The study of amino acid composition and oxidation processes in meat of Aberdeen-Angus bull-calves in chilled and supercooled state

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Abstract. Improving the safety and quality of meat raw materials from animals of special-purpose beef cattle helps to meet the demand for foods at high nutritional value. In the Russian Federation, the Aberdeen-Angus breed is one of the leading beef cattle, from which high quality marbled beef is obtained. Investigations have been carried out to study the qualitative indices of beef from this breed at storage under temperature conditions of cooling medium of 1 ± 0.5 ºС and minus 2.5 ± 0.5 ºС in cold chambers during 12 and 24 days. A decrease in the content of free amino acids in meat during storage was noted. It was found that after 24 days of storage, the biological value of the product (ratio of essential amino acids to nonessential ones) at minus 2.5±0.5 ºС was 1.14 times higher as compared with storage at 1.0±0.5 ºС temperature. After 12 days of storage at 1.0±0.5 ºС temperature, the values of acid and peroxide numbers increased about 3 times in comparison with the initial values, while at minus 2.5±0.5 ºС two times only. After 24 days of storage, the increase of acid number in chilled meat was 42% higher than in super-cooled meat; the values of the peroxide number in meat at 1.0±0.5 ºС was 1.3 times higher than in meat at minus 2.5±0.5 ºС. The researches resulted in recommendations of the temperature conditions providing the storage of super-cooled vacuum-packed meat without phase transition of water into ice as well as ensuring better preservation of biologically relevant components and nutritional value of meat.

1. Introduction
A significant expansion of production of domestic foods and ensuring their safety are the major concerns of the state policy in the field of healthy nutrition of the population of Russia [1, 2]. Therefore, the task of maximal preservation of food raw materials and food products at all stages of their production, storage, transportation and realization is of primary importance [3]. It is known that meat has a high nutritive value and plays an important role in a man living as a source of proteins of full value because it contains all essential amino acids as well as fats, vitamins, mineral and extractive substances easy to digestion [4].
At present, the Aberdeen-Angus strain of cattle, bred in the Russian Federation, is one of the leading beef cattle. These strains of cattle are famous not only for its productivity but for beef high quality [5, 6].

Cooling is one of the methods of meat and meat products preservation that allows stopping microbiological deterioration and keeping the structure and consumer properties with their minimal changes [7].

The selection of the optimal temperature conditions to be applied during meat refrigeration processing and cold storage is very important. In the case of the odd choice or non-compliance with the temperature parameters of technological processes, the growth of the pathogenic microflora occurs, which leads to a decrease of quality and safety of products [8].

The shelf life of meat and meat products may be prolonged by increasing their resistance to the microorganism activity, which is observed when applying near-cryoscopic temperatures [9]. For example, refrigeration processing using the super-cooling technology and providing meat temperature decrease by (1–2) °C below the cryoscopic temperature that results in a partial ice formation, together with further storage at subcryoscopic temperatures can ensure the preservation of meat and meat product quality, reducing mass losses. In this case, the degree of transition of water into ice is the dominant parameter determining the quality of a super-cooled product, since the ice content in the product of more than 30% provokes a significant exudation [10].

One of the promising trends is cold meat storage at the temperature below the cryoscopic one without phase transition of water into ice (effect of super-cooling). Super-cooled foods of animal origin have the same quality indices as chilled ones, but they are better as regards some indices, besides, the shelf life may be prolonged [11]. Therefore, there is a need for obtaining objective data on the change of meat quality indices depending on the species and breed characteristics during storage in a super-cooled state.

The purpose of these investigations was to study the impact of temperature parameters and duration of storage of vacuum-packed meat from Aberdeen Angus bull-calves on changes in the amino acid composition of proteins, peroxide and acid numbers of fats.

2. Materials and methods

The object of this study was vacuum-packed meat (muscle L. Dorsi) weighing (0.2 - 0.3) kg from Aberdeen Angus bull-calves.

The cold chambers that could provide automatic maintaining of different temperature conditions were used to store the vacuum-packed meat. The vacuum-packed meat samples were stored during 24 days at two temperature conditions of cooling medium: 1.0±0.5 °C (chilled state) and minus 2.5±0.5 °C (super-cooled state). The air and vacuum-packed meat temperatures were recorded by electronic recorders designed for measurement, registration and storage of data, with further the data transfer to a personal computer and plotting diagrams (DV2TSM air temperature and humidity recorder at ±0.3 °C absolute measurement error; iBDL-R thermograph at ±0.5 °C absolute measurement error).

During storage the next qualitative indices of initial samples after 12 and 24 days of storage were defined: total number of free amino acids of proteins according to GOST 34132-2017; changes of peroxide and acid numbers of fats according to GOST R 55480-2013 and GOST 34118-2017. The studies were performed at the Testing centre of the V.M. Gorbatov Federal Scientific Center of Food Systems RAS.

Statistical processing of the data obtained was carried out according to the methodological instructions on reporting measurement results using Microsoft Excel operating system [12]. The difference veracity was adopted at B1 = 0.95 reliability threshold (significance level P<0.05).

3. Results and discussion

Figures 1 and 2 show the changes in the air and product temperatures depending on storage duration and temperature regime is chosen. During studies, the values of air temperature in cold chambers were
measured every 10 minutes using the DV2TSM recorder and every 5 minutes in the meat centre using iBDL-R logger.

![Figure 1](image1.png)

**Figure 1.** Thermogram of chilled meat storage conditions

After data processing the average air temperature during chilled meat storage was 0.87 °C, and the average temperature in the vacuum-packed meat centre was 0.92 °C at standard deviations ±0.16 °C and ±0.05 °C correspondingly (Figure 1).

![Figure 2](image2.png)

**Figure 2.** Thermogram of meat storage in a super-cooled state

The air average temperature during storage in a super-cooled state was minus 2.71 °C, but the temperature in the vacuum-packed meat centre was 2.62 °C. Standard deviations were ±0.13 °C and ±0.02 °C, respectively (Figure 2).

The thermograms in Figures 1 and 2 demonstrate the product temperature stability throughout the experiment that is an essential condition to preserve quality and increase chilled meat shelf life [9].

The changes in the total number of amino acids during meat storage in the chilled and super-cooled state are given in Table 1.
The analysis of the data cited in Table 1 indicates a common tendency in decreasing the number of free amino acids during storage of meat both in chilled and super-cooled states as compared with fresh meat.

Table 1. Changes of the total number of amino acids during vacuum-packed meat storage under different temperature conditions

| №  | Amino acids | Content (g/100g of product) |
|----|-------------|-----------------------------|
|    |             | Fresh meat | Free amino acids after 12 days of storage at a temperature | Free amino acids after 24 days of storage at a temperature |
|    |             | Common amino acids | Free amino acids | minus 2.5±0.5 ºC | 1.0±0.5 ºC | minus 2.5±0.5 ºC | 1.0±0.5 ºC |
| 1  | Asparaginic | 2.16 ±0.06 | 0.0103 ±0.0003 | 0.0179 ±0.0005 | 0.128 ±0.0004 | 0.0136 ±0.0004 | 0.0091 ±0.0003 |
| 2  | Glutaminic | 3.27 ±0.10 | 0.0446 ±0.0013 | 0.0808 ±0.0024 | 0.0184 ±0.0018 | 0.0120 ±0.0017 | 0.0114 ±0.0014 |
| 3  | Serine     | 0.84 ±0.03 | 0.0062 ±0.0000 | 0.0184 ±0.0006 | 0.0120 ±0.0004 | 0.0117 ±0.0004 | 0.0114 ±0.0003 |
| 4  | Histidine  | 0.76 ±0.02 | 0.0007 ±0.0000 | 0.0099 ±0.0003 | 0.0093 ±0.0001 | 0.0112 ±0.0003 | 0.0084 ±0.0002 |
| 5  | Glycine    | 1.32 ±0.04 | 0.0315 ±0.0000 | 0.0380 ±0.0003 | 0.0377 ±0.0003 | 0.0382 ±0.0003 | 0.0336 ±0.0003 |
| 6  | Threonine  | 1.00 ±0.03 | 0.0058 ±0.0000 | 0.0011 ±0.0000 | 0.0030 ±0.0001 | 0.0062 ±0.0001 | 0.0028 ±0.0002 |
| 7  | Arginine   | 1.32 ±0.04 | 0.0893 ±0.0000 | 0.0694 ±0.0000 | 0.0689 ±0.0000 | 0.0738 ±0.0000 | 0.1296 ±0.0000 |
| 8  | Alanine    | 1.32 ±0.04 | 0.0536 ±0.0000 | 0.0698 ±0.0000 | 0.0760 ±0.0000 | 0.0841 ±0.0000 | 0.0753 ±0.0000 |
| 9  | Tyrosine   | 0.70 ±0.02 | 0.0113 ±0.0000 | 0.0018 ±0.0000 | 0.0024 ±0.0000 | 0.0099 ±0.0000 | 0.0083 ±0.0000 |
| 10 | Cystine    | 0.29 ±0.01 | 0.0136 ±0.0000 | 0.0248 ±0.0000 | 0.0249 ±0.0000 | 0.0281 ±0.0000 | 0.0167 ±0.0000 |
| 11 | Valine     | 1.13 ±0.03 | 0.0090 ±0.0000 | 0.0174 ±0.0000 | 0.0195 ±0.0000 | 0.0179 ±0.0000 | 0.0182 ±0.0000 |
| 12 | Methionine | 0.59 ±0.02 | 0.0003 ±0.0000 | 0.0000 ±0.0000 | 0.0005 ±0.0000 | 0.0005 ±0.0000 | 0.0005 ±0.0000 |
| 13 | Phenylalanine | 0.88 ±0.03 | 0.0143 ±0.0000 | 0.0147 ±0.0000 | 0.0205 ±0.0000 | 0.0185 ±0.0000 | 0.0094 ±0.0000 |
| 14 | Isoleucine | 1.01 ±0.03 | 0.0151 ±0.0000 | 0.0527 ±0.0000 | 0.0447 ±0.0000 | 0.0488 ±0.0000 | 0.0586 ±0.0000 |
| 15 | Leucine    | 1.77 ±0.05 | 0.0302 ±0.0000 | 0.0413 ±0.0000 | 0.0466 ±0.0000 | 0.0487 ±0.0000 | 0.0586 ±0.0000 |
| 16 | Lysine     | 1.72 ±0.05 | 0.1926 ±0.0000 | 0.0601 ±0.0000 | 0.0518 ±0.0000 | 0.0495 ±0.0000 | 0.0400 ±0.0000 |
| 17 | Proline    | 1.08 ±0.03 | 0.0051 ±0.0000 | 0.0049 ±0.0000 | 0.0048 ±0.0000 | 0.0052 ±0.0000 | 0.0059 ±0.0000 |
|    | Total      | 21.16 ±0.63 | 0.5450 ±0.0000 | 0.5344 ±0.0000 | 0.5110 ±0.0000 | 0.5411 ±0.0000 | 0.5268 ±0.0000 |
It was defined that during the first 12 days of the storage there was a decrease of the content of free amino acids by 0.034 g/100 g at cooling medium temperature of 1.0±0.5 °C and by 0.0106 g/100 g at a temperature of minus 2.5±0.5 °C. At the same time, throughout all storage period of 24 days, the decrease was by only 0.0182 g/100 g (chilled meat) and by 0.0039 g/100 g (super-cooled meat). Hence, the content of free amino acids decreased by the end of 12 days of storage and increased slightly by the end of 24 days of storage.

After 24 days of storage at cooling medium temperature of 1.0±0.5 °C, an increase of the content of the next nonessential amino acids was marked: arginine by 0.0403 g/100 g and alanine by 0.0217 g/100 g, and the essential ones: isoleucine by 0.0435 g/100 g and leucine by 0.0166 g/100 g. At a cooling temperature of minus 2.5±0.5 °C the increase of the content of the following nonessential acids was established: glutamic acid by 0.0180 g/100 g, alanine by 0.0305 g/100 g, cystine by 0.0145 g/100 g; and the essential ones: histidine by 0.0105 g/100 g, isoleucine by 0.0337 g/100 g and leucine by 0.0185 g/100 g.

The results obtained may be explained by the change of the amino acid content due to fermentative processes [4]. The process of amino acid disintegration and decrease of their content in meat is catalyzed by the activity of oxidases and decarboxylases, which is the highest in fresh meat as well as during the initial period of its refrigeration processing. Proteolysis contributes to the increase of the cathepsin activity, that’s why the increase of amino acid content occurs later during cold storage, as far as they are released from disintegrating lysozymes. The initial decrease and further increase of the free amino acid content in meat under different conditions of storage may be explained by the difference of speeds of these fermentative processes.

So, the total content of amino acids is determined by two processes: proteolysis, leading to amino acid accumulation, and disintegration that decreases their content. It was found that after 24 days of storage the biological value of the product (ratio of essential amino acids to nonessential ones) at minus 2.5±0.5 °C temperature was 1.4 times higher than when storing at 1.0±0.5 °C temperature.

| Shelf life  | Storage temperature, °C | Indices | Acid number, (Mg KOH)/g | Peroxide number, (mmol act. oxygen) / kg |
|------------|-------------------------|---------|------------------------|---------------------------------------|
| Initial materials raw | - | 0.4 | 0.3 |
| 12 days | 1.0±0.5 °C | 1.3 | 0.9 |
| minus 2.5±0.5 °C | 0.8 | 0.6 |
| 24 days | 1.0±0.5 °C | 2.7 | 1.7 |
| minus 2.5±0.5 °C | 1.9 | 1.3 |

The data obtained and showed in Table 2 indicate the hydrolytic and oxidation changes during storage. It was established that the increase of the fat acid number in samples, resulted from the hydrolysis of triglycerides, was accompanied by the formation of free fatty acids. So, after 12 days of storage in the chamber at 1.0±0.5 °C, the acid number increased 3.25 times as compared with the initial values, while at minus 2.5±0.5 °C temperature storage it increased two times only. After 24 days of storage, the increase of the acid number in the chilled meat samples was 42% higher than in the super-cooled meat ones.

Also during storage, the increase of the peroxide number due to the fat oxidation was observed. After 12 days of storage, it increased 3 times at 1.0±0.5 °C temperature and 2 times at minus 2.5±0.5 °C. After 24 days of storage, this index reached a value of 1.7 (mmol act. oxygen)/kg for chilled meat, which was 0.4 (mmol act.oxygen)/kg higher than the value for super-cooled meat.

The rate of acid and peroxide number increase was 0.096 (mg KOH)/g and 0.058 (mmol act. oxygen)/kg on the average, and 0.063 (mg KOH)g and 0.042 (mmol act.oxygen)/kg per day at minus 2.5±0.5 °C temperature of storage respectively.
4. Conclusion

Based on the biochemical studies of the vacuum-packed meat samples from Aberdeen-Angus bull-calves during storage of 24 days in cold chambers at 1.0±0.5 °C and minus 2.5±0.5 °C temperatures, it was established that the decrease of the medium cooling temperature below the cryoscopic one but not higher a maximum permissible super-cooling temperature, at which the crystallization of intramuscular moisture occurs, allowed retarding hydrolytic and oxidation changes in lipids, as well as providing the best quality preservation of vacuum-packed meat as regards its amino acid composition.

Compared to chilled meat, the storage of super-cooled vacuum-packed meat of Aberdeen-Angus breed provided the best preservation of its quality, as regards the amino acid composition, 1.03 times, acid and peroxide numbers 1.4 and 1.3 times respectively, on condition that the given temperature parameters of storage were strictly observed.

The results obtained may be used to improve cold storage conditions of vacuum-packed meat of both Aberdeen-Angus breed and other beef cattle.

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