

## SECTION X. BIOLOGY AND BIOTECHNOLOGY

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### INDIVIDUAL FEATURES OF THE BOVINE COLOSTRUM PROTEOME

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**Ivanov Ievgen**

PhD, Associate professor, Department of Molecular Biology and Biotechnology  
*V. N. Karazin Kharkiv National University*

**Kozheshkurt Valentyn**

PhD, Senior lecturer, Department of physical and biomedical electronics and  
integrated information technology  
*V. N. Karazin Kharkiv National University*

*UKRAINE*

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One of the urgent tasks of biotechnology is obtaining of biologically active compounds of natural origin. Colostrum is a promising object for solving such problems, since it contains a large number of various natural compounds. Bovine colostrum proteins are of particular interest in this respect. As is known, most farm animals (calves, foals, piglets, camels) are born agammaglobulinemic [1] and receive passive immunity due to the first intake of colostrum, which is extremely rich not only in immunoglobulins, but also in various protein factors regulating both the immune system and the general metabolism of the body. Of great interest as factors regulating metabolism are low molecular weight fractions of proteins and peptides, which include the so-called transfer factors [2]. These are peptides with a molecular weight from 3500 Da to 6000 Da, which are inherent in all animals, are not species specific, they carry antigenic fragments and stimulate the activity of the immune system [3]. As is known, a cow gives from 5 to 10 liters of colostrum per milking, which significantly exceeds the needs of a calf [4], and the remaining amounts of colostrum can be used for industrial processing in order to obtain various biologically active compounds. Despite the fact that according to Chaudhary et.al. 2016 colostrum is "liquid gold" and has long been used in traditional medicine, it has not yet found proper use in modern industrial processing. This is due to a number of features and unresolved problems: the difficulty of storage and the peculiarity of processing; incomplete knowledge of its effect on the functional characteristics of an adult organism; instability and high individual variability of composition. A number of experts believe that the potential of biologically active compounds of colostrum, including its low molecular weight components, is not fully used [5]. In this regard, a part of the proteomic composition was studied, which includes proteins and peptides with a molecular weight from 4500 kDa to 9500 kDa, in different cows kept in the same farm, receiving the same feed, of the same breed and of the same age. To do this, fat was removed from fresh colostrum by repeated sequential centrifugation at 3000 g for 20 min, followed by casein precipitation at the isoelectric point. High molecular weight fractions of proteins were removed by membrane filtration; for this, filters with a pore diameter of 10  $\mu\text{m}$  were used. The analysis of the obtained samples

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was carried out on a mass spectrometer Autoflex II LRF 20 “Bruker Daltonics” (Germany).

It was found that the cow named “Aurora” had 27 proteins in the studied range of proteins (Fig. 1a) and the cow “Barynia”, respectively, 32 proteins (Fig. 1b). It should be noted that all protein fractions had a pronounced individual character, in the sense that these proteins differed not only in molecular weight, but also in the number of similar proteins obtained from different cows (Fig. 1a, b).

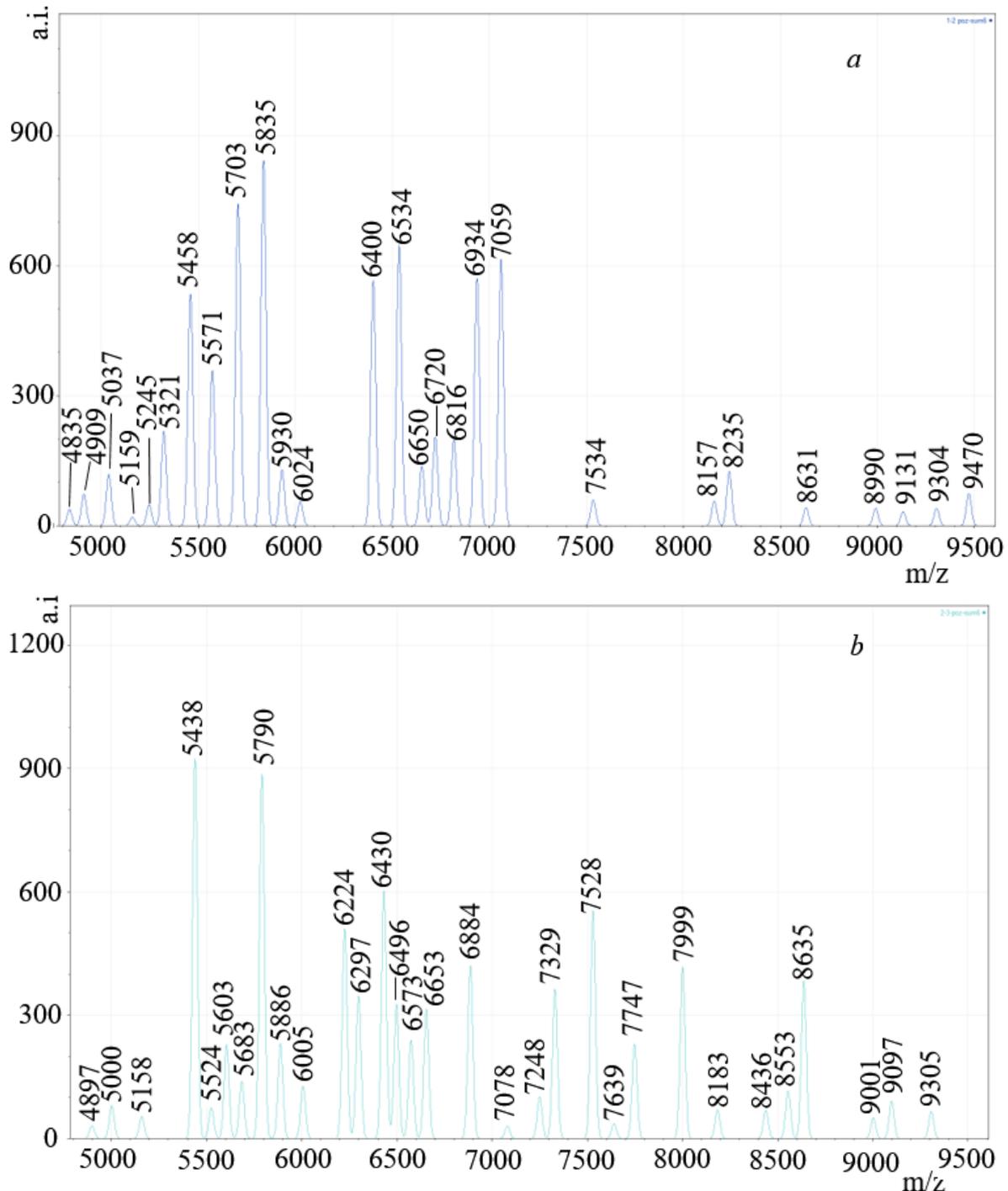


Fig. 1. Proteome of low molecular weight fractions of colostrum proteins according to the results of mass spectroscopy: colostrum obtained from the cow “Aurora” (a); colostrum obtained from “Barynia” (b). Representative spectra of proteins are shown.

Attention should be paid to the fact that proteins with a molecular weight, which are characteristic of the transfer factor, there are up to 12 fractions in “Aurora”, and 10 proteins were detected in this range in “Barynia” (Fig. 1a, b). It is known that the components of the transfer factor are formed in the body in the presence of certain pathogens, and in this sense, they are inducible proteins. Therefore, individual differences in the composition of the transfer factor can be explained by the fact that for each organism there are always different options for “encounter-infection” with different pathogens and, as a result, they always have an individual form of spectrum of inducible proteins.

We believe that in the industrial processing of colostrum, in particular, in the production of low molecular weight components that have the functions of a transfer factor, it is necessary to use a “selective-integrative” approach.

The essence of the approach is that at the first stage, colostrum of the first milking is taken from cows in the same farm, of the same age, of the same breed and similar in physiological characteristics. At the second stage, colostrum obtained from at least 10 cows is combined, which is processed as a separate batch immediately after it is received, and if this is not possible, it is stored at a temperature of -30 C until a complete batch is formed with subsequent processing.

The important issue in obtaining biotechnologically active compounds is the standardization and quality assessment of complex, multicomponent substances. Taking into account the fact that transfer factor molecules are relatively small and may have a charge, it can be assumed that such a biophysical method as the measurement of electrical conductivity can be a good approach to solving the issue of standardization of such multicomponent mixtures. It was found that the electrical conductivity of low molecular weight components of colostrum, which include molecules of transfer factors, also had a pronounced individual character (Fig. 2).

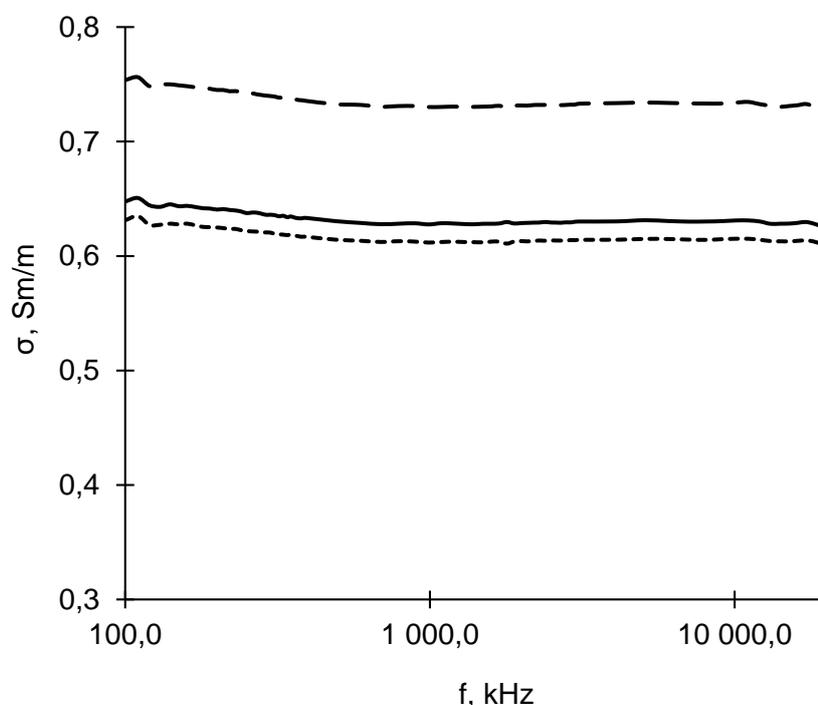


Fig. 2. Electrical conductivity of low molecular weight components of colostrum obtained from “Aurora” (—), “Barynia” (— —) and “Mukha” (----) in the frequency range of 100 kHz – 100 MHz.

Thus, the electrical conductivity of the low molecular weight fractions of proteins obtained from “Aurora” was 0.65 S/m and remained unchanged with increasing current frequency, for the “Barynia” it was significantly higher (0.78 S/m) and for “Mukha” it was similar to that of “Aurora” (Fig. 2). After combining the fractions of low molecular weight components of colostrum, an average electrical conductivity characteristic was obtained, which characterized the resulting batch of low molecular weight components of colostrum.

The results obtained allow us to conclude that the measurement of the electrical conductivity of the obtained colostrum fractions can be used as a fast and effective integrative method for assessing the quality of colostrum.

Consequently, the low molecular weight fraction of colostrum, which includes transfer factor, is characterized by a pronounced individual composition, which has a similar individual character with electrical conductivity. A “selective-integrative” method for the standardization of complex multicomponent biotechnological substances in the preparation of biologically active compounds is proposed.

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