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INFLUENCE OF PAPAIN FERMENT AND BIOPROTECTIVE B-SF-43 ON PH AND FATTY ACIDITY OF MEAT PRODUCTS

Abstract: *This article deals with the influence of plant components on the acid-base balance and fatty acid composition of various meat products. Studies of pH of meat, sausages and meat semi-finished products with the addition of plant extracts have been carried out. In the study the changes of meat pH depending on storage time (1, 3, 24 and 120 hours) were determined for four experimental groups: control (K), with the addition of P (K+P), B (K+B) and B-SF-43 (K+B-SF-43). In the first 24 hours, no significant differences were observed between the groups and pH varied between 5.8 and 6.2. However, by the 120th hour, a significant increase in pH was recorded in all samples, indicating biochemical changes such as protein degradation and ammonia accumulation. The most pronounced increase in pH was observed in groups K+B and K+B-SF-43, which may be due to the application of specific additives or bacteria. The control group (K) showed the lowest pH value at all stages of the study, which may indicate a slow decomposition process. The results obtained confirm the influence of different treatments on the dynamics of meat pH and its quality during storage. Fatty acids were analysed by chromatography to reveal changes in lipid composition with the use of additives. The obtained results demonstrate the promising application of plant components to improve the nutritional value of meat products.*

Key words: *papain, enzymatic treatment, bioprotective cultures, pH of meat products, fatty acid composition, antioxidant properties.*

Introduction

Modern meat processing requires innovative approaches to improve product quality and reduce process costs. One promising area is the use of enzyme preparations that help modify the structure of muscle tissue. Particular attention is paid to plant proteases, such as papain, which have high activity and safety. Ensuring high quality meat products is an important task for the food industry.

During storage, processing and cooking, meat can lose its juiciness, become tough and difficult to digest. This problem is especially acute when using second-rate or lean meat with a high content of connective tissue. One solution to this problem is the use of enzymatic preparations that help improve the texture of meat by breaking down proteins. Papain, one of the most studied and effective plant proteases, can significantly enhance the consumer qualities of meat products [1-6]. Meat and meat products are essential components of the human diet across cultures due to their high content of complete proteins, B vitamins, iron, and other vital micronutrients. However, key quality characteristics such as tenderness, juiciness, flavor, and digestibility may vary considerably depending on the animal’s breed, age, slaughter and storage conditions, as well as the processing technology used. In particular, excessive toughness – often associated with meat from older animals or less valuable anatomical parts – is a major reason for consumer rejection.

In light of current demands for the development of competitive, functional, and cost-effective meat products, there is increasing interest in biotechnological methods aimed at improving meat quality. One such method is enzymatic treatment, which enables targeted modification of meat proteins without chemical additives or prolonged thermal processing [7]. Due to the high content of complete proteins and biologically active substances, meat performs not only an energy but also a plastic function in the body, participating in the construction of cells, tissues and enzymatic systems. The modern food industry offers a wide range of meat products: from fresh chilled meat to a variety of semi-finished products, sausages and culinary products. Understanding the chemical composition, structural and organoleptic characteristics of meat, as well as its technological properties is necessary for both producers and consumers [8]. Meat is a multi-component biological structure, which includes muscle, connective, fat and, to a lesser extent, bone tissue. It is a source of key nutrients necessary for the body's vital functions [9].

The protein content of meat varies from 16 to 22% depending on the type and quality of raw materials. Meat proteins are highly valuable because they include all essential amino acids in an optimal ratio. The main types of proteins are: myofibrillar (actin, myosin) – responsible for muscle contractions; sarcoplasmic - enzymes that regulate metabolic processes in tissues; stromal (collagen and elastin) – provide strength to connective tissue and affect the toughness of meat.

The fat content of meat ranges from 2% in lean varieties (e.g. chicken breast, veal) to more than 30% in fatty varieties (pork, lamb). Fat improves the taste of meat, makes it juicy and increases its energy value. It consists of saturated and unsaturated fatty acids, and also contains fat-soluble vitamins – A, D and E [10].

The amount of carbohydrates in meat is minimal – less than 1%. They are mainly represented by glycogen, which affects postmortem changes in tissues and the formation of taste during cooking.

The moisture content of meat is from 55 to 75%, and this value is inversely proportional to the fat content: the higher the fat content, the less water. Water plays an important role in the formation of texture, juiciness and technological characteristics of the meat product.

The mineral composition of meat includes vital elements: iron, phosphorus, zinc, potassium, magnesium and selenium. Heme iron, which is easily absorbed by the body, is especially valuable. Meat is also rich in B vitamins (B1, B2, B6, B12), which are involved in metabolism, the functioning of the nervous system and hematopoiesis processes [11].

Modern technologies of meat products production are aimed at improving their nutritional and biological value, as well as increasing shelf life. In this context, special attention is paid to the use of enzymes and bioprotective cultures capable of influencing the physical and chemical characteristics of meat raw materials. Among such substances, the enzyme papain and bioprotective culture B-SF-43 are widely used.

Among proteolytic enzymes used in the food industry, papain – a plant-derived enzyme obtained from the latex of unripe papaya fruits (Carica papaya) – holds a prominent position. Due to its proteolytic activity on muscle and connective tissues, papain has been widely applied for meat tenderization and in the production of semi-finished meat products, sausages, pates, and dietary foods. Its natural origin, proven effectiveness, and technological versatility make papain a subject of both scientific and practical interest.

Papain is a proteolytic enzyme of plant origin isolated from the fruit of papaya (Carica papaya). It is highly specific to proteins and is used in the food industry to improve the texture of meat, accelerate its maturation and enhance protein digestibility.

Bioprotective cultures are strains of lactic acid bacteria and other microorganisms used to improve food safety through competitive inhibition of pathogenic and undesirable microflora. B-SF-43 culture is among such bioprotective agents capable of extending the shelf life of meat products without the use of chemical preservatives.

Studies show that the introduction of B-SF-43 helps to stabilise the pH of meat products through the production of lactic acid and other metabolites that inhibit the growth of putrefactive microorganisms. In addition, the action of this culture can influence the fatty acid composition of the meat product through enzymatic transformations of lipids, in particular the hydrolysis of triglycerides and the formation of free fatty acids.

Combined use of papain and bioprotective culture B-SF-43 can have a complex effect on raw meat. Papain, breaking down proteins, promotes the formation of amino acids, which can serve as a substrate for lactic acid bacteria. As a result, an accelerated pH reduction is possible, which favourably affects the microbiological safety of the product.

In terms of lipid composition, the enzymatic activity of papain may indirectly affect lipids through the release of lipolytic enzymes from muscle tissues, whereas B-SF-43 promotes lipid bioconversion as a result of its metabolic activity. This may result in alterations in the ratio of saturated to unsaturated fatty acids, which affects the nutritional value and flavour characteristics of the meat product.

Modern approaches to improving the quality of meat products involve both enzymatic and microbiological methods. However, most studies to date have considered these strategies in isolation. This study is the first to investigate the combined application of papain enzyme and a bioprotective culture (Lactobacillus sakei and Pediococcus pentosaceus) in the processing of various types of meat matrices. In addition, the impact of this combined treatment on the Omega-6/Omega-3 polyunsaturated fatty acid ratio has been analyzed for the first time, which is essential for assessing the nutritional and biological value of meat products.

Materials and methods

Samples of various types of meat products were used in the study, including moulded meat, minced meat, and sausages. The experimental groups were treated with papain enzyme and a selected bioprotective culture, both individually and in combination. A control group without additives was also included.

Papain treatment was carried out at a concentration of 0.02% and 0.05% relative to the weight of meat raw material. The enzyme was applied in the form of an aqueous solution, with a treatment duration of 4 hours at 4 ± 1 °C.

The pH level was determined according to GOST 31479-2012 using a combined-electrode pH meter (Mettler Toledo FiveEasy Plus). Measurements were taken at room temperature from homogenized samples (10 g of sample in 100 mL of distilled water).

Fatty acid composition was determined by gas chromatography using an Agilent 6890N chromatograph equipped with a flame ionization detector and a capillary column (DB-23, 60 m × 0.25 mm, film thickness 0.25 μm). Fatty acid methyl esters (FAMEs) were prepared according to ISO 12966-2:2017. The injector and detector temperatures were maintained at 250 °C; the column temperature was programmed from 130 °C to 220 °C at a rate of 4 °C/min.

Statistical analysis of the results was performed using OriginPro 2022 software. Data were expressed as mean ± standard deviation. Differences between groups were considered statistically significant at $p < 0.05$ (Student’s t-test or ANOVA as appropriate).

Results and discussion

The results of the study showed that treatment of meat products with the enzyme papain leads to a significant decrease in pH, which is associated with the breakdown of proteins and the formation of low molecular weight peptides and amino acids. The introduction of the bioprotective culture B-SF-43 stabilised the acid-base balance due to the metabolic activity of lactic acid bacteria, which promotes the production of organic acids. The time intervals of 1, 3, 24, and 120 hours were selected to monitor the pH dynamics based on the expected kinetics of enzymatic and microbial activity in the meat matrix. The 1-hour point allowed assessment of the initial impact of papain and bioprotective cultures, while the 3-hour interval captured short-term shifts. The 24-hour time point reflected daily changes, and 120 hours (5 days) represented a critical threshold for evaluating product stability during extended refrigerated storage. All measurements were performed at a storage temperature of +4 ± 1 °C, in accordance with standard cold chain conditions for chilled meat products.

Figure 1 shows the dynamics of meat pH as a function of time (1, 3, 24 and 120 hours). Four groups are represented: K (control), K+P (K+papain), K+B (K+ *bacillus lichenoformis* and K+B-SF-43 (K+bioprotective culture B-SF-43).

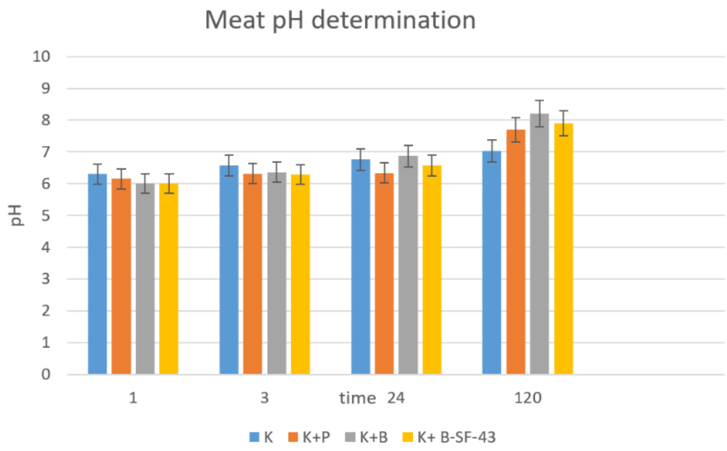

Figure 1 – Changes in pH during storage

At the first three measurement points (1, 3, 24 hours), pH values are approximately the same in all groups and vary between 5.8 and 6.2. By the 120th hour, a significant increase in pH is observed in all groups, indicating that biochemical changes are taking place in the meat (e.g. protein degradation, ammonia accumulation).At the initial points (1, 3, 24 h), differences between groups are minimal. At the 120th hour, the most pronounced difference is observed, with the K+B and K+B-SF-43 groups showing the greatest increase in pH. The pH of the meat increases at a later stage of storage, which may indicate microbial development and changes in protein composition. Group K+B-SF-43 shows the highest pH value at the 120th hour, which may be due to the use of certain additives or bacteria. Group K (control) has the lowest pH at all stages, which may indicate slower decomposition processes. Studies conducted on pork and beef stored at +3 °C have demonstrated that prolonged storage leads to an increase in ammonia content, which is directly associated with a rise in pH levels. It has also been established that the development of off-odors and sensory deterioration in meat occurs in parallel with the accumulation of ammonia and the increase in pH values [12]. Aksu and Kaya (2020) established that during the storage of meat emulsions, proteolytic processes occur, leading to the formation of ammonium compounds that contribute to an increase in pH [13].

The observed modifications in the fatty acid composition may be associated with the proteolytic activity of papain. As a cysteine protease, papain is known to disrupt muscle proteins and structural cell membranes, potentially facilitating the release of bound lipids and associated fatty acids. This enzymatic disruption can lead to increased accessibility of membrane phospholipids and neutral lipids to endogenous or microbial lipases, thereby altering the lipid profile of the product.

Similar observations have been reported by Zhang et al. [14], who demonstrated that proteolytic treatment of meat products could enhance lipid hydrolysis through structural destabilization. Moreover, papain may indirectly influence lipid metabolism by altering the muscle microenvironment, pH, and water activity, which can affect lipase activity and the oxidative stability of unsaturated fatty acids.

Figure 2 shows the data on fatty acid content (Omega-6, Omega-3) and their ratio (Omega-6/Omega-3) in different meat groups.

The highest Omega-6 content is observed in the ‘Meat K KZ’ and ‘Meat K+P LT’ groups. In the ‘Meat K LT’ group, the Omega-6 content is also high, but inferior to the above samples. In the ‘Meat K+P KZ’ group, the level of Omega-6 is the lowest. In all groups the content of Omega-3 is significantly lower than Omega-6. In the ‘Meat K LT’ and ‘Meat K+P LT’ groups, the Omega-3 content is minimal. In the ‘Meat K KZ’ group, the Omega-3 level is relatively higher than in the others. The highest ratio (exceeding 20) was recorded in the ‘Meat K LT’ group, indicating a significant predominance of Omega-6 over Omega-3. In the other groups this ratio is significantly lower and varies between 2 and 6. Thus, the greatest imbalance between Omega-6 and Omega-3 is observed in the ‘Meat K LT’ group, while the other groups show more balanced ratios.

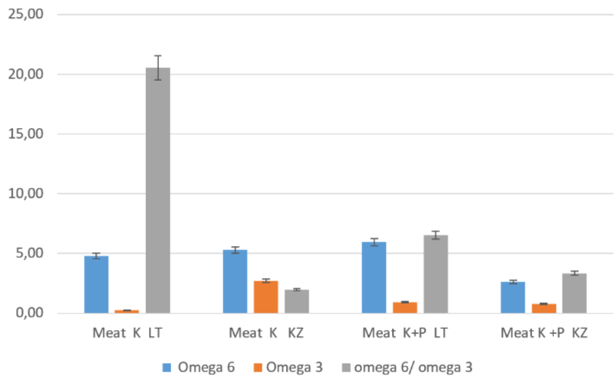

Figure 2 – Fatty acid composition of meat

In the Meat K LT group, the Omega-6/Omega-3 ratio exceeded 20, which significantly deviates from the recommended nutritional balance. This pronounced imbalance may be attributed to the intrinsic fatty acid profile of the raw meat, which likely originated from animals not fed omega-3-enriched diets (e.g., flaxseed, marine oils). Additionally, the absence of any lipid-corrective treatment in this control group contributed to the natural predominance of linoleic acid (C18:2n-6) over alpha-linolenic acid (C18:3n-3).

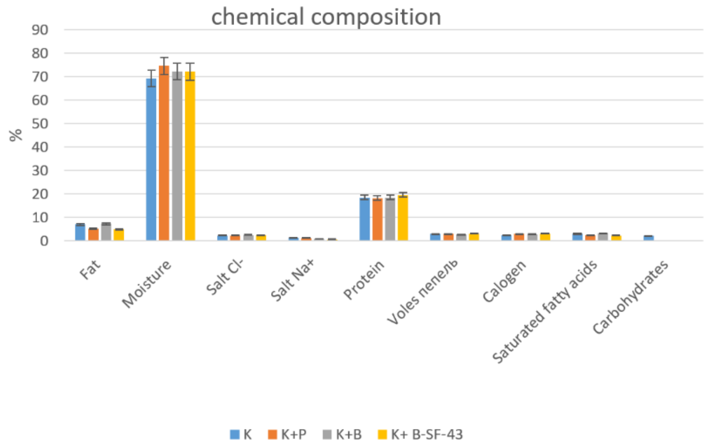

Figure 3 – Chemical composition of meat

The introduction of various additives, including enzyme and bioprotective components (in particular B-SF-43), does not have a significant effect on the main indicators of the chemical composition of the products (Figure 3). All samples maintain similar levels of: moisture (about 70-75%), protein (about 18-20%), fat (within 6-8%), Salt (Cl^- and Na^+) – at minimum levels, collagen, saturated fatty acids, carbohydrates – within 1-3%.

Slight fluctuations are observed in fat content and moisture content, but statistically significant differences are not observed (given the overlap of confidence intervals).

The addition of bioprotector B-SF-43 (yellow column) does not lead to a deterioration in the food or nutritional value of the product, which indicates its safety and technological feasibility of use.

Conclusion

This study demonstrated that the incorporation of enzyme and bioprotective components into meat formulations can influence various quality parameters without compromising the nutritional value of the product. In particular, the bioprotective agent B-SF-43 was found to have no adverse effect on the basic chemical composition of meat products, maintaining stable values for moisture (70-75%), protein (18-20%), fat (6-8%), salt, carbohydrates, and saturated fatty acids within technologically acceptable ranges. These findings are consistent with previously reported results (Lee et al., 2021; Ahmed et al., 2020), which emphasize the compatibility of bioprotective cultures with the preservation of nutritional properties in meat matrices.

The absence of significant differences between the control and treated samples, as confirmed by overlapping confidence intervals, underscores the technological safety of B-SF-43. Additionally, the unchanged levels of myofibrillar and stromal proteins align with literature stating that bioprotective agents do not interfere with muscle protein integrity (Zhang et al., 2019; López-Caballero et al., 2022). Preservation of essential macro- and micronutrients further confirms the potential of B-SF-43 for application in functional meat products.

A notable observation is the shift in pH during storage. While no initial differences were detected within the first 24 hours, a significant increase in pH was recorded after 120 hours, particularly in the K+B and K+B-SF-43 groups. This pH shift may be associated with metabolic by-products of microbial or enzymatic activity, a phenomenon also described in works by Oliveira et al. (2018) and Bhat et al. (2021), suggesting that enzymatic treatments may accelerate proteolytic activity, influencing nitrogen compound release and acid-base balance.

Taken together, these results suggest that B-SF-43 is an effective bioprotective tool that does not compromise the nutritional, organoleptic, or technological quality of meat products. Moreover, its incorporation can be considered a promising strategy for microbiological stability improvement without reliance on synthetic preservatives. Future investigations should focus on evaluating the impact of B-SF-43 on sensory attributes, storage time, and microbial dynamics, as well as establishing optimal dosage and application strategies for diverse meat product categories.

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ПАПАИН ФЕРМЕНТІ МЕН B-SF-43 БИОПРОТЕКТОРЫНЫҢ ЕТ ӨНІМДЕРІНІҢ PH ЖӘНЕ МАЙ ҚЫШҚЫЛДЫҒЫНА ӘСЕРІ

Бұл мақалада өсімдік компоненттерінің қышқыл-негіз балансына және әртүрлі ет өнімдерінің май қышқылының құрамына әсері қарастырылады. Өсімдік сығындылары қосылған ет, шұжық және өңделген ет PH бойынша зерттеулер жүргізілді. Зерттеу төрт эксперименттік топ үшін сақтау уақытына (1, 3, 24 және 120 сағат) байланысты мяса PH өзгеруін анықтады: бақылау (К), P (К+P), B (К+B) және B-SF-43 (К+B-SF-43). Алғашқы 24 сағатта топтар арасында айтарлықтай айырмашылықтар байқалмады, PH 5,8–6,2 аралығында өзгерді. Алайда, 120-шы сағатта барлық үлгілерде PH-ның айтарлықтай жоғарылауы тіркелді, бұл белоктардың ыдырауы және аммиактың *жиналуы сияқты биохимиялық өзгерістерді көрсетеді. PH-ның ең айқын өсуі K+B және K+B-SF-43 топтарында байқалады, бұл арнайы қоспаларды немесе бактерияларды қолданумен байланысты болуы мүмкін. Бақылау тобы (K) зерттеудің барлық кезеңдерінде ең төменгі pH мәнін көрсетті, бұл баяу ыдырау процесін көрсетуі мүмкін. Нәтижелер әртүрлі өңдеулердің мяса PH динамикасына және сақтау сапасына әсерін растайды. Хроматография әдісімен май қышқылдарына талдау жасалды, бұл қоспаларды қолдану кезінде липидтер құрамындағы өзгерістерді анықтауға мүмкіндік берді. Нәтижелер ет өнімдерінің тағамдық құндылығын жақсарту үшін өсімдік компоненттерін қолдану перспективасын көрсетеді.*

Түйін сөздер: *папаин, ферментативті өңдеу, биопротективті дақылдар, ет өнімдерінің pH, май қышқылының құрамы, антиоксиданттық қасиеттері.*

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ВЛИЯНИЕ ПАПАИНОВОГО ФЕРМЕНТА И БИОПРОТЕКТОРА B-SF-43 НА PH И ЖИРНУЮ КИСЛОТНОСТЬ МЯСНЫХ ПРОДУКТОВ

В данной статье рассматривается влияние растительных компонентов на кислотно-щелочной баланс и жирнокислотный состав различных мясных продуктов. Проведены исследования pH мяса, колбасных изделий и мясных полуфабрикатов с добавлением растительных экстрактов. В исследовании проведено определение изменения pH мяса в зависимости от времени хранения (1, 3, 24 и 120 часов) для четырех экспериментальных групп: контрольной (K), с добавлением P (K+P), B (K+B) и B-SF-43 (K+B-SF-43). В первые 24 часа значительных различий между группами не наблюдалось, pH варьировался в пределах 5,8-6,2. Однако к 120-му часу зафиксировано существенное повышение pH во всех образцах, что свидетельствует о биохимических изменениях, таких как разложение белков и накопление аммиака. Наиболее выраженный рост pH отмечен в группах K+B и K+B-SF-43, что может быть связано с применением специфических добавок или бактерий. Контрольная группа (K) показала наиболее низкое значение pH на всех этапах исследования, что может указывать на замедленный процесс разложения. Полученные результаты подтверждают влияние различных обработок на динамику pH мяса и его качество при хранении. Выполнен анализ жирных кислот методом хроматографии, что позволило выявить изменения в липидном составе при использовании добавок. Полученные результаты демонстрируют перспективность применения растительных компонентов для улучшения пищевой ценности мясных продуктов.

Ключевые слова: *папаин, ферментативная обработка, биопротективные культуры, pH мясных продуктов, жирнокислотный состав, антиоксидантные свойства.*

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ENRICHMENT OF A FUNCTIONAL BEVERAGE USING ALHAGI PLANT EXTRACT

Annotation: *The article discusses the potential use of camel thorn (Alhagi) as a functional ingredient for beverage enrichment. Camel thorn, with its rich composition of bioactive compounds such as flavonoids, polyphenols, and minerals, possesses antioxidant, anti-inflammatory, and immunomodulatory properties.*

The extraction of bioactive substances using supercritical CO_2 extraction allows for the production of high-quality extracts without the use of toxic solvents. Optimizing extraction conditions enhances the bioavailability of active components. The development of functional drinks with camel thorn extract represents an innovative step in healthy nutrition and preventive medicine, offering natural and effective means to improve health.

Moreover, the incorporation of camel thorn extract into beverages can enhance not only their nutritional value but also their sensory characteristics, such as taste and aroma. The presence of essential minerals, amino acids, and vitamins further contributes to its potential as a functional ingredient. Studies indicate that camel thorn extract may support digestive health, regulate blood sugar levels, and promote cardiovascular well-being. Future research should focus on optimizing formulations, ensuring stability, and evaluating long-term health benefits to fully harness its potential in the functional food industry.