JIVS JOURNAL OF ISTANBUL VETERINARY SCIENCES

Determination of collagen and pH measurement in beef: Modern laboratory techniques

Sedef Keleş[1]*, Nezir Yaşar Toker[2]

1. Graduate Education Institute, Istanbul University-Cerrahpasa, Istanbul, Turkey . **2.** İstanbul University-Cerrahpaşa, Faculty of Veterinary Medicine, Department of Biochemistry, İstanbul, Turkey. Keleş, S. ORCID ID: 0009-0005-4995-6956; Toker, N. Y. ORCID ID: 0000-0003-4522-991X

Review Article
Volume: 8, Issue: 2
August, 2024
Pages: 126-130

Article History
Received: 07.05.2024
Accepted: 15.05.2024
Available online: 06.06.2024

ABSTRACT

Determining the pH value and collagen value in beef is very important in terms of healthy and quality nutrition. Collagen value is of great importance in meat-like products in order to offer healthy products to consumers. Today, various measurement techniques are used to measure pH value and collagen value. However, the success and reliability of each measurement varies. Therefore, more sensitive and reliable measurement methods need to be developed. Within the scope of this study, modern techniques used in the food industry to measure the pH value and collagen value of cut meat were examined. The reliability and acceptance level of each technique varies. This research aims to contribute to the development of more accurate methods for measuring collagen and pH values. In this way, it is aimed to increase the quality of beef products and offer healthier and more delicious products to consumers.

Keywords: pH, beef, collagen, measurement, techniques

DOI: https://doi.org/10.30704/http-www-jivs-net. 1479918

To cite this article: Keleş, S., & Toker, N. Y. (2024). Determination of collagen and pH measurement in beef: Modern laboratory techniques. ***Journal of Istanbul Veterinary Sciences, 8*(2), 126-130.** **Abbreviated Title*: J. İstanbul vet. sci.***

Introduction

The increasing human population in the world causes people to migrate from rural to urban areas. As a result, the demand for animal foods is gradually increasing (FAO, 1998). People meet their nutritional needs by consuming protein-rich foods, and animal foods such as meat (cattle, sheep, goats, poultry, pigs, fish and seafood/shellfish), milk and eggs are the main ones (Geletu et al., 2021).

Meat and meat products are the most valuable animal products and sources of high quality protein due to their amino acid composition. While the need for protein is increasing day by day, the rich protein content in these products offers an important source for a healthy diet. For the food industry, the safety of products and consumer satisfaction are as important as meeting protein needs. In the food industry, collagen content and pH value are two important parameters that determine the quality of beef products. Collagen is one of the building blocks of meat tissue and is a critical component that determines the texture, tenderness and durability of meat. Therefore, determining the right amount of collagen is a factor that affects the quality and cooking characteristics of meat. In addition, pH value is another important parameter affecting meat quality. pH value indicates the acidity level of meat and affects its color, taste and storage time. Correct pH values ensure that the meat remains fresh and durable and offers a better product to the consumer.

Beef is an important source of protein consumed worldwide and consumer demands are consistently high. Therefore, ensuring quality control in beef production is a critical imperative for the industry. Collagen and pH are two determining factors that

*Corresponding Author: Sedef Keleş
E-mail: sedefkeles2009@hotmail.com

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affect success in this process. Too little or too much collagen can adversely affect the texture of the meat and provide an unpleasant experience for the consumer. Likewise, an incorrect pH value can shorten the shelf life of the meat and reduce the consumability of the product. Therefore, accurately measuring and regulating collagen and pH in cattle is a fundamental step to improve the quality of meat products and gain the trust of consumers.

In this study, we will examine in detail why the measurement of collagen and pH in cattle is important and the laboratory techniques used to determine these parameters. Laboratory methods are indispensable tools to optimize the quality of bovine meat products and to provide safe and healthy products in the food industry.

1.1. Collagen: structure, variety and role in food products

Collagen is the most abundant protein in mammals and birds and is found in all tissues, especially skin, tendons and bone. Connective tissue, which is mainly composed of collagen, serves to support, separate, protect and provide bedding for vascular and neural tissue and prevents excessive elongation of the muscle and damage to its contractile structure (Weston et al., 2002). Collagen is found in various parts of the muscle. Within the muscle, it acts to coordinate and transmit the forces generated in individual muscle fibers so that a reaction occurs as a result. In the form of tendons, collagen connects muscle to bone and plays a role in moving a body part through muscle contraction (Fratzl, 2008). Naturally synthesized collagen molecules consist of three long helicoid chains of amino acid residues with non-helical terminals at both ends. At least 46 unique polypeptide chains have been found in collagen from various animals (Matinong et al., 2022).

Collagen chains are most commonly composed of a Gly-X-Y motif. Here, Gly represents the amino acid glycine, while X and Y usually refer to the amino acids proline and 4-hydroxyproline, respectively. This motif is different from other ECM components (Matinong et al. 2022). The α chains of different types of collagen differ in composition depending on the frequency of repetition and the length of the segment containing the Gly-X-Y motif, whether it is interrupted or continuous, and the amino acid residues occurring at the X and Y positions (Holmes et al., 2018).

The arrangement of polypeptide chains and the diversity of terminals give both fibrillar and non-fibrillar collagen types their distinctiveness. Collagen types vary in conformations resulting in a distribution of different lengths of helices and non-helical segments. These criteria are used to group collagens into various groups. General groups include fibrillar collagens, FACIT (fibril-associated collagens with interrupted triple helices), FACIT-like collagen, basement membrane collagen, beaded filament collagen, transmembrane collagen, short-chain collagen and unclassified collagen (Sherman et al., 2015). At least 29 types of collagen are currently recognized (Sorushanova et al., 2021). Fibrillar collagens are the most abundant ECM proteins in vertebrates and provide stability, connectivity and form to tissues and organs. The most abundant fibrillar collagen in most tissues is type I collagen. It is found primarily on the fibrillar surfaces of skin and bones and in connective tissues. Collagen type I has a rod-like structure consisting of three helical chains. It has a molecular weight of approximately 300 kDa, a length of 280 nm, a diameter of 1.4 nm and contains approximately 1020 amino acid residues per chain (Matinong et al., 2022).

There are also some commercial collagen-based by-products used in various meats and meat products, including hides (skins) and skin trimmings, tendon, bovine shank, sinus remnants and bones separated manually or mechanically from bovine, porcine and poultry carcasses (Zarkadas and Maloney, 1998). Inauthentic meat products may include the substitution of meat from a high-value species with meat from a lower-value species, the augmentation of a meat product with connective tissue or fat (so that these ingredients are present in greater quantities than those naturally associated with the meat used), and the use of non-meat proteins or other substances. Many countries therefore specify the maximum allowable level of collagen in comminuted meat products, such as Protected Designation of Origin (PDO) and Protected Geographical Indication (PGI) products (Messia et al., 2008).

Collagen content in slaughtered beef products may vary depending on various factors. In one study, it was found that the total collagen content in the longissimus lumborum (LL) muscle of cattle breeds such as Limousine and Charolais was around 287-368 mg/100 g, while the insoluble collagen content was 215-278 mg/100 g (Szałkowska and Modzelewska-Kapituła, 2017).

Another study by Modzelewska-Kapituła et al. (2016) examined the effect of crossbreeding on collagen solubility and tenderness in beef. The researchers found that crossbreeding had an effect on collagen content and solubility, as well as on the shear strength and eating quality of beef. This suggests that crossbreeding can affect the collagen properties and tenderness of beef.

A meta-analysis conducted by Blanco et al. (2013) investigated the influence of animal and management factors on collagen properties in beef. The study revealed that the development of total and insoluble collagen content with degree of maturity (DOM) differed between dairy and beef breeds, especially in bulls. However, the relationships between collagen content and DOM were not precise enough for prediction. The study suggested the development of a dynamic mechanistic model to better understand changes in collagen content in beef.

Bruce and Roy (2009) investigated the production factors affecting the contribution of collagen to beef toughness. They found that factors such as age of the animal at slaughter, steroid and beta-adrenergic agonist use and breed of cattle can affect the contribution of collagen to beef quality. In particular, mature collagen cross-link pyridinoline (PYR) concentrations were positively correlated with cutting force and age at slaughter.

1.2. Importance of pH value in determining meat quality

The pH level of meat is one of the most important criteria in determining quality. While the pH level in meat is 7.3 when the animal is alive, the pH level drops to 7.0 after the animal is slaughtered and bled. The decrease in oxygen level after slaughter and the increase in lactic acid formed as a result of aneorobic glucose in the muscles cause the pH level in the meat to decrease. Within the first 1 hour after slaughter, the pH value of the meat drops to between 5.6-6.2. As a result of the decrease in pH, the meat absorbs more water and becomes crisp. It is a known fact that the rearing conditions of the animals and the ill-treatment at the time of slaughter and the resulting stress are effective on the pH values measured after slaughter. Among these treatments, stress (beating, handling, exercise), electrical stimulation and stunning before slaughter are known to affect the pH levels of meat. It is also known that the quality of feeding applied to the animal is also a factor affecting the pH level after slaughter. Animals fed high energy diets have lower pH levels compared to animals fed low energy diets (Şireli, 2018).

We have stated that the pH value in beef can be affected by various factors. In one study, a relationship was found between the increase in the number of days in the feedlot and the increase in carcass weight and the increase in the temperature predicted at pH 6 and the occurrence of high rigor temperature (Warner et al., 2014). In another study, it was observed that pH decreased significantly as the number of freeze-thaw cycles increased, indicating a deterioration in the physicochemical quality of bovine muscle (Rahman et al., 2015). Research has also shown that instrumental precision measurements such as pH and shear force can be used to predict beef eating quality with a high degree of accuracy. However, the success of these predictions can vary greatly and R2 values for sensory precision can be as low as 0.01 (Farmer and Farrell, 2018). Furthermore, pH values of beef patties did not show clear trends and were not significantly affected by time (Gómez et al., 2014). In terms of meat quality, the expression of heat shock proteins has been associated with both the sensory quality and meat quality characteristics of highly marbled beef (Oh et al., 2019). Heat shock proteins play a role in protecting cells from stress and can affect the tenderness and flavor of beef.

Modern laboratory tests used in the field of food engineering

Some important modern laboratory techniques used in food engineering to determine collagen and pH in beef are listed below:

1.3.a. Near infrared spectroscopy (NIRS): Near infrared spectroscopy (NIRS) is increasingly used to monitor fermentation processes using NIR absorption spectroscopy ranging from 12-820 to 4000 cm-1. Chemical structures contribute to the characteristic position, shape and size of the analyte's absorption bands. Nutrients, metabolites, product formation or biomass concentrations can be monitored simultaneously with process-related changes in the NIR spectra of complex culture media. NIRS is a frequently used non-destructive technique for collagen determination in the meat industry (Hills, 2017; Monago-Maraña et al., 2021).

1.3.b. Fluorescence Spectroscopy: Fluorescence spectroscopy is used to determine the changes that occur in food products as a result of technological process and storage. This method can determine various properties of foods (such as functional component, composition, nutritional component) without the use of chemical reagents. White and red meat products have a composition rich in protein, polyunsaturated fatty acids, vitamins and minerals. However, the quality of red and white meat deteriorates rapidly during storage due to microbial growth, oxidation and enzymatic autolysis. Therefore, rapid analysis methods are important to ensure food safety and quality (Karoi and Blecker, 2011; Ankaralıgil and Güneşer, 2021).

1.3.c. FTIR-ATR Spectroscopy: FTIR, also known as the Fourier transform method, is a chemical analytical method that measures the wavelength of light against its infrared intensity. FTIR-ATR method is used in quantitative and qualitative analysis in food research. This method allows for rapid evaluation of ingredients to ensure quality, safety and traceability in foods. The scale of infrared spectroscopy can be extended for applications such as food classification, sorting, authentication, tracking of contaminants and adulteration. FTIR-ATR has been used to determine fatty acid content in meat and meat products. This method has the ability to determine the ratio of saturated fatty acids to monounsaturated and polyunsaturated fatty acids (Lucarini et al., 2018).

1.3.d. High Performance Liquid Chromatography (HPLC): HPLC is an efficient technology used in analytical chemistry for the separation, identification and quantification of components. This technique is based on the principle of moving a pressurized liquid solvent (mobile phase) at high pressure through a column filled with a solid adsorbent material (stationary phase). Each component experiences unique molecular interactions with the adsorbent material, and since these interactions are different for each component, the components have different flow rates, which allow them to separate as they exit the column. Analysis is supported by multiple detectors such as a variable wavelength ultraviolet (UV) detector, fluorescence detector and refractive index detector (RID). HPLC is a versatile and widely preferred technique in industry that can quantify target components using calibrations prepared with standard substances (Sheppard and O'Dell, 2003; Shockcor, 2017).

1.3.e. pH meter: A pH meter is a critical tool used to measure levels of acidity or alkalinity. For example, the specially designed Skin pH Meter® PH 905 is used to measure the pH of the skin and includes a special electrode to increase skin contact (Ariffin and Hasham, 2020). pH meters were used in a study evaluating the efficacy of proton pump inhibitor (PPI) injections by measuring the pH of gastric fluids (Diesner et al., 2016).

The pH meter is an important tool in research and analysis (Ariffin and Hasham, 2020). In the food industry, pH meters are used to measure the pH of different food products and for quality control. These modern techniques offer valuable assistance to food engineers in the quality, nutritional value and safety of beef (Sheppard and O'Dell, 2003; Shockcor, 2017).

Conclusion

In the food industry, it is important to provide consumers with high quality and healthy products. This high product quality is a combination of a number of factors, including the amount of collagen in meat. Meat contains collagen, a connective tissue protein, which has a decisive effect on its texture and durability. The amount of collagen, its maturation state and pH value are factors that directly affect the tenderness and flavor of meat. Collagen provides structure and durability by forming intermolecular cross-links between the muscle fibers in the meat. These bonds change during the processing of meat with temperature. Collagen fibrils shrink as they heat up, which causes the loss of water in the meat, resulting in meat that contains less water and is less tender. However, this process also serves to hold the muscle fibers of collagen together to preserve the texture of the meat after cooking.

Collagen is more abundant in older animals and mature cross-links increase with age, resulting in tougher meat than in younger animals. Therefore, the content of collagen, its maturation state and pH value is an important factor to determine the quality of meat products. Furthermore, the post-mortem degradation of collagen and the use of collagenase enzymes are also used to control the tenderness of meat. These enzymes are used to alter the structure of collagen to provide the desired tenderness and texture.

In the food industry, the detection, analysis and pH determination of collagen plays a critical role in improving the quality of meat products and providing consumers with more flavorful and tender meat. Factors such as collagen content, maturation status and pH value are important determinants of the quality of meat products and therefore need to be carefully monitored. There is therefore a great interest in developing tools for the rapid determination of collagen in meat and for controlling its pH value.

In this study, modern laboratory techniques used for collagen determination and pH measurement in cattle were discussed. These techniques include tools such as near infrared spectroscopy (NIRS), fluorescence spectroscopy, FTIR-ATR spectroscopy, high pressure liquid chromatography (HPLC) and pH meters. These techniques play an important role in assessing and analyzing the quality of meat products, and their ability to precisely measure critical properties such as collagen content and pH value has been highly successful in ensuring quality control in beef production and providing consumers with more palatable, healthy and durable meat products. Nowadays, modern laboratory techniques are frequently used in the meat industry to improve product quality. Our study makes an important contribution to improve the quality standard of meat products and meet consumer demands.

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