A STUDY ON THE CHEMICAL AND MINERAL COMPOSITION OF THE PROTEIN-MINERAL PASTE FROM POULTRY AND CATTLE BONE RAW MATERIALS

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Abstract
When processing cattle and poultry, a large quantity of secondary slaughter products in the form of bone raw materials are formed in enterprises of the meat and poultry processing industry. Nowadays, its use is not optimal and rational. One of the promising directions of using bone raw materials in enterprises is production of finely ground meat and bone paste. The aim of this research was to study the chemical and mineral compositions, as well as the content of toxic elements in meat and bone paste from poultry and cattle bones obtained after complex grinding on a grinder and ultra-fine grinder. Chicken bone products (chicken necks, drumsticks, wings, breasts) and cattle costal and vertebral bones with residues of muscle tissue were used for investigation. The comparative analysis of the nutritional value of the meat and bone paste showed the following results: protein mass fraction was 18.5% in the meat and bone paste obtained from poultry bones, and 12.1% in the meat and bone paste obtained from cattle bones. The mass fraction of fat was two times higher in the meat and bone paste from poultry bones. As for the mineral composition, it has been established that meat and bone paste is a rich source of calcium: the calcium content was 1,654.02 mg/100g in the poultry meat and bone paste, and 5,318.13 mg/100g in the cattle meat and bone paste. In regard to the toxic element content, the normed values of lead and arsenic, cadmium and mercury were not revealed in the poultry and cattle meat and bone paste. The obtained meat and bone paste can be used for food purposes as a food additive in meat product manufacture, which will allow rational and economic use of bone raw materials in cattle and poultry processing.

Introduction
Nowadays, most meat processing enterprises face an acute problem of the maximum and rational use of secondary products of processing farm animals and poultry. When processing initial raw materials, valuable kinds of secondary raw material resources are obtained, such as blood, bone, by-products of the 2nd category, crude fat, nonedible by-products and others. They can be used for manufacturing additional food, feed and technical products [1,2]. Wider introduction of complex processing of secondary raw materials will enable its rational use as the main components in the meat product technology increasing product output and assortment [3].

A carcass or half-carcass obtained by slaughter of farm animals and poultry is a complex of muscle, fatty, connective and bone tissues. The rational use of bones from slaughter animals and implementation of non-waste and low-waste technological processes into production has assumed great importance allowing exclusion or minimization of losses and assurance of high-quality products. Moreover, bone raw materials is a source of mineral, protein and fat substances [4,5].

In the Republic of Kazakhstan and CIS states, there are few technologies for processing cattle and poultry meat and bone raw materials to the state of finely ground paste. In the foreign countries, meat and bone paste production is common in Japan and China [6,7,8,9].

At present, there are many technological solutions for bone raw material processing that differ from each other by technological parameters, equipment, process duration and so on. Their common feature is striving for maximum extraction of edible components, edible and technical fat, collagen, mineral substances (components) from raw materials by the mechanical, physical, chemical and thermal impact on bones [10].

L. V. Antipova et al. [11] studied the poultry bone residue regarding the nutritional value and proposed a recipe and technology for dry concentrate production based on broth from poultry bone residue. The authors noted that in terms of the chemical composition, the bone residue contained 25% of protein, 18.9% of fat and 11.1% of ash. In regard to the mineral composition, the high content of calcium (3900 mg/kg), phosphorus (2000 mg/kg) and iron (101.8 mg/kg) was observed.

Meat and bone raw materials are used for food purposes as protein hydrolysates and mineral additives, bone broth and fat are produced after corresponding technological processing. For example, an effective and cost efficient technology for processing poultry meat and bone raw materials was created [12]. By hydrolysis of chicken necks in...
the presence of an inorganic catalyst, a protein product was obtained, which can be used in sausage production, in production of minced semi-finished products, meat fillings and delicacies. Addition of the protein mixture enables increasing the nutritional value and yield of finished delicacy products, improving their sensory properties and rheological characteristics.

Kutsakova et al. [13] developed technologies of three-stage hydrolysis of meat and bone residue of broiler chickens and carcasses of laying hens that allow obtaining food-grade protein hydrolysates that differ by composition and functional-technological properties. Recommendations on the use of hydrolysates as protein additives in meat product manufacture were developed.

The study [14] was devoted to investigation of bone raw material hydrolysis and production of hydrolysates using bone waste from meat, poultry and fish with food additive production. As a result of the hydrolysis of protein mixture from meat and bone raw materials and blood with enzymes of the activated yeast biomass and pancreas, the hydrolysate with the amino acid composition close to optimal was obtained.

Mezenova et al. [15] developed and determined modification parameters for meat and bone raw materials by the hydrolytic method under an action of high temperatures (140 °C) and increased pressure in the aqueous medium (0.62 MPa) to remove valuable protein, fatty and mineral substances. The method of high-temperature modification of meat and bone raw materials allows the complex use of all organic components of raw materials (proteins, minerals, fatty acids) while preserving their native nature and ensuring their sanitary safety. Products of modification of meat and bone raw materials are recommended for the use as food and feed additives, in the composition of biologically active food additives with the osteotropic and gerodietic direction, microbiological media, feeds for aquaculture, fatty compositions.

Wang et al. [16] used the method of hot-pressure extraction (HPE) for extraction of nutrients (protein, collagen, and minerals) from chicken bone residues. High ratios of protein (83.51%) and collagen (96.81%) were obtained at 135 °C and 120 min. Essential amino acids accounted for 31.03% to 47.73% of total amino acids in the extract of chicken bones. However, the calcium content in the extract of chicken bones (4.2 to 4.8 mg/100 g) was relatively low compared to that in the chicken bone residue (1078 mg/100 g). Extracts of chicken bones were obtained by heating at 130±0.5 °C for 120 min. with following filtration, sedimentation, defatting and concentration. Extracts contain ideal protein and fat, which makes them an excellent substrate for hydrolysis. Extract hydrolysates are potentially suitable food additives in the food industry [17]. It can be concluded that hot-pressure extraction is a method for transformation of chicken bone residues into a nutritional substrate with a flavoring agent; however, it is not an effective way for calcium extraction.

Zhang et al. [18] obtained collagen from chicken bone. Fat, minerals and hybrid protein were removed using ethyl acetate, hydrochloric acid and calcium hydroxide, respectively. The optimal process of collagen extraction envisaged the solid-liquid ratio 1: 8, extraction temperature 100 °C, extraction time 4h and pH 4.0. The content of extracted protein was 88.75%, collagen 86.02%. An ultra-fine bone powder was prepared from bone residues using an ultra-fine pulverizer. Then, the content of nutrients was determined. This method not only solves the environmental problems of livestock and poultry processing industry but also opens new channels and new areas for studying functional foods, and has good social and economic benefits.

Another direction of using bone raw materials is collagen production. Cansu et al. [19] developed a multi-step procedure for collagen isolation from chicken bones that allowed removing 87.5% of minerals and 57.1% of fats with protein losses of about 18.6% and hydroxyproline losses of 14.9%. The collagen yield was about 85% of the initial content, its quality and functional properties were assessed.

Suparno et al. [20] and Kodous et al. [21] described isolation of collagen from chicken feet in the acetic acid solution at 4 °C for 24 hours using papain and pepsin with the yield of 18.16% and 22.94%. The obtained collagen contained large amounts of glycine, glutamic acid, proline and hydroxyproline.

These methods of bone raw material processing are based on thermal, acid treatment, which main disadvantage is the loss of nutrients. Moreover, there are significant energy expenditures and labor intensity in several processes.

In this connection, it is necessary to emphasize particularly such direction in bone processing for food purposes as its processing into the finely dispersed mass for using in production of different meat product types. Processing of meat and bone raw materials into finely dispersed paste allows adding this product into sausages, meat semi-finished products such as pelmeni, cutlets, meat balls and so on enriching them with valuable mineral and protein components. The proposed technology for processing meat and bone raw materials allows obtaining meat and bone paste with the high proportion of mineral substances, in particular, calcium, magnesium, iron satisfying the daily requirement up to 50%. The obtained meat and bone paste is recommended to use as a food additive in production of combined meat products.

Meat product manufacture using meat and bone paste ensures an increase in the product yield, broadens an assortment of manufactured products, contributes to an improvement of the ecological state of production and enables the rational use of secondary raw materials, thereby increasing economic efficiency. Moreover, this will give a noticeable economic effect with an increase in profitability of a product in the market [22]. With that, it is necessary to note that the ecological problem is solved simultaneously as all raw materials including those of low value are
subjected to processing. Therefore, the proportion of industrial waste generated in case of consumption of poultry meat as carcasses or meat and bone semi-finished products is significantly reduced.

The above-mentioned suggests the topicality of the development of the theoretical foundations and practice for deep non-waste processing of meat and bone raw materials and their use in new technologies for functional meat products to enrich them with valuable macro- and microelements and food nutrients.

The aim of the work was to study the chemical and mineral composition of the meat and bone paste from poultry and cattle bone raw materials.

Materials and methods

The objects of the research were samples of frozen meat and bone paste from poultry and cattle bones. Bones with residues of poultry and cattle muscle tissue were used for grinding. Meat and bone raw materials were obtained from meat processing enterprises and large meat trading pavilions in Semey city (Eastern Kazakhstan region, Kazakhstan Republic).

Production of meat and bone paste for investigations

At the first stage of the experiment, the scheme for meat and bone raw material processing was developed, which consisted in successive grinding of meat and bone raw materials preliminarily frozen to a temperature of –18 °C…–20 °C in freezers. For the experiment on grinding of meat and bone raw materials, chicken bone products (chicken necks, drumsticks, wings, breasts) and cattle costal and vertebral bones with residues of muscle tissue were used for investigation.

Cattle costal and vertebral bones were preliminarily ground to a size of 50–70 mm. Then, meat and bone raw materials were preliminarily frozen in freezers at a temperature of –18 °C…–20 °C for 60 min. After that, frozen raw materials were fed into a bin of a grinder with a diameter of plate holes of 8 mm. After grinding, the obtained meat and bone mass was frozen to a temperature of –18 °C…–20 °C and was ground again in a grinder with a diameter of plate holes of 5 mm. Ice water was added to the meat and bone mass with the raw materials: water ratio of 1:0.5 and was mixed until obtaining a homogeneous mass [23, 24].

Then, meat and bone forcemeat was successively ground using a Supermasscolloider ultra-fine grinder with the following clearance between the nonporous grinders: 0.25 mm, 0.1 mm, 0.02 mm. Meat and bone paste with tender spreadable consistency without a sense of toughness on the tongue was obtained at the exit of the grinder. The obtained meat and bone paste was stored at a temperature of 2–4 °C until the further experiments.

Determination of the total chemical composition was carried out by the method of a single weighed portion of the test sample. The method consisted in the successive determination of moisture\(^1\), protein\(^2\) and fat\(^3\) in a single weighed portion.

Minerals were determined according to the normative documents R 4.1.1672–03 “Guidelines for quality control and safety of biologically active food additives”, GOST 26928–86 \(^5\) “Foodstuffs. Method for determination of iron”, GOST 33824–2016 “Foodstuffs and food ingredients. Stripping voltammetric method for determination of toxic elements (cadmium, lead, copper and zinc)”. Toxic safety was studied by the standard methods according to TR CU021/2011 Technical Regulations of the Customs Union “On the safety of food”. Heavy metals (Pb, As, Cd, Hg) were determined according to the normative documents: GOST 30178–96 \(^6\) “Raw material and foodstuffs. Atomic absorption method for determination of toxic elements”, GOST R 51766–2001 \(^7\) “Raw material and food-stuffs. Atomic absorption method for determination of arsenic”, MUC 4.1.1472–03 \(^8\) “Atomic absorption determination of the mass concentration of mercury in biomaterials of animal and plant origin (food, feed, etc.)”.

Statistical analysis was carried out using the software package Statistica 6.0 and Excel 2007.

Results and discussion

At the first stage, the chemical composition of the meat and bone paste from poultry and cattle raw materials was studied. The bone tissue consists of the cellular elements and intercellular substance, which includes intermediate substance, formed particles — collagen fibers and inorganic salts [25]. The results of the chemical composition are presented in Table 1. The results of the comparative analysis of the chemical composition show that the poultry and cattle meat and bone paste is characterized by the high protein content (18.3% and 12.1%),

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1 GOST 33319–2015, “Meat and meat products. Method for determination of moisture content”. Moscow: Standartinform, 2018. — 14 p. (In Russian)
2 GOST 25011–2017 “Meat and meat products. Protein determination methods”. Moscow: Standartinform, 2018. — 14 p. (In Russian)
3 GOST 23042–2015 “Meat and meat products. Methods of fat determination”. Moscow: Standartinform, 2019. — 8 p. (In Russian)
4 R 4.1.1672–03 “Guidelines for quality control and safety of biologically active food additives”. Retrieved from https://www.rospotrebnadzor.ru/upload/iblock/33e/r-4.1.1678_03.pdf Accessed December 11, 2020. (In Russian)
5 GOST 26928–86 “Food-stuffs. Method for determination of iron”. Moscow: Standartinform, 2010. — 6 p. (In Russian)
6 GOST 33824–2016 “Foodstuffs and food ingredients. Stripping voltammetric method for determination of toxic elements (cadmium, lead, copper and zinc)”. Moscow: Standartinform, 2016. — 23 p. (In Russian)
7 TR CU021/2011 Technical Regulations of the Customs Union “On the safety of food”. Retrieved from http://docs.cntd.ru/document/902320560 Accessed December 11, 2020. (In Russian)
8 GOST 30178–96 “Raw material and food-stuffs. Atomic absorption method for determination of toxic elements”. Moscow: Standartinform, 2010. — 20 p. (In Russian)
9 GOST R 51766–2001 “Raw material and food-stuffs. Atomic absorption method for determination of arsenic”. Moscow: Standartinform, 2011. — 12 p. (In Russian)
10 MUC 4.1.1472–03 “Atomic absorption determination of the mass concentration of mercury in biomaterials of animal and plant origin (food, feed, etc.)”. Retrieved from http://docs.cntd.ru/document/1200034851 Accessed December 11, 2020. (In Russian)
respectively). This suggests the expediency of using meat and bone paste in products aimed towards compensation of deficiency in the human protein ration. This allows regarding meat and bone paste as a valuable raw material for food manufacture. The main protein of bone tissue is collagen [26]. Bone tissue collagen is assigned to fibrillar collagens of type I. Collagen is an important component in the composition of food products with a favorable effect on the state of beneficial microflora. A distinctive feature of collagen is the high content of proline and oxyproline. Moreover, collagen facilitates an increase in the emulsifying capacity of the system [27].

Significant differences in the fat content are seen. For example, the poultry meat and bone paste contains 11.8% of fat, which is higher than that in the cattle meat and bone paste (5.3%). Bone fat is concentrated mainly in bone marrow. The peculiar characteristic of bone fat is the high content of lecithin compared to other types of animal fats. The high content of lecithin determines the good emulsifying capacity of this fat type and predetermines its use in production of emulsions [28].

The ash part of the meat and bone paste from poultry was equal to 4.35%, from cattle — 6.80%. Bones have very high stiffness and resilience, which is achieved by a peculiar combination of the organic base with minerals. Bone tissue is a source of mineral salts. It contains 98% of all inorganic substances in the body including 99% Ca, 87% P, 58% Mg, 46% Na [29].

The chemical composition of bones is variable and depends on an animal breed, age and fatness, as well as on a bone type: the fat and mineral content is increased and water content is decreased with an increase in fatness [30].

The results of the mineral composition of poultry and cattle meat and bone paste are shown in Table 2.

Table 1. Chemical composition of meat and bone paste from poultry and cattle bone raw materials

| Type of meat and bone paste | Moisture, % | Protein, % | Fat, % | Ash, % |
|---------------------------|------------|------------|--------|--------|
| Poultry meat and bone paste | 65.55 ± 1.41 | 18.3 ± 0.36 | 11.8 ± 0.28 | 4.35 ± 0.12 |
| Cattle meat and bone paste | 75.8 ± 1.91 | 12.1 ± 0.21 | 5.3 ± 0.17 | 6.80 ± 0.19 |

Table 2. Mineral composition of poultry and cattle meat and bone paste compared to the mineral composition of by-products

| Minerals, mg/100g | Type of meat and bone paste | Calcium | Magnesium | Iron | Zinc |
|-------------------|----------------------------|---------|-----------|------|------|
| Poultry meat and bone paste | 1,654.02 ± 330.80 | 14.54 ± 2.91 | 3.83 ± 0.77 | 0.070 ± 0.020 |
| Cattle meat and bone paste | 5,318.13 ± 1063.63 | 207.62 ± 41.52 | 8.35 ± 1.67 | Not detected |

Cattle by-products (literature data) [31, 32]

| Minerals, mg/100g | Type of meat and bone paste |
|-------------------|----------------------------|
| Liver | 5.00 ± 0.67 |
| Heart | 8.00 ± 1.67 |
| Kidney | 13.00 ± 0.22 |
| Tongue | 6.39 ± 1.07 |
| Brain | 43.00 ± 22.50 |

Poultry by-products and eggshell (literature data)

| Minerals, mg/100g | Type of meat and bone paste |
|-------------------|----------------------------|
| Chicken liver [33] | 15 |
| Eggshell [34] | 33400–37300 |

Calcium is one of the most important elements for the human body ensuring its normal vital activities. Calcium improves the blood coagulability and nervous system function [35]. The human body cannot produce calcium by itself. Therefore, to maintain the sufficient level of calcium, it is necessary to intake it from different food sources [36]. The best method for meeting the need for this mineral is balanced nutrition with consumption of foods rich in calcium. However, food additives can also be a calcium source.

Analysis of the mineral composition of the meat and bone paste from poultry and cattle bones indicates the considerable calcium content. For example, the calcium content was 1,654.02 mg/100g in the meat and bone paste from poultry bone raw materials and 5,318.13 mg/100g in the meat and bone paste from cattle bones. The significant difference in the calcium content is explained by the structure of cattle and poultry bone tissue. It is known that chicken bones are less hard and thinner than those of mammals; tubular bones do not contain bone marrow and are filled with air [37].

The comparative analysis of the mineral composition of the meat and bone paste and by-products from cattle (liver — 5.00 mg/100g, heart — 8.00 mg/100g, kidney — 13.00 mg/100g, tongue — 6.39 mg/100g, brain — 43.00 mg/100g) and poultry (chicken liver — 15 mg/100g) revealed a significant excess of the calcium content in the meat and bone paste, excluding eggshell.

Another important macro-element for the human body is magnesium. Magnesium normalizes metabolic processes and prevents the development of allergy [38]. The magnesium content was 14.54 mg/100g in the meat and bone paste from poultry bones and 207.62 mg/100g in the meat and bone paste from cattle bones. The increased magnesi-
sium content in cattle bones is explained by its important role in balance maintenance and retention of calcium and phosphorous in bone tissue. Compared to other by-products, the magnesium content in the meat and bone paste from cattle bones is much higher than that in cattle and poultry by-products. Only eggshell contains more magnesium (406–412.9 mg/100 g).

Besides calcium and phosphorous compounds, bone tissue contains iron, copper and zinc. Iron in bone tissue facilitates calcification of bones of the skeleton. In addition, it is necessary for the cytochrome oxidase activity, which plays an important role in the bone cell function [39]. The high metabolic role of zinc is conditioned by the fact that it is an activator of several enzymes including alkaline phosphatase of bone tissue and others. An excess in calcium and copper inhibits zinc metabolism, processes of growth and differentiation of bone tissue, especially in tubular bones [40].

As for microelements, the iron content was the highest in the cattle meat and bone paste (8.35 mg/100g); it was two times lower in the poultry meat and bone paste (3.83 mg/100 g) upon the reference daily intake of 1–2 mg. Among by-products, chicken liver was characterized by significant amounts of iron (17.5 mg/100 g) compared to cattle liver (4.80 mg/100 g). Significant differences were observed in the zinc content. For example, the highest zinc content was recorded in the chicken liver (6.6 mg/100 g) and cattle liver (4.00 mg/100 g). Lower amounts of zinc (0.070 mg/100g) were observed in the poultry meat and bone paste.

At the next stage, toxic safety of the meat and bone paste was determined (Table 3). The content and level of accumulation of heavy metals in the animal body depend on the kind of consumed feed and type of feeding, zone of raising, climatic conditions and so on. Heavy and toxic metals entering the animal body are accumulated mostly in bones, liver and kidneys.

As lead is widely distributed in nature in relatively high amounts, it is accumulated in the animal body, mainly in bones (90%), comparatively quickly. Lead negatively affects the hematopoietic, nervous, digestive systems and kidneys [41].

Cadmium has high toxicity. Exposure to cadmium causes the oxidative stress in the animal body disturbing the oxidative and antioxidative balance [42]. Excessive accumulation of cadmium in the human body leads to the impaired renal function, dizziness, nausea, skin disorders, reduced appetite, increased arterial pressure, changes and pain in bones and joints. The presence of arsenic compounds in foods results in serious human diseases later on. Long-term exposure to arsenic can lead to cancer development and skin lesions [43].

The other dangerous element is mercury. Mercury is toxic for the central and peripheral nervous system (mercurial erethism). Chronic poisoning causes predisposition to tuberculosis, atherosclerotic events, liver and gall bladder disorders, hypertension [44].

The critical levels of heavy metals were not revealed in the meat and bone paste: the lead content was 0.071 mg/kg and 0.056 mg/kg, the arsenic content was 0.016 mg/kg and 0.011 mg/kg. It corresponded to the norms of MACs by the content of toxic elements.

Therefore, animal and poultry bones are a rich source of mineral substances. The use of mineral constituents in the food technology upon proper technological and mechanical processing allows enriching products with mineral additives, in particular, calcium, magnesium and other elements.

**Conclusion**

As a result of the performed experiments, complex processing of meat and bone raw materials was proposed for production of meat and bone paste. The proposed technology for production of meat and bone paste from poultry and cattle bone products differs from existing ones due to the complex scheme of meat and bone processing including stepwise grinding with freezing and following processing to produce finely dispersed meat and bone paste suitable for the use with food purposes as food additives. Analysis of the chemical composition of meat and bone paste shows that meat and bone paste from poultry and cattle bones is a source of protein (18.3% and 12.1%, respectively), fat and minerals, which indicates its nutritional value. Analysis of the mineral composition revealed that the main constituent of the meat and bone paste was calcium, which content was 1,654.02 mg/100 g in the poultry meat and bone paste and 5,318.13 mg/100g in the meat and bone paste from cattle bones. In regard to the toxic element content, critical levels of lead (0.071 mg/kg) and arsenic (0.016 mg/kg) were not revealed in the poultry and cattle meat and bone paste. The content of toxic elements in the meat and bone paste corresponds to food safety standards. The produced meat and bone paste from poultry and cattle meat and bone raw materials is a valuable source of minerals. Production of combined meat products with addition of meat and bone paste into a recipe allows improving the nutritional value of final products.

**Table 3. Content of toxic elements in meat and bone paste**

| Toxic elements mg/kg, not more: | Norms according to normative documentation | Poultry meat and bone paste | Cattle meat and bone paste |
|---------------------------------|------------------------------------------|----------------------------|----------------------------|
| Lead                            | 0.5                                      | 0.071                      | 0.056                      |
| Arsenic                         | 0.1                                      | 0.016                      | 0.011                      |
| Cadmium                         | 0.05                                     | Not detected               | Not detected               |
| Mercury                         | 0.03                                     | Not detected               | Not detected               |
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