

Обґрунтовано способи виділення білків із овечої та коров'ячої сироваток для виробництва сиру «Урда». Встановлено однакову залежність впливу способу виділення білків як з овечої, так і з коров'ячої сироваток, на вихід білкової маси. Найбільший вихід білкової маси був при хлоркальцієвому способі виділення. Найменший вихід відзначено при тепловому способі. Вихід білкової маси з овечої сироватки у 1,60 раз вищий, ніж з коров'ячої сироватки. Отже, для виробництва альбумінового сиру «Урда» може бути використаний тепловий спосіб виділення білків

Ключові слова: овеча сироватка, коров'яча сироватка, альбуміновий сир «Урда», способи виділення білків, білкова маса

Обоснованы способы выделения белков из овечьей и коровьей сыворотки для производства сыра «Урда». Установлена одинаковая зависимость влияния способа выделения белков как из овечьей, так и с коровьей сыворотки, на выход белковой массы. Наибольший выход белковой массы был при хлоркальциевом способе выделения. Наименьший выход отмечен при тепловом способе. Выход белковой массы из овечьей сыворотки в 1,60 раз выше, чем из коровьей сыворотки. Для производства альбуминового сыра «Урда» может быть использован тепловой способ выделения белков

Ключевые слова: овечья сыворотка, коровья сыворотка, альбуминовый сыр «Урда», способы выделения белков, белковая масса

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SUBSTANTIATION OF THE METHOD OF PROTEIN EXTRACTION FROM SHEEP AND COW WHEY FOR PRODUCING THE CHEESE "URDA"

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1. Introduction

Biological and nutritional value of milk whey is predetermined by the content of nitrogen-containing components, lipids, carbohydrates, minerals, vitamins, organic acids, enzymes in it [1]. One of the most valuable components of milk whey is the serum proteins, the content of which amounts to 1.5 % [2]. This is a group of globular proteins, different by structure and properties. The main representatives of serum proteins are β -lactoglobulin, α -lactalbumin, serum albumin and immunoglobulins [3]. By their properties, in contrast to caseins, they are similar to the proteins of women's milk and, therefore, are important for the nutrition of children, the elderly, and athletes. Serum proteins contain all the essential amino acids and the ratio corresponds to a full value protein that provides the vital needs of the human body [4]. Albumin milk, albumin sour-milk cheese, albumin mousse, curds «Caucasus», etc. are made from serum proteins [5]. However, these products are not widely used in Ukraine. At the same time, in the Carpathian region, under home conditions, the cheese "Urda" from serum proteins is traditionally made from the sheep milk [6]. However, the production of sheep

milk in Ukraine is limited and quite expensive, and the volume of sheep's milk whey is limited accordingly. At the same time, a combination of whey from the sheep milk and whey from the cow milk will allow reducing the cost and increase the volumes of production of this valuable product. However, the use of such combination of raw materials predetermines certain technological features in the production of the cheese "Urda", which requires scientific research and substantiation of the technology for implementing it industrially.

2. Literature review and problem statement

There has been a significant interest lately in milk whey and the products of its processing, considering their use as functional ingredients for the food and pharmaceutical industry, which contains valuable nutrients for healthy food [7]. Whey is a valuable byproduct in the manufacture of cheeses, cottage cheese, milk protein concentrates and it can be attributed to secondary raw material resources of milk subcomplex in agroindustrial sector [8]. Milk whey is an important food product required by the human organism

to meet energy needs, for the normal flow of biochemical and microbiological processes in it. An analysis of data in the scientific literature [9] confirms a growing trend of using the milk whey and the products of its processing.

In order to produce the albumin cheese "Urda", the extraction of serum proteins from sheep whey (in line with classical technology of the product) or blended serums, proposed in article [6], is a necessary stage of the technological process. Therefore, a research into ways of extraction of proteins from cow, sheep whey and mixtures became a necessary stage.

For the extraction of serum proteins from whey, the following methods are employed: thermal, acid, acid-alkaline and chlorine-calcium. According to data in the scientific literature [10], the maximum percentage of proteins is extracted by the acid-alkaline method; slightly less – by the chloro-calcium method. Coagulation of the fraction of β -lactoglobulin is the largest at thermal denaturation. Coagulation of the fraction of α -lactalbumin is the largest at chloro-calcium method of deposition. The extraction of serum proteins by the thermal coagulation method has some advantages. First, the cost of it, both capital and operational, is insignificant compared to other techniques. Second, the method is suitable for use at dairy enterprises of any capacity. Third, the extraction of proteins by thermal coagulation is a part of technological processes for traditional products, which are made from cow whey at the enterprises of this industry. Fourth, this method is the easiest and most affordable.

It was established that serum proteins in the process of extraction by the method of thermal coagulation are exposed to the influence of high temperatures. Digestibility remains virtually identical to the natural ones [11]. That is why it appears relevant and timely to substantiate the method of protein extraction from the sheep and cow whey for manufacturing the cheese "Urda".

3. The aim and tasks of research

The aim of present study is to substantiate the way of protein extraction from sheep and cow whey for the production of the cheese "Urda".

To achieve this aim, it is necessary to solve a number of tasks:

- to investigate the effect of the method of protein extraction from cow whey on the yield and chemical composition of received protein;
- to examine the effect of the method of protein extraction from sheep whey on the yield and chemical composition of received protein;
- to explore the effect of thermal method of protein extraction from a mixture of sheep and cow whey on the yield and chemical composition of received protein.

4. Material and methods for examining the method of protein extraction from sheep and cow whey for the production of the cheese "Urda"

The research was conducted under conditions of scientific laboratory at the Department of Technology of Milk and Dairy Products of Lviv National University of Veterinary Medicine and Biotechnology named after S. Z. Gzhytskyy (Ukraine).

Raw materials for the production of the albumin cheese "Urda" were selected on the farms of SVS "Service" in the villages of Kostichany and Malynivka, Novoselytsky region, Chernivetsky oblast (Ukraine). For this purpose, dairy whey of sheep milk and dairy whey of cow milk were used.

There were four samples of cheese prepared for the research, repeated three times:

- control – albumin cheese, produced from sheep milk whey (Fig 1);
- variant 1 – albumin cheese, produced from the mixture of whey from sheep and cow milk in the ratio 1:3;
- variant 2 – albumin cheese, produced from the mixture of whey from sheep and cow milk in the ratio 1:1;
- variant 3 – albumin cheese, produced from the mixture of sheep and cow milk in the ratio 3:1 (Fig. 2).



Fig. 1. Albumin cheese "Urda" [12]



Fig. 2. Examined variants during the extraction of proteins by different ways [12]

It is possible to find more information about methodology of the research conducted in article [12].

5. Results of research into substantiation of the methods of protein extraction from sheep and cow whey for the production of the cheese "Urda"

Results of research into effect of the method of protein extraction from cow whey on the yield and chemical composition of the received protein are given in Table 1. It was established that the largest yield of protein (3.47 ± 0.10 %) is at chloro-calcium method of the protein extraction, a bit less – 3.41 ± 0.08 % at the acid-alkaline method. However, the acidity of the protein, obtained by the chloro-calcium and acid-alkaline methods, is not high – 37.2 ± 1.8 and 45.6 ± 1.4 %, respectively, which negatively affects the taste qualities of the product. The albumin cheese, produced from such protein, is savorless, the taste is not pronounced.

Table 1
Effect of the method of protein extraction from cow whey on the yield and chemical composition of bulk protein

Method of protein extraction	Yield of bulk protein, %	Mass fraction of protein in bulk protein, %	Mass fraction of moisture in bulk protein, %	Titred acidity of bulk protein, °T
Thermal	3.17±0.11	17.5±0.6	80.0±0.1	71.3±0.8
Acid	3.32±0.05	17.3±0.5	79.7±0.2	95.8±1.7
Acid-alkaline	3.41±0.08	17.4±0.4	79.8±0.3	45.6±1.4
Chlorine-calcium	3.47±0.10	17.4±0.3	80.1±0.4	37.2±1.8

When employing the acid method of protein extraction from cow whey, the yield of bulk protein is somewhat smaller than when applying the chloro-calcium and acid-alkaline methods. Albumin cheese, produced from bulk protein, received by the acid method, has an elevated titrated acidity (98...104 °T), which predetermines excessive lactic sour taste. Such cheese will not be competitive in the consumer market of Ukraine because its shelf life is very short.

Regarding the thermal method of protein extraction, the lowest yield of bulk protein was achieved (3.17±0.11 %), however the albumin cheese, received from such bulk protein was the best by its taste properties.

Results, received in the course of research, regarding the effect of the method of protein extraction from cow whey on the yield and chemical composition of bulk protein are fully consistent with data in the scientific literature [13].

There is no description in the scientific literature of the methods of protein extraction from sheep whey. Therefore, a necessary stage of the research is to determine the effect of the method of protein extraction from sheep whey on the yield and chemical composition of bulk protein. It could become a basis for the production of the albumin cheese "Urda" in future. We examined the same methods of protein extraction as in the case with cow whey, in particular thermal, acid, acid-alkaline, chlorine-calcium.

Research results are given in Table 2.

Table 2
Effect of the method of protein extraction from sheep whey on the yield and chemical composition of bulk protein

Method of protein extraction	Yield of bulk protein, %	Mass fraction of protein in bulk protein, %	Mass fraction of moisture in bulk protein, %	Titred acidity of bulk protein, °T
Thermal	5.31±0.15	18.8±1.1	80.4±0.1	90.0±2.4
Acid	5.50±0.18	18.5±1.0	79.8±0.2	115.5±1.5
Acid-alkaline	5.54±0.20	18.7±0.5	79.9±0.3	57.5±1.3
Chloro-calcium	5.56±0.15	18.8±0.6	80.5±0.4	48.0±2.5

We established the same dependence of the effect of the method of protein extraction from sheep whey on the yield of bulk protein as was the case with the cow whey. The largest

yield of bulk protein (5.56±0.15 %) was achieved when applying the chloro-calcium method of extraction. The lowest yield was achieved when employing the thermal method – 5.31±0.15 % while it was somewhat less when applying the acid-alkaline (5.54±0.20 %) and acid (5.50±0.18 %) methods. It should be noted that the yield of bulk protein from sheep whey is 1.60...1.67 times larger than that from cow whey, which is caused by the higher content of proteins in sheep whey when compared to the cow whey.

Mass fraction of proteins in the bulk protein received from sheep whey is 6.9...8.0 % larger compared with that received from the cow whey. This is due to the higher content of proteins in the original raw material – sheep whey. Thus, combining of sheep and cow whey in different proportions in the technology of the cheese "Urda" will make it possible to receive a product with the required content of proteins.

The bulk protein, obtained from sheep whey by the chloro-calcium and acid-alkaline methods, has low titrated acidity (48.0±2.5 and 57.5±1.3 °T, respectively). The cheese produced from the bulk protein received from sheep whey by the acid method has an extremely high value of acidity (115.5±1.5 °T). We also found an excessive lactic acid taste and smell. That is why, for the extraction of proteins from sheep whey, in the technology of the albumin cheese "Urda", using the thermal method is recommended, similar for the extraction of proteins from the cow whey.

For the production of the cheese "Urda", we proposed using a mixture of cow and sheep whey (variants 1...3). The next stage of experimental research was determining the yield and chemical composition of the bulk protein received by thermal method. Research results are shown in Fig. 3.

Received data indicate that in the experimental samples of bulk protein the yield is lower than in the control, due to the lower concentration of proteins in the mixtures of whey compared to the sheep whey. With an increase in the mass fraction of sheep whey in the mixture, the yield of bulk protein increases (the highest yield is for variant 3 – 4.76±0.18 %). This is explained by the highest content of proteins in this variant of the raw material. In variant 2, we also observe a rather high yield of bulk protein – 4.25±0.22 % (12.0, 12.2 % lower than in variant 1).

The mass fraction of proteins in bulk protein of variants 2 and 3 (18.4±1.2 and 18.6±1.1, respectively) is close to that in the control (18.8±1.1 %), while for variant 1 this indicator is 4.7...5.0 % lower than that in the control.

Moisture content in all examined samples of bulk protein was at the normalized level, which proves the possibility of producing the albumin cheese "Urda" from all compiled variants of whey mixtures. It should be noted that the consistency of bulk protein in variant 1 is very greasy, not characteristic for the target product, but in variants 2 and 3 matched that in the control.

The value of titrated acidity of bulk protein depends on the content of cow whey in the mixture. With an increase in the mass fraction of cow whey in the mixture, the titrated acidity of bulk protein decreases. This is due to the lower content of proteins. Bulk protein of variants 2 and 3 has acidity of 85.0±2.3 and 87.0±2.2 °T, respectively, which will make it possible (in compliance with high sanitary conditions of production) to provide for the longer shelf life of the product.

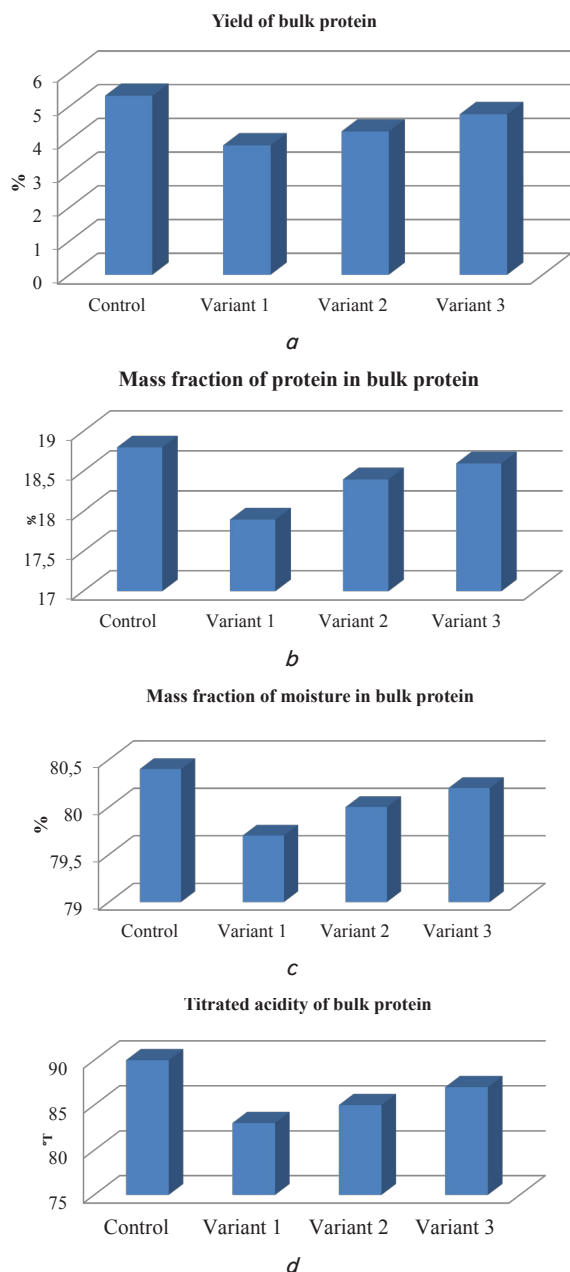


Fig. 3. Effect of the thermal method of protein extraction from the mixture of sheep and cow whey on the yield and chemical composition of bulk protein: *a* – the yield of bulk protein; *b* – mass fraction of protein in bulk protein; *c* – mass fraction of moisture in bulk protein; *d* – titrated acidity of bulk protein

6. Discussion of results of substantiation of the methods for protein extraction from sheep and cow whey for the production of the cheese “Urda”

Manufacturing the cheese “Urda” under industrial conditions may take a significant place in the range of dairy products because cheese has high biological value and rich taste. Obtaining the cheese “Urda” depends on the amount of fresh sheep milk whey. It is known that it is possible to receive 450...500 g of the cheese “Urda” from 10 dm³ sheep milk because this product is made exclusively of sheep whey. It is much more expensive compared with other kinds of

cheese. However, by using the whey from cow milk as an additional raw material, it is possible to reduce the price of the finished product significantly. As well as to use rationally the secondary raw material, which forms during production of cheese from cow and sheep milk.

After the extraction of casein from milk, a certain amount of soluble proteins, known under the name serum, are left in whey [14]. There are known ways of the extraction of serum proteins based on the physical and chemical properties [15]. There are four ways of protein extraction from milk whey widely used in production: thermal, acid, acid-alkaline and chloro-calcium [16].

Fresh whey is degreased, separated at a temperature of 35–40 °C, heated to 90–95 °C and sent to a container for coagulation. For a more complete coagulation of proteins of cheesy whey, one of the following methods (in combination with thermal treatment) is recommended:

- acid – with whey acidification, heated to 95 °C, acidic whey prepared in advance (with acidity not less than 150 °T) or concentrated hydrochloric acid to 30–35 °T (pH 4.4–4.6);

- acid-alkaline – with the subsequent deoxidation of whey, acidified to 30 °T, to pH 6–6.5 (10–15 °T) by adding a 10 % solution of sodium bicarbonate;

- chloro-calcium, which is the addition of a 18–20 % solution of calcium chloride to the whey, heated up to 90–95 °C, calculated as 1 % of the whey amount.

By organoleptic and physical-chemical requirements, bulk protein should have clean smell, fresh taste with a specific taste of albumin. In addition, it should have homogenous consistency, delicate, the presence of grains is allowed. The color should be white with yellowish tinge; the content of dry matter should not be less than 20 %; acidity – not exceed 95 °T.

We established a dependence of the effect of the methods of protein extraction from cow and sheep whey on the yield of bulk protein. The largest yield of bulk protein was achieved when using the chloro-calcium method of extraction, slightly less when employing the acid-alkaline and acid methods. The lowest yield is observed when applying the thermal method.

While studying the thermal coagulation of proteins from sheep whey, it was found that the maximal extraction of proteins is observed in the isoelectric point of albumin fraction of serum proteins at acidity 37 °T and pH 4.55. Optimal temperature in this case is 95 °C; the duration of holding is not less than 25 min. That is why it is recommended in the technology of the albumin cheese “Urda” to use a method of thermal coagulation for the extraction of serum proteins.

Thus, for the production of the albumin cheese “Urda” it is necessary to use the thermal method of protein extraction from a mixture of sheep and cow whey in the ratio of 1:1 or 3:1.

7. Conclusions

1. We studied the effect of different methods of protein extraction from cow whey on the yield and chemical composition of bulk protein. It was established that the largest yield of bulk protein was received when using the chloro-calcium method. The albumin cheese, produced from bulk protein, received by the acid technique, has an elevated titrated acidity (98...104 °T). The lowest yield of bulk protein was observed while using the thermal method of protein extraction, but the albumin cheese, received from such bulk protein, was the best by its taste.

2. We examined the effect of different methods of protein extraction from sheep whey on the yield and chemical composition of bulk protein. It was found that the largest yield of bulk protein was achieved when employing the chloro-calcium technique of extraction; somewhat less – when applying the acid-alkaline and acid methods. The lowest yield of bulk protein was registered while using the thermal method when it was $5.31 \pm 0.15\%$, respectively. The bulk protein, received from sheep whey by the chloro-calcium and acid-alkaline techniques, had low titrated acidity. The cheese, produced from bulk protein

obtained from sheep whey by the acid method, has extremely high values of acidity (115.5 ± 1.5 °T). Therefore, for the extraction of proteins from sheep whey, we recommend using the thermal method in the technology of the albumin cheese “Urda”.

3. We scientifically substantiated applying the thermal method of protein extraction from a mixture of sheep and cow whey to yield and chemical composition of bulk protein. It was found that for the production of the albumin cheese “Urda”, the thermal method of protein extraction from the serum mixtures can be employed.

References

1. Lasik, A. The fermentation dynamics of sheep milk with increased proportion of whey proteins [Text] / A. Lasik, J. Pikul, R. Dank, D. Cais-Sokolinska // *Acta Scientiarum Polonorum: Technologia Alimentaria*. – 2011. – Vol. 10, Issue 2. – P. 155–163.
2. Bilyk, O. Ya. Doslidzhennya biolohichnoyi tsinnosti syru urda [Text] / O. Ya. Bilyk, H. V. Dronyk // *Kharchova nauka i tekhnolohiya*. – 2014. – Issue 4. – P. 24–27.
3. El-Hatmi, H. E.-H. Comparison of composition and whey protein fractions of human, camel, donkey, goat and cow milk [Text] / H. E.-H. El-Hatmi // *Mljekarstvo*. – 2015. – Vol. 65, Issue 3. – P. 159–167. doi: 10.15567/mljekarstvo.2015.0302
4. Tsiryak, O. Y. Sklad ta vlastyvoli ovechoho moloka otrymanoho vid ovets' riznykh porid Bukovyny [Text] / O. Y. Tsiryak, I. M. Slyvka, O. Ya. Bilyk, O. B. Lesyk // *Sil's'kyi gospodar*. – 2013. – Issue 11-12. – P. 2–5.
5. Bilyk, O. Developing technologies for producing urda albumin cheese [Text] / O. Bilyk, G. Dronyk // *Eastern-European Journal of Enterprise Technologies*. – 2014. – Vol. 3, Issue 10 (69). – P. 49–53. doi: 10.15587/1729-4061.2014.25255
6. Hramcov, A. G. Innovacionnye priority ispol'zovaniya molochnoy syvorotki na principah logistiki bezothodnoy tekhnologii [Text] / A. G. Hramcov, I. A. Evdokimov, P. G. Nesterenko // *Molochnaya promyshlennost'*. – 2008. – Issue 11. – P. 28–32.
7. Ostroumova, T. L. Belkovyy produkt iz vtorichnogo molochnogo syr'ya [Text] / T. L. Ostroumova, I. G. Kulipchik, N. A. Panasenko // *Molochnaya promyshlennost'*. – 2007. – Issue 2. – P. 54.
8. Yukalo, A. V. Bilky kazeyinovoho kompleksu moloka korovy (Bos taurus) yak poperednyky biolohichno aktyvnykh peptydiv [Text] / A. V. Yukalo, L. A. Storozh, V. H. Yukalo // *Biotekhnolohiya*. – 2012. – Vol. 5, Issue 4. – P. 21–33.
9. Tkachenko, N. A. Fraktsiynny sklad bilkiv u pastakh dlya dytyachoho kharchuvannya [Text] / N. A. Tkachenko, Yu. S. Ukrayintseva, A. S. Avershyna // *Naukovyy visnyk LNUVMBT*. – 2015. – Vol. 17, Issue 4 (64). – P. 164–172.
10. Yukalo, V. H. Vyznachennya umov otrymannya pryrodnykh bioaktyvnykh kazeyinovykh fosfopeptydiv [Text] / V. H. Yukalo, L. A. Storozh, M. O. Shtokalo // *Naukovyy visnyk LNUVMBT imeni S. Z. Hzhys'koho*. – 2014. – Vol. 16, Issue 3 (60). – P. 192–200.
11. Volkova, T. A. Al'buminnaya massa i pasty na ee osnove [Text] / T. A. Volkova, E. F. Kravchenko // *Pererabotka moloka*. – 2008. – Issue 8. – P. 38–39.
12. Bilyk, O. Study of the different ways of proteins extraction from sheep and cow whey for “urda” cheese production [Text] / O. Bilyk, N. Slyvka, B. Gutyj, H. Dronyk, O. Sukhorska // *EUREKA: Life Sciences*. – 2017. – Issue 3. – P. 3–8. doi: 10.21303/2504-5695.2017.00333
13. Ha, M. In-Depth Characterization of Sheep (*Ovis aries*) Milk Whey Proteome and Comparison with Cow (*Bos taurus*) [Text] / M. Ha, M. Sabherwal, E. Duncan, S. Stevens, P. Stockwell, M. McConnell et. al. // *PLOS ONE*. – 2015. – Vol. 10, Issue 10. – P. e0139774. doi: 10.1371/journal.pone.0139774
14. Fong, B. Y. Fractionation of bovine whey proteins and characterisation by proteomic techniques [Text] / B. Y. Fong, C. S. Norris, K. P. Palmano // *International Dairy Journal*. – 2008. – Vol. 18, Issue 1. – P. 23–46. doi: 10.1016/j.idairyj.2007.06.005
15. El-Agamy, E. I. The challenge of cow milk protein allergy [Text] / E. I. El-Agamy // *Small Ruminant Research*. – 2007. – Vol. 68, Issue 1-2. – P. 64–72. doi: 10.1016/j.smallrumres.2006.09.016
16. Gutyj, B. The influence of cryopowder “garbuz” on the technology of curds of different fat content [Text] / B. Gutyj, Y. Hachak, J. Vavrysevych, V. Nagovska // *Eastern-European Journal of Enterprise Technologies*. – 2017. – Vol. 2, Issue 10 (86). – P. 20–24. doi: 10.15587/1729-4061.2017.98194