



Received: 20.9.2022
Revised: 30.10.2022
Accepted: 1.11.2022
Published: 29.11.2022

Potravinarstvo Slovak Journal of Food Sciences
vol. 16, 2022, p. 810-823
<https://doi.org/10.5219/1794>
ISSN: 1337-0960 online
www.potravinarstvo.com
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Processing of beef rumen with ultrasonic waves

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ABSTRACT

The article discusses the main trends in processing animal products, the development of technologies to improve their quality and technologies to preserve the quality indicators of the product over time. A review of the effects of ultrasound treatment on beef rumen is presented, and the main directions of ultrasound application are determined. The advantages of ultrasonic processing and its influence on the characteristics of raw meat were researched. The modes and parameters (frequency, intensity and duration) of ultrasound treatment of muscle tissue were established based on the results. This study evaluated the effect of ultrasound treatment on beef rumen's physical, microstructural and organoleptic characteristics. The physicochemical, mineral, microbiological, vitamin and amino acid composition of beef rumen and reticulum were also studied. Based on the results of the presented review, it can be concluded that the development of technologies for processing beef rumen with ultrasound is of potential interest. The optimal parameters are 400 and 600 W/m², with a frequency of 40 kHz, for 50-60 minutes.

Keywords: resource conservation, offal, beef rumen, ultrasound treatment, cutting efforts

INTRODUCTION

The increase in production rates and output volumes of the meat industry is inextricably linked with the improvement and development of new resource-saving technologies and the integrated use of livestock raw materials, including the processing of offal. Offal is an interesting food resource that is not always fully utilized.

There is a need to conduct studies on the quality of offal obtained during animal slaughter to establish indicators like organoleptic, physicochemical and morphological properties. Also, freshness is an important indicator that must be considered in considering using beef rumen for food production. Food offal is of great importance in supplying meat products to the population, being an additional resource of protein nutrition. This product presents 10-12% of the mass of meat on bones and occupies an important place in the country's food balance [1].

The tradition of eating rumen has ancient roots. In various parts of the world, it is possible to find traditional rumen dishes. Today, ready pre-prepared and processed rumen can be purchased in many countries.

The beef stomach consists of four chambers: the rumen is the first chamber covered with small 'hairs', the reticulum is the second chamber of the stomach, which has a lining similar to a honeycomb, and the omasum is the third chamber, covered with large and thin leaflets, like the pages of a book. The last chamber is the abomasum, which is very much appreciated as a delicacy worldwide. All these chambers are used to prepare delicious dishes that have become traditional in many countries and are known worldwide [2].

The chemical composition of beef rumen contains vitamins B1, B2, B12, H and PP and minerals necessary for the human body: potassium, calcium, magnesium, zinc, selenium, copper, manganese, iron, phosphorus and sodium. The rumen is rich in iron (100 grams of the finished product contains 16.7% of the daily value of this substance), sulphur (14.8% of the daily requirement), potassium (13%), iodine (4.7%) and sodium (5%). In addition, it contains calcium, phosphorus, zinc, a small amount of manganese, copper and selenium [3], [4].

One of the most effective ways to use such raw materials is the pretreatment of beef rumen with ultrasonic waves.

Ultrasound is acoustic energy, considered a mechanical, non-ionizing, environmentally friendly energy with immense potential for use in high-quality food production processes. Ultrasound causes changes in the physical, chemical and functional properties of food products. This, therefore, can affect the quality of various food systems, increasing their productivity and efficiency [5], [6], [7].

Ultrasound is classified as non-thermal processing and is used in the food industry in the frequency range from 20 kHz to 1 MHz. Currently, ultrasound is widely used in the meat industry to improve procedures such as meat tenderizing, mass transfer emulsification, marinating, freezing, homogenization, crystallization, drying and inactivation of microorganisms. In addition, combining ultrasonic energy with a disinfectant has a synergistic effect on reducing the number of microbes.

Ultrasound is used in 'Green Food Production' to produce high-quality and safe food products. Ultrasound is a modern technology that improves meat products' biochemical and functional characteristics.

The use of ultrasound in the food industry, including with meat, can significantly accelerate several technological processes, increase the utilization rate of raw materials, and improve the quality and safety of products [8], [9], [10].

In this regard, it is of scientific and practical interest to conduct special studies on the use of beef rumen in the quality of meat products and the rational use of meat raw materials.

The relevance of this study lies in the fact that being an additional resource primarily for protein nutrition, beef rumen has a high nutritional value and is used both in the form of natural products and as an ingredient for the manufacture of various products.

Scientific Hypothesis

Theoretically, we assume that ultrasonic exposure's modes and parameters (frequency, intensity and duration) significantly affect the quality of the beef rumen, reticulum, abomasum, omasum and caecum. Consequently, their shelf life is increased. This paper includes the results of a study of ultrasonic treatment for qualitative indicators of the rumen, reticulum, abomasum, omasum and caecum of beef, which was our goal.

MATERIAL AND METHODOLOGY

Samples

The objects of the study were: the beef rumen, reticulum, abomasum, omasum and caecum. For the experiment, the rumen of cattle and other samples were purchased from the company 'Sozak et'.

Chemicals

All reagents were of analytical grade and were purchased from Laborfarm (Kazakhstan) and Sigma Aldrich (USA).

Animals and Biological Material

The rumen of cattle was used for the experiment.

Instruments

We used AquaLab 4TE water activity analyzer (Decagon Devices Inc, USA), Ultrasonic homogenizer Bandelin SONOPULS HD 2200.2 (Bandelin, Germany), automatic amino acid analyzer AAA-881 (Mikrotechna, Czech Republic), atomic emission spectrometer ICP-OES (Spectro, Boschstr, Germany), MX-50 weight moisture meter (LTD A&D Co, Japan).

Laboratory Methods

All analyses were carried out in an accredited laboratory of the Almaty Technological University and Kazakh Research Institute of Processing and Food Industry LLP.

The moisture binding capacity (WCC): The moisture binding capacity was determined by the method proposed by Grau and Hamm [11]. A sample with the same weight was placed between discs of filter paper. A press (a load of 1 kg) was placed on top for 10 minutes.

Determination of the cutting force by Warner Bratzler: To determine the Warner-Bratzler cutting force, the samples were pre-cut into a tube-shape with a diameter of 1.27 mm. The measurement was carried out on a Brookfield texture analyzer. Samples were measured at a temperature of 10-12 °C.

Water activity: Water activity was determined using the AquaLab 4TE water activity analyzer (Decagon Devices Inc). The samples were pre-crushed in a blender (KangQi, Singapore), weighed and evenly distributed over the device's cup.

Determination of humidity: The moisture content in the rumen was determined on the MX-50 weight moisture meter. All samples for determining the moisture content were weighed by 5 g and evenly distributed inside the device cup.

Determination of the rumen pH: The rumen's active acidity (pH) was determined by the potentiometric method. The crushed sample was mixed twice with distilled water in a ratio of 1:10, followed by stirring on a magnetic stirrer for 30 minutes. The pH after extraction was determined on a HI 99163 device (Hanna Instruments Inc.).

Determining collagen content in raw materials: The determination of collagen content in raw materials was quantified according to GOST 33692-2015 [12]. The method is based on the release of oxyproline in the acid hydrolysate of the sample, and neutralization of the hydrolysate. Its oxidation with chloramine T, with the formation of a red compound and photometric measurement of optical density at a wavelength of (558 ± 2) nm and subsequent conversion to collagen.

Microbiological indicators: All studies were conducted in a microbiological box under aseptic conditions. The number of mesophilic aerobic and facultative anaerobic microorganisms and bacteria of the *E. coli* group in the pate samples was determined. All samples were stored in a refrigerator at 4 °C before sampling. 20 g of samples from each batch were transferred to a sterile flask with 90 ml of 0.1% peptone water. By the method of serial dilution, the resulting initial solution was diluted in sterile test tubes with 9 ml of peptone water. Each analysis was carried out three times. The average number of microbial colonies in Petri dishes was multiplied by the dilution factor and expressed in colony-forming units (CFU). The following culture media were used for the study: Lactose broth with bile and brilliant green (selective medium) and Tryptone water.

Determination of amino acid composition: The ion exchange chromatography method was used on an automatic amino acid analyzer AAA-881 (Mikrotechna, Czech Republic) following the instructions for the device. The method includes mandatory preliminary hydrolysis of proteins with acid or alkali to obtain free amino acids. They were then determined using ion exchange chromatography on columns filled with a solid carrier chemically coupled with charged groups that provided electrostatic interaction with the object under study. The coloured solution was passed through a spectrophotometer with a wavelength of 570 nm [13].

Determination of mineral composition: The mineral composition of the samples was determined according to the AOAC (2000) method [14]. All samples to determine the mineral content were weighed by 5 g, placed in a container, and heated in a microwave muffle oven for 12 hours to a final temperature of 600 °C. After microwave splitting, the samples were diluted with 10 ml of hydrochloric acid HCl solution in distilled water (1:1), mixed with a glass rod and passed through a paper filter. The contents of Ca (wavelength 422.7 nm) and P (470 nm) were determined on the atomic emission spectrometer ICP-OES.

Description of the Experiment

The advantage of ultrasonic treatment and its influence on the characteristics of raw materials were studied, and the modes and parameters of ultrasonic treatment of muscle tissue were determined: frequency, intensity and duration. The influence of ultrasonic treatment on the physical, microstructural and organoleptic characteristics of the beef tripe was studied in detail, and the physicochemical, mineral and microbiological properties, vitamin, and amino acid composition of the beef tripe and mesh were also studied.

Number of samples analyzed: We analyzed 5 samples.

Number of repeated analyses: All tests were performed in triplicate.

Number of experiment replications: 3 times.

Design of the experiment: Experimental studies were carried out in the Kazakh Research Institute of Processing and Food Industry LLP laboratory. Beef rumen was chosen for the experiment. Under laboratory conditions, beef rumen samples were cut into pieces (approximately 300 g) for subsequent use after removing visible connective tissue and fat. The tenderizing effect of various ultrasonic treatments on the characteristics of muscle fibre and connective tissue of beef rumen has been studied. The beef rumen sample test experiment was exposed to the technological ultrasound device Bandelin Sonopuls UW2200. Beef tripe samples are shown in Figure 1. The technological process of beef rumen processing is shown in Figure 2.

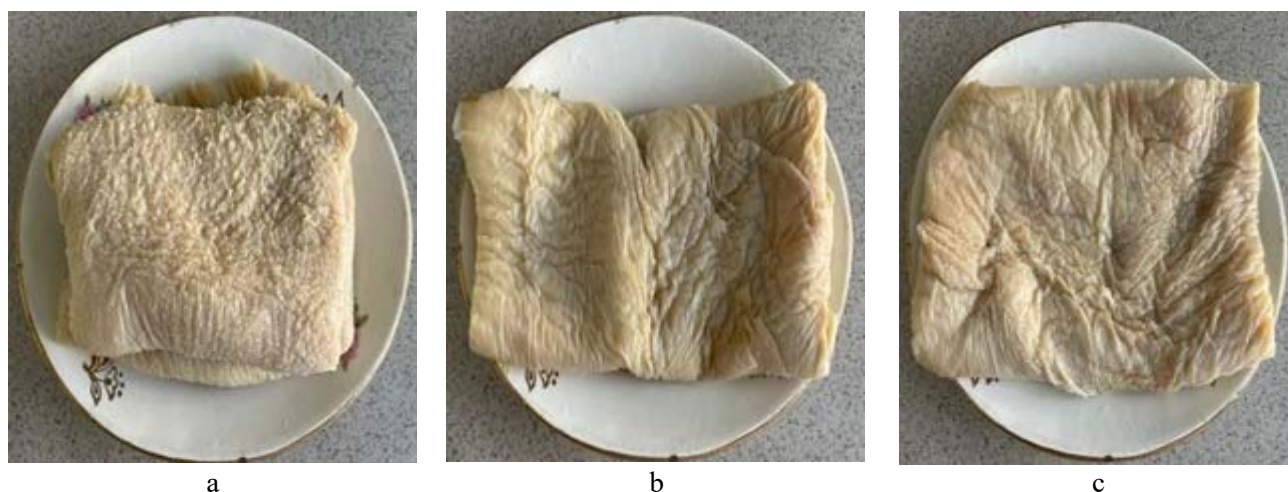


Figure 1 Beef tripe samples: a – Treatment No. 1, b – Treatment No. 2, c – Treatment No. 3.



Figure 2 The technological process of beef rumen processing.

Statistical Analysis

The experiments were carried out in threefold repetition. The values of the standard deviation are indicated for all measurements. The differences in the measurements of the experimental and control groups were calculated using a variation analysis (one-way ANOVA) using the Tukey test. The measurement value $p < 0.05$ was considered significant. Also, the data were analyzed using MS Excel for Windows version 10 Pro, 2010.

RESULTS AND DISCUSSION

The technological properties of raw materials play a key role in the creation of meat products. In this regard, the content of collagen, BCC and the cutting force in beef offal were studied [16], [17], [18]. Structural and mechanical studies show that the connective tissues of beef offal increase the cutting forces, and offal has greater rigidity [19], [20], [21], [22]. The technological properties of beef offal are given in Table 1.

Table 1 Technological properties of beef offal.

| Name | WBC, % | Cutting forces, 10^2 N/m | Collagen, % |
|-----------|-------------------|-------------------------------|-------------------|
| Rumen | 56.6 ^a | 6.70 ^b | 10.2 ^c |
| Omasum | 57.9 ^a | 6.8 ^b | 8.5 ^c |
| Abomasum | 55.4 ^a | 5.73 ^b | 7.3 ^c |
| Reticulum | 55.4 ^a | 5.95 ^b | 9.2 ^c |
| Caecum | 59.3 ^a | 5.30 ^b | 7.8 ^c |

Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

The data obtained allow us to conclude that beef rumen has high-strength characteristics. In this regard, it is necessary to provide for the modification of collagen-containing raw materials by mechanical methods – cavitation of its properties. Due to the diversity of the structure of the collagen protein molecule, it is necessary to choose the processing parameters for each type of collagen-containing raw material [23], [24], [25]. The collagen content in beef offal is shown in Figure 1. Data analysis (Figure 3) showed that beef rumen and reticulum have a high collagen content (rumen – 10.2% and reticulum – 9.2%), and have high strength characteristics.

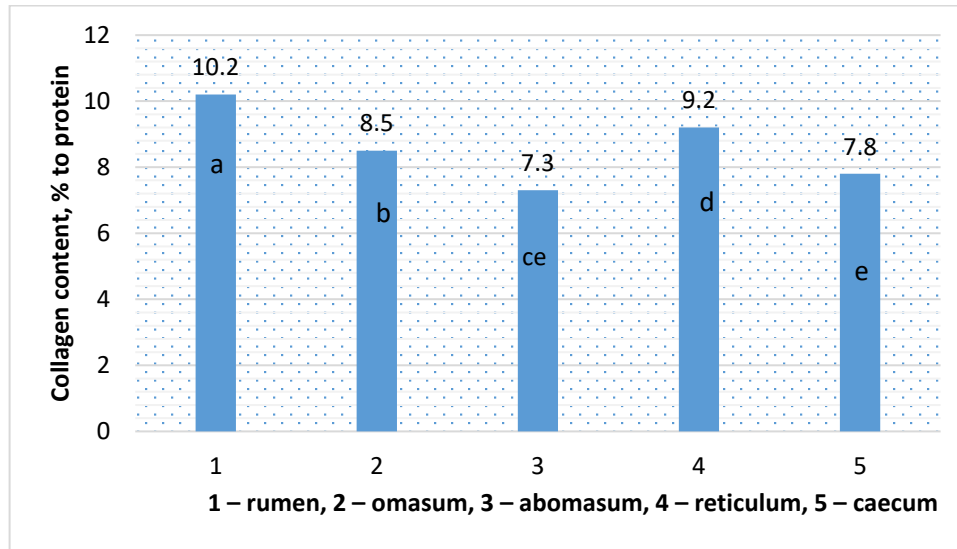


Figure 3 Collagen content in beef offal. Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

To analyze the effect of ultrasonic treatment on consistency, we have analyzed the shear cutting force by texturometer. This method is based on the penetration of the cutting organ through a sample (speed of 50 mm/min). A parallelepiped with a length of 5 mm and a square shape with a side length of 20 mm was formed. The shear forces of cutting beef offal are presented in Figure 4.

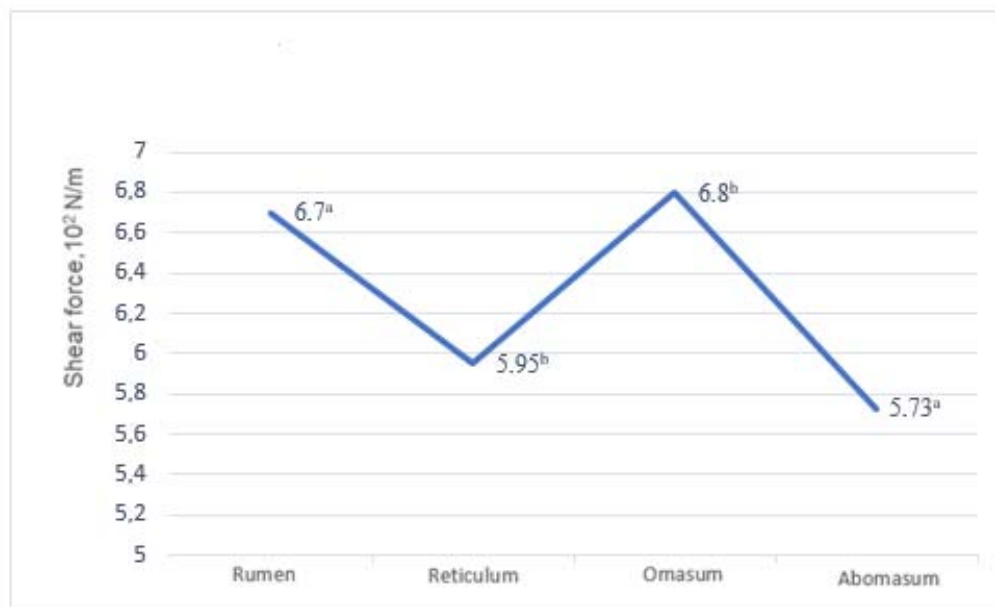


Figure 4 Cutting force of beef offal. Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

Other studies have shown that the possibility of using secondary animal raw materials makes it possible to obtain combined meat products for functional purposes [26], [27], [28], [29]. For this purpose, the chemical composition, mineral elements, vitamin and amino acid composition, and microbiological parameters of beef rumen and reticulum were studied.

The research was carried out in the accredited testing laboratory of the Scientific Research Institute of Food Safety at the Almaty Technological University JSC. The results of physicochemical, mineral and microbiological indicators of beef rumen and reticulum are shown in Table 2.

Table 2 Physico-chemical, mineral and microbiological parameters of beef rumen and reticulum.

| Name of indicators, units of measurement | Rumen | Reticulum |
|--|--------------------|--------------------|
| - pH | 6.82 ^a | 6.78 ^b |
| - calcium, mg/100 g | 16.55 ^a | 14.55 ^b |
| - potassium, mg/100 g | 22.53 ^a | 17.74 ^b |
| - phosphorus, mg/100 g | 81.97 ^a | 64.71 ^b |
| - chlorine, % | 0.53 ^a | 0.40 ^b |
| - Quantity and facultative anaerobic microorganisms of mesophilic aerobic, CFU/cm ³ (g) | 7×10 ³ | 7×10 ³ |
| - Bacteria of the <i>E. coli</i> group (coliforms) in 0.01 g of product | Not detected | Not detected |
| - pathogenic, including <i>Salmonella</i> sp. in 25 g of the product | Not detected | Not detected |
| - <i>S. aureus</i> in 0.1 g of product | Not detected | Not detected |
| - yeast, CFU/cm ³ (g) | Not detected | Not detected |
| - mould, CFU/cm ³ (g) | Not detected | Not detected |

Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

Comparative analysis (Table 2) of the results of the physicochemical and mineral parameters of the rumen and reticulum shows that the highest values were found in beef rumen. Microbiological indicators of quantity and facultative anaerobic microorganisms of mesophilic aerobic, CFU/cm³ (g) of the rumen are 7×10³, and the reticulum are 5×10³.

Studies have found that the rumen and reticulum contain vitamins B5, B2, B3 and B6 (Table 3).

Table 3 Vitamin composition of beef rumen and reticulum.

| No. | Component | Reticulum conc., mg/100g | Rumen conc., mg/100g | SEM | Prob > F |
|-----|-----------------------|--------------------------|----------------------|---------|----------|
| 1 | B2 (riboflavin) | 0.12 | 0.11 | 0.01764 | 0.72 |
| 2 | B6 (pyridoxine) | 0.72 | 0.062 | 0.00886 | 2.77 |
| 3 | B3 (pantothenic acid) | 0.28 | 0.26 | 0.01374 | 0.12 |
| 4 | B5 (nicotinic acid) | 0.50 | 0.057 | 0.01158 | 2.78 |

Note: SEM - Standard Error of the Mean.

As a result of the conducted studies (Table 3, Figure 5 and Figure 6), it was found that beef rumen and reticulum contain vitamins B5, B2, B3 and B6. The quantitative composition of B vitamins in the rumen has the following indicators B2-0.11±0.046, B3-0.26±0.052, B5-0.057±0.010, B6-0.062±0.012, the reticulum also contains B2-0.12±0.05, B6-0.72±0.14, B3-0.28±0.06, B5 in terms of the number of B vitamins-0.50±0.09. The content of vitamins B2, B6 and B3 in the reticulum was higher than in the rumen.

Figure 3 and Figure 4 shows a chromatogram of B vitamins in the rumen and reticulum.

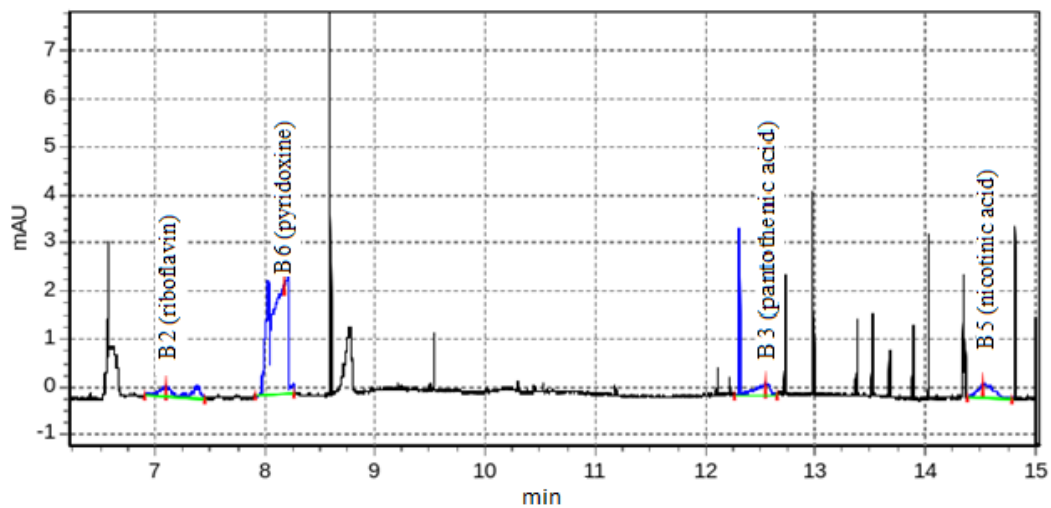


Figure 5 Chromatogram of B vitamins of the beef rumen.

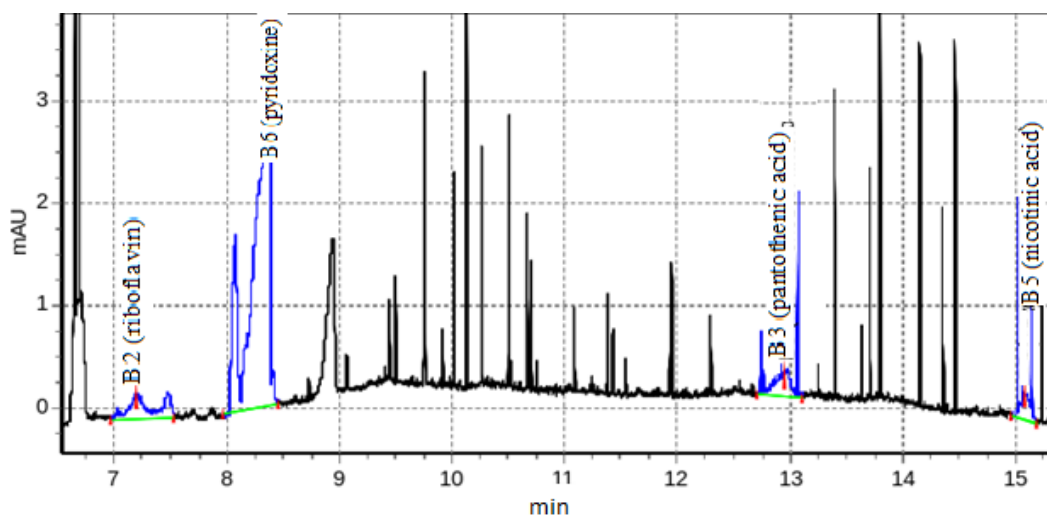


Figure 6 Chromatogram of B vitamins of beef reticulum.

Next, the amino acid composition of beef rumen and reticulum were investigated. The results of the studies on the amino acid composition of the beef rumen and reticulum are presented in Table 4.

Table 4 Amino acid composition of beef rumen and reticulum.

| No. | Components | Rumen | Reticulum |
|-----|----------------------|-------------------|-------------------|
| 1 | arginine | 0.77 ^a | 1.41 ^b |
| 2 | lysine | 0.74 ^a | 0.75 ^b |
| 3 | tyrosine | 0.35 ^a | 0.34 ^b |
| 4 | phenylalanine | 0.41 ^a | 0.41 ^b |
| 5 | histidine | 0.29 ^a | 0.21 ^b |
| 6 | leucine = isoleucine | 0.70 ^a | 0.61 ^b |
| 7 | methionine | 0.36 ^a | 0.32 ^b |
| 8 | valine | 1.27 ^a | 1.42 ^b |
| 9 | proline | 1.23 ^a | 1.34 ^b |
| 10 | threonine | 0.51 ^a | 0.53 ^b |
| 11 | serine | 0.71 ^a | 0.60 ^b |
| 12 | alanine | 0.92 ^a | 0.92 ^b |
| 13 | glycine | 1.51 ^a | 1.83 ^b |

Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

Figure 7 and Figure 8 shows chromatograms of the amino acid composition in the rumen and the reticulum.

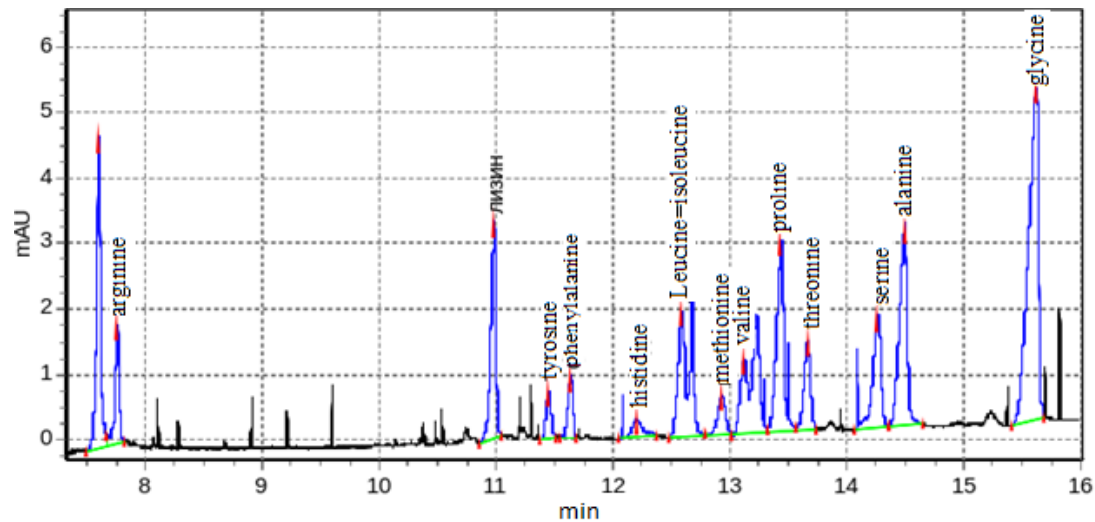


Figure 7 Chromatogram of amino acid composition in the rumen.

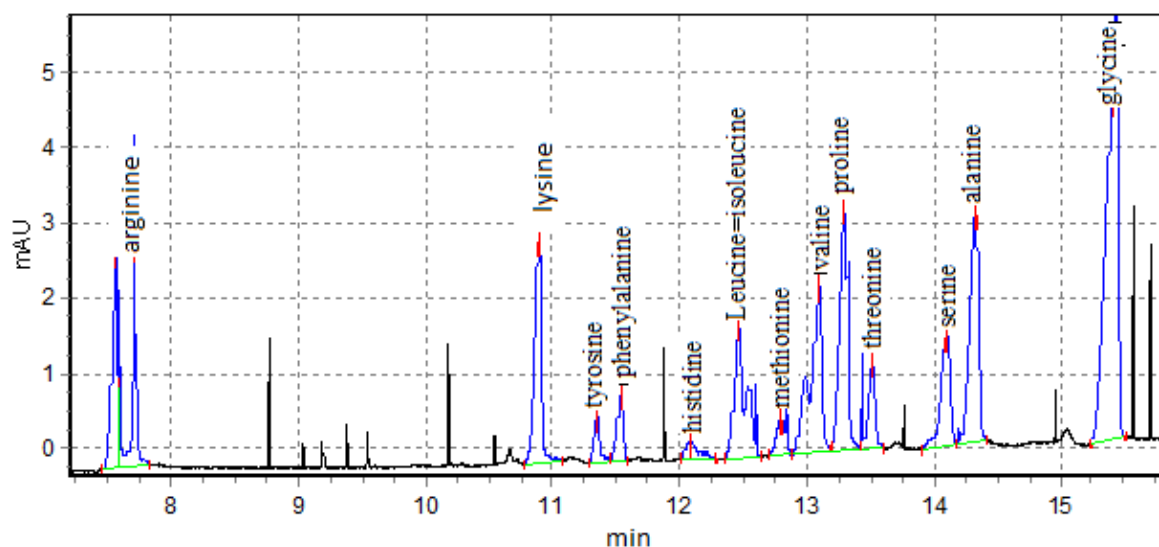


Figure 8 Chromatogram of amino acid composition in the reticulum.

Figure 7 and Figure 8 show that the quantitative amino acid composition of beef rumen is alanine 0.92%, arginine 0.77%, lysine 0.74% and serine 0.71%.

The arginine content in the reticulum was 1.43% higher than in the rumen.

Before the study, beef rumen was washed with cold water to remove dirt and blood and cleaned from tendons and membranes. For the experiment, 6 samples of 300 grams of rumen were prepared.

The rumen samples were stored at 4 °C for 7 days and then immersed in a glass dish filled with water, and an ultrasound field was applied (frequency 10, 20, 30, 40, 50, 60, 70 kHz and intensity/power/ 80, 100, 200, 400, 600, 800, 1000 W/cm²) for 20, 30, 40, 50, 60, 70, 80 minutes.

Ultrasound can be used at high frequencies with low intensity (>1 MHz, <1 W/cm²) or at low frequencies with high intensity (from 20 to 100 kHz from 10 to 1000 W/cm²). Both types are useful for the food industry [30]. In this study, the ultrasound was applied after storage of the offal. We have analyzed the physical and organoleptic properties of the product. Parameters like ultrasound frequency and intensity, duration and temperature during the treatment determine the degree of the desired result achieved by ultrasonic treatment.

Ultrasonic treatment was used at different times, frequencies and intensities. In an experimental study of ultrasound exposure with a frequency of 10, 20 and 30 kHz and an intensity of 80, 100, 200, 300 W/cm², no significant ($p < 0.05$) softening was observed in the rumen.

The advantages of ultrasonic treatment of beef rumen were observed at 40 kHz and 400, 500, 600 W/ cm² for 40, 50 and 60 min. The results showed that the hardness decreased compared to the control, and the consistency was better in the 400 W and 600 W. The qualitative characteristics of the ultrasound-treated beef rumen were evaluated by digital equivalent from 0 to 1, and by colour background from light brown to grey-white.

To visualize the results of experiments on a computer, we have created software program that generates files with the extension.dat; then these digital data were converted into the SURFER graphic editor, which automatically generates a file with the extension.grd and plots the surfaces of changes in the qualitative properties of the beef rumen. Figure 9 and Figure 10 show graphs of the surface change in the qualitative characteristics of the rumen.

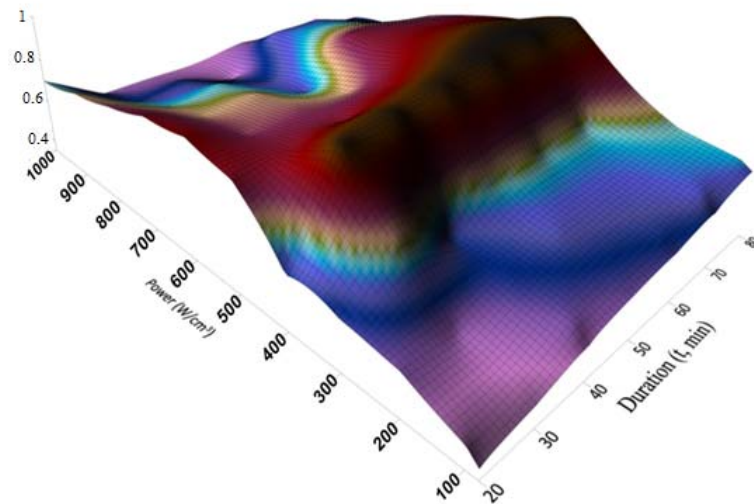


Figure 9 Graph of the surface change in quality characteristics of the rumen.

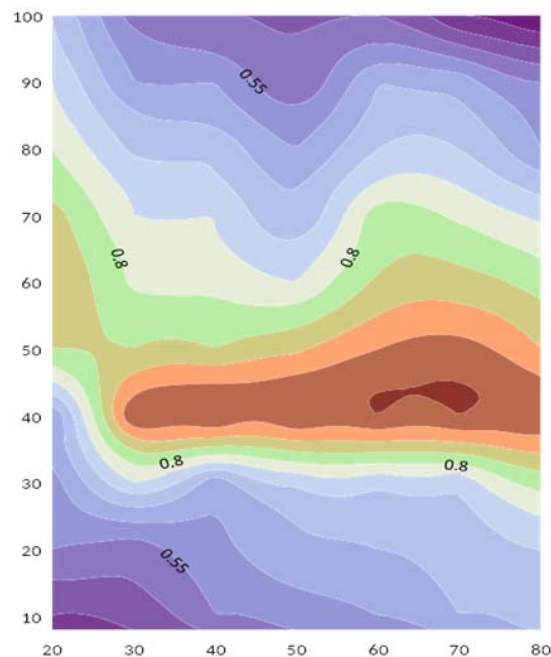


Figure 10 Isolines of the qualitative characteristics of the rumen.

Figure 9 and Figure 10 show that the best quality characteristics (close to 1, the colour is grey-white) were obtained when exposed to ultrasound with power from 400 W/m² to 600 W/m².

The rumen, stored for 7 days before ultrasound treatment, had a more intense pungent odour and a light brown colour.

After treatment with ultrasonic waves, an improvement in the appearance of the rumen was observed. The untreated rumen had a yellowish-light brown colour and a dense structure. There was a sharp, specific smell of the rumen.

After ultrasound treatment, the mechanical strength of the connective tissue decreased in the rumen samples, which had a grey-white colour, but there was still a specific smell of the rumen.

A comparative analysis of the change in the rumen tissue cutting force values after ultrasound treatment was carried out (Figure 11).

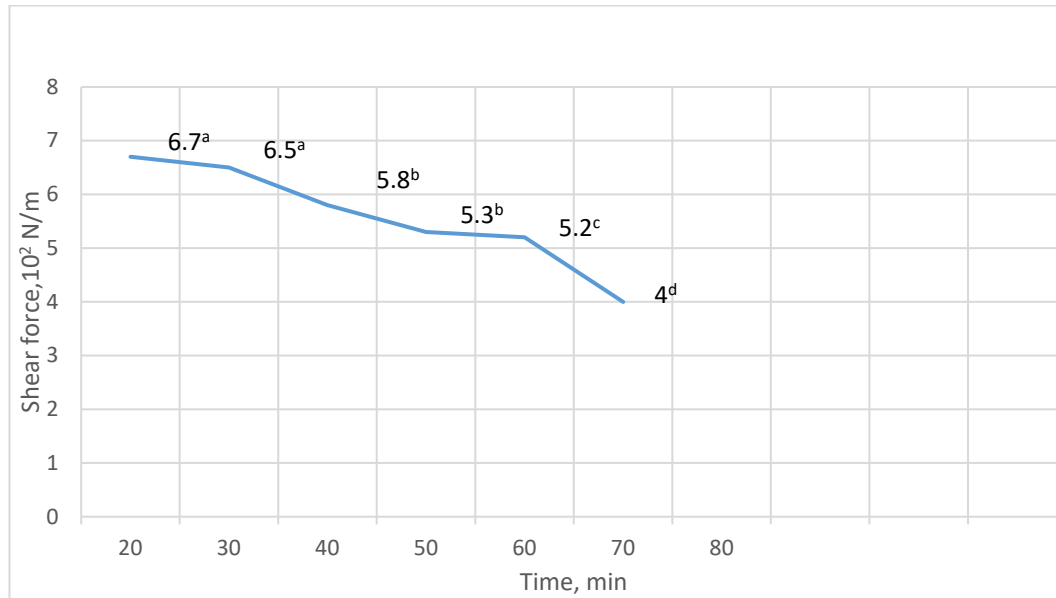


Figure 11 Rumen tissue cutting forces.

Note: ^{a-c} Values with different letters on the graph mean a difference between distinct types of muscles ($p < 0.05$).

Based on the data obtained (Figure 11), it can be assumed that ultrasonic processing significantly reduces the cutting forces of the prototypes relative to the control ones, as evidenced by the values of the cutting force of the rumen samples.

Studies have shown that ultrasound significantly reduces the microbial load and increases the tenderness of the rumen because cavitation destroys cell walls, which leads to the destruction of living cells and thereby contributes to food preservation.

Ultrasonic vibrations have a lethal effect on bacteria and viruses at an intensity of 1 W/cm^2 with an oscillation frequency of hundreds of kilohertz (almost instantaneous rupture of cell membranes, destruction of the internal contents of a microbial cell, up to its complete dissolution. Rod bacteria die faster than cocci.

Spore forms are more stable than vegetative cells. The sterilizing effect of ultrasound depends on the microbial suspension's density and the medium's temperature. The higher the viscosity and acidity of the medium, the less effective the action of ultrasound. The best results of ultrasonic sterilization are achieved in liquid media.

Under the action of ultrasound, partial mechanical destruction of the fibres of muscle and connective tissues occurs, and favourable conditions are created for the action of meat enzymes and the acceleration of chemical processes in tissues.

Numerous studies have been conducted to develop methods for improving tenderness during a series of experiments. Among them is the method of ultrasound application, where various times, frequencies and intensity of ultrasound were used. Most authors agree that ultrasound significantly increases the tenderness of meat products [31], [32], [33], [34] and other qualitative parameters [35], [36]. Thus, the use of ultrasound is an effective alternative technology. This method is used in the processing and preservation of meat as an additional or auxiliary technology [37], [38], [39].

According to numerous studies, it has been revealed that ultrasound does not affect the colour of meat since the heat generated cannot be sufficient to denature proteins and pigments [40]. On the contrary, in our studies, when assessing the effect of ultrasound (400 W/cm^2) on the rumen, it was found that the colour changed to a lighter and

more greyish white, which is less bright than the control. Consequently, ultrasound accelerates the overall colour change, limits the formation of oxymyoglobin and slows down the synthesis of metmyoglobin [41].

Ultrasonic salting makes it possible to obtain tender, evenly coloured pieces of the product without their preliminary intramuscular injection and, accordingly, to obtain final products (for example, ham) without tissue damage.

The best results are obtained when salting 100-200 g of meat according to the following technology: 10 minutes of ultrasonic treatment, cooling to a temperature of 10-15 degrees, subsequent processing for 10 minutes and holding for a day in brine for complete preparation of the product.

CONCLUSION

1. Low-intensity ultrasound is currently used to improve quality, taste and tenderness, which are consumers' most important quality attributes. This review aims to study the effect of ultrasound on the physicochemical, mineral and microbiological, vitamin, amino acid, rheological, functional properties, and nutritional value of the rumen.

2. The rumen treated with ultrasound showed a more delicate structure, but there was still a specific smell. In general, the ultrasound parameters are 400, 600 W/m² for 50-60 minutes duration. They can be selected as the best conditions for processing beef rumen.

3. The use of ultrasound after storage can be a safe method of tenderizing beef rumen when used under the conditions described in this study.

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Funds:

The presented studies were carried out within the framework of the project "Development of a draft standard for slaughter products (bile, internal fat, intestinal raw materials, etc.) in accordance with international standards for the production of export-oriented products", program-targeted financing for 2021-2023 "Development of high-tech technologies for deep processing of agricultural raw materials in order to expand the range and output of finished products with units of raw materials, as well as reducing the share of waste in the production of products" BR10764970, funded by the Ministry of Agriculture of the Republic of Kazakhstan and within the framework of the project funded by the Science Committee of the Ministry of Education and Science of the Republic of Kazakhstan No. AP09058083.

Acknowledgments:

The authors express their gratitude to the administration of the Kazakh Research Institute of Processing and Food Industry and the executors of the project "Development of a draft standard for slaughter products (bile, internal fat, intestinal raw materials, etc.) in accordance with international standards for the production of export-oriented products".

Conflict of Interest:

The authors have declared that there are no conflicts of interest.

Ethical Statement:

This article does not contain any research that would require an ethical statement.

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