EFFECT OF ELASTIN HYDROLYSATE ON BACTERIA AND SOME SENSORY TRAITS OF CHILLED GROUND BEEF

[1] Ghufran M.M. Al-ghanimi, Alrubeii A. M. S.
Researcher Prof.
[1]Agricultural Engineer at the Iraqi Ministry of Agriculture
[2]Coll. Agri. Engin. Sci., University of Baghdad; Baghdad, Iraq.
[1]gh.m20123991@gmil.com [2]alrubeii@yahoo.co.uk

ABSTRACT

This study was aimed to evaluate the effect of elastin hydrolysates on the microbial content and sensory evaluation of ground beef refrigerated at 2°C for 12 days. The study included 10 treatments: T_1 without addition, T_2 addition (1000ppm/kg meat) and T_3 addition (500ppm/kg meat) of elastin hydrolysates produced by the elastase enzyme (E.H.E).T_4 addition (1000ppm/kg meat) and T_5 addition (500ppm/kg meat) of Elastin hydrolysates produced by the collagenase enzyme (E.H.C), T_6 addition (1000ppm/kg meat) and T_7 addition (500ppm/kg meat)of elastin hydrolysates produced by the trypsin enzyme (E.H.T), T_8 addition and (1000ppm/kg meat) T_9 addition (500ppm/kg meat) of elastin hydrolysate produced by the pepsin enzyme(E.H.P) and T_{10} added to the antioxidant BHA (0.01) Per kg of meat. The treatments were stored cold for periods of 1, 3, 6, 9 and 12 days, and some laboratory tests were conducted on them that showed the effect of elastin hydrolysates when their effectiveness was tested against *Escherichia coli Staphylococcus aureus* bacteria. When hydrolysates were added to cold-stored minced beef, the lowest number of bacteria was recorded, as they decreased the logarithm. Total Plate Count TPC37, total psychrophilic count TPC4, and *E.coli* bacteria. Addition treatments also recorded significant increase in degrees of tenderness, flavor, juiciness, and general acceptability.

Keywords: meat spoilage, shelf life of meat, bacterial contamination.
Part of Ph.D. Dissertation of the 1st author.

أثير متحللات الإيلاستين على البكتيريا وبعض الصفات الحسية للحوم البقر المفروم المبرد

غفران منصور محمد الغانمي [1] اميرة محمد صالح الربيعي[2]
باحث استاذ
[1] مهندس زراعي في وزارة الزراعة العراقية
[2]قسم الانتاج الحيواني/ كلية علوم الهندسة الزراعية، جامعة بغداد

المستخلص

هدفت هذه الدراسة إلى تقييم تأثير متحللات الإيلاستين على المحتوى الميكروبي والتقييم الحسي للحم البقري المفروم المخزن بالتبريد بدرجة 2 درجة مئوية لمدة 12 يوم. شملت الدراسة 10 معاملات:T_1 بدون إضافة، T_2 إضافة (1000 جزء في المليونل/كغم لحم) و T_3 إضافة (500 جزء في المليون/كغم لحم) من متحللات الإيلاستين المنتجة بأنزيم الايلاستيز(E.H.E)، T_4 إضافة (1000 جزء في المليون/كغم لحم) و T_5 إضافة (500 جزء في المليون/كغم لحم) من متحللات الايلاستين المنتجة بأنزيم الكولاجينيز(E.H.C)، T_6 إضافة (1000 جزء في المليون/كغم لحم) و T_7 إضافة (500 جزء في المليون/كغم لحم) من متحللات الايلاستين المنتجة بأنزيم التربسين(E.H.T)، T_8 إضافة (1000 جزء في المليون/كغم لحم) و T_9 إضافة (500 جزء في المليون/كغم لحم) من متحللات الايلاستين المنتجة بأنزيم البيبسين(E.H.P) و T_{10} إضافة مضاد الأكسدةBHA (0.01) لكل كغم لحم .خزنت معاملات الاضافة بالتبريد على فترات 1، 3، 6، 9 و 12 يوم، وأجري لها بعض الفحوصات المختبرية اذ أظهرت هذه المتحللات تأثيرا واضحا عند اختبار فعاليتها ضد بكتريا *E-coli* و*Staphylococcus aureus*، وعند اضافة هذه المتحللات للحم البقر المفروم المخزون بالتبريد سجلت أقل عدد من البكتيريا اذ خفضت لوغاريتم العد كلي للبكتريا الاعتيادية، وعدد البكتريا المحبة للبرودة وبكتريا القولون *E.coli*، كما سجلت معاملات الاضافة تفوقاً معنوياً في كل من درجات الطراوة، النكهة، العصيرية والتقبل العام.

الكلمات المفتاحية: فساد اللحوم، اطالة العمر الخزني للحوم، التلوث البكتيري.
جزء من أطروحة الدكتوراه للباحث الاول

Received: 13/7/2023, Accepted:29/10/2023

INTRODUCTION

Meat and its products are distinguished by their high nutritional value and constitute a major source of the essential amino acids that the human body needs, in addition to being a source of vitamin B complex and some mineral elements such as iron (3). Because of the chemical and biological nature of meat, it is subject to spoilage during the storage period as a result of oxidation, spoilage, and bacterial contamination, which are among the factors that most affect the quality of meat (12, 28). The increasing demand for meat has increased competition and encouraged the innovation of new methods in the meat industry, as the meat industry is focusing around the world on developing new manufacturing, production and processing methods to meet consumer demand (4). Many recent studies are largely directed towards discovering compounds that preserve meat from damage and spoilage. These are biologically active peptides from natural sources (32), the use of which can have a significant impact on the meat industry through their use in reducing spoilage resulting from microorganisms. It acts as an antibacterial, in addition to improving the sensory qualities of meat such as tenderness, juiciness, flavor and general acceptability. Likewise, adding biologically active compounds to raw meat during meat processing and meat products has a positive effect on the meat and thus on the health of the consumer (15). Bioactive peptides can be classified into antimicrobial, anticancer, antithrombotic, antihypertensive, immune-enhancing, mineral-binding, and antioxidant (39). Many peptides produced from fermentation and enzyme-induced hydrolysis have shown physiological benefits in humans (8). Bioactive peptides can also be produced from meat proteins and then incorporated into meat products to improve the functional properties of meat products (9). Therefore, the meat industry relies heavily on methods that work to improve the quality and safety of products, such as improving taste, flavor, and texture, reducing fat and salt content, and packaging techniques (42). This study aims to prolong the shelf life of meat, reduce the number of bacteria, improve tenderness, flavor, and juiciness, and increase acceptability General.

MATERIALS AND METHODS

Elastin hydrolysates are produced by the enzymes elastase (E.H.E), collagenase (E.H.C), trypsin (E.H.T), and pepsin (E.H.P). According to the method described by (30, 41) the antibacterial activity was measured according to the method described by Hussein (26). Locally isolated bacterial strains E-coli and Staphylococcus aureus were obtained from the Department of Food Sciences/College of Agricultural Engineering Sciences/University of Baghdad with some modifications. The bacterial strains used were activated by taking cultures of the bacterial strains and injecting them into tubes containing TSB medium and incubating them at 37°C for 24 h, and the total number of bacteria was counted using the molded plate method after incubation. Bacterial growth was confirmed using TSA soy agar medium (25). Brain Heart Infusion BHI agar was prepared and poured into sterile Petri dishes and left to solidify. These bacteria were then wiped onto the media using sterile cotton swabs, and filter paper discs (0.5 cm in diameter) were placed. Soaked with 40, 30, 20, and 10 µl of elastin hydrolysate (until saturation), placed on the surface of the dish, and incubated at 37°C for 24 h (26). The diameters of the zones free of bacterial growth were measured (18). The activity of elastin hydrolysates and the size of the inhibition halo formed against E. coli and staphylococci were compared to the difference between the diameter of the zone of inhibition (clear zone) and the diameter of the disc after 24 hours. As for the number of bacteria in the addition treatments, the total number of bacteria in the meat samples was calculated using the Robert and Greenwood method (35). Total Plate Count, total psychrophilic count of bacteria was determined according to the method mentioned by Andrew (13). And E.coli bacteria using the Prince method (23). Sensory evaluation scores for flavor, tenderness, juiciness and general acceptability were also calculated for meat samples prepared for sensory evaluation by cooking them in an electric oven at a temperature of 165°C until the internal temperature of the meat reached 70°C (19). SAS statistical

analysis software was used to analyze the data according to a completely randomized design (CRD), and significant differences between means were compared using Duncan's multinomial test (22).

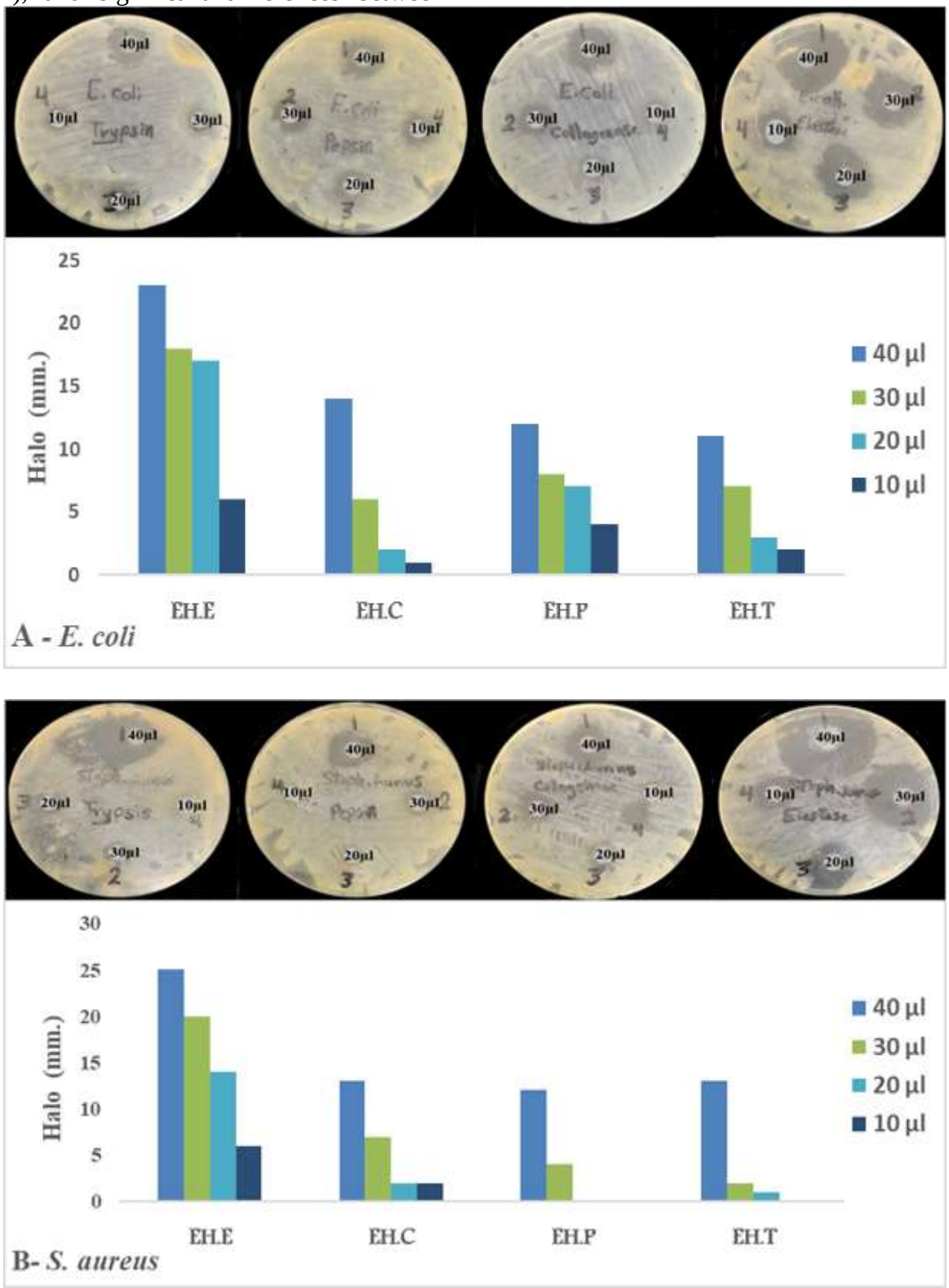

Figure 1. The effect of elastin hydrolysates on inhibiting bacteria : A- E. coli B- Staphylococcus bacteria

RESULTS AND DISCUSSION

Antibacterial activity of elastin hydrolysates

Figure (1) shows the effect of elastin hydrolysates produced by elastase (E.H.E), collagenase (E.H.C), trypsin (E.H.T), and pepsin (E.H.P) in inhibiting *E.coli* and *Staphylococcus* bacteria. It is noted that the size of the corona resulting from the inhibition of (E.H.E) was clear at a concentration of 40 µl, as it reached 25 and 23 for each of the

E.coli and *Staphylococcus* bacteria, respectively. Also, the concentrations of the rest of the elastin hydrolysates inhibited the numbers of bacteria in a varying manner, which explains the ability of elastin hydrolysates to inhibit These two types of bacteria, as well as collagenase (E.H.C), trypsin (E.H.T), and pepsin (E.H.P) *E.coli*, had a clear inhibition at high concentrations on *E.coli* and *Staphylococcus* bacteria, also, the inhibition of (E.H.E) was superior to other degraders. This is consistent with Pereira, et al (33). who indicated the ability of elastin peptides to inhibit *Staphylococcus aureus* and *E.coli* bacteria. Some studies also indicated that biologically active peptides produced from animal sources are considered to have anti-*E.coli* and *Staphylococcus* activity, as studies have indicated. The decomposers produced from pepsin possess antibacterial activity, and when monitoring the antibacterial ability of these decomposers, a decrease in the growth of *Escherichia coli* and *Staphylococcus aureus* bacteria, respectively, was observed (46). These results are also consistent with the findings of Theolier (29, 43)..

Effect of elastin hydrolysates added to ground beef stored in cold storage on bacteria: Chart (1) shows the effect of elastin hydrolysates added to fresh, chilled minced beef on Total psychrophilic count, *E.coli* bacteria, and Total Plate Count, as it was observed that there was a significant increase ($P<0.01$) in the number of bacteria for T_1 6.67, 3.05, and 8.15 CFU/gm meat in TBC4, *E.coli*, and TBC37, respectively, while elastin hydrolysates reduced the number of bacteria, especially with the T_2 and T_3 treatments, as the number of bacteria in T_2 reached 4.86, 2.34, and 5.58, and T_3 reached 5.21, 2.55, and 5.83 CFU/gm. meat for TPC4, *E.coli*, and TPC37 bacteria, respectively. The reason for the decrease in bacterial numbers is due to the effectiveness of antioxidant elastin hydrolysates in preventing the decomposition of both protein and fat and thus reducing oxidative stress due to microorganisms. Di Bernardini, et al (11, 21, 40). This is consistent with Alrubeii and Alalaq (8, 10), who indicated the effect of using Lactobacillus plantarum as a preservation agent in reducing the number of bacteria in basturma. Made from buffalo meat and also consistent with the results of the study of Araújo et al (14). Who indicated that the use of essential oils and niacin in meat sausages reduces the microbial load. It agrees with Uma and Ravi (43). who studied the effectiveness of some carotenoids extracted from shrimp shells, which showed antibacterial activity, especially *Bacillus sp, Achromobacter sp*. As for the effect of periods, chart (2) shows a significant increase ($P < 0.01$) in the logarithm of the total number of bacteria in the periods of 12, 9, 6 and 3 days compared to the period of one day. The number of bacteria begins to increase as the cold storage period progresses and reaches its highest level in a period of 12 days, and this is consistent with the study of Hać-Szymańczu et al (24) and Naveena et al. (31). The action of elastin hydrolysates as antibacterial substances reduces the ability of bacteria to attach to surfaces and embed themselves in mucous membranes, as peptides showed antibacterial activity in the lactoferrin peptide by binding to lipopolysaccharides, purines and outer membrane proteins, as the antimicrobial activity leads to an acceleration of membrane permeability in E. coli (49). This agrees with Quintieri, who indicated that the effect of lactoferrin peptide hydrolysates resulting from digestion with pepsin prevents the growth of bacteria contaminating mozzarella cheese during cold storage *Pseudomonas fragi, Pseudomonas gessardi, Serratia proteamaculans, Aeromonas salmonicida and Rahnella aquatilis* (34). These results are also consistent with ahmed Al-Zubaidi (5). who indicated that adding extracts of antioxidants to ground beef stored refrigerated for different storage periods led to a decrease in the logarithm of total plate count, total psychrophilic count bacteria, and E.coli, compared to control treatments in ground beef and refrigerated stock.

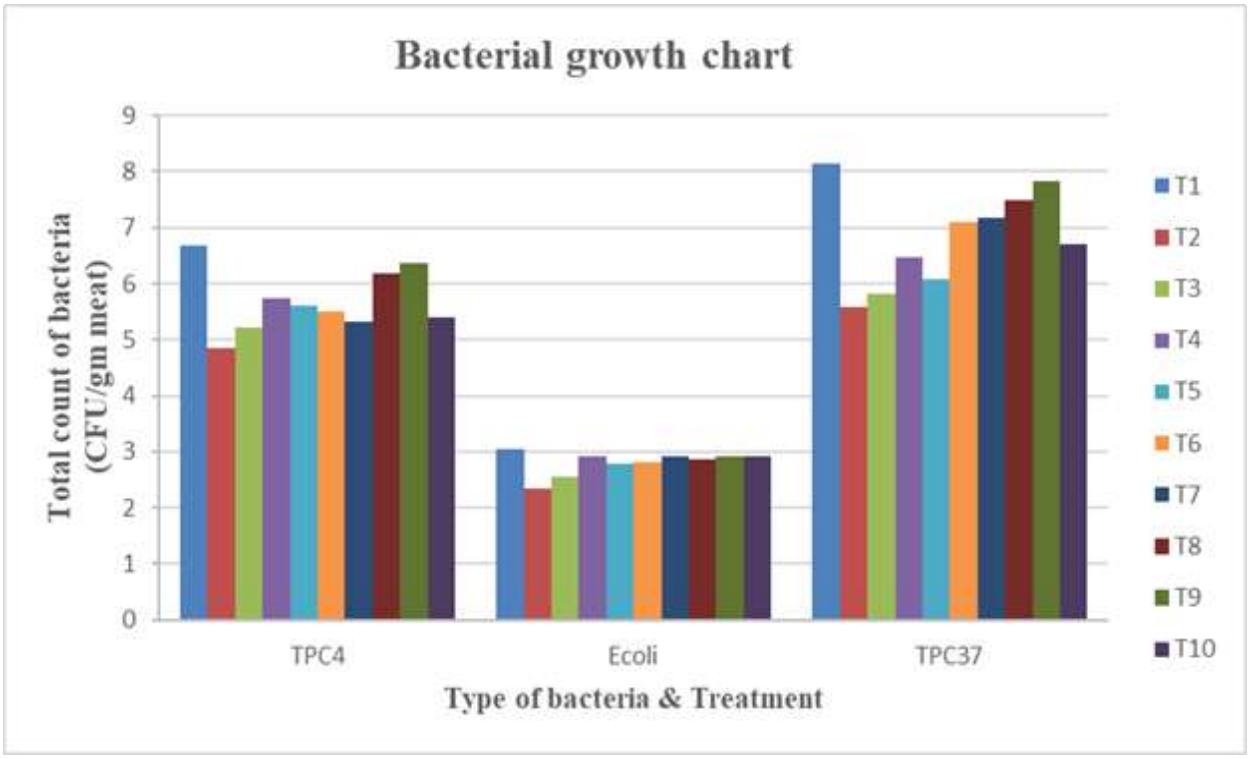

Chart 1. The effect of adding elastin hydrolysates to ground beef stored in cold storage on the total psychrophilic count, E.coli bacteria, and the total plate count.

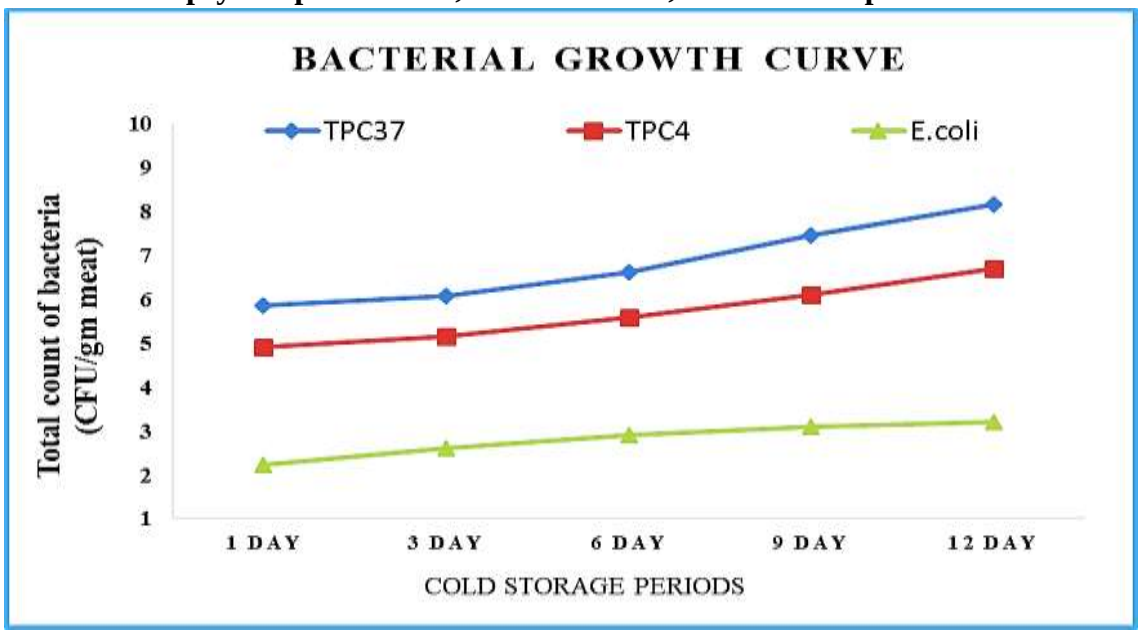

Chart 2. The effect of storage periods on the numbers of bacteria.

Sensory evaluation of cold-stored ground bee: Table (1, 2) shows the sensory evaluation scores (Tenderness, flavor, juiciness, general acceptability) for all treatments and for cold storage periods of 1, 3 and 6 days only. Days 9 and 12 were excluded for fear of high microbial content and to preserve the health of the assessors, as Table 1 shows the effect of adding the above-mentioned elastin hydrolysates on the sensory evaluation of ground beef stored in cold storage. The coefficients indicate a significant superiority ($P<0.01$) in the sensory evaluation scores (Tenderness, flavor, juiciness, and general acceptability) for T2, which recorded 44.5, 4.31, 4.09, and 4.52, and T3, which recorded 4.23, 3.98, 3.93, and 4.14. For tenderness, juiciness, flavor and overall acceptability, respectively. Compared with the control T1, which scored 3.34, 3.31, 3.35, and 3.47 in tenderness, juiciness, flavor, and overall acceptability, respectively. The reason for the high tenderness in the addition treatments is due to the high moisture content, the increased

solubility of the meat protein, the ability of the meat to retain water, and the decrease in loss during cooking. As for the low tenderness in the control treatment, it is due to the high rate of loss during cooking, as the cooking process leads to the loss of fat and juices ,it also leads to shrinkage of the meat and thus a decrease in the tenderness of the meat (7).

Table 1. Effect of adding elastin hydrolysates to cold-stored ground beef on sensory evaluation scores ± SE

Treatment	Sensory evaluation scores			
	Tenderness	Juice	Flavor	General
T_1	3.34 ± 0.04^{G}	3.31 ± 0.07^{F}	3.35 ± 0.05^{G}	3.47 ± 0.12^{E}
T_2	4.54 ± 0.09^{A}	4.31 ± 0.19^{A}	4.09 ± 0.2^{A}	4.52 ± 0.18^{A}
T_3	4.23 ± 0.12^{B}	3.98 ± 0.17^{B}	3.93 ± 0.15^{B}	4.14 ± 0.2^{B}
T_4	3.95 ± 0.16^{C}	3.85 ± 0.18^{C}	3.94 ± 0.15^{B}	4.15 ± 0.1^{B}
T_5	3.81 ± 0.11^{D}	3.50 ± 0.06^{D}	3.79 ± 0.10^{C}	3.73 ± 0.09^{D}
T_6	3.43 ± 0.05^{G}	3.36 ± 0.04^{F}	3.43 ± 0.05^{F}	3.64 ± 0.07^{DE}
T_7	3.38 ± 0.05^{G}	3.35 ± 0.04^{F}	3.38 ± 0.06^{FG}	3.59 ± 0.21^{DE}
T_8	3.57 ± 0.06^{F}	3.48 ± 0.05^{DE}	3.73 ± 0.11^{D}	3.67 ± 0.12^{DE}
T_9	3.43 ± 0.03^{G}	3.43 ± 0.05^{E}	3.62 ± 0.11^{E}	3.72 ± 0.11^{D}
T_{10}	3.67 ± 0.09^{E}	3.49 ± 0.07^{DE}	3.84 ± 0.10^{C}	3.93 ± 0.14^{C}

- Means with similar letters are not significantly (P<0.05) different between them.
- The means with different letters are significantly different (P<0.05) among them.
- T_1 control treatment, T_2(1000ppm/kg meat of E.H.E), T_3(500ppm /kg meat of E.H.E), T_4(1000ppm/kg meat of E.H.C), T_5(500ppm/kg meat of E.H.C), T_6(1000ppm/kg meat of E.H.T), T_7(500ppm /kg meat of E.H.T), T_8(1000ppm/kg meat of E.H.P), T_9(1000ppm/kg meat of E.H.P), T_{10} (0.01ppm/kg meat of BHA

Table 2. Effect of cold storage periods on the average sensory evaluation scores for cold-stored fresh ground beef samples to which elastin hydrolysates were added ± SE

Treatment	Cold storage periods		
	1 day	3 day	6 day
Tenderness	3.94 ± 0.11^{A}	3.73 ± 0.09^{B}	3.53 ± 0.07^{C}
Juice	3.85 ± 0.11^{A}	3.61 ± 0.08^{B}	3.36 ± 0.04^{C}
Flavor	4.01 ± 0.09^{A}	3.69 ± 0.05^{B}	3.43 ± 0.04^{C}
General	4.18 ± 0.09^{A}	3.88 ± 0.09^{B}	3.51 ± 0.06^{C}

- Means with similar letters are not significantly (P<0.05) different between them.
- The means with different letters are significantly different (P<0.05) among them.

While the increase in flavor levels in the addition treatments may be due to the effect of elastin hydrolysates on oxidation and microbial content compared to the control treatment, which recorded a significant decrease in sensory evaluation scores due to proteolysis, fat decomposition, and fatty acid production by increasing the duration of refrigerated storage, which causes unacceptable flavor and odors in the meat. The addition of elastin hydrolysates act as antioxidants, and antioxidants from natural sources delay rancidity and fat oxidation without any effect on sensory properties or nutritional value (47), which leads to preserving the quality of meat and prolonging its shelf life (2, 9, 37). The high degree of general acceptance in the addition treatments for elastin hydrolysates contributed to preserving the quality of the meat and reducing the damage caused by oxidation, proteolysis, microbial degradation, etc., as Abbas and Shakir indicated (1). It is noted that there is an increase in the degrees of general acceptance in the addition parameters, and this increase is due to the effect of elastin hydrolysates in preserving the quality of meat and reducing damage from oxidation, proteolysis, microbial degradation, etc., which leads to improving other characteristics, such as an increase in the percentage of tenderness and juiciness, and an improvement in flavor, and this is reflected in the general acceptance. Table (2) also shows the effect of the periods on the sensory evaluation scores of chilled ground beef, as it is noted that the degree of tenderness decreases in periods 1, 3, and 6, respectively .This decrease may be attributed to enzymatic hydrolysis that occurs in muscle tissue and protein degradation, and this degradation results from proteolytic enzymes such as calpains and cathepsins in addition to

storage conditions and others (20, 27). The effect of the cold storage period on the juiciness was significant (P<0.01), as the meat juiciness levels were highest during the 1 day period 3.85, while they gradually decreased as the storage periods progressed on days 3 and 6 (3.44 and 3.36), respectively. While the periods of cold storage showed a significant decrease in the degree of flavor in the period of 1, 3 and 6 days, respectively, the effect of the period of cold storage is evident in the significant decrease in the degrees of general acceptability, as it decreased significantly in the period of 6 days (3.50) compared to the period of one day. (4.18) Adding collagen hydrolysates extracted from the skin of wild fish to minced meat stored for 10 days at a temperature of 4°C showed antioxidant activity. The improvement of the above-studied characteristics, such as tenderness, juiciness and flavor improvement, was reflected in the general acceptance of the meat. . This is consistent with (8), who indicated the effect of using *Lactobacillus plantarum* as a preservative and antioxidant in improving the qualitative and sensory characteristics and prolonging the shelf life of pastarma made from buffalo meat. The use of antioxidants in the meat industry and In many applications, it has led to improving the quality and safety of products, such as improving taste, flavor, and texture, reducing the percentage of fat and salt in products, packaging techniques, and improving the pathogen detection system (6, 38). The use of antioxidants in the meat industry and in many applications has led to improving the quality and safety of products, such as improving taste, flavor, and texture, reducing the percentage of fat and salt in products, packaging techniques, and improving the pathogen detection system (17, 21, 48).

CONCLUSION

From the results obtained from this study, it can be concluded that elastin hydrolysate possesses antimicrobial properties and has a high ability to inhibit bacteria, and that adding elastin hydrolysate to ground beef stored in refrigeration contributed to reducing the logarithm of the total number of bacteria in ground beef represented by Total Plate Count TPC37, Total psychrophilic count TPC4 Coldness, and spoilage E.coli bacteria. The decomposers also improved the sensory evaluation scores of chilled ground beef.

REFERENCES

1. Abbas, A. A., and K. A. Shakir. 2021. Evaluation of antioxidant functionality of fish collagen enzymic hydrolysate. Iraqi Journal of Agricultural Science, 52(4): 876-884. https://doi.org/10.36103/ijas.v52i4.1395

2. Abed, I. J., M. E. Ahmed, and MH. S. AL-Shimmary. 2021. Rosemary volatile oil as a preservative agent in some canned meat fods. Iraqi Journal of Agricultural Sciences, 52(1); 155-162. https://doi.org/10.36103/ijas.v52i1.1247

3. Ahmad, D. E., and A. S. Al-Hani. 2016."Estimating function supply for red meat in Iraq for the period 1995-2013." Iraqi Journal of Agricultural Sciences 47.6. doi:www.iasj.net/iasj/article/127360

4. Ahmed Al-Zubaidi, L., Al-Rubeii, A. M., and A. S. Al-Salmany. 2021. Effect of Cinnamon and Turmeric Nanoparticles Extract on microorganisms of Fresh Ground Beef During Cold Storage. In IOP Conference Series: Earth and Environmental Science 910(1):012058. IOP Publishing. DOI 10.1088/1755-1315/910/1/012058

5. Ahmed S. M. Al-Salmany, and A. M. S. AL-Rubeii. 2020. Effect of Cinnamon and Turmeric Nanoparticles Extract In Quality Characteristics Of Ground Beef During Freeze Storage. Plant Archives 20(1):350356. DOI 10.1088/1755-1315/910/1/012058

6. AL-Ghanimi, G. M. M., and A. S. AL-Rubeii. 2020. Effect of antioxidant potential of Astaxanthin and Allyl isothiocyanate in quality characteristics of raw ground beef meat during cold storage. Plant Archives, 20(1):673-679. DOI: 10.1088/1755-1315/910/1/012074

7. Al-Ghanimi, G. M., and A. M. Al-Rubaii. 2023. Study of some Functional Properties of Elastin Extracted from Bovine Leather. In IOP Conference Series: Earth and Environmental Science 1214(1):012036. IOP Publishing. doi:10.1088/1755-1315/1214/1/012036

8. Alrubeii, A. M., and M. M. Alalaq 2018. The bio-preservation of buffalo meat manufactured (pastrama) by using lactobacillus plantarum bacteria. The Iraqi

journal of agricultural sciences, 49(1):152-159. https://doi.org/10.36103/ijas.v49i1.219

9. Al-Rubeii, A. M., and H. G. Zahir. 2019. Effect of adding different Levels of Flaxseed powder as a source of Omega-3 on the chemical characteristics of Karadi Carcass Lambs. Basrah Journal of Agricultural Sciences, 32, 194-206. DOI:10.37077/25200860.2019.160

10. Al-Wasidi, A. S., A. M., Naglah, M., Refat, S. M., El-Megharbel, Kalmouch, A., and G. O. M. Moustafa, 2020. Synthesis, spectroscopic characterization and antimicrobial studies of Mn (II), Co (II), Ni (II), Cr (III) and Fe (III) melatonin drug complexes. Egyptian Journal of Chemistry, 63 (4): 1469-1481. https://doi.org/10.21608/ejchem.2020.21193.2263

11. Alwash, B. M. J., Saed, N. H. J., and ALqusay, S. A. 2017. The effect of alcoholic extract Eriobotrya japonica leaves on some bacterial genes and some chemical and microbiological properties of beef. The Iraqi Journal of Agricultural Science, 48(5):1231-1238. https://doi.org/10.36103/ijas.v48i5.332

12. Andres, S., L. Huerga, J. Mateo, M. L. Tejido, R. Bodas and L. Moran 2014. The effect of quercetin dietary supplementation on meat oxidation processes and texture of fattening lambs. Meat Science, 96,806–811. https://doi.org/10.1016/j.meatsci.2013.09.020

13. Al-Sudani, H. M. Khalaf, and A.M. S. Al-Rubeii. 2021. The effect of nano bentonite supplementation in some carcass characteristics of awassi lambs." Systematic Reviews in Pharmacy 12(1):1103-1113. Print. doi:10.31838/srp.2021.1.153

14. Araúj M. K., A. M. Gumiela, K Bordin,., F. B. Luciano and R. E. F. de Macedo. 2018. Combination of garlic essential oil, allyl isothiocyanate, and nisin Z as bio-preservatives in fresh sausage. *Meat Science*, 143, 177-183. http://dx.doi.org/10.1016/j.meatsci.2018.05.002

15. Arihara, K. 2004. Functional foods. In W. K. Jensen, C. Devine, and M. Dikeman (Eds.), Encyclopedia of Meat Sciences pp:492−499. Oxford: Elsevier

16. Arihara, K. 2006. Strategies for designing novel functional meat products. Meat Science, 74, 219−229.

17. Baker, I. A., and J. E. Alkass, 2020. A comparative study on the effectiveness of adding rosemary and ginger extracts on meat quality during freezing storage. Iraqi Journal of Agricultural Sciences, 51(1):333-338. https://doi.org/10.24126/jobrc.2023.17.1.693

18. Bauer A.W, D.M. Perry, W.M.M. Kirby. 1959. Single-disk antibiotic-sensitivity testing of staphylococci. AMA Arch Intern Med 1959;104:208. https://doi.org/10.1001/archinte.1959.00270080034004

19. Alfawaz, M., J. S. Smith, and I. J. Jeon. 1994. Maillard reaction products as antioxidants in pre-cooked ground beef. Food Chemistry, 51(3):311-318. https://doi.org/10.1016/0308 8146(94)90032-9

20. Daniela F. O., B. Ruth, C. Gladys, C. Fernanda, M. Nora 2013. Kinetics of color and texture changes of beef during stor age J Food Sci Technol. 50 (4): 821-825'. doi: 10.1007/s13197-012-0885-7

21. Di Bernardini, R., P. Harnedy, D. Bolton, J. Kerry, E. O'Neill, A. M. Mullen and M. Hayes (2011). Antioxidant and antimicrobial peptidic hydrolysates from muscle protein sources and by-products. Food Chemistry, 124(4): 1296-1307. https://doi.org/10.1016/j.foodchem.2010.07.004

22. Duncan,B.D. 1955. Multiple range and multiple F test .Biometric.11:1-24.

23. Elmer, W. K, D. A. Stephen, V.R. Dowell, Jr. Herbert,.and M. Sommer. 1979. Color Atlas and text book of Diagnetic Microbiology.Priinted in the united starter of America.

24. Hać-Szymańczuk, E., E. Lipińska, and M. Stasiuk. 2011. 'The effect of rosemary preparations on the microbial quality and TBARS value of model pork batters', Acta Scientiarum Polonorum Technologia Alimentaria, 10(2):165–174. 10.1016/j.meatsci.2008.09.004

25. Harrigan, W. F. and M. E. McCance.1976. Laboratory Methods in Food and Dairy Microbiology. Academic Press, London. DOI: 10.4236/ce.2016.714189

26. Hussein, S. I.; A. F. ,Kaluf; Y. Ahmed ;B. Ahmed; and A.Iyad .2020. Determination of inhibition activity of α- amylase enzyme, antioxidant activity, antibacterial activity and phenolic compounds by using some medical plants. Iraqi Journal of Agricultural Sciences:51(1):411-421. https://doi.org/10.36103/ijas.v51i1.940

27. Khalef, A. A. 2017. 'Effect of pomegranate peel extract on some physical, microbial and sensory properties of birker made from frozen chicken meat. Tikrit University Journal of Agricultural Sciences. Special Issue of Proceedings of the Sixth Scientific Conference of Agricultu',1813–1646. DOI: 10.1088/1755-1315/788-6

28. McCarthy, T. L., , J. P. Kerry, J. F. Kerry, P. B. Lynch, and D. J. Buckley. 2001. Evaluation of the antioxidant potential of natural food/plant extracts as compared with synthetic antioxidants and vitamin E in raw and cooked pork patties. Meat Science, 57:45–52. DOI:10.1080/19476330903572945

29. Mcclean, Stephen; BEGGS, B. Louise Welch, W. Robert. 2014. Antimicrobial activity of antihypertensive food-derived peptides and selected alanine analogues. Food Chemistry. 146: 443-447. https://doi.org/10.1016/j.foodchem.2013.09.094

30. Nadalian, M., N. Kamaruzaman, M. S. M. Yusop, A. S. Babji and S. M. Yusop. 2019. Isolation, purification and characterization of antioxidative bioactive elastin peptides from poultry skin. Food Science of Animal Resources, 39(6):966. DIO:10.5851/kosfa.2019.e90

31. Naveena, B. M., Vaithiyanathan, S., Muthukumar, M., A. R. Sen, Y. P. Kumar, M. Kiran, and K. R. Chandran. 2013. Relationship between the solubility, dosage and antioxidant capacity of carnosic acid in raw and cooked ground buffalo meat patties and chicken patties. Meat Science, 95(2): 195-202.https://doi.org/10.1016/j.meatsci.2013.04.043

32. Patel S. 2015. Functional food relevance of whey protein: A review of recent findings and scopes ahead. J. Funct. Foods:19(A):308-1936. https://doi.org/10.1016/j.jff.2015.09.040

33. Pereira, A. M., A. D. Costa, S. C. Dias, M. Casal and R. Machado. 2021. Production and purification of two bioactive antimicrobial peptides using a two-step approach involving an elastin-like fusion tag. Pharmaceuticals, 14(10):956.https://doi.org/10.3390/ph14100956

34. Quintieri, L., L. Caputo, L. Monaci, D. Deserio, M. Morea and F. Baruzzi. 2012. Antimicrobial efficacy of pepsin-digested bovine lactoferrin on spoilage bacteria contaminating traditional Mozzarella Cheese. Food Microbiology, 31(1):64-71. https://doi.org/10.1016/j.fm.2012.02.015

35. Roberts, D. and M. Greenwood 2003. Practical food microbiology, 3ed. Blackwell Publishing Ltd Blackwell Publishing Inc., 350 Main Street, Malden, Massachusetts 02148-5018, USA. DOI:10.1002/9780470757512

36. Saiga, A., T. Okumura, T. Makihara, S. Katsuta, T. Shimizu, R. Yamada. 2003. Angiotensin I-converting enzymes inhibitory peptides in a hydrolyzed chicken breast muscle extract. Journal of Agricultural and Food Chemistry, 51: 174−1745. https://doi.org/10.1021/jf020604h.

37. Saleh, A. M., and G. M. Mohammed. 2020. Studying the effect of adding different concentrations of Ataxanthin and allyl isothiocyanate and their synergistic action in lipid oxidation and some quality characteristics for minced veal meat at cold storage. Diyala Agricultural Sciences Journal, 12(special Issue): 664-674. https://doi.org/10.52951/dasj.20121056

38. Salman, Z. O., B. M. J. Alwash, and E. J.Kadhim. 2019. Effect of essential oil of Cestrum nocturnum flowers cultivated in Iraq as antioxidant and elongation cold storage period of minced meat. Iraqi Journal of Agricultural Sciences, 50(2); 601-607. https://doi.org/10.36103/ijas.v2i50.659

39. Szmańko, T., Lesiów, T., and J. Górecka, 2021. The water-holding capacity of meat: A reference analytical method. Food Chemistry, 357, 129727. https://doi.org/10.1016/j.foodchem.2021.129727

40. Sánchez, A., and A. Vázquez 2017. Bioactive peptides: A review. Food Quality and Safety, 1(1): 29-46. https://doi.org/10.1093/fqsafe/fyx006

41. Seligman, M., R. G., Eilberg and L. Fishman. 1975. Mineralization of elastin

extracted from human aortic tissues. Calcified tissue research, 17:229-234 https://doi.org/10.1007/BF02546686

42. Singh, P. K., G. Jairath and S. S. Ahlawat. 2016. Nanotechnology: a future tool to improve quality and safety in meat industry. Journal of Food Science and Technology, 53(4): 1739-1749. https://doi.org/10.1007/s13197-015-2090-y

43. Taghipour, M. J., H. Ezzatpanah, and M. Ghahderijani. 2023. In vitro and in silico studies for the identification of anti-cancer and antibacterial peptides from camel milk protein hydrolysates. Plos one, 18(7): e0288260. https://doi.org/10.1371/journal.pone.0288260

44. Théolier J, R. Hammami, P. Labelle, I. Fliss and J.Jean .2013. Isolation and identification of antimicrobial peptides derived by peptic cleavage of whey protein isolate. J Funct Foods.:5(2):706–14. https://doi.org/10.1016/j.jff.2013.01.014

45. Vercruysse, L., J. van Camp and G. J. Smagghe. 2005. ACE Inhibitory peptides derived from enzymatic hydrolysates of animal muscle protein: A review. Journal of Agricultural and Food Chemistry, 53, 8106−8115.https://doi.org/10.1021/jf0508908

46. Wafaa, N., H. A., E. M. A. Elbarbary , H. A. Ibrahim, Mohamed and H. Jenssen. 2022. Effect of enzyme type and hydrolysis time on antibacterial and antioxidant activity of whey protein hydrolysates. Iraqi Journal of Agricultural Sciences, 53(6):1340-1357. https://doi.org/10.36103/ijas.v53i6.165

47. Zainy, Z. I., and A. M. S. Alrubeii. 2023. Effect of replacement nitrite by beetroot and silybum marianum powder in physical characteristics and lipid oxidation for pasterrma. Iraqi Journal of Agricultural Sciences, 54(4):1131-1136. https://doi.org/10.36103/ijas.v54i4.1806

48. Zangana, B. S. R., andJ. S. H , Al-Shamery. 2016. Effect of partial replacement of some plant sources in quality and sensory characteristics of processed of goose meat burger. Iraqi Journal of Agricultural Sciences, 47(4):1089-1100. https://doi.org/10.36103/ijas.v47i4.548

49. Zarzosa-Mreno, D., C. Avalos-Gómez, L. S. Ramírez-Texcalco, E. Torres-López, R. Ramírez-Mondragón, - J. O Hernández. Ramírez, and de la M. Garza, 2020. Lactoferrin and its derived peptides: an alternative for combating virulence mechanisms developed by pathogens. Molecules, 25(24): 5763. https://Doi:10.3390/molecules25245763.