements. It is known that mineral substances perform a plastic function in human body participating in the metabolism of almost any human tissue. But often not only their content, but also their ratio is important. Thus, the optimal ratio of calcium and magnesium is 4:1, and an increase in the concentration of magnesium may impair the digestibility of calcium [36]. However, we found that meat pates are characterized by a high magnesium content. In this connection, a decrease in the concentration of magnesium and an increase in the amount of calcium in the experimental pates led to a more optimal ratio of these elements. Thus, Ca: Mg ratio in the control sample was 0.72:1, and for samples 1 and 2 it was 0.74:1 and 0.89:1, respectively. Selenium is an essential mineral for humans, as its deficiency is associated with an increased risk of mortality, poor immunity, and decreased cognitive function. At the same time, it is known that selenium has antiviral and antioxidant effects and is necessary for the normal functioning of the reproductive system of men and women, and also reduces the risk of developing autoimmune thyroid diseases [37,38]. At the moment, the problem of selenium deficiency is observed all over the world [39], and therefore, increasing the amount of this microelement by 8.6% ($P < 0.10$) in experimental pates due to the addition of onion peels extract is especially important. Copper is considered a redox metal and is an essential nutrient for all species studied to date. It is known that deficiency of this microelement may contribute to the development and progression of a number of diseases, including cardiovascular diseases and diabetes [40]. In addition, the biological role of copper is to cross-link collagen via Cu-

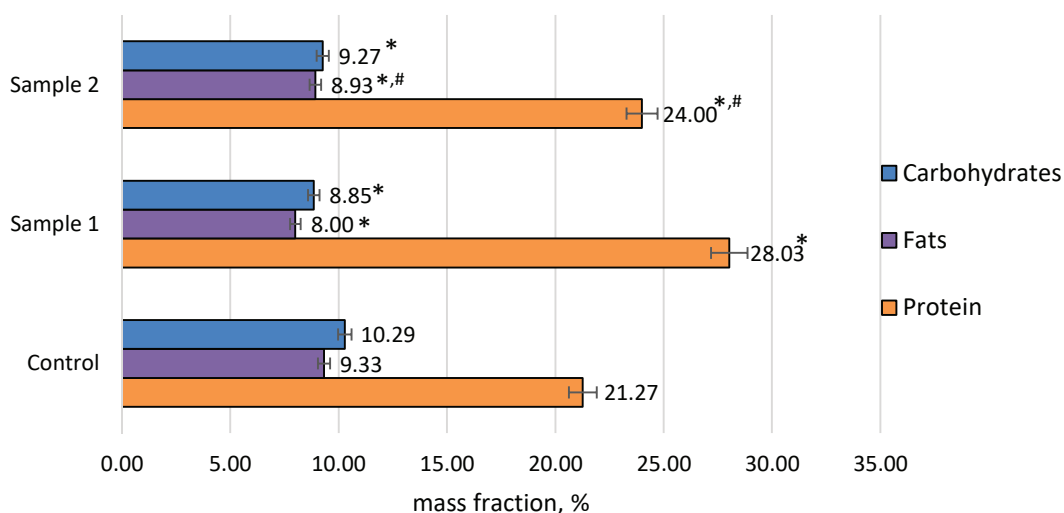
dependent lysyl oxidase and maintain the activity of superoxide dismutase (CuZn SOD) [41,42]. The high copper content in experimental pates is due to the use of beef liver in the formulation in the amount of 23% of all main raw materials, which is characterized by a high content of not only protein and iron, but also copper and vitamins [43].

Figure 1 shows the mass fractions of the main nutrients in the experimental meat pates with and without the addition of OHE to their formulations.

The diagram clearly shows that the addition of onion peel extract to meat pates contributed to a significant increase in the amount of protein and a decrease in fats and carbohydrates, with more addition of OHE leading to greater changes. Thus, the mass fraction of protein in the control product was inferior to samples 1 and 2 by 6.76% and 2.73% ($P < 0.10$), respectively. The concentration of carbohydrates in the experimental pates decreased by no less than 1.04% ($P < 0.10$), and concentration of fat decreased by no less than 0.4% ($P < 0.10$). The noted changes in the main indicators of the nutritional value of pates were associated with the replacement of beef broth with ethanol extract of onion peels by 34% in Sample 1 and 17% in Sample 2. It is known that ethyl alcohol is a volatile compound, the vaporization of which increases with increasing temperature. Since the pate preparation technology included a hot chopping step, during which the minced pate was homogenized for 5 minutes at a temperature above 40 °C, the ethanol contained in the extract evaporated. In addition, in [24], we demonstrated that the addition of OHE to the formulation of meat pates led to a decrease in the mass fraction of moisture by 4.17% (Sample 1) and by 1.47% (Sample 2).

Experimental data characterizing the fatty acid and amino acid compositions of the developed meat pates are presented in Table 5 and Table 6, respectively.

Analysis of the fatty acid composition of the experimental meat pates presented in Table 5 showed that the addition of OHE contributed to a slight change in the



*, significant difference between the control and test samples, Mann-Whitney U-test, P -value < 0.10 ; #, significant difference between test samples, Mann-Whitney U-test, P -value < 0.10

Figure 1. Main indicators of the nutritional value of pates

Table 5. Fatty acid composition of meat pates

No.	Fatty acid	Control	Sample 1	Sample 2
8	Myristic acid C14:0	2.67 ± 0.09	2.73 ± 0.04	2.70 ± 0.08
9	Myristoleic acid C14:1	0.37 ± 0.02	0.44 ± 0.02*	0.42 ± 0.04
10	Pentadecanoic acid C15:0	0.40 ± 0.01	0.38 ± 0.01	0.37 ± 0.01*
12	Palmitic acid C16:0	22.01 ± 0.21	22.56 ± 0.21*	22.12 ± 0.16 #
13	Palmitoleic acid C16:1	3.20 ± 0.15	3.50 ± 0.03*	3.24 ± 0.08 #
14	Margaric acid C17:0	2.52 ± 0.03	2.4 ± 0.03*	2.30 ± 0.01*,#
16	Stearic acid C18:0	20.84 ± 0.42	19.14 ± 0.42*	18.36 ± 0.61*
17	Oleic acid C18:1	33.37 ± 0.47	36.17 ± 0.70*	34.50 ± 0.85 #
18	Elaidic acid C18:1	2.33 ± 0.07	2.10 ± 0.17	1.98 ± 0.11*
19	Linoleic acid C18:2ω6	9.72 ± 0.14	8.54 ± 0.36*	11.69 ± 0.25*,#
20	Linolelaidic acid C18:2ω6	0	0	0.05 ± 0.09
21	Linolenic acid C18:3ω3	0.30 ± 0.04	0.27 ± 0.04	0.28 ± 0.06
22	Arachidic acid C20:0	0.07 ± 0.12	0	0.19 ± 0.02 #
23	Arachidonic acid C20:4ω6	1.28 ± 0.24	1.0 ± 0.01	0.96 ± 0.01*,#
25	Dihomo-γ-linolenic acid C20:3ω6	0.68 ± 0.14	0.59 ± 0.02	0.54 ± 0.03 #
28	Gondoic acid C20:1ω9	0.25 ± 0.01	0.18 ± 0.16	0.30 ± 0.01*,#

*, significant difference between the control and test samples, Mann-Whitney U-test, P-value < 0.10; #, significant difference between test samples, Mann-Whitney U-test, P-value < 0.10.

concentration of some fatty acids, which is also explained by the presence of myristic, palmitic, palmitoleic and other acids in onion waste [35]. Based on the results of the studies, atherogenic index (AIP) of all pates was calculated, which was 0.52 for the Control and Sample 1 and 0.50 for Sample 2. It was determined that the AIP of the experimental pates was 29.7% below the average AIP of the human diet for the Control and Sample 1 and 32.4% below the average AIP of the human diet for Sample 2, which is 0.74 according to A. B. Lisitsyn et al. [44].

Table 6. Amino acid composition of meat pates

No.	Amino acid	Content of bound amino acids (g/100 g of product)		
		Control	Sample 1	Sample 2
1	Aspartic acid	2.75 ± 0.28	2.86 ± 0.09	2.61 ± 0.32
2	Glutamic acid	4.75 ± 0.75	4.97 ± 0.40	4.47 ± 0.85
3	Serine	1.07 ± 0.23	1.14 ± 0.12	0.98 ± 0.26
4	Histidine	1.0 ± 0.07	0.96 ± 0.07	1.02 ± 0.02
5	Glycine	1.20 ± 0.15	1.24 ± 0.08	1.14 ± 0.17
6	Threonine	1.24 ± 0.17	1.3 ± 0.08	1.18 ± 0.18
7	Arginine	1.72 ± 0.49	1.77 ± 0.41	1.65 ± 0.53
8	Alanine	1.49 ± 0.25	1.55 ± 0.15	1.42 ± 0.28
9	Tyrosine	0.87 ± 0.15	0.92 ± 0.08	0.83 ± 0.16
10	Cystine	0.19 ± 0.04	0.20 ± 0.02	0.18 ± 0.04
11	Valine	1.1 ± 0.17	1.16 ± 0.07	1.02 ± 0.19
12	Methionine	0.79 ± 0.10	0.79 ± 0.08	0.78 ± 0.09
13	Phenylalanine	0.91 ± 0.14	0.94 ± 0.10	0.89 ± 0.15
14	Isoleucine	0.70 ± 0.09	0.71 ± 0.07	0.69 ± 0.10
15	Leucine	1.66 ± 0.22	1.71 ± 0.15	1.60 ± 0.23
16	Lysine	1.12 ± 0.21	1.13 ± 0.19	1.10 ± 0.22
17	Proline	2.45 ± 1.80	1.44 ± 0.06	1.46 ± 0.06
18	Oxyproline	0.14 ± 0.01	0.17 ± 0.01*	0.09 ± 0.01*,#
19	Tryptophane	0.32 ± 0.01	0.34 ± 0.01*	0.38 ± 0.01*,#
	Total	25.48 ± 5.08	25.33 ± 2.24	23.48 ± 3.85

*, significant difference between the control and test samples, Mann-Whitney U-test, P-value < 0.10; #, significant difference between test samples, Mann-Whitney U-test, P-value < 0.10

When studying the amino acid composition of experimental pates, a slight increase in tryptophane and a decrease in proline and hydroxyproline were noted. However, no significant differences were found between the control and test samples, as evidenced by the same amount of total amino acid content. The results of the calculated biological value of the protein in the experimental products relative to the standard FAO/WHO scale are presented in Table 7 and Table 8.

Table 7. Amino acid score for essential amino acids in pates

Amino acid	Amino acid concentration, g/100 g of protein			Amino acid score, %			
	Reference protein, FAO/WHO	Control	Sample 1	Sample 2	Control	Sample 1	Sample 2
Isoleucine	4	3.29	2.53	2.88	82.25	63.25	72
Leucine	7	7.8	6.1	6.67	111.43	87.14	95.29
Lysine	5.5	5.27	4.03	4.58	95.822	73.27	83.27
Methionine + cysteine	3.5	4.61	3.53	4	131.71	100.86	114.29
Phenylalanine + tyrosine	6	8.37	6.64	7.17	139.5	110.67	119.5
Threonine	4	5.83	4.64	4.92	145.75	116	123
Tryptophane	1	1.5	1.21	1.58	150	121	158
Valine	5	5.17	4.14	4.25	103.4	82.8	85

Table 8. Indicators of meat pate biological value

Indicators	Control	Sample 1	Sample 2
Minimum amino acid score, C _{min}	0.83	0.63	0.72
Amino acid score difference coefficient, AASDC, %	25.47	17.76	22.40
Biological value of pate, %	74.54	82.24	77.60
Rationality coefficient of amino acid composition, Rc, fractions of units	0.71	0.69	0.72
Indicator of comparable excess content of EAAs, σ, mg/g of reference protein	148.69	158.89	140.69

According to the results of the AAS calculation, it was revealed that the control product had 2 limiting amino acids, i. e. isoleucine and lysine, while the experimental products had 4 limiting amino acids, which also include leucine and valine. Moreover, for all samples, isoleucine was the first limiting amino acid. It was noted that the AASs of all essential amino acids in the experimental products decreased, which indicated a decrease in the biological value of the protein, presumably associated with replacing part of the beef broth with OHE. With the maximum addition of the extract, a greater decrease in AAS was observed. According to Uniprot database [29], beef flank contains water-soluble proteins such as myoglobin (17.0 kDa), myosin (16.9 kDa), triosephosphate isomerase 1 (23.0 kDa) and hemoglobin (15.4 kDa). These proteins are characterized by a high content of essential amino acids, in particular leucine, lysine, valine, threonine and isoleucine. Presumably, when blanching beef flank, these proteins pass into the broth, which, according to the formulation of the pates, was used as a liquid, and in the case of Samples 1 and 2 was replaced by OHE in the amount of 34% and 17%, respectively. This fact led to a decrease in AAS of EAAs in studied samples. Amino acid scores of essential amino acids were used to calculate indicators of the biological value of pates.

It was found that the addition of OHE to the meat pate reduced the minimum AAS along with an increase in the amount of limiting amino acids. Greater addition of the extract contributes to more significant changes. However, it was noted that the experimental pates were characterized by a smaller excess amount of essential amino acids not used for plastic needs, as evidenced by a decrease in the AAS difference coefficient by 7.71% in the case of Sample 1 and by 3.07% in the case of Sample 2. In addition, biological value of the experimental pates increased by 7.7% (Sample 1) and by 3.06% (Sample 2), which was also comparable to the change in the added volume of OHE. The slight variability in rationality coefficient of amino acid composition of all samples indicated an equal balance of essential amino acids in relation to the physiologically necessary norm (reference) in both the Control and Samples 1 and 2. Despite the fact that C_{\min} , biological value and AASDC of Sample 2 were better than that of Sample 1, the control product and pate with a lower content of OHE (Sample 2) were characterized by lower σ by 10.11% and 18.2%, respectively, which indicated a smaller mass of essential amino acids not used for anabolic purposes.

As a result of studying samples of meat pate using one-dimensional electrophoresis with equal application (all samples were added in volume of 15 μ l) and various staining options, electropherograms were obtained, presented in Figure 2.

The difference in protein visualization between the control and test samples was insignificant. There were no significant differences, such as the absence or, conversely, the appearance of *de novo* protein fractions between the samples. All the main muscle proteins were preserved. The

formulation of the pate used pork heart, lean pork, beef liver and beef flank. The highest intensity of proteins was detected in the control sample, less pronounced intensity was in Sample 2 and the lowest concentration of proteins was in Sample 1. Such a visualization of proteins in the electropherogram is due to the fact that Sample 1 has the highest content of alcohol extract (6.8 ml per 100 g of product) instead of beef broth (in the control sample), so the concentration of proteins present in the broth decreases accordingly.

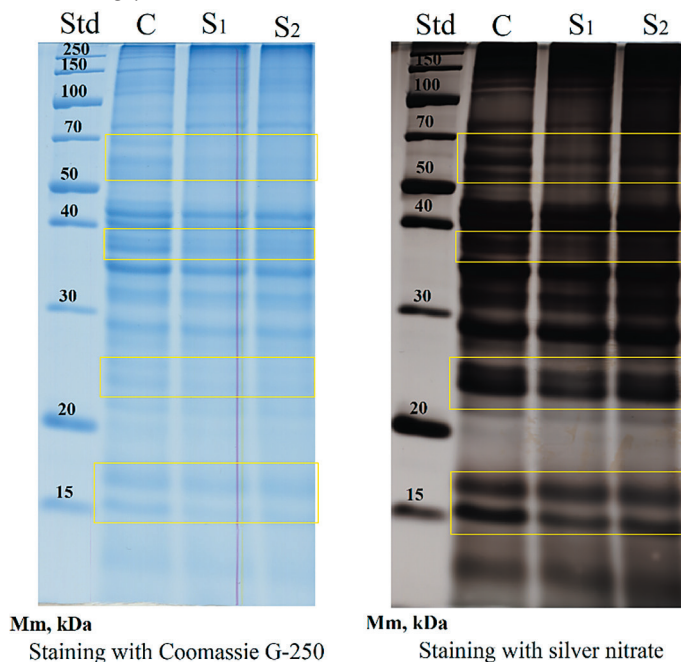


Figure 2. Electropherogram of 10% PAGE of meat pates on day 0: Std is molecular weight standard: 250, 150, 100, 70, 50, 40, 30, 20, 15 (from top to bottom); C is Control; S1 is Sample 1; S2 is Sample 2

When conducting a bioinformatic analysis of electropherograms, differences were identified in the intensity of staining of proteins present in skeletal muscles, with a molecular weight in the range of 15 to 17 kDa and 23 to 25 kDa, probably being fractions of hemoglobin (15.4 kDa) [45], myosin light chains (16.9 kDa) [46], myoglobin (17.0 kDa) [47], triosephosphate isomerase 1 (23.0 kDa) [48], troponin T (23.0 kDa) [49] and troponin I (25.0 kDa) [50]. Differences were also found in protein fractions in the range of 37 to 38 kDa, presumably being fractions of cathepsin B (36.9 kDa) [51] and beta-2-glycoprotein 1 (38.2 kDa) [52], which are present in pork heart and beef liver, respectively. The intensity of protein fractions with molecular weights of 55 kDa, 61 kDa, 68 kDa differed in staining intensity between the Control and Samples 1 and 2, and presumably may correspond to aldehyde dehydrogenase 1A1 (54.8 kDa) [53], leucine-rich protein 1 (61.1 kDa) [54] and 2-hydroxyacyl-CoA lyase 2 (68.1 kDa) [55], which are expressed in beef flank and beef liver.

Histological analysis revealed that all samples were characterized by the same structure and a high degree of constituent component grinding. The main part was represented by individual fragments of cross-striated muscle fibers and hepatocytes located singly or in small groups of

2 to 20 pieces, between which there was a small amount of fine-grained protein mass (Figure 3).

The samples also contained fragments of liver and loose fibrous connective tissue that retained their structural organization, individual fat drops evenly distributed throughout the minced meat, single fragments of heart muscle tissue and plant components related to wheat flour, natural spices (mustard, black pepper and allspice, nutmeg) and onions. The dispersion of the main part of the samples was 10 to 170 μm . All samples were characterized by a dense arrangement

of structural elements; no differences in microstructural organization were identified. Thus, the histological analysis of the test samples showed that the introduction of OHE into the pate did not lead to a change in its main structural characteristics and did not affect the microstructure of the components included in the product. Studying the effect of OHE on the microstructure of test samples was of practical interest, due to the lack of similar studies in the scientific literature, where the focus is on the antioxidant and antimicrobial aspects, as well as the sensory characteristics of the finished

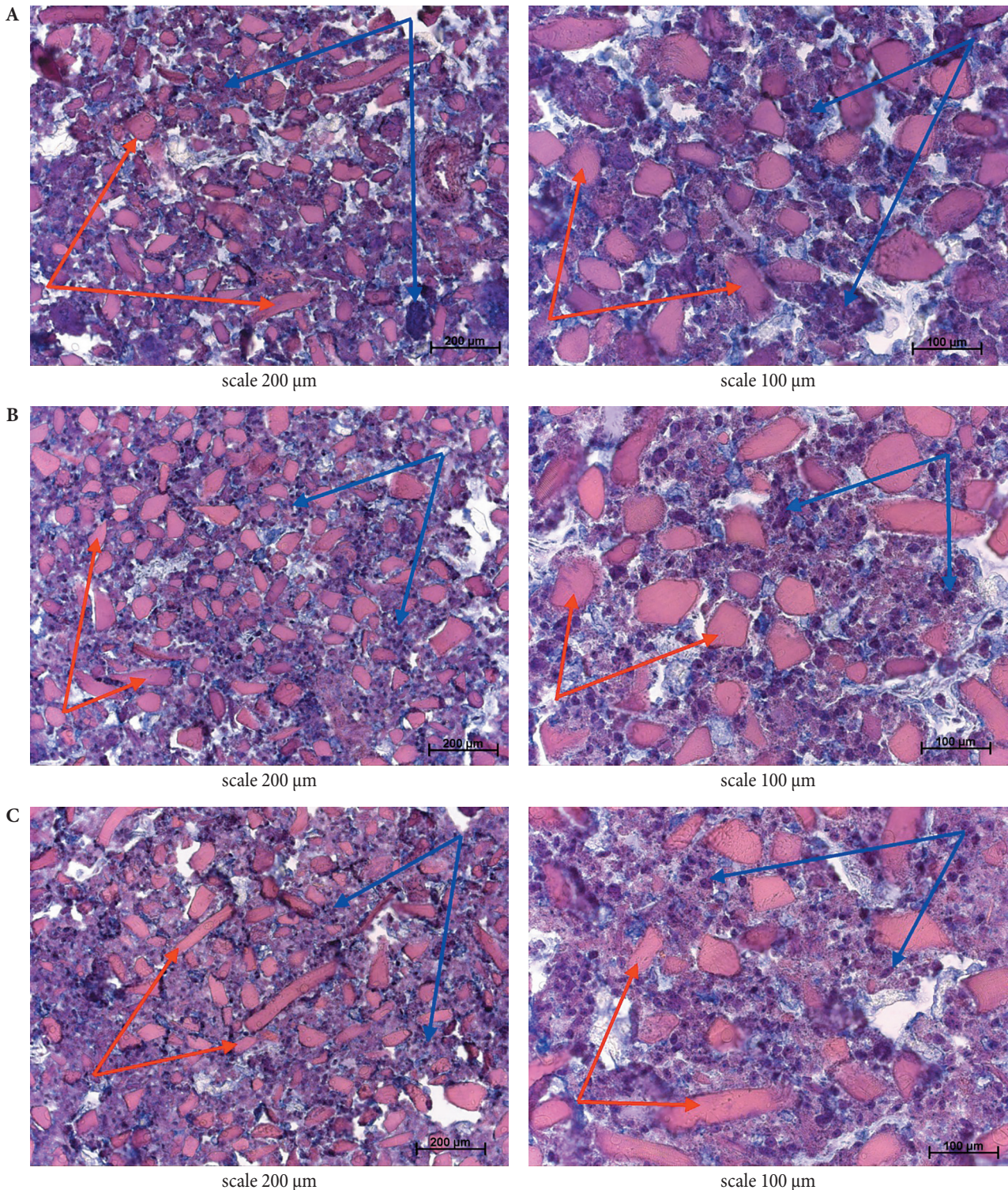


Figure 3. Microstructure of the studied samples (A is Control, B is Sample 2, C is Sample 2). Hematoxylin and eosin staining. Red arrows denote fragments of muscle fibers; blue arrows denote liver fragments and individual hepatocytes

products [56,57]. At the same time, the revealed absence of potential differences in the structure of animal tissue fragments, as the main source of protein, confirms the obtained results of proteomic analysis.

Conclusion

The introduction of 70% ethanol extract of yellow onion peels into the standard formulation of meat pate contributed to the enrichment of the product with plant-based antioxidants, which within 14 days, significantly increased the total antioxidant capacity against nitrogen-containing free radicals by no less than 2.32 times ($P < 0.10$) compared to the control sample. The addition of the extract to the meat product led to an increase in the percentage of adequate daily intake of B vitamins, a decrease in the concentrations of zinc, manganese and magnesium by no more than 14% ($P < 0.10$), simultaneously with an increase in the amount of selenium, copper, potassium and calcium from 8% to 17.35% ($P < 0.10$) depending on the microelement. Fatty acid and amino acid compositions of the experimental products showed minor changes. All pates were characterized by a reduced atherogenic index in comparison with the average AIP of the human diet by 29.7% for the control sample and by 32.4% for the experimental products. The noted increase in the mass

fraction of protein in Samples 1 and 2 by 6.76% and 2.73% ($P < 0.10$), respectively, together with a slight change in amino acid composition, led to a decrease in the minimum amino acid score simultaneously with a decrease in the excess amount of essential amino acids not used for plastic needs. This was evidenced by a decrease in the amino acid score difference coefficient by 7.71% in the case of Sample 1 and by 3.07% in the case of Sample 2, which led to an increase in the biological value of the experimental pates by 7.7% when added the maximum amount of extract and by 3.06% for Sample 2. According to the results of proteomic analysis, it was revealed that the addition of the extract did not lead to significant changes in the protein composition: such fundamental differences as the absence or, conversely, the appearance of *de novo* protein fractions between the control and experimental pates was not detected, while all the main muscle proteins were preserved. Histological analysis revealed that all samples were characterized by the same structure and a high degree of constituent component grinding. The addition of onion peel extract to meat pate, both in the maximum and in half of the maximum volumes, did not affect the density of the structural element arrangement, and there were no differences in the microstructural organization for all experimental products.

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