

# STUDY OF CHEMISTRY AND HYDROLYSATES DRYING PARAMETERS OF FEATHER-DOWNY RAW MATERIAL

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Received February 07, 2016; Accepted in revised form March 20, 2016; Published June 27, 2016

**Abstract:** The article describes chemical and amino acid composition of feather-downy raw material. It determines the mass fraction of crude protein, crude fiber, ash, calcium, phosphorus, sodium in the samples of feather-downy raw material. It is stated that the waste from poultry processing obtained from hens of all the studied species are characterized by a high content of crude protein and low in crude fiber and ash. The most valuable feather-downy raw material regarding protein is waste containing keratin obtained from the Lohmann Brown hens. We have studied the composition of the peptide fractions of feather-downy raw materials by polyacrylamide gel electrophoresis according to Laemmli in order to understand chemistry of feather-downy raw material better. The obtained results show the presence of a wide variety of protein fractions with different molecular weights in tested keratin raw material. Half of all the proteins are fractions with a molecular weight of 60.0–56.0 kDa. It is found that feather and downy raw material has a sufficient number of low-molecular peptides. It is proved that this fraction corresponds to alpha-keratin. An important indicator for animal feed is their amino acid content, so we studied the quantitative content of essential and nonessential amino acids in feather-downy raw materials. The results of research indicate that poultry processing waste is rich in sulfur-containing amino acids such as cysteine and methionine. It is proved that the data on the physical and chemical composition of feather-downy raw material obtained from three different breeds of chickens, allow recommending the processing of poultry waste as a promising object for high-protein feed for farm animals with a balanced content of macro and microelements.

**Keywords:** Feather-downy raw materials, mass fraction of protein, fiber, ash, waste from poultry processing

DOI 10.21179/2308-4057-2016-1-44-50

*Foods and Raw Materials*, 2016, vol. 4, no. 1, pp. 44–50.

## INTRODUCTION

The most interesting of all the waste from poultry processing is feather-downy raw material as it constitutes 30% of all waste weight. For its processing it is necessary to study the structure and origin of the raw materials in details and to find an alternative source of its degradation [1, 2, 3]. O.F. Chernova, who works at A.N. Severtsov Institute of Ecology and Evolution, in her article, gives the broad and substantive review of hypotheses of raw materials origin containing keratin, such as feathers and hair. Moreover she makes a list of similarities and differences in their structure, analyzes some signs of convergence [4, 5].

In this study physical and chemical properties of the feathers are of special interest for the selection of the optimal conditions of total hydrolysis. Feathers and hair are developed from the follicle with the ectoderm and mesoderm, they have multi-layered keratin structures capable of regeneration. Regeneration is provided by stem cells located within the follicle [6, 7].

It was found that the development of feathers and hair is governed by a set of similar proteins which work in uniform signaling cascades. Protein SNN is the alarm center, Noggin is responsible for the division of feather and hair cells, WNT $\beta$ -11 catenin determines the differentiation of stem cells in buds. The appearance of skin structures is determined by gradients of signal proteins. It is shown experimentally that all the skin appendages appear as a result of the interaction of ecto- and mesoderm. The degree of participation of ecto- and mesoderm in this case may differ, i.e. the development and differentiation of skin appendages are “different variations” [8, 9, 10].

The similarity of functions and a hypothetical common origin of feathers and hair suggest a lot of similarities in their structure. Under the common origin we understand participation of the same tissue structures in the production of feathers and hair. It means some common histological features and similar biochemical regulation.

Feathers and hair evolved separately. It is seen from different composition of feathers and hair keratins. Feathers and scales of modern reptiles are composed by two types of keratins:  $\alpha$ - and  $\beta$ -keratins, including the specific form of  $\beta$ -keratin –  $\phi$ -keratin (analyzed by the appropriate tissue of an alligator). Modern mammals' hair have only  $\alpha$ -keratin [11, 12].

Keratins (from Greek *keras*, Gen.case *keratos* – a horn) are structural fibrous proteins consisting of parallel polypeptide chains which have the conformation of  $\alpha$ -helix or  $\beta$ -structure (the structure of the pleated sheet).  $\alpha$ -Keratins (often simply called keratines) are the main type of protein, which form the outer protective coating of vertebrates. Keratin is a typical representative of fibrous proteins. It is found in the tissue of epidermal origin that is wool, hair, horns, claws, feathers, hooves, whalebone, and others [13]. All of the above tissues are complex multi-component biological formation composed of separate cells which form their various histostructural elements [14].

The aim of this research is to study the chemistry of feather-downy raw material of chickens of various breeds and the development of technological parameters of hydrolysates drying of feather-downy raw material.

#### OBJECTS AND METHODS OF STUDY

The object of the research is feather-downy raw material obtained from hens of different breeds: the French breed F-15 from LLC “Breeding Poultry State Fram Kolmogorov” (Kemerovo region, Yashkinsky district, village Kolmogorovo); the Lohmann Brown breed from JSC “Kuzbass Poultry Factory” (Kemerovo region, Novokuznetsk district, village Stepnoi); The Lohmann LSL-Classic from Ltd “Inskaya Poultry Factory” (Kemerovo region, village Inskoi).

At different stages of work we used the following materials and reagents: distilled water (GOST 6709-72); sodium chloride (GOST 4233-77, 99.8%, reagent grade); soluble starch (GOST 10163-76, 98.0%, analytical grade); acrylamide (“Sigma”, USA); N, N'-methylene-bisacrylamide (“Sigma”, USA); ethidium bromide (Sigma, USA).

At different stages of work we used the following equipment: a spectrophotometer UV 1800 (Shimadzu, Japan), ultracentrifuges Beckman J2-HS (Beckman, USA), a nitrogen analyzer Rapid N Cube (Elementar,

Germany), a liquid chromatograph LC-20 (Shimadzu, Japan), an amino acid analyzer ARACUS (Analytical Systems Gmb, Germany), the camera for vertical electrophoresis and power source PowerPack HC (Bio-Rad, USA), a UV-transilluminator TCP-20M (Vilber Lourmat, USA), gel documentation system Doc XR plus (Bio-Rad, USA), a fermenter Biostat A plus MO, 5 l, Sartorius (Sartorius, Germany), a refractometer HI 96801 (HANNA, Romania), a centrifuge CV-50 (ELMI, Latvia), analytical balance AND HR-202 i (A & D, Japan), a pH-meter Sevev Compact (Mettler Toledo, USA), a laboratory microbiological incubator ILM-170-01 “Laminar-C” (JSC “Laminar systems” Russia), freeze-drier “HOARFROST-6M” (Russia).

Theoretical and experimental studies were carried out in accordance with modern research methodology of complex phenomena by means of conventional, standard and original methods of biochemical, physical, chemical, structural and mechanical analysis.

#### RESULTS AND DISCUSSION

The results of the mass fraction of crude protein, crude fiber, ash, calcium, phosphorus, sodium in the samples of feather-downy raw materials are shown in Table 1.

Table 1 shows that the waste of poultry processing, obtained from chickens of all studied breeds, are characterized by a high content of crude protein (79.53–90.11%) and a low crude fiber content (less than 0.87%) and ash (max 0.20%). Most valuable feather-downy raw material regarding protein content are keratin-wastes obtained from the Lohmann Brown hens (mass fraction of crude protein is 90.11%).

The results of studying microelement composition of feather-downy raw material suggest that test samples satisfy the requirements of normative documents for poultry feed regarding calcium (0.90–0.97%), phosphorus (0.70–0.71%) and sodium (0.16–0.31%) content.

The obtained data indicate that poultry processing waste can be used as a promising feedstock for producing high-protein feed for agricultural animals.

In order to understand chemistry of feather-downy raw material better we studied the composition of the peptide fractions by polyacrylamide gel electrophoresis according to Laemmli. The results are shown in Table 2.

**Table 1.** Chemistry of feather-downy raw materials obtained from hens of different breeds

| Indicator   | Indicator values of raw material from hens of different breeds |                   |                         |
|---|--|-------------------|-------------------------|
|   | “F-15 Isa”   | The Lohmann Brown | The Lohmann LSL-Classic |
| Mass fraction of crude protein, %                       | 79.53 ± 5.33   | 90.11 ± 5.56      | 89.08 ± 5.51            |
| Mass fraction of crude fiber, %                         | 0.87 ± 0.04  | 0.49 ± 0.02       | 0.69 ± 0.03             |
| Mass fraction of ash. insoluble in hydrochloric acid, % | 0.20 ± 0.02  | 0.31 ± 0.02       | 0.30 ± 0.02             |
| Mass fraction of calcium, %                             | 0.90 ± 0.10  | 0.97 ± 0.10       | 0.89 ± 0.10             |
| Mass fraction of phosphorus, %                          | 0.66 ± 0.05  | 0.71 ± 0.05       | 0.57 ± 0.04             |
| Mass fraction of sodium, %                              | 0.16 ± 0.02  | 0.31 ± 0.02       | 0.20 ± 0.03             |

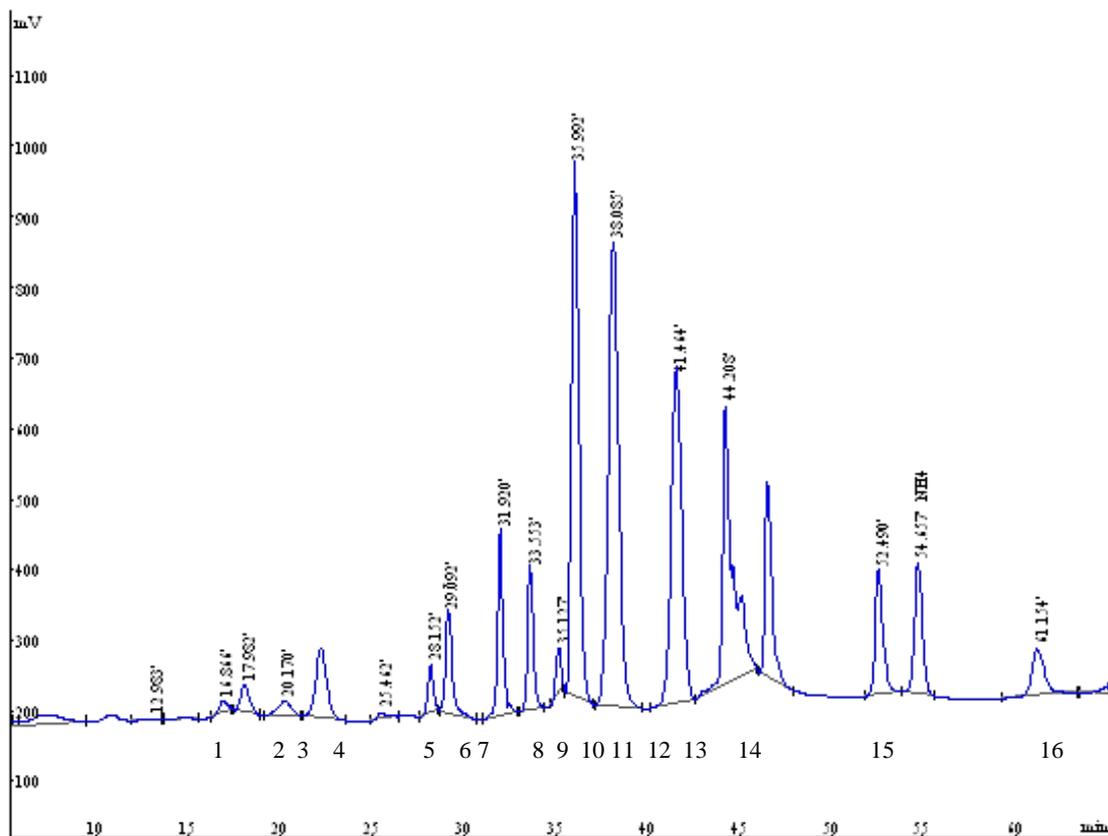
**Table 2.** Molecular weight distribution of proteins in feather-downy raw material

| Molecular weight range, % | Relative content of fractions, % |                   |                         |
|---------------------------|----------------------------------|-------------------|-------------------------|
|                           | “F-15 Isa”                       | The Lohmann Brown | The Lohmann LSL-Classic |
| 71.0–66.0                 | 4.1                              | 4.1               | 3.9                     |
| 66.0–60.0                 | 3.6                              | 3.9               | 4.1                     |
| 60.0–56.0                 | 48.6                             | 50.1              | 49.1                    |
| 56.0–46.0                 | 16.1                             | 15.6              | 13.9                    |
| 46.0–36.0                 | 5.5                              | 4.7               | 4.9                     |
| 36.0–30.0                 | 15.9                             | 13.8              | 14.1                    |
| 30.0–15.0                 | 9.7                              | 6.8               | 9.2                     |
| less than 15              | 3.4                              | 2.0               | 3.1                     |

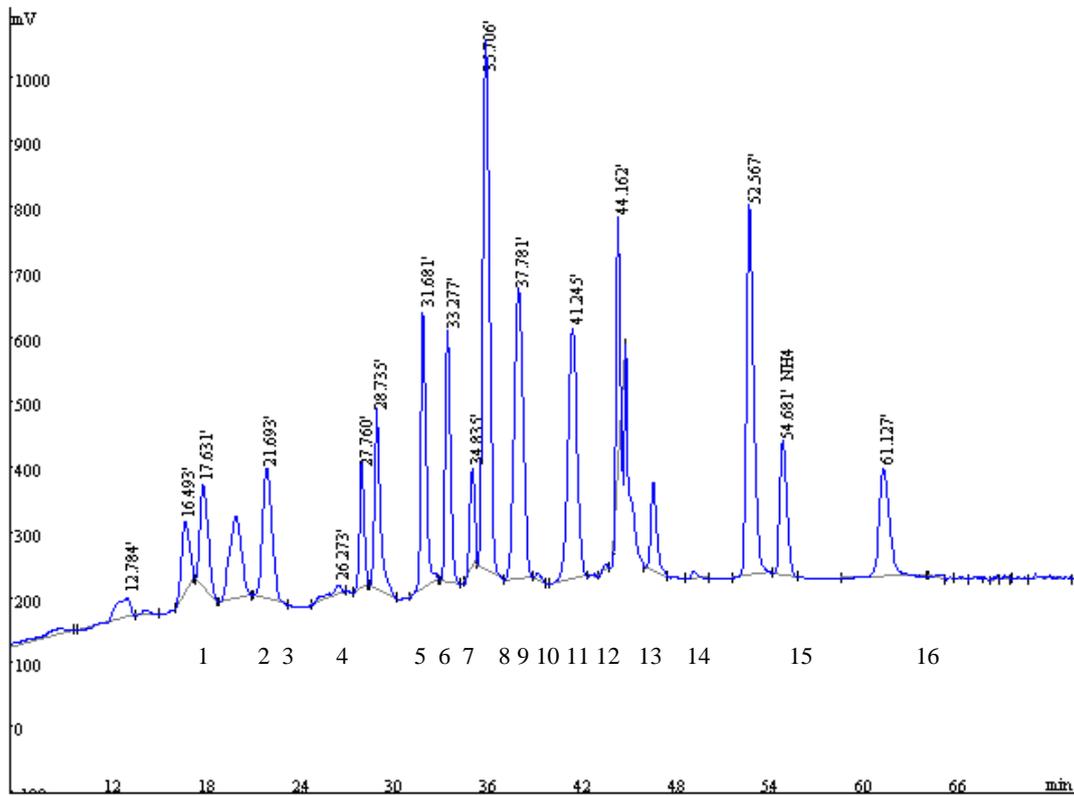
The results presented in Table 2 indicate a wide variety of protein fractions with different molecular weights in the test raw material containing keratin. Half of all the proteins are fraction with a molecular weight of 60.0–56.0 kDa.

Feather-downy feedstock has a sufficient number of low-molecular peptides with molecular weight less than 10 kDa. According to the literature, this fraction corresponds to the  $\alpha$ -keratin. In addition, the electrophoretogram identified protein fractions with a molecular weight of 43.0–33.0 kDa which are probably representatives of  $\beta$ -keratins.

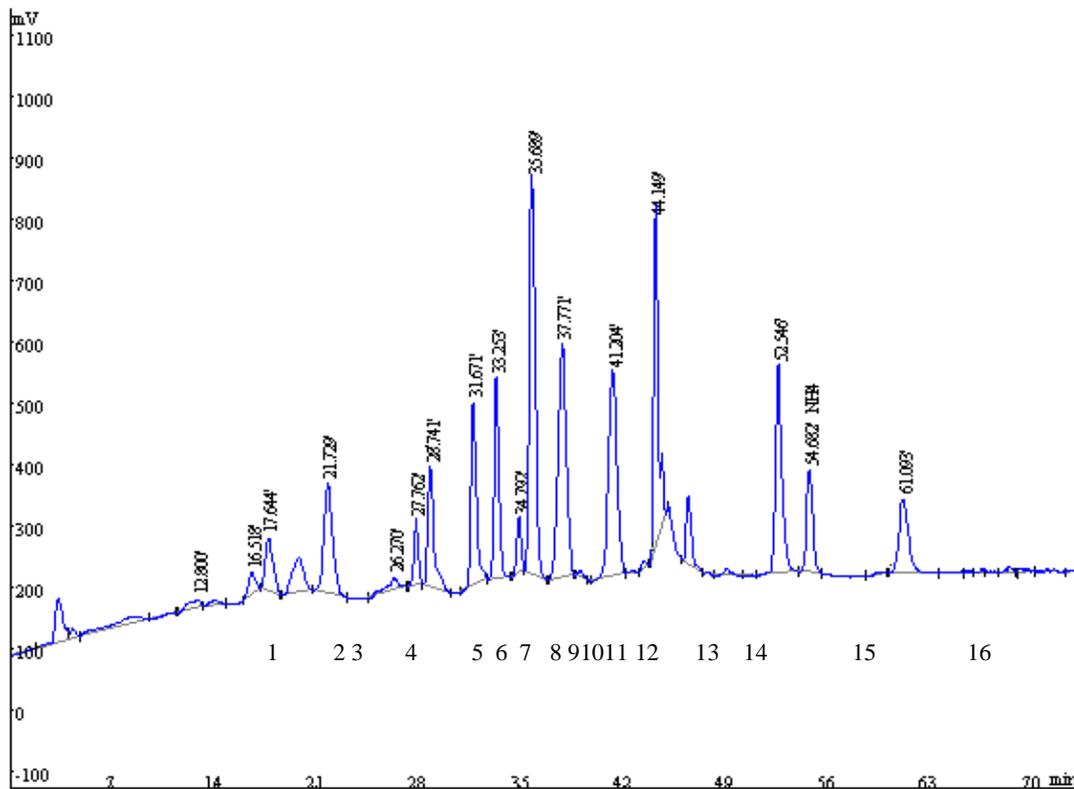
An important indicator for animal feed is its amino acid composition. Therefore, we studied the quantitative content of essential and nonessential amino acids in feather-downy raw material. The obtained data are shown in Fig. 1–3.



**Fig. 1.** The chromatogram determining amino acid composition of feather-downy raw material obtained from the breed “F-15 Isa”: 1 – aspartic acid; 2 – serine; 3 – threonine; 4 – glutamic acid; 5 – proline; 6 – glycine; 7 – alanine; 8 – cysteine; 9 – methionine; 10 – isoleucine; 11 – leucine; 12 – tyrosine; 13 – phenylalanine; 14 – histidine; 15 – lysine; 16 – arginine.



**Fig. 2.** The chromatogram determining amino acid composition of feather-downy raw material obtained from the Lohmann Brown breed: 1 – aspartic acid; 2 – serine; 3 – threonine; 4 – glutamic acid; 5 – proline; 6 – glycine; 7 – alanine; 8 – cysteine; 9 – methionine; 10 – isoleucine; 11 – leucine; 12 – tyrosine; 13 – phenylalanine; 14 – histidine; 15 – lysine; 16 – arginine.



**Fig. 3.** The chromatogram determining amino acid composition of feather-downy raw material obtained from the Lohmann LSL-Classic breed: 1 – aspartic acid; 2 – serine; 3 – threonine; 4 – glutamic acid; 5 – proline; 6 – glycine; 7 – alanine; 8 – cysteine; 9 – methionine; 10 – isoleucine; 11 – leucine; 12 – tyrosine; 13 – phenylalanine; 14 – histidine; 15 – lysine; 16 – arginine.

The obtained results of amino acid composition of feather-downy raw material indicate that poultry processing waste is rich in sulfur containing amino acids such as cysteine and methionine. The cysteine content is at the average 9.14 mg / 100 g of sample, the methionine content is 12.48 mg / 100 g of sample. As for other amino acids, samples had a high content of aspartic acid (6.88 mg / 100 g), serine (5.39 mg / 100 g), glycine (6.51 mg / 100 g), alanine (5.36 mg / 100 g), leucine (6.24 mg / 100 g), tyrosine (5.14 mg / 100 g), lysine (5.08 mg / 100 g), arginine (5.98 mg / 100 g).

An important step in the production of feed for farm animals based on hydrolysates of secondary raw materials is their dehydration (drying).

Among hundreds of engineering solutions and processes which are applied nowadays there are two basic directions: atmospheric drying and vacuum drying. Atmospheric drying has a significant drawback: it involves a prolonged contact of the product with a high temperature oxygen ambient air composition. It leads to intense oxidation reactions and, as a result, to low-quality of most dry food. Therefore at present vacuum drying at pressure below the triple point (freeze-drying) or vacuum evaporation are more widespread.

Analysis of modern methods of drying leads to the conclusion that the gentlest method of dehydration of biological objects is freeze-drying (lyophilization).

Lyophilization is dehydration of biological objects in a frozen state under vacuum. When freeze drying method is used the removing moisture is carried out through transition ice - vapor. Most moisture (65–85%)

is removed by ice sublimation at temperatures below 0°C, and residual moisture removes only when it is heated up to 50–70°C.

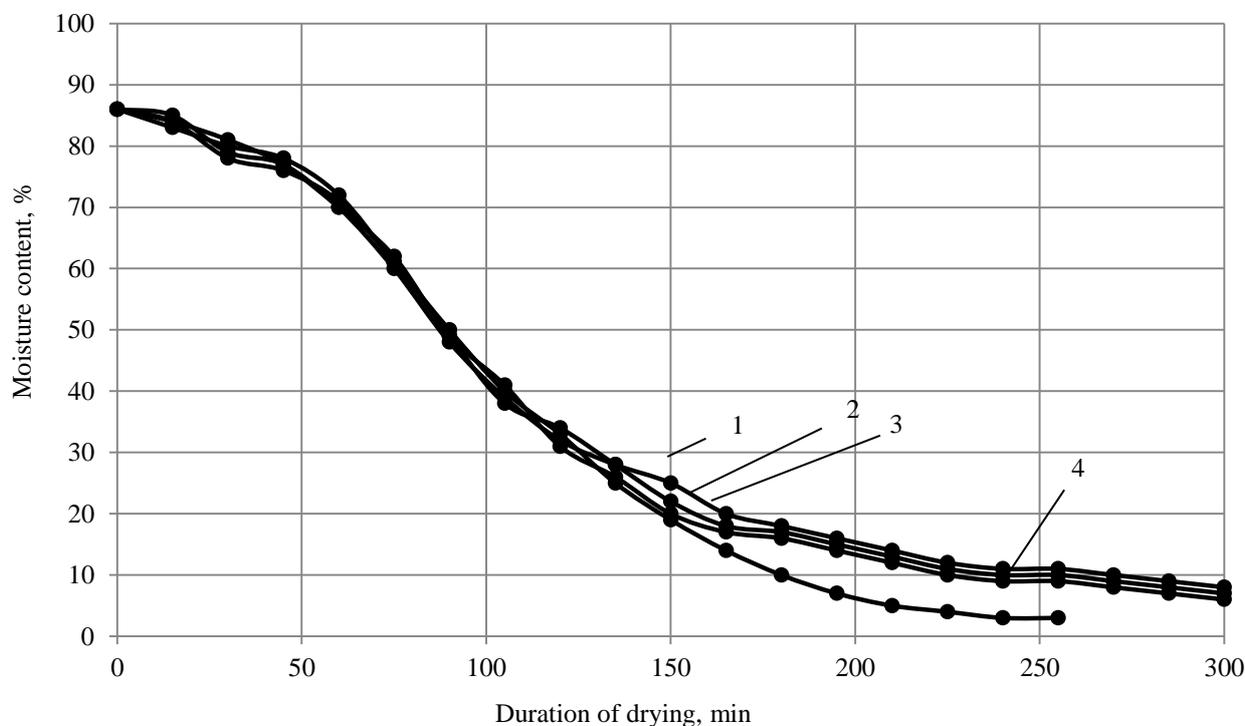
Freeze drying has three stages. The first process step of freeze drying is to freeze biological material. In the process of object freezing 20–25% of the moisture vaporizes through the allocation of ice melting heat when water freezes.

The second period (sublimation) is characterized by the constant speed of object drying. At this stage the bulk of moisture is removed. The third period of removing residual moisture is characterized by falling of drying speed; the object temperature becomes positive. During this period the bound moisture not frozen in the object is removed. The drying speed depends on the intensity of heat input. The object temperature is gradually increased to ambient temperature.

Freeze drying provides a longer shelf life of products (up to 10 years) and the maximum degree of recoverability.

At this stage we set parameters of freeze drying (temperature and layer thickness) of waste hydrolysates of feather-downy raw material in order to produce feed for farm animals.

Scores of the temperature and freeze drying duration of waste hydrolysates of feather-downy raw material were carried out at the drying layer thickness of 6.0 mm and at different drying temperatures:  $26 \pm 1^\circ\text{C}$ ,  $31 \pm 1^\circ\text{C}$ ,  $36 \pm 1^\circ\text{C}$  and  $41 \pm 1^\circ\text{C}$ . Curves of lyophilized hydrolysates of feather-downy raw material at different temperatures are shown in Fig. 4.



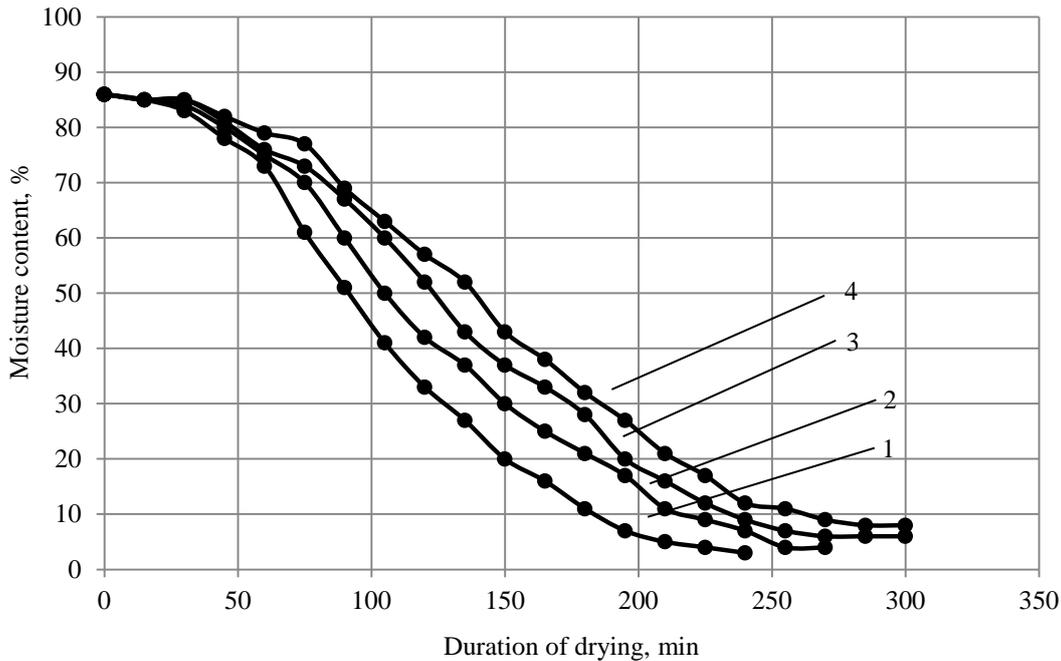
**Fig. 4.** The curves of lyophilized hydrolysates of feather-downy raw material at different temperatures: 1 –  $26 \pm 1^\circ\text{C}$ ; 2 –  $31 \pm 1^\circ\text{C}$ ; 3 –  $36 \pm 1^\circ\text{C}$ ; 4 –  $41 \pm 1^\circ\text{C}$ .

Fig. 4 shows that an increase of the heating temperature decreases drying time and moisture content in the hydrolysates at temperatures of  $26 \pm 1^\circ\text{C}$ ,  $31 \pm 1^\circ\text{C}$  and  $36 \pm 1^\circ\text{C}$ . The duration of lyophilization is 300 min; the moisture content in the final product is equal to 9.5%. Drying time at temperature  $41 \pm 1^\circ\text{C}$  is 255 min; the moisture content of lyophilized hydrolysates is 2.5%. Therefore, for further studies we selected temperature  $41 \pm 1^\circ\text{C}$  of freeze drying of hydrolysates waste from poultry industry and lyophilization duration was 255 min.

An important parameter of freeze drying is the layer

thickness. We obtained curves of lyophilization hydrolysates of feather-downy raw material at temperature  $41 \pm 1^\circ\text{C}$  and with different thickness values (Fig. 5).

Fig. 5 demonstrates that the increase in layer thickness of waste hydrolysates of feather-downy raw material leads to duration of drying and to moisture content in the final hydrolysates. Thus, when a layer thickness is 6.0, 9.0, 16.0 and 21.0 mm the length lyophilization is 240, 270, 300 and 300 minutes, and the moisture content is 2.5, 3.5, 5.5 and 7.5%, respectively. 9.0 mm is selected as an optimal drying layer thickness.



**Fig. 5.** Curves of lyophilization hydrolysates of feather-downy raw material at temperature of  $41^\circ\text{C}$  and of products with different layer thickness: 1 – 6.0 mm; 2 – 9.0 mm; 3 – 16.0 mm; 4 – 21.0 mm.

## CONCLUSION

So, we set the freeze-drying parameters of waste hydrolysates from poultry industry: the heating temperature is  $41 \pm 1^\circ\text{C}$ ; the duration of drying is 4.5 hours; the layer thickness is 9.0 mm.

Thus, the research of physical and chemical composition of feather-downy raw material and the study of freeze drying parameters allow recommending the poultry waste processing as a promising object for high-protein balanced high-quality feed.

## ACKNOWLEDGEMENTS

The work was completed under an integrated project funded by the Ministry of education and science of the Russian Federation within the framework of government resolution 218 “Organization of high-tech manufacturing high-protein fodder additives and bio-fertilizers on the basis of complex technology of processing of feather down raw materials and other low value waste of the poultry industry” (agreement № 02.G25.31.0151 from 01.12.2015 year).

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**Please cite this article in press as:** Lisitsyn A.B., Kriger O.V., and Mitrokhin P.V. Study of chemisrty and hydrolysates drying parameters of feather-downy raw material. *Foods and Raw Materials*, 2016, vol. 4, no. 4, pp.44–50. doi: 10.21179/2308-4057-2016-1-44-50.

