MSc THESIS

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EVALUATION OF SOUS-VIDE CHICKEN BREAST PROPERTIES DURING STORAGE

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1. INTRODUCTION

The Food and Agriculture Organization (FAO) reports that between 2000 and 2020, the world's consumption of meat, eggs, and milk increased by 23–30%, with notably high increases of 2–6% in developed countries and up to 60% in developing countries. In 2019, there were about 207 million tons of meat produced worldwide from a variety of sources, including poultry, cattle, sheep, goats, and buffalo. At 57% of the total, poultry meat was the largest category (FAO, 2021). It is anticipated that production of poultry will expand most in Indonesia, Russia, Brazil, Japan, and India. According to Uzundumlu and Dilli (2021), global per capita consumption of chicken meat is anticipated to rise from 15.0 kg in 2018 to 17.0 kg in 2025. Because of its nutritional advantages and affordable price compared to other meats, poultry meat is growing in popularity across the globe. It is a healthy option for a variety of demographics due to its high protein content, complete amino acid composition, vital micronutrients, and high amount of PUFA (polyunsaturated fatty acids) fat (Marangoni et al., 2015).

In recent years, the majority of poultry (80%) has been sold as processed or cut-up parts. In contrast, 80 years ago, only a small part of chicken was cut up at local butcher shops while most poultry (80%) was sold as whole birds. There has been a movement toward cut-up and processed poultry goods, which can be attributed to several things, including rising consumer income levels and growing demand for quick-to-prepare dishes like ready-to-eat chicken. The trend of buying prepared foods has also been influenced by demographic changes, such as an increase in single-income households, as people today spend much less time preparing meals at home than they did 60 years ago (15 minutes versus 90 minutes, respectively) (Barbut, 2020). In the last few years, sous-vide cooking has gained popularity in the food industry, among producers of ready-to-eat meals, and in households, due to the demand for minimally processed, convenient food with enhanced natural qualities and high nutritional value (Zavadlav et al., 2020). Meat is cooked at a lower temperature for an extended period of time in sous-vide cooking, which can enhance certain qualities of the meat and preserve its natural moisture, nutritional value, and flavor (Gil et al., 2022). Sous-vide cooking differs from traditional methods in that the raw food is placed in a vacuum-sealed bag and heated between 55 and 70 degrees Celsius in a carefully controlled environment. The length of cooking time depends on the meat's characteristics, such as its type, size, and shape. This method has several advantages,

including a more efficient transfer of heat from water or steam to the food, a longer shelf life due to the elimination of the risk of recontamination during storage, and the prevention of oxidation and loss of flavour and moisture during heating (Baldwin, 2012).

There are two methods for sous-vide cooking: the double-step method, which uses two different temperature conditions, and the single-step method, which uses only one temperature. According to Ismail et al. (2019), a two-stage sous vide method using temperatures between 45-50°C resulted in enhanced textural characteristics, such as reduced shear force, increased suppleness, and enhanced chewiness, compared to the conventional single-stage method. Meat product treated with the double-step temperature method exhibited improved texture (including shear force, hardness, gumminess, and chewiness), decreased cooking loss, acceptable redness values, decreased lipid oxidation levels, and increased total protein solubility, compared to the single-step method (Christensen et al., 2011; Biykli et al., 2020;. Karpinska-Tymoszczyk et al., 2020). In addition, meat contains endogenous proteolytic enzymes with the highest activity reported between 40 and 50°C; the increased activity of these enzymes at these temperatures is responsible for higher desmin (a muscle-specific intermediate filament that is essential for proper muscle structure and function) degradation, indicating improved meat tenderness (Akoglu et al., 2018).

Both one-step and two-step sous vide treatments at 50°C and 60°C are safe based on the thermal inactivation of the heat-resistant microorganism Enterococcus faecalis at the utilized pasteurization levels. (Hasani et al., 2023). This indicates that both methods can be used without endangering food safety. Chilling is an effective method of preserving the chemical, organoleptic, and nutritional properties of the product while preventing the harmful impacts of microorganisms and enzymes. (Fernandes et al., 2016). Meat products require proper storage conditions to prevent deterioration, including the loss of moisture, texture, colour, flavor, and aroma (Barbut and Leishman, 2022). The purpose of this study was to compare the effects of storage duration and temperature on the quality of sous-vide chicken breasts.

2. AIM OF THE STUDY

The overall objective of this research is to assess the impact of the storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C) on different quality properties of three sous-vide treatment (based on cooking temperature and time) of chicken breast sample.

The specific objectives of this research are:

- To analyze the moisture content (%) of three type sous-vide cooked chicken breast under storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C).
- To analyze the cooking loss (%) of three type sous-vide cooked chicken breast under storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C).
- To analyze the color (lightness, redness, yellowness, and total colour different) of three type sous-vide cooked chicken breast under storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C).
- To analyze the lipid oxidation of three type sous-vide cooked chicken breast under storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C).
- To analyze the odor acceptability of three type of sous-vide cooked chicken breast under storage time (0, 7, 14, and 21 days) and different storage temperature (4°C and 10 °C).

3. LITERATURE REVIEW

3.1 Poultry Meat

3.1.1 Definition, Market, and Production

Poultry is a term used to explain a variety of birds, including chickens, turkeys, pheasants, geese, ducks, ostrich, emu, and quail, that are raised for commercial meat production purpose The chicken, also known as *Gallus gallus domesticus*, is a domesticated version of the native Southeastern Asian grey and Ceylon junglefowl. There are two primary uses for chicken: flesh production (broilers) and egg laying (layers). Due to its low price and delicious flavour, chicken has become a popular alternative to traditional red meats such as beef and swine. This is largely due to advancements in production techniques that have reduced the time required to raise a broiler chicken for slaughter to approximately six weeks, making poultry a more efficient and cost-effective option for meat production (Mozdziak, 2019).

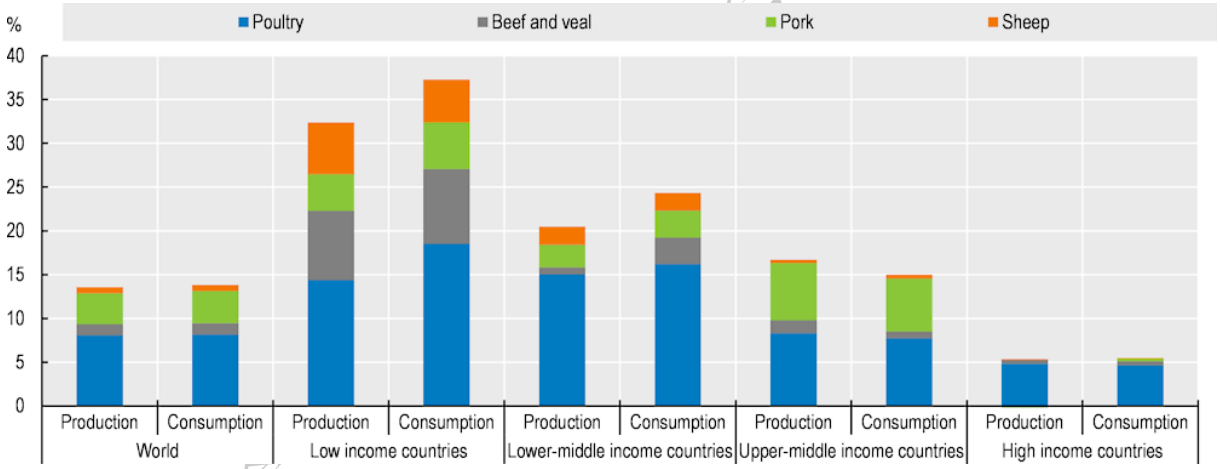

Figure 1. Growth in meat production and consumption on a protein basis, 2021 to 2030 (FAO, 2021)

Between 2000 and 2020, the global consumption of meat, eggs, and milk increased significantly, with the highest increases occurring in developing countries (up to 60%). In 2019, the world produced approximately 207 million tons of meat from a variety of sources, including cattle, livestock, goats, and buffalo, with poultry accounting for 57% of this total (FAO, 2021). In the approaching years, Indonesia, Russia, Brazil, Japan, and India are anticipated to experience the greatest increases in poultry production. The consumption of chicken meat per

capita is projected to increase from 15.0 kg in 2018 to 17.0 kg in 2025 (Uzundumlu and Dilli, 2021).

Several factors, including the high nutritional value of poultry, low production costs, product consistency, cultural and religious considerations, and the use of advanced technology and automation in the poultry industry, contribute to this projection. It is expected that the development of commercial technology and the incorporation of robotic equipment into the poultry supply chain will further stimulate the industry's expansion. It is anticipated that artificial intelligence, sensors, robotics, and transportation systems will play key roles in the future of the broiler industry and breeding management (Park et al., 2022).

Individuals may interpret the notion of poultry quality in a variety of ways. In the case of poultry, for instance, a farmer may prioritize rapid growth, good health, and efficient feed conversion, while a processor may be more concerned with uniformity, high meat yield, and the absence of defects such as bruises and fractured bones. When determining the quality of poultry, consumers may place more importance on characteristics such as texture, flavour, moistness, and appearance (Barbut, 2020).

3.1.2 Nutritional Properties of Poultry Meat

Poultry meat is an excellent source of protein, vitamins, and fat. It also contains essential amino acids, vitamins, and fat. In comparison to red meat, it is often considered a healthier option due to its higher content of unsaturated fatty acids and lower fat content in certain cuts. For instance, the skinless chicken breast fillet contains only 2.5% fat and no marbling. The majority of the fat in poultry is located beneath the skin; therefore, skinless fillets contain very little fat, which consists primarily of polyunsaturated fatty acids found in cell membranes. On the other hand, skinless broiler drum meat has an average fat content of 6%, while skin-on drum meat contains approximately 10% fat (USDA, 2019).

Table 1. Nutritive value of the whole and breast of the poultry meat, per 100g of an edible portion (Barroeta, 2007).

	Whole	**Breast**		**Whole**	**Breast**		**Whole**	**Breast**
Water (%)	70.3	75.4	Iodine (µg)	0.4	0.4	Vitamin B_2 (mg)	0.15	0.15

Energy (kcal)	167	112	Magnesium (mg)	22	23	Niacin eq. (mg)	10.4	14
Protein (g)	20	21.8	Zinc (mg)	1	0.7	Vitamin B_6 (mg)	0.3	0.42
Total fat (g)	9.7	2.8	Selenium (µg)	6	7	Biotin (µg)	2	2
SFA (g)	2.6	0.76	Sodium (mg)	64	81	Folic acid (µg)	10	12
MUFA (g)	4.4	1.3	Potassium (mg)	248	320	Vitamin B_{12} (µg)	0.4	0.4
PUFA (g)	1.8	0.52	Phosphorus (mg)	147	173	Vit. A: Eq. Retinol (µg)	9	16
PUFA/SFA	0.69	0.69	Sodium (mg)	64	81	Vitamin D (µg)	0.2	0.2
Cholesterol (mg)	110	69	Potassium (mg)	248	320	Vitamin E (mg)	0.2	0.29
Calcium (mg)	13	14	Phosphorus (mg)	147	173	Vitamin C, K (µg)	—	—
Iron (mg)	1.1	1	Vitamin B_1 (mg)	0.1	0,1	Vit. A: Eq. Retinol (µg)	9	16

Breast meat from poultry is not a particularly rich source of iron. This is reflected in the meat's very light colour, which is a result of its low myoglobin content. In contrast, duck breast meat has a higher concentration of myoglobin and appears darker due to the abundance of red muscle fibres adapted for extended flights in migratory birds. In general, poultry products are perceived to be leaner, lower in fat, and healthier than red meat products (Barbut, 2020).

3.1.3 Phyisical and Chemical Properties of Poultry Meat

3.1.3.1 Color

When purchasing food, such as chicken, color is an essential factor that can attract or detract consumers. various markets favor various skin tones, including yellow, white, and intermediate tones. The skin pigmentation of chickens can be altered by feeding them feedstuffs containing carotenoids, such as corn or spirulina (Altmann and Rosenau, 2022). Numerous factors can influence the color of poultry meat, including the age (increase the amount of heme pigment in the muscle), the genotype of the bird (affect its capacity for pigment fixation and fatness), the

glycogen storage in the muscle (affect the glycolytic potential), and the level stress that the bird experienced prior to slaughter (which can also influence the color of the meat) (Barbut, 2020).

$$\Delta E = \sqrt{(L^* - L_0^*)^2 + (a^* - a_0^*)^2 + (b^* - b_0^*)^2} \quad (1)$$

The CIELAB colour space diagram is a three-dimensional representation of colours based on their luminance (L*) and chromaticity values (a* and b*). In colour science and technology, it is frequently used to quantify and compare the perceived hue differences between samples, as well as to standardise colour measurements in industries including food, cosmetics, and textiles. The L* axis represents the lightness or darkness of a colour, with 100 representing white and 0 representing black. Positive values indicate red and negative values indicate green along the a* axis. On the b* axis, positive values represent yellow and negative values represent blue. Using equation (1), the total colour difference (E) can be calculated (CIE, 1986). Tomasevic et al. (2019) categorized the total colour difference (E) into four separate categories. Category I with $0 < \Delta E < 1$ indicates that no discernible difference exists, whereas Category II with $1 < \Delta E < 2$ indicates that only an experienced observer can detect a difference. Category III with $2 < \Delta E < 3.5$ indicates that the colour difference can be perceived even by an inexperienced observer, whereas Category IV with $3.5 < \Delta E < 5$ indicates that the colour difference is obvious. L* = 53.71 1.90, a* = 7.98 1.30, and b* = 15.67 0.32 are the CIELAB values for fresh chicken breast, according to Nyam et al. (2023).

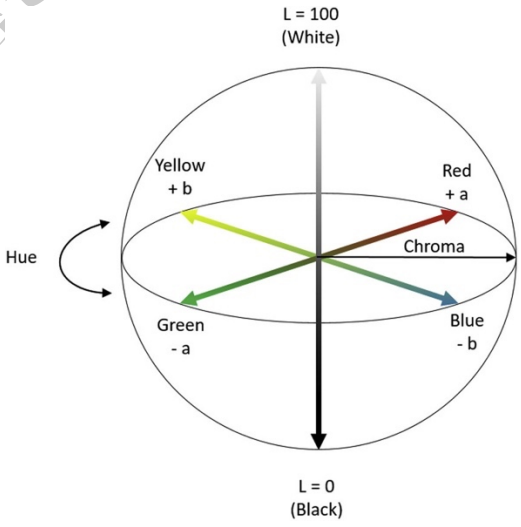

Figure 2. CIELAB colour space diagram (CIE, 1986)

Pizato et al. (2015) analysed the color variations of cooked chicken breasts during storage at temperatures of 4°C and 10°C. It was observed that the chicken breast gradually lost its lightness and redness during storage. However, the yellowness of the chicken breast increased.

Table 2. Values of colour for cooked chicken breast meat stored at 4°C (Pizato et al., 2015)

Storage Time (hours)	L *	a*	b *
0	83.61±0.02	1 .04±0.38	14.22±0.43
24	83.49±0.39	0.73±0.08	14.44±0.71
72	82.49±0.39	0.50±0.05	15.32±0.33
120	79.86±0.54	0.29±0.08	17.31±0.40b
168	78.39±0.75	0.18±0.17	17.97±0.54
216	77.49±0.99	0.13±0.20	18.53±0.02
264	76.73±0.57	0.12±0.30	19.71±0.37
312	76.35±0.79	0.10±0.02	23.34±0.31
360	75.97±0.52	1 .18±0.12	24.14±0.28

Table 3. Values of colour for cooked chicken breast meat stored at 10°C (Pizato et al., 2015)

Storage Time (hours)	L *	a*	b *
0	82.0±1.03	1.22±0.43	12.18±1.16
24	81.85±0.2	-0.2±o.10	13.74±0.71
72	81.55±0.62	-0.47±0.24	14.76±0.28
120	81.28±0.94	-0.58±o.12	15.01±l.12
168	81.08±0.37	-0.81±0.31	15.55±1.28
216	80.66±0.56	-0.95±o.16	15.70±0.35
264	80.25±0.68	1.0±0.49	15.86±0.11

3.1.3.2 Appearance

The appearance of poultry, whether as individual parts or the entire bird, can have a significant impact on a consumer's purchasing decision. Visibly damaged poultry, such as those with bruises, fractured bones, or discoloration, is less likely to sell or may sell for a lower price due to the perception that it is of inferior quality (Kennedy et al., 2004). Numerous poultry grading systems emphasize the appearance of the flesh, including the absence of defects. For instance, in Canada, poultry with bruises or discoloration encompassing more than 6.5 cm2 in the breast

area or 8.0 cm2 elsewhere is downgraded from Grade A to Utility Grade (B), similar to U.S. regulations (Barbut, 2020).

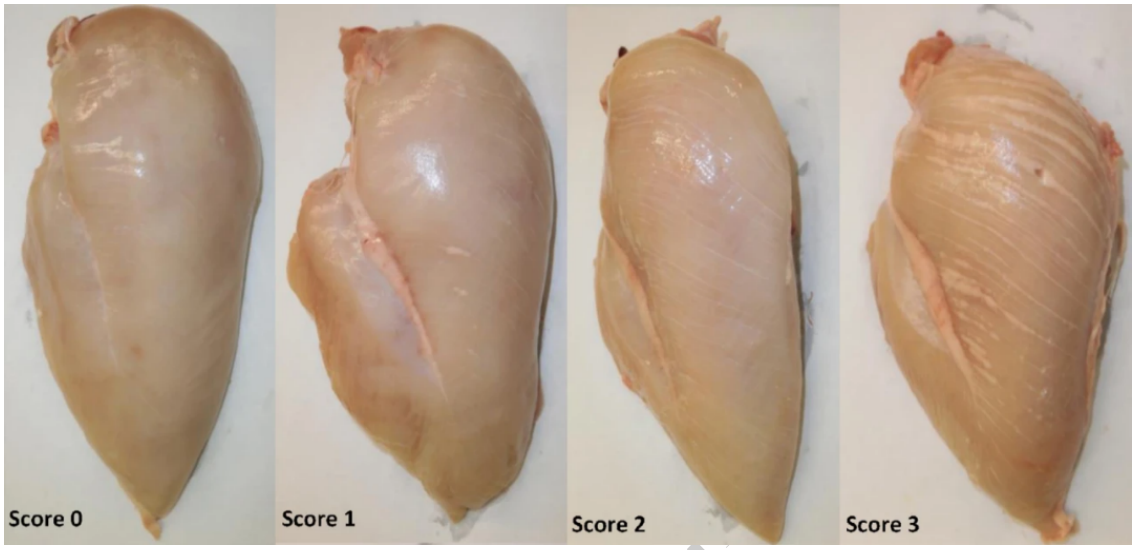

Figure 3. Breast fillets can be classified based on the presence of white striping, with a score of 0 indicating no white striping and a score of 3 indicating a high degree of white striping (Bailey et. al., 2015)

Fresh, uncooked chicken is typically pale pink with white fat and is tender and juicy. It should not odour strongly. The chicken is likely deteriorated if it is slimy, has an unpleasant smell, or has changed colour to yellow, green, or gray. Chicken breasts can be graded based on the presence of white marbling, which appears as white lines running parallel to the muscle fibres. The quantity and thickness of white stripes can vary between chickens. White striping is caused by muscle injury that is replaced by fat and collagen(Bailey et al., 2015; Huang and Ahn, 2019).

3.1.3.3 Texture

Unless there are issues with the initial processing stages, chicken meat that is between 5 and 7 weeks old has a tender texture. These problems can occur if the meat is deboned too early or chilled too rapidly prior to rigor mortis (the natural process by which the muscles of a deceased body become stiff and rigid). It typically begins a few hours after death and can last anywhere from one to three hours for poultry before progressively dissipating, which can cause cold-shortening and make the meat tougher (Barbut, 2015).

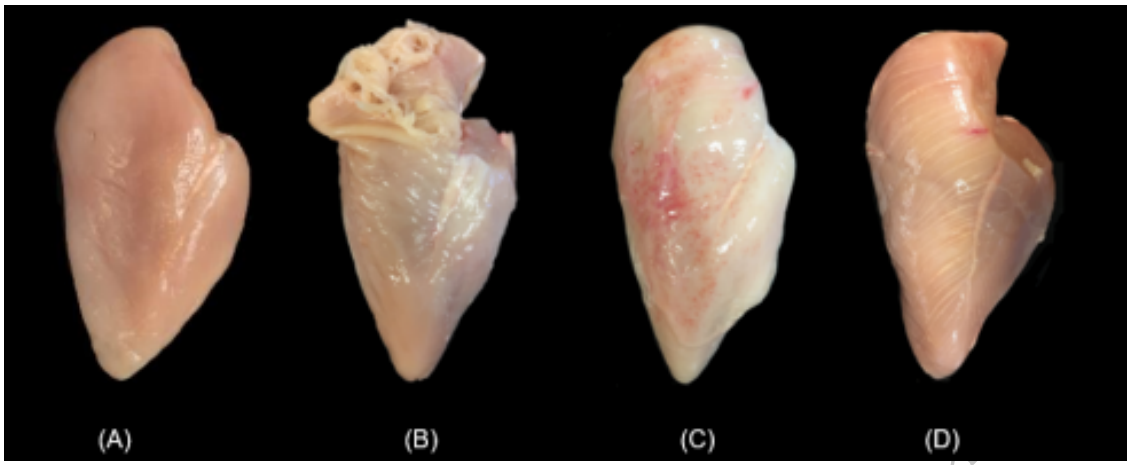

Figure 4. Myopathies in young broilers can manifest in different ways, including a normal fillet (A), spaghetti meat (B), the wooden breast syndrome (C), and fillet with white striping (D) (Barbut, 2020)

Sometimes, the processing of young animals can result in meat that is too soft or has minimal binding. This issue is from the myopathies, which are neuromuscular disorders where muscle weakness due to dysfunctional of the muscle fibre. Such as the spaghetti meat syndrome, which causes the separation of muscle fibres in certain young, rapidly-growing broilers. There are also cases of tough breast fillet meat in young broilers, known as the wooden breast syndrome, in which the Pectoralis major muscle demonstrates degeneration, necrosis, and the accumulation of connective tissue fibres and fat (Petracci et al., 2019). Up to 90% of broiler chickens display woody breast (WB) and white striping (WS), resulting in annual economic losses between $200 million and $1 billion for the U.S. poultry industry (Tijare et al., 2016).

3.1.3.4 Juiciness

The juiciness of meat is the sensation of hydration and lubrication when we chew the product. Chewing accomplishes this by generating saliva and releasing juice (consisting of water and lipids). A higher moisture and/or lipid content enhances the perception of juiciness in meat. If both are prepared in the same fashion, a chicken leg will be more juicy than a chicken breast. This is due to the fact that chicken breast fillets (without skin) contain an average of 74% moisture and 2% fat, whereas chicken leg meat contains 72% moisture and 8% fat (Barbut, 2020).

Using dry heat methods, such as in the oven or on the grill, to cook chicken breast fillets can result in a dry and tough final product. To enhance the moisture content and texture of these cuts, many pre-marinated fresh poultry breast meat products with added water and spices are sold. This can help make the prepared meat more flavourful and juicy. Additionally, the method of storage and chilling (either water or air chilling) can affect the ultimate product's juiciness. Chicken that has been water-chilled may be sold with additional moisture added during the procedure. Depending on the amount of moisture retained (some of which may be lost during storage as drip loss), cooking method, and cooking time, the final product may be perceived as juicier than air-chilled birds (Demirok et al., 2013).

3.1.3.5 Flavour and Aroma

Meat obtains flavour and aroma during cooking as a result of chemical reactions between its various components (such as proteins, fat, minerals, and seasonings). These reactions decompose these constituents and liberate volatile small molecules that interact with other small molecules generated by the elevated heating temperature. The Maillard reaction is a crucial chemical reaction that occurs during the cooking of chicken meat and other types of flesh. It involves the interaction of amino compounds and reducing carbohydrates, which results in the formation of numerous compounds responsible for the flavour of meat (Jayasena et al., 2013). This reaction is a significant factor in the development of the flavour and aroma of prepared meat.

Meat's flavour and aroma can be affected by storage conditions and duration. During meat maturation, endogenous enzymes (such as protease and lipolytic enzymes) degrade various components within the muscle, resulting in tenderization and the formation of small molecules that contribute to the development of aroma. Microorganism activity can also result in the release of enzymes that degrade large molecules (such as proteases that degrade proteins, resulting in an unpleasant odour and discoloration) and polypeptides that show as slime on the product. Due to a chemical reaction between fatty acids and reactive oxygen, rancidity can occur at room temperature (Haugen et al., 2006).

3.1.4 Stability and Safety Aspect of Poultry Meat

As infectious illness can have severe consequences for public health, everyone involved in food production and distribution has a responsibility to prioritize food safety. Meat that contains or is contaminated with microorganisms can deteriorate and transmit diseases such as *E. coli, Salmonella, Listeria, and Campylobacter.* This is a problem because microorganisms can be found on the skin, feathers, and digestive and respiratory tracts of birds entering processing facilities. One gram of intestinal content can contain as many as 108 microorganisms. These bacteria use the nutrients in the meat for energy and produce enzymes and acids as by-products (Bruckners et al., 2012). The effect of temperature on the shelf life of fresh poultry meat is depicted in Figure 5. It demonstrates that as temperature or storage time increases, so does the rate of bacterial growth.

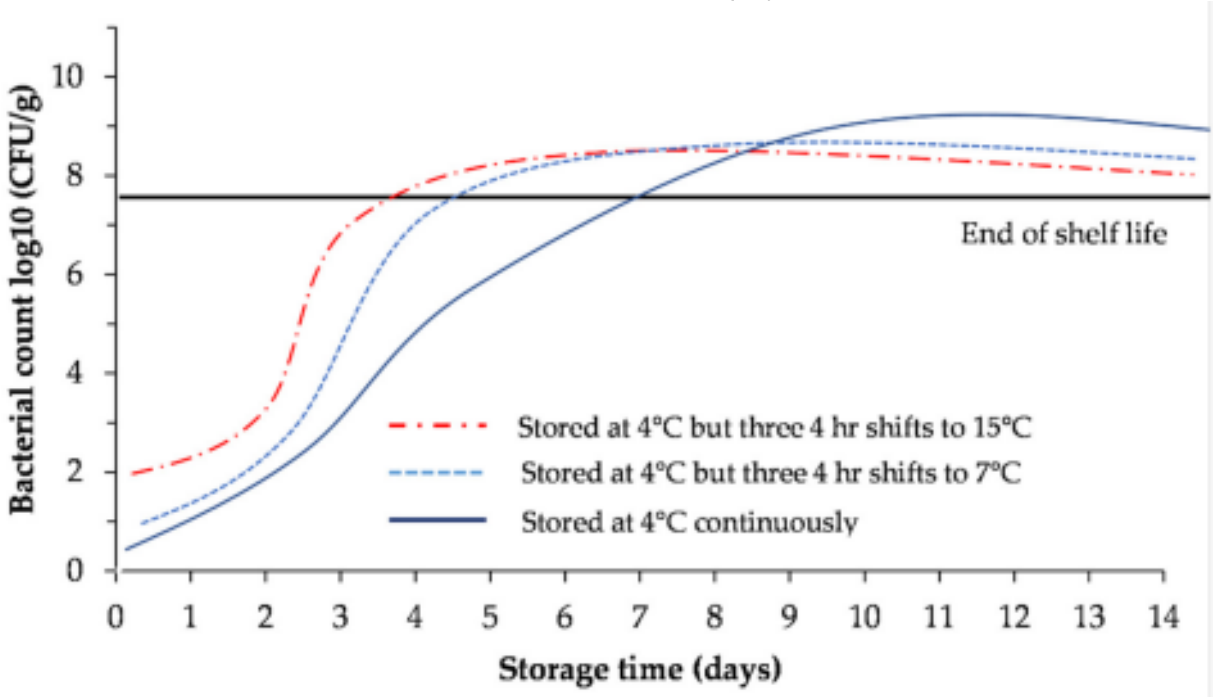

Figure 5. The growth of *Pseudomonas spp*. on poultry during storage was described using the Gompertz model (Bruckner et. al., 2012)

Pale Soft Exudative (PSE) and Dark Firm Dry (DFD) meats are two of the most common quality defects in the meat industry. These defects reduce the consumer acceptability, expiration life, and yield of meat, which has a significant impact on profits. The breed, sex, species, and pre- and post-slaughter management of animals are among the most influential factors for the

presence of PSE and DFD in meats. Premortem tension is the leading cause of both PSE and DFD in meat. The development of PSE is because of the pre-slaughter aggression in animals subjected to brief stress prior to slaughter. DFD meats may be generated when animals are subjected to prolonged or long-term stress prior to slaughter, such as when they are transported long distances. Due to its low water-binding capacity, PSE meat has a pH below 5.5, a pale colour, a soft texture, and an excess of fluid in the muscle tissue. As the rate of glycolysis increases at high temperatures, the pH decreases, which increases protein denaturation and water loss due to structural protein degradation. In contrast, DFD meat has a pH greater than 6.2, is darker in colour, has a firm texture, and contains less water in the muscle tissue. This condition is caused by protracted stress in the animal, which reduces its carbohydrate energy reserves prior to slaughter, resulting in insufficient lactic acid production (Adzitey and Nurul., 2011; Gotardo et al., 2015). These flaws can result in substantial monetary losses for poultry producers. (Leishman et al., 2021) It is crucial that producers manage conditions in their facilities to minimize the risk of meat quality issues.

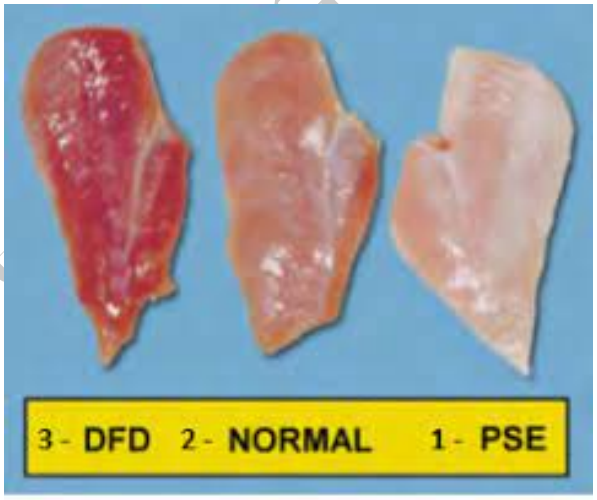

Figure 6. Comparison of DFD, normal, and PSE meat (Gotardo et al., 2015)

Poultry may contain pathogenic microorganisms that can cause illness. To ensure that poultry is suitable for consumption, it must be cooked to the proper internal temperature. The USDA recommends cooking poultry to an internal temperature of 74°C for at least 15 seconds, whereas the Canada Health Authority recommends 85°C (Kozak et al., 2010). However, if the temperature of the heat treatment exceeds 65 degrees Celsius, the meat may lose some of its

juiciness and tenderness. To ensure that poultry meat is safe to consume and has an acceptable texture, it must be properly cooked (Thompson et al., 2005). When deciding what to purchase, shelf life is an essential consideration for consumers because products with a longer shelf life are more convenient. Fresh poultry meat, like other meats, is perishable because it supplies the nutrients necessary for the growth of microorganisms and lacks natural defences such as lysosomes in eggs or low pH in citrus fruits. According to Adamski et al. (2013), many consumers prefer to purchase fresh meat rather than preserved.

There are many techniques for extending the shelf life of flesh. The use of high levels of sodium, drying to reduce water activity, cooking at high temperatures (such as for canning to create shelf-stable products), and smoking (which introduces antimicrobial compounds such as phenols and aldehydes) are examples. Meat can also be irradiated using an electron beam or isotopes such as cobalt, but this requires the use of expensive, specialized facilities (Barbut, 2015). In addition, processed meat products (such as prepared, ready-to-eat meat) are frequently packaged to preserve their quality and extend their shelf life. This can be accomplished by utilizing packaging techniques that generate an oxygen barrier or prevent lipid oxidation. Examples of methods for enhancing the barrier include vacuum packaging and modified atmosphere packaging. Various processed poultry products are sold in this type of packaging, which can be advantageous for consumers due to its reseal ability, microwave safety, and options for single servings or family-sized portions (Barbut and Leishman, 2022).

3.15. Convenience Aspect of Poultry Meat

Today's consumers value convenience and are willing to pay a premium for products that are simple to use and require minimal preparation. Consumers spend significantly less time preparing food nowadays. In particular, they spend approximately one-sixth as much time preparing food as they did 50 years ago (Barbut, 2020). This demand for convenience has encouraged innovation in the food industry, with a concentrate on developing products that can be eaten on the go. Due to their versatility and fast cooking time, poultry products such as chicken fillets are frequently viewed as convenient options (Kennedy et al., 2004). The availability of pre-prepared sauces and marinades only adds to their convenience. Moreover, poultry meat is widely accepted in all societies and is not subject to the same religious restrictions as other types of meat.

There are several reasons why poultry is less expensive than other types of flesh. The feed conversion rate, which refers to the efficacy with which an animal converts feed into weight gain, is one of the most important factors. In general, poultry has a higher feed conversion rate than other livestock. For example, chicken has a conversion rate of 2.5, while pork and beef have conversion rates of 5.0 and 10.0, respectively. Additionally, poultry has a much shorter growth period than other livestock, which serves to reduce production costs (Smil, 2002).

3.2 Sous-vide Technology

3.2.1 Definition of Sous-vide Technology

Sous-vide is a method of cooking that entails vacuum-sealing uncooked or partially cooked food in heat-resistant, food-grade plastic bags and cooking it with precisely controlled heat. This technique differs from conventional cooking methods in that the food is vacuum-sealed and cooked at a precise temperature. Sous-vide cookery involves vacuum-sealing raw or partially cooked ingredients in plastic bags without additives or preservatives to preserve their freshness and minimize processing. The pouches are vacuum-sealed to eliminate air, and then the food is cooked using precise temperature control, typically through hot air, steam, or water immersion. This method has a number of benefits, including: 1) reduced risk of contamination, 2) efficient heat transmission and cooking in the food's own juices, 3) minimal loss of flavours, aromas, and nutrients, and 4) avoiding oxidation due to the near-complete removal of oxygen from the pouch (Baldwin, 2012).

In recent years, sous-vide cooking has become increasingly widely used. This trend includes the use of sous-vide cookery at lower, milder temperatures (between 42 and 60 degrees Celsius). This method of cooking is commonly considered to be more considerate of the nutritional properties of food because it helps to preserve the natural flavours, aromas, and hues of the ingredients. In addition, the textures of food prepared using sous-vide techniques at lower temperatures are frequently more tender than those of food cooked using conventional methods. As a result of the low temperature of sous-vide cooking, the survival of microbes in food is a concern in terms of the microbial safety of the products (Stringer et al., 2012).

Figure 7 illustrates the various phases of sous-vide cooking from a cooking standpoint. These stages can be categorized into two major groups: "cook-serve" and "cook-chill," also referred to as "direct" and "indirect" cooking. The term "cook-serve" refers to the processing of foods

that require low cooking temperatures and quick cooking periods and are intended for immediate consumption. In contrast, "cook-chill" entails heating ingredients at low temperatures (60-65°C) for extended periods of time (2-8 hours) in order to soften tough foods and allow for their storage in a chiller. If tender foods, such as chicken breast, must be preserved, shorter cooking periods may be used for "cook-chill." In all stages of sous-vide cooking, precise heating is necessary to guarantee food safety and prevent overcooking (Brugalla, 2019).

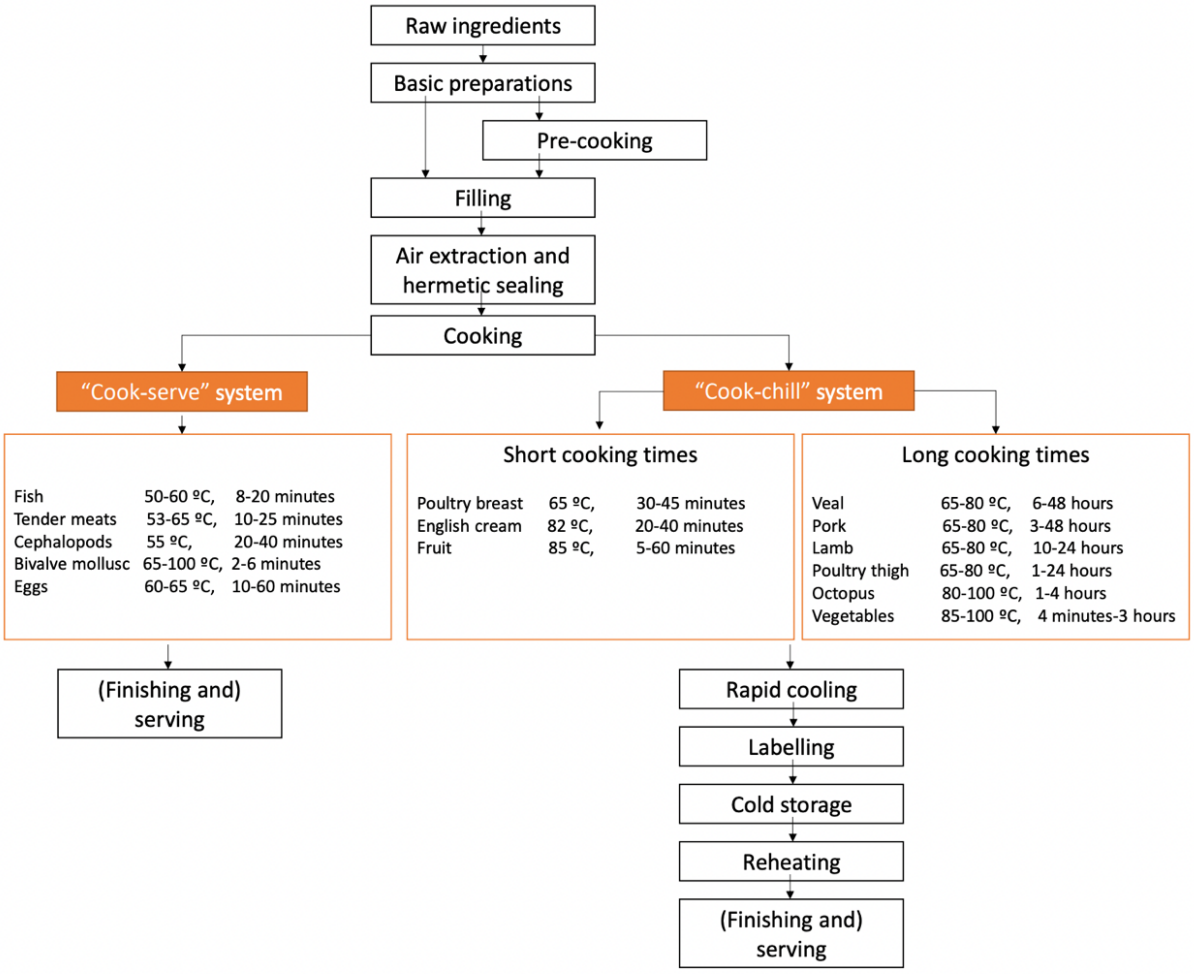

Figure 7. Flow diagram of sous-vide cooking (Brugalla, 2019)

Another method for implementing the sous-vide treatment is known as the double-step method. The double-step method requires the food to be cooked at two different temperatures, whereas the standard method only requires one temperature. Studies have demonstrated that the double-

step method improves texture and reduces cooking loss in chicken breast, while also reducing lipid oxidation and increasing protein solubility (Hasani et al., 2022). Hasani et al. (2023) also concluded that the two-step sous vide cooking method for chicken breasts demonstrated acceptable oxidative and microbiological stability during both chilled and frozen storage, similar to the one-step method. These results suggest that the two-step heat treatment may be a viable alternative cooking method for enhancing the quality properties of chicken breasts without diminishing their shelf-life.

3.2.2 Effect of Sous-vide Treatment on Moisture Content and Cooking Loss

The moisture content of a substance is the quantity of water it contains. It is frequently expressed as a percentage of the substance's moist weight to its dry weight. The moisture content of food products is an essential consideration, as it can impact the product's quality and shelf life. Temperature and duration of cooking can affect the moisture content of a food product. Higher cooking temperatures can alter the structure of the meat's protein molecules, altering the meat's porosity and moisture content. When making purchasing judgments, the moisture content of a food product can be crucial from the consumer's perspective. For instance, consumers may prefer meat products with a higher moisture content that results in juiciness (Dominguez-Hernandez et al., 2018).

Cooking loss is the weight reduction of a dietary item that occurs during cooking. This can be caused by a number of factors, including the loss of moisture or lipids, volatile compounds, or other components through leaching or evaporation. Cooking loss can be a significant factor in the preparation and processing of food products, as it can impact the final product's yield and quality. Due to the loss of water and various water-soluble compounds, such as salts, proteins, and polyphosphates, as well as aromatic compounds, heating loss increases as the cooking temperature rises (Offer et al., 1984).

Table 4. Comparation of moisture content (%) and cooking loss (%) of sous-vide treatment with other cooking methos (%) (Ayub and Ahmad, 2019)

	Sous-vide	Boiling	Deep Frying	Hot plate cooking	Oven	Pan frying
Moisture (%)	81	65.9	64.9	68.5	65.9	68.7
Cooking Losses (%)	19	50.2	44.64	32.86	35.45	33.03

As sous-vide cooking requires cooking food at low temperatures, moisture and cooking loss in meat products can be minimized by minimizing protein denaturation. In comparison to conventional cooking methods, sous-vide cooking reduces chicken's moisture loss by 15 to 30 percent, as opposed to 30 to 40 percent for other cooking methods. (Botinestean et al., 2016) Sous-vide reduces cooking loss in chicken samples by four times compared to conventional cooking methods. Additionally, this procedure increases product yield by 19%. Sous-vide preparation is frequently used to preserve flavours and volatile compounds. While moisture loss can concentrate the nutrients in meat, it can also result in the loss of heat-sensitive nutrients and fluids containing specific nutrients (Ayub and Ahmad, 2019).

3.2.3 Effect of Sous-vide Treatment on Colour

The colour of chicken meat can vary depending on the diet and living conditions of the chicken. Generally, the meat of a chicken is pale in colour when raw, with a slightly pink or yellow tinge. Cooked chicken meat can vary in colour, ranging from white to brown, depending on the cooking method and temperature. Chicken meat that has been cooked to a safe internal temperature should be white or pale in colour. Da Silva et al. (2019) classified cooked chicken breast into three categories based on lightness: pale (L* > 53), normal (L* = 46–53), and dark (L* < 46). If the chicken meat is dark or pink, it may not be fully cooked and should be returned to the heat for further cooking. It is important to properly cook chicken to ensure that it is safe to eat (Nyam et al., 2023).

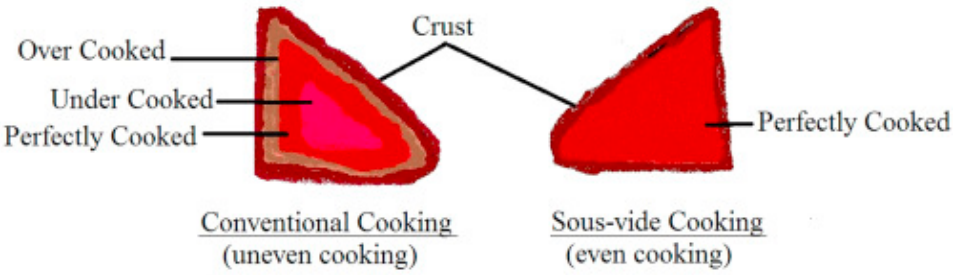

Figure 8. Meat cooked illustration in conventional cooking comparing with sous-vide cooking (Ayub and Ahmad, 2019).

Meat prepared to a higher level of doneness tends to be somewhat dry and appear greyish-brown in colour. In contrast, meat prepared to a moderate degree at low temperatures tends to be moist and appear pinkish-red. (Ayub and Ahmad, 2019) Sous-vide cooking enables a greater degree of doneness to be accomplished when meat is cooked for a longer amount of time (6-24 hours), resulting in even cooking. As heating temperature increases, redness values in meat, such as lamb longissimus and beef semitendinosus, tend to decrease. Meat prepared for longer periods of time tends to have slightly greater yellowness (Dominguez-Hernández et al., 2018). Due to the fact that it endures less browning during the cooking process, sous-vide chicken is typically lighter in colour than raw chicken or chicken cooked via grilling or frying.

Maillard reactions, which contribute to the browning of meat, typically occur at temperatures above 140°C, so they are more unlikely to occur in sous-vide chicken than in grilled or fried chicken (Silva et al., 2016). Due to the existence of deoxy myoglobin, a heat-resistant form of meat protein, Naveena et al. (2017) discovered that sous-vide-cooked chicken has a higher redness (a*) value than other cooking techniques. Some consumers may find sous-vide-cooked chicken's lighter colour alluring. According to Ayub and Ahmad (2019), sous-vide-cooked meat typically has greater b* values. This increase in b* values may be attributable to an increase in metmyoglobin levels, which can result in the production of meat with a brownish hue.

3.2.4 Effect of Sous-vide Treatment on Texture Properties

Depending on the muscle and the cooking procedure, the texture of cooked chicken meat can vary. Pectoral muscle-derived (chicken breasts) are typically lean and have a firm, fine-grained texture. Thighs and drumsticks, which are derived from the leg muscle, are typically more juicy and tender. Temperature is a crucial element to consider when preparing meat, as it can alter the protein structure and texture. When meat is cooked at higher temperatures, the proteins can denature, resulting in a texture change. Myofibrillar proteins, which are responsible for muscle contraction and are found in the muscle fibres of meat, denature between 55 and 60 degrees Celsius. The denaturation temperature of sarcoplasmic proteins, which are present in the fluid surrounding muscle fibres and play a role in muscle metabolism, is between 50 and 70 degrees Celsius (Yu et al., 2017).

Five parameters are typically used to determine texture properties: rigidity, springiness, gumminess, cohesiveness, and chewiness (toughness). The hardness of a product is its resistance to compression. Hard foods are difficult to bite or chew and may require additional

force to break down. A food's springiness is its ability to regain its original structure after being deformed. The texture of springy foods is springy or elastic. The gumminess of a product indicates its viscoelasticity, or its ability to flow and resist deformation. Gummy foods have a sticky consistency and may feel dense or weighty in the mouth. The cohesiveness of a product indicates how well it retains its shape between the first and second chew. Chewiness (toughness) refers to the resistance of a food to being broken down by the molars. Chewy foods require additional chewing or biting to be broken down and may have a dense or firm texture. According to Ayub and Ahmad (2019), when compared to conventional cooking, sous-vide improves texture characteristics by increasing springiness and decreasing hardness, cohesiveness, and toughness.

3.2.5 Effect of Sous-vide Treatment on Lipid Oxidation

There are a variety of lipids, or fats, in chicken meat, including saturated fats, monounsaturated fats, and polyunsaturated fats. Depending on the part of the bird and the method of preparation, the specific types and quantities of lipids in chicken meat can vary. Chicken is an excellent source of polyunsaturated fats, such as omega-3 fatty acids, and contains less saturated fat than red meat (Marangoni et al., 2015).

Lipid oxidation is a chemical process that occurs in poultry and other meats in which the lipids (fats and oils) are decomposed and become rancid. This procedure has the potential to alter the flavour, texture, and nutritional value of the meat, as well as produce harmful compounds. Lipid oxidation is affected by a variety of factors, including the presence of oxygen, heat, light, and metal ions, as well as the lipid type and quantity. Due to the application of sous-vide at low temperatures and under vacuum conditions, this technique can be used to effectively reduce lipid oxidation in chicken products, thereby extending their expiration life. The changes in fatty acid content of sous-vide chicken are typically less pronounced than those resulting from conventional cooking methods such as frying and grilling (Silva et al., 2016).

3.2.6 Effect of Sous-vide Treatment on Protein Solubility

Solubility of proteins refers to their capacity to dissolve in aqueous solutions. The solubility of proteins in chicken meat can be affected by a number of factors, including the type and quantity of proteins present, the pH and temperature of the surrounding environment, and the presence of other substances. Protein solubility can affect the texture, flavour, and functional properties

of chicken meat, as well as its nutritional value and potential food processing applications. Proteins with a high solubility may be easier digested and absorbed by the body, whereas proteins with a low solubility may contribute to the toughness and chewiness of meat (Nahar et al., 2014).

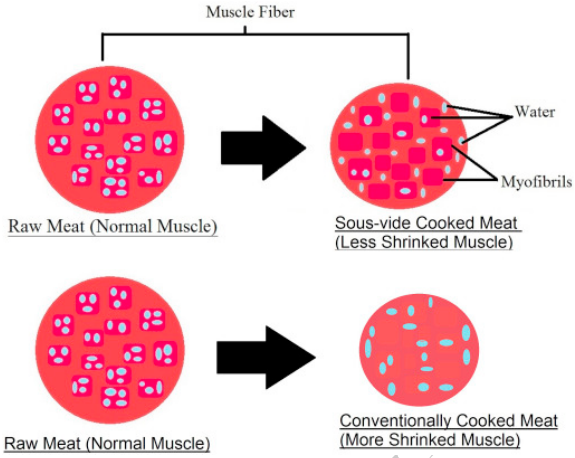

Figure 9. Comparison of muscle between WHC (water holding capacity) of raw meat with high water holding capacity) against sous vide cooked meat and conventional cooked meat (Ayub and Ahmad, 2019).

Meat's texture, colour, flavour, and juiciness are significantly determined by protein modifications. Some proteins are denatured at temperatures above 60°C, which causes the meat structure to contract, resulting in weight and moisture loss. Nonetheless, low-temperature sous-vide cooking results in less protein denaturation and meat structure shrinkage compared to conventional cooking, where high-temperature protein denaturation reduces the water-holding capacity and causes muscle shrinkage (as shown in Figure 9). Changes in myofibrils affect the tenderness of meat, whereas intramuscular connective tissues determine its rigidity. Due to the higher water-holding capacity of intramuscular connective tissues, sous-vide cooking typically yields more tender flesh. Nevertheless, protein denaturation can occur at any temperature if exposed for a longer amount of time (Dominguez-Hernandez et al., 2018; Ayub and Ahmad, 2019).

Myofibrillar and sarcoplasmic proteins are two major categories of proteins that contribute to the structure and function of muscle tissue. The sarcoplasm, which is the fluid-filled space surrounding the myofibrils in a muscle cell, contains the sarcoplasmic proteins. The primary

function of sarcoplasmic proteins within the muscle cell is to store and transport nutrients and other substances. Enzymes, structural proteins, and transport proteins are all sarcoplasmic proteins. Myofibrillar proteins, on the other hand, are located within the myofibrils, which are long, thin filaments of protein that comprise the contractile portion of muscle cells. Myofibrillar proteins' primary function is to generate force and movement through the process of contraction. The principal myofibrillar proteins are actin and myosin, which are organized in a repeating pattern along the length of myofibrils (Dominguez-Hernández et al., 2018). As sous-vide treatment occurs at lower temperatures than conventional cooking, the denaturation of these two types of proteins will be reduced, resulting in an increase in protein solubility in meat products (Croptova et al., 2019).

3.2.7 Effect of Sous-vide Treatment on Sensory

3.2.7.1 Effect of Sous-vide Treatment on Colour Acceptance

Colour is a vital factor of consumer acceptance. It indicates the product's doneness and the consumer's perception of the product's safety and quality (King and Whyte, 2006). The colour of meat is due to the presence of myoglobin, which exists in three forms: oxymyoglobin (brilliant red), deoxymyoglobin (purple-red), and metmyoglobin (brown) (Kurp et al., 2022). (Naveena et al., 2017) found that the sous-vide technique has a higher redness (a* value) than traditional cooking techniques. The reason for this is that myoglobin begins to denature between 55°C and 65°C, and is completely denatured at 80°C (Geileskey et al., 1998).

As the temperature increases, the redness of the meat decreases. Despite the red colour associated with undercooked meat, the sous-vide cooking colour was still tolerated in pork chops products (Honegger et al., 2022). The pink hue of meat may be interpreted by consumers as a sign of undercooked or unsafe flesh, resulting in dissatisfaction. Hong et al. (2015) reported that consumers do not generally favour this colour. Holownia et al. (2003) established a pink colour threshold with an a* value of 3.80.

3.2.7.2 Effect of Sous-vide Treatment on Flavour and Aroma

The cooking temperature and time are essential for the formation of flavour and aroma volatiles in cooked meat products, as these volatile compounds originate from two primary reactions: the degradation of amino acid components (including the Maillard reaction) and the thermal degradation of lipids (Elmore and Motta, 2006). As sous-vide is an LTLT (long time and low

temperature) technique, the Maillard reaction does not occur because the minimum temperature for sous-vide cooking is 140°C, so there is no browning and no development of flavour based on Maillard reaction. The sous-vide technique is more conducive to the hydrolysis of proteins and peptides, which increases water-soluble free amino acids and adenosine monophosphate concentrations, which are then set to be released into the a liquid, resulting in an abundance of umami flavour-active compounds in the meat. As sous-vide cooking occurs under vacuum conditions, it prevents fat oxidation and retains flavour volatiles and moisture during the preparation process (Gil et al., 2022). Thus, minimal flavour and aroma loss occurred (Offer et al., 1986; Baldwin, 2012. The sensory evaluation comparing sous vide to other cooking techniques for meat concluded that sous vide-prepared flesh was more delicate to the consumer (Honegger et al., 2022).

3.2.7.3 Effect of Sous-vide Treatment on Tenderness and Juiciness

Tenderness is how the consumer perceived ease of consumption, as well as the disorganization of the flesh structure during mastication. The heat treatment is associated with the denaturation of myofibrils and solubilization of connective tissue proteins (Ayub and Ahmad, 2019). According to Akoglu et al. (2018), meat contains endogenous proteolytic enzymes that are most active between 40 and 50 degrees Celsius. The higher desmin (a muscle-specific filament that is essential for maintaining the proper structure and function of muscles) degradation observed at these temperatures is attributable to the increased activity of these enzymes, which in turn improves the tenderness of the meat. Ismail et al. (2019) discovered that a two-stage sous vide method employing temperatures ranging from 45 to 50°C produced better texture characteristics, such as decreased shear force, increased softness, and enhanced chewiness, when compared to the conventional single-stage sous vide cooking method.

Moreover, juiciness is strongly correlated with the moisture content and water distribution of samples, which will be released by flesh after the initial bite and during the chewing process (Alvarez et al., 2021). Sous-vide cooking increases consumer adoption by delivering a juicier sensation while retaining traditional sensory characteristics and flavour (Cheng et al., 2022). The sensory characteristics that had the greatest impact on the overall acceptability of sous-vide-cooked meat were tenderness and juiciness. Due to its tenderness and juiciness, cooking at a sous-vide temperature received a higher rating than other heat treatments (Honegger et al.,

2022). As little as two hours of low-temperature cooking produces favourable results in these sensory attributes (Gil et al., 2022).

3.2.8 Safety Aspect of Sous-vide Technology

Salmonella and *Listeria monocytogenes* are two main foodborne pathogens from poultry that can cause illness in humans, according to EFSA (2018). *Salmonella* is the second most frequently reported zoonosis in the European Union, with *S. Enteritidis* being the most prevalent serotype. Listeriosis, on the other hand, has the highest hospitalization and mortality rates of all zoonotic diseases and is most prevalent in elderly individuals, especially those over 84 years old. 2017 hospitalization and mortality rates for salmonellosis and listeriosis among confirmed human cases in the EU are presented in Table 5.

Table 5. Reported *Salmonella* and *Listeria monocytogenes* cases in the European Union in 2017 (EFSA, 2018)

	Salmonelosis	**Listeriosis**
Confirmed cases	91,662	2,480
Notification rate per 100,000 population	19.7	0.48
Status available (%)	43.1	40.4
Reporting Member States	14	16
Reported hospitalised cases	16,796	988
Proportion hospitalised (%)	42.5	98.6
Outcome available (%)	67.8	65.8
Reporting Member States	17	18
Reported deaths	156	225
Case fatality (%)	0.25	13.8

The European Chilled Food Federation recommends that sous-vide foods be pasteurized by heating to at least 70°C for 2 minutes in order to achieve a 6-log reduction in *Listeria monocytogenes* (z-value = 10 C) (SVAC, 1991). According to Doyle et al., (2001), *Listeria monocytogenes* can withstand higher temperatures (has greater heat resistance) than many other non-sporulating foodborne pathogens, such as *Salmonella spp.* and pathogenic *Escherichia coli.*

Therefore, eliminating *Listeria monocytogenes* suffices to eradicate *Salmonella spp*. Using a heat treatment of 60°C for 90 minutes to control the proliferation of *Salmonella spp.* and *Listeria monocytogenes* in "cook-chill" sous-vide meat products may be safe and effective. (Brugalla, 2019). Endrit et al. (2023) explored the effects of Enterococcus species on sous vide treatment in a recent study. Compared to other pathogenic bacteria, such as *Escherichia coli* O157: H7, *Salmonella enterica*, and *Listeria monocytogenes*, it is known that these microorganisms are highly resistant to heat. Both one-step and two-step sous vide treatments were found to be safe for pasteurization levels, as the heat-resistant microorganism *Enterococcus faecalis* was inactivated. Specifically, the one-step sous-vide treatment at 60°C for 120 minutes eradicated *Enterococcus faecalis* from chicken breast. Additionally, chicken breasts prepared at 50°C and 60°C for 120 minutes met the pasteurization outcome criterion of a 3-log decrease in *Enterococcus faecalis* in chicken breast.

3.2.9 Effect on Storage Stability of Sous-vide Technology.

During sous-vide cooking, the combination of time and temperature during heat treatment and storage must be precisely regulated to ensure that the food remains microbiologically safe for the duration of its shelf life. In order to accomplish this, the food's temperature is monitored throughout the cooking procedure to determine the level of bacterial inactivation. This ensures that the food is safe to consume at the expiration of its shelf life. Table 6 presents time and temperature combinations for heat treatment and storage of food, based on the microorganisms that must be eliminated to assure the food safety of sous-vide products.

Table 6. Heat treatments and shelf-life recommended or suggested for cooked chilled foods (Brugalla, 2019)

Organism	**Target log reduction**	**Core heat treatment**		**shelf-life**	
		°C	**Min**	**days**	**°C**
Listeria monocytogenes	6	65	15	10	4
				4	8
		70	2	5	7
				10	6

Chilling and freezing meat as a method of preservation is an effective means to preserve the chemical, organoleptic, and nutritional properties of the product as closely as possible to their original state and to prevent the harmful effects of microorganisms and enzymes. Bacteria can multiply quickly if chicken is stored at temperatures above 4°C, increasing the risk of foodborne illness. When meat is stored at low temperatures (0-4°C), its water-holding capacity and thawing-cooking loss are enhanced. Bacterial growth is slowed, and the percentage of occurrence of *Salmonella spp*. showed inconsistencies (Fernandes et al., 2016).

Nyati (2000) monitored the quality of certain sous-vide products during a five-week storage period at both cold (3°C) and mildly cold (8°C) temperatures. When stored at the recommended cold temperature, the sous vide products demonstrated minimal microbial growth and maintained their flavour throughout the entire storage period for all 19 products studied, and none of the processed samples contained harmful microorganisms such as *Listeria monocytogenes, Salmonella, Clostridium perfringens, Bacilluscereus, and Enterobacteriaceae.* However, after three weeks of storage at mildly cold temperatures and exposure to temperature abuse, some products had unacceptable levels of microorganisms. Hasani et al. (2023) discovered that chicken breasts prepared using the two-step sous vide method remained microbiologically stable for 21 days when tested for *Enterococcus faecalis* and total mesophilic aerobic counts at 4°C and 20°C, respectively. However, chicken breasts preserved at 10 degrees Celsius were not as stable. Therefore, it can be concluded that the two-step sous vide cooking method produced chicken breasts with acceptable oxidative and microbiological stability during both refrigerated and frozen storage, similar to those produced by the one-step sous vide cooking method.

Freezing chicken meat can cause texture and appearance changes. Freezing causes the formation of ice crystals within the flesh, which can damage the muscle fibres. This can cause the meat to lose moisture and become less tender when thawed. Due to alterations in their structure and function, the freezing of chicken meat can reduce the solubility of proteins, including actomyosin. However, in some instances, the chilling process may increase the solubility of certain types of proteins, such as sarcoplasmic proteins. Various factors, such as the specific properties of the proteins, the storage conditions, and the interactions with other molecules or substances present in the meat, can contribute to this (Nahar et al., 2014).

4. MATERIALS AND METHODS

4.1 Experimental Design

The objective of this study was to investigate the impact of using a two-step temperature ratio on various time ratios during sous vide cooking of chicken breast, as well as the impact of storage time and temperature. Samples of chicken breast were cooked at one temperature treatment of 60°C and two temperature treatments consisting of a lower temperature treatment of 50°C as the first step and a higher temperature treatment of 60°C as the final step, in different time ratios of the same total treatment time (see Table 7). The study was established using a two-way complete randomization strategy, with three replicates of each sous vide treatment. Chicken pectoralis major musculature (24 h post-mortem, pH24 = 5.83 0.12) were obtained from a Budapest, Hungary slaughterhouse. The chicken breasts were then trimmed of excess fat and connective tissue and cut into 2-centimeter-thick segments weighing approximately 105±10 g each. By applying a vacuum sealer (Multivac C100, MULTIVAC Sepp Haggen muller SE & Co. KG, Germany), chicken breasts were vacuum packaged in PA/PE packaging (200x250 mm^2).

Table 7. Group of treatment of the samples

Group	Time at the Temperature of 50 ◦C (min)	Time at the Temperature of 60 ◦C (min)	Treatment Time Ratio 50 ◦C/60 ◦C	Total Treatment Time (min)
T1	0	120	0:1	120
T2	40	80	1:2	120
T3	60	60	1:1	120

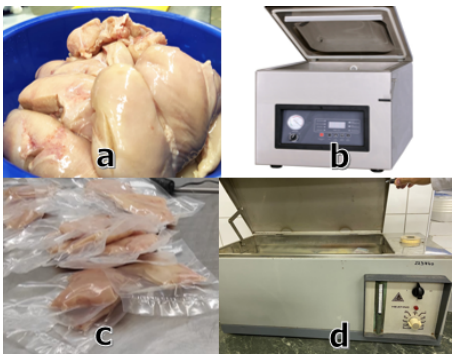

Figure 10. a) raw materials of chicken breast, b) vacuum sealer, c) vacuumed raw materials, and d) water bath.

In this investigation, the sous vide process was carried out using two thermostatic water baths (Laboratory Water Bath LP507/01). Using a Type-T thermocouple positioned in the centre of the chicken breast samples, the internal temperature was monitored throughout the sous vide cooking procedure. As recommended by the Centres for Disease Control and Prevention (2017), the chicken breast samples were chilled in ice-water (1°C) and stored in cold conditions (2°C) for 6 hours to attain a temperature below 4°C. By rapidly reducing the chicken's temperature in ice water, the development of bacteria can be inhibited. Moreover, ice shocking can halt the cooking process, preventing the poultry from becoming overcooked and dry.

The stability of storage for sous vide-cooked chicken breast samples at both one-step and two-step temperature methods was evaluated while they were stored in chilled temperature (4±0.5°C) and mild chilled temperature (10±0.5°C) for a maximum of 21 days, with the storage temperature constantly monitored. On days 0, 7, 14, and 21, samples were collected for analysis of moisture content, cooking loss, colour, lipid oxidation, and aroma acceptability.

4.2 Moisture Content and Cooking Loss

The standard AOAC International 950.46 method (2005) was utilized three times to determine the moisture content of chicken breasts. Approximately 4 grams of the cooked chicken breast were weighed and then dehydrated in an air-forced oven ((Labor Müszeripari Müvek, Hungary) at 105 degrees Celsius for sixteen hours. The relative humidity was calculated using equation (2).

$$\text{Moisture content (\%)} = (\text{dried sample weight} / \text{raw sample weight}) * 100 \quad (2)$$

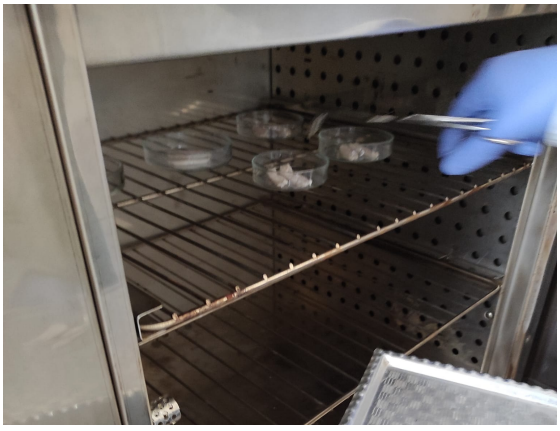

Figure 11. Dry oven process of the samples.

For the cooking loss, the initial weight of chicken breast samples was determined. The sample's water was removed from its packaging and weighed following treatment. Equation 3 was used to calculate the weight loss percentage.

$$\text{Cooking loss (\%)} = (\text{initial weight-final weight})/\ \text{initial weight} \times 100 \qquad (3)$$

4.3 Colour Measurement

Using the CIELAB grading system, the colour characteristics of the chicken breasts were evaluated. After calibrating the instrument, samples were measured for their luminosity (L*), redness (a*), and yellowness (b*) using a Konika Minolta Sensing CR-410 colorimeter. Five parallel measurements were performed on each sample (three samples total).

Figure 12. CR-410-type colorimeter

4.4 Lipid Oxidation

The TBARS (thiobarbituric acid reactive substances) method was modified slightly from Dias et al. (2013). sample in order to measure the lipid oxidation in chicken breasts. Using the TBARS method, the following procedures were taken to measure the lipid oxidation in chicken breasts:

1. Using a Digital Ultra-Turrax homogenizer, a 5-g sample of chicken breast was homogenized with 20 mL of 5% TCA and 0.5 mL of the antioxidant BHT for 2 minutes.
2. 10 minutes were spent centrifuging the mixture
3. The supernatant was filtered with No. 1 filter paper and adjusted to 25 mL in volumetric flasks with 5% TCA.
4. In glass tubes, combine 2 mL of the filtrate with 2 mL of 0.08% TBA.
5.Heated the glass tubes for 30 minutes at 95°C in a water bath.
6. cooled and vortexed the glass tubing.

7. Measuring the absorbance at 532 nm with a UV-visible spectrophotometer type U 2900 against a blank mixture of 5% TCA and 0.08% TBA in 2 mL.

8. The TBARS (mg MDA/kg sample) results are expressed as milligram malondialdehyde per kilogram of meat sample based on the average of triplicate measurements.

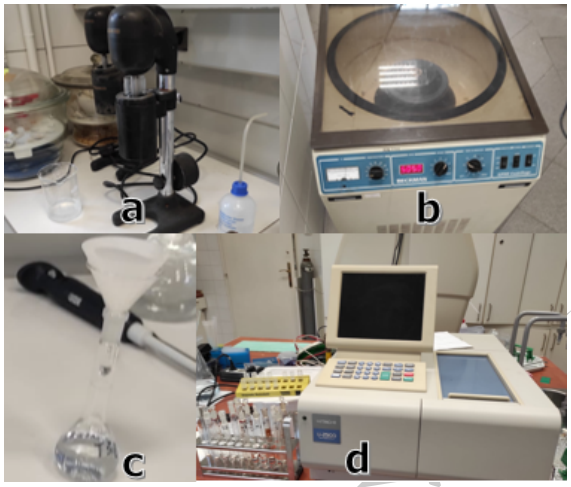

Figure 13. a) homogenizer, b) centrifuge machine, c) filtration process, and d) UV-Vis spectrophotometer

4.5 Odour Acceptability

The samples were opened under aseptic settings, and the odour was evaluated using the technique outlined by Youssef, Gill, and Yang (2014). The samples were graded on a five-point scale for their odour acceptability: 1 for acceptable, 2 for slightly acceptable, 3 for neither acceptable nor unpleasant, 4 for slightly unacceptable, and 5 for unacceptable. The panelists were consisted of five people.

4.6 Statistical Analysis

The statistical analysis of the data was performed with IBM SPSS (27.0). The research applied descriptive analysis and analysis of variance (ANOVA) to evaluate the effects of treatment, storage time, and storage temperature on a number of variables, such as moisture content, cooking loss, color parameters (L*, a*, b*, and E), TBARS, and odor acceptability. Using post hoc Tukey's, further analysis was conducted to determine the differences between groups. The error bar was represented by standard deviations in a bar chart designed in Microsoft Excel 2021 software.

5. RESULT AND DISCUSSION

5.1 Moisture Content and Cooking Loss

Moisture content refers to the quantity of water present in a substance or material, expressed as a percentage of the weight of the dried sample relative to the weight of the raw sample. It is a crucial parameter in many sectors, including the food industry, because it can impact product quality, shelf life, and safety. Using the AOAC International 950.46 standard method (2005), the moisture content of the sous-vide cooked chicken breasts was determined. Cooking loss, also known as weight loss or drip loss, is the weight of food wasted during cooking. Typically, it is expressed as a percentage of the difference between the initial and final weights, divided by the initial weight. Figures 14 and 15 represent the findings for moisture content, while figures 16 and 17 represent the results for cooking loss.

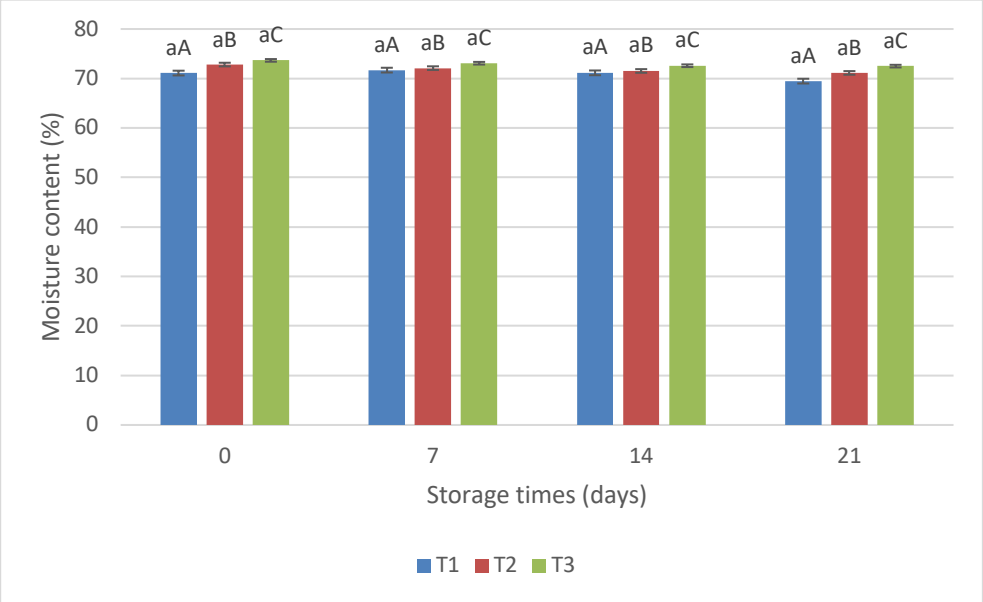

Figure 14. The moisture content (%) of sous-vide cooked chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

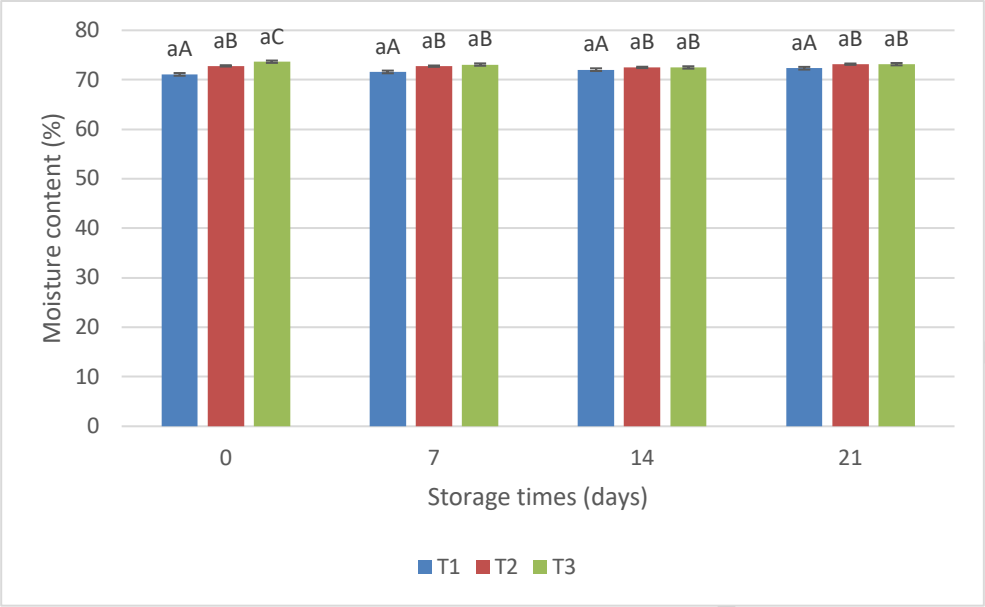

Figure 15. The moisture content (%) of sous-vide cooked chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

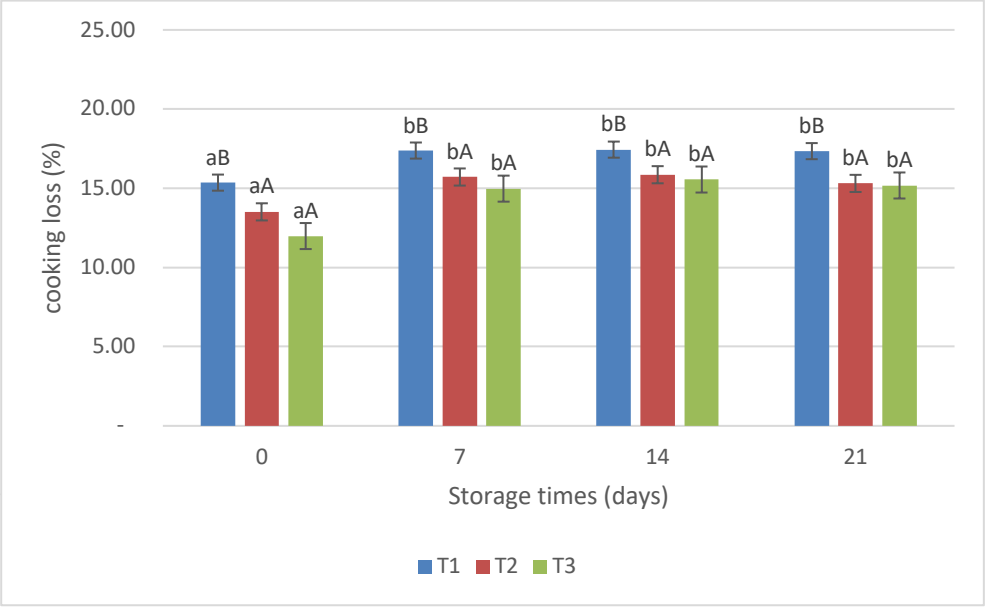

Figure 16. The cooking loss (%) of sous-vide cooked chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

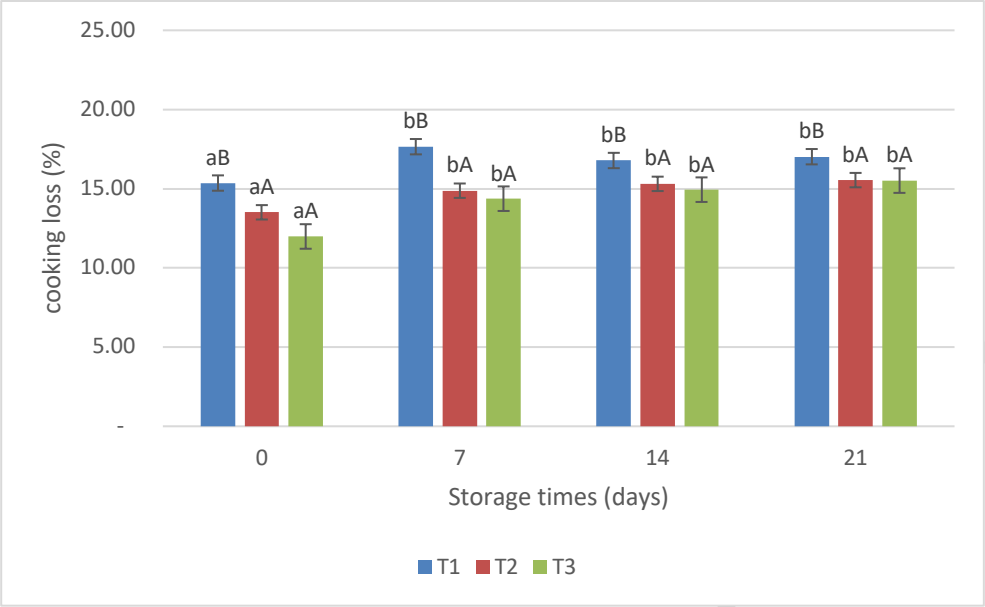

Figure 17. The cooking loss (%) of sous-vide cooked chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$)..

According to the results of the ANOVA and post-hoc Tukey test, each treatment (T1, T2, and T3) differs significantly in terms of moisture content (%) and cooking loss (%) ($p<0.05$). In addition, the double step treatments (T3 and T2) had substantially lower cooking loss (percent) and higher moisture content (percent) than the single step treatment (T1). These results are consistent with those of Hwang et al. (2019), Biyikli et al. (2020), and Endrit et al. (2022), who examined pork loin, turkey, and chicken breast samples and discovered that as cooking temperature increased, cooking loss increased as moisture content diminished. Meat's moisture content is dependent on the degree of thermal protein denaturation, which shrinks muscle fibres, resulting in a volume reduction and consequent water loss at high cooking temperatures (Offer et al., 1984). When meat is cooked, collagen denatures and becomes soluble, leading to gel formation and an increase in the meat's water content. This can result in cooking loss, as water and gelatinous material are lost during cooking (Zielbauer, 2015). Moisture loss can concentrate nutrients in meat, but it can also result in the loss of heat-sensitive nutrients and fluids containing specific nutrients (Ayub and Ahmad, 2019).

There was no significant difference in cooking loss (%) based on storage temperature ($p = 0.542$), but there was a significant difference in moisture content (%) ($p = 0.016$). The moisture content (%) during storage at 4°C was notably greater than at 10°C. Moisture content (%) did not differ significantly ($p>0.05$) during the storage period, whereas cooking loss (%) did ($p<0.05$) for both storage temperatures (4°C and 10°C). The temperatures of both storage containers varied significantly between day 0 and days 7, 14, and 21. Previous research by Kaewthong et al. (2019) discovered that storage time affects chicken breast weight loss, with a significant increase after day 9. During storage, meat can lose weight due to evaporation, which occurs when water vapor departs from the surface of the meat, and microbial growth and activity, which can result in the breakdown of muscle fibres and loss of moisture. These factors contribute to an increase in cooking loss as the meat dries out and loses moisture during storage. When meat is stored at low temperatures (0-4°C), its water-holding capacity and thawing-cooking loss are enhanced (Fernandes et al., 2016). Weight loss during storage is associated with water loss, which can have an impact on the quality and yield of raw and cooked flesh. In addition, storage temperature and time, as well as microbiological development, can impact the capacity of meat myofibrils to retain water during cold storage (Zhang et al., 2015).

5.3 Colour measurement

Using three indicators, the hue of sous-vide chicken breast samples was measured: lightness (L*), redness (a*), and yellowness (b*). The L* axis ranges from 0 to 100, with 0 representing black and 100 representing white, and represents the colour's lightness or darkness. Positive values represent red and negative values symbolize green along the a* axis. Positive values indicate yellow and negative values indicate blue along the b* axis, which represents yellow and blue, respectively. To determine the total colour difference (E), the equation (1) was utilized.

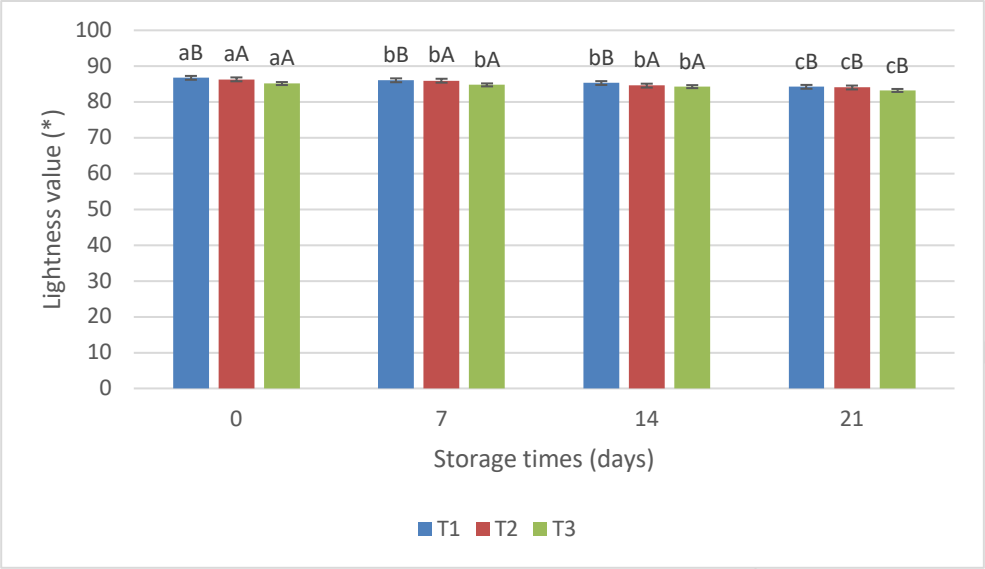

Figure 18. Lightness value (L*) of of sous-vide cooked chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$)..

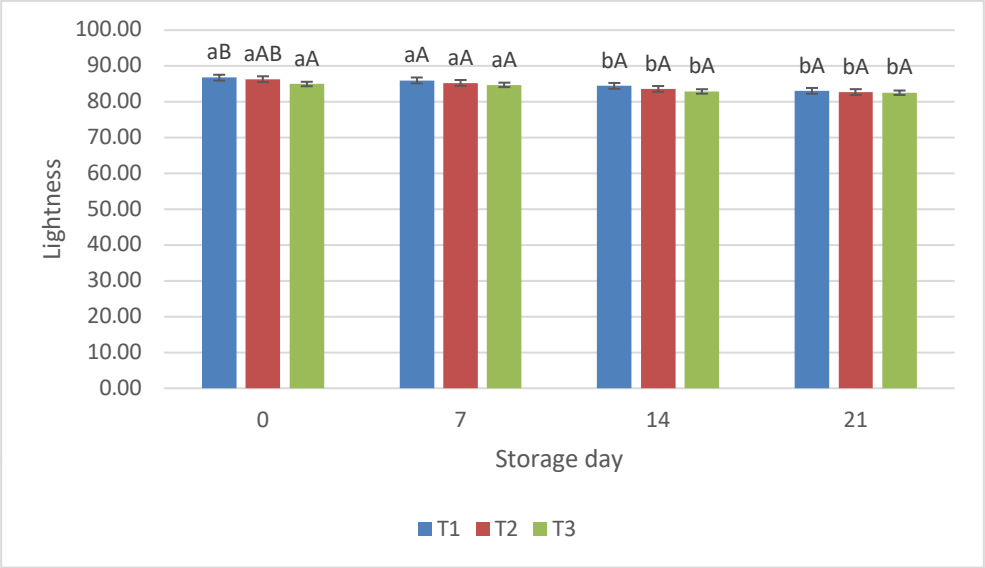

Figure 19. Lightness value (L*) of sous-vide cooked chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

Figures 18 and 19 represent the results of the lightness value (L*) of chicken breast samples. On day 0, the ANOVA analysis with post hoc Tukey test revealed significant differences ($p<0.05$) between the treatments. Single step (T1) and double step (T2 and T3) sous-vide treatments revealed significant differences, with T1 having higher lightness (L*) values, which can be attributed to the myoglobin alterations that play a role in determining lightness during denaturation. (Christensen et al., 2011).

Higher temperatures can increase protein denaturation, resulting in brighter meat. No significant differences ($p>0.05$) were observed on day 21 during storage at 4°C after 14 days of observation. From day 7 to day 21 of storage at 10°C, there were no significant differences ($p>0.05$) between the three treatments (T1, T2, and T3). According to the results of this study's lightness measurements, all sous vide treatments examined by Da Silva et al. (2019) belonged to the category of pale appearance ($L^* > 53$).

In terms of storing period of time, significant differences ($p<0.05$) were observed beginning on day 7 for storage at 4°C and day 14 for storage at 10°C. With increasing storage duration, the lightness value (L*) decreased. Regarding storage temperature, however, there was no significant difference between 4°C and 10°C ($p = 0.054$). This result is supported by Pizato et al. (2015), who found no significant differences in the lightness of cooked chicken breast between mild cold storage (10°C) and cold storage (4°C), with a significant decrease in lightness with time. Saláková et al. (2009) discovered a correlation between the lightness value of cooked chicken breast and the cooking loss, indicating that lightness can be affected by variety of factors.

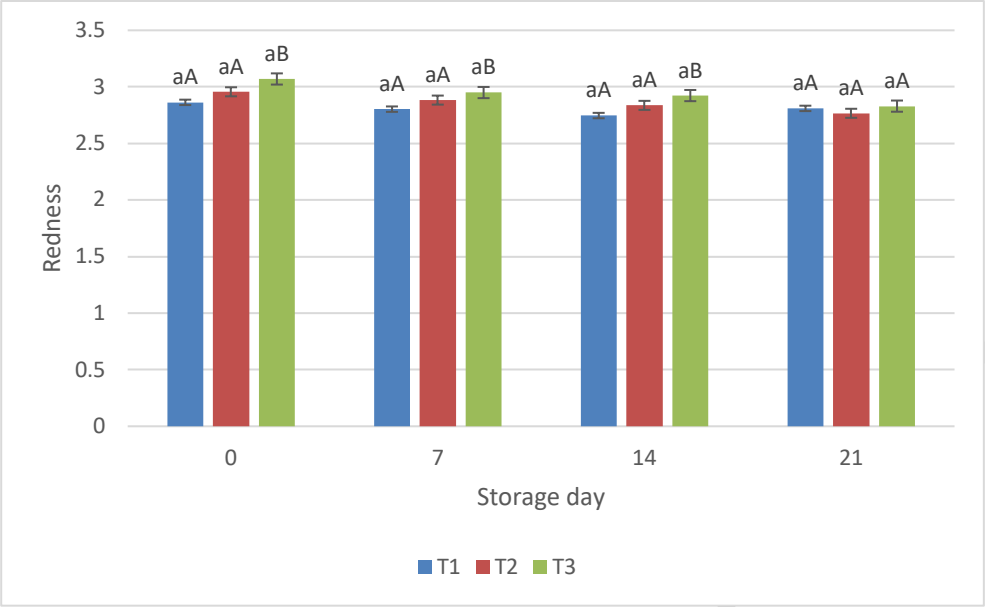

Figure 20. Redness value (a*) of sous-vide cooked chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).).

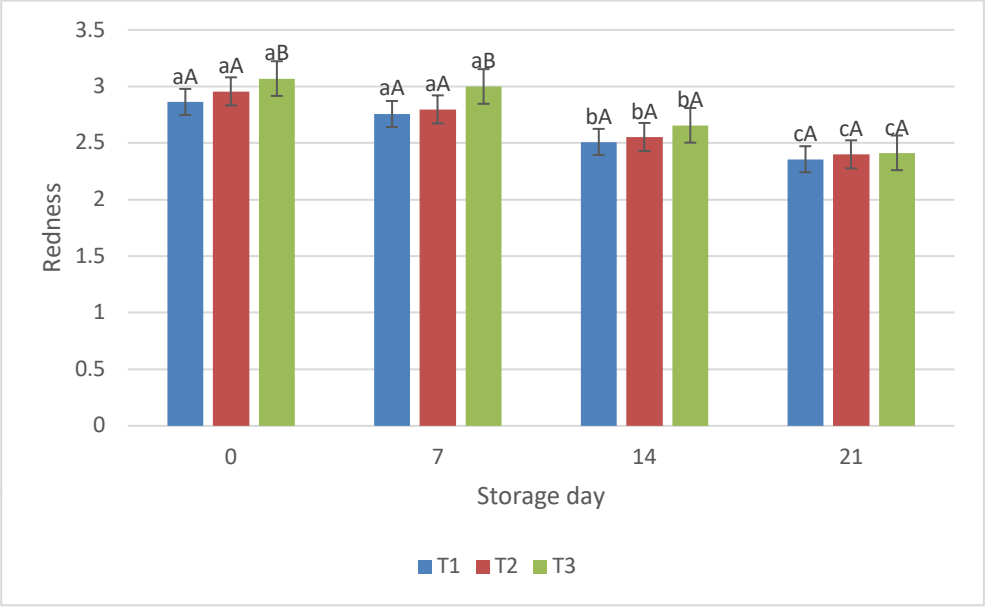

Figure 21. Redness value (a*) of sous-vide cooked chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

The redness value (a*) measurements (presented in Figures 20 and 21) indicate that on day 0, T3 had a significantly higher redness value ($p<0.05$) compared to treatments T2 and T1, reflected by a higher redness value than other treatments. This is likely because T3 was cooked at 60°C for a lesser duration and at 50°C for a longer duration than T2 and T1. Myoglobin pigment concentration, which influences redness values, begins to denature between 55 and 65°C (Hunt et al., 1999) and denatures completely at 80 C (Geileskey et al., 1998). These results remained the same until day 14 for storage at 4°C and until day 7 for storage at 10°C, at which point they did not differ significantly ($p>0.05$).

It should be noted that the pink colour of meat can be distracting to consumers, as it may be interpreted as an indication of undercooked meat or a safety hazard, which could result in complaints. The majority of consumers dislike this colour (Hong et al., 2015). Holownia et al. (2003) determined the pink colour threshold to be $a^* = 3.8$. According to the results of the study, all samples had erythema values below this threshold.

There were significant differences ($p<0.05$) in redness value over time for storage at 10°C, specifically on days 14 and 21, when the redness value decreased. For storage at 4°C, however, the results were insignificant ($p>0.05$). Regarding storage temperature, the p-value of 0.003 indicates a statistically significant difference between storage at 4°C and 10°C, with the redness value being greater for 4°C storage. Similar to the decrease in lightness, the decrease in redness is caused by the activity of deteriorating microorganisms that result in colour changes (Pizato et al., 2015).

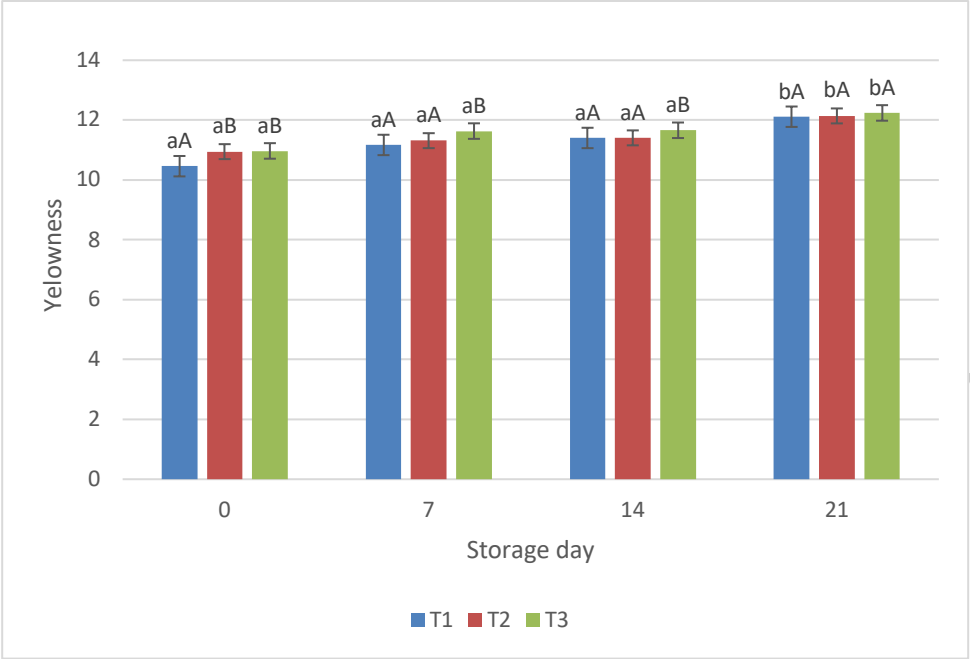

Figure 22. Yellowness value (b*) of sous-vide cooked chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$)..

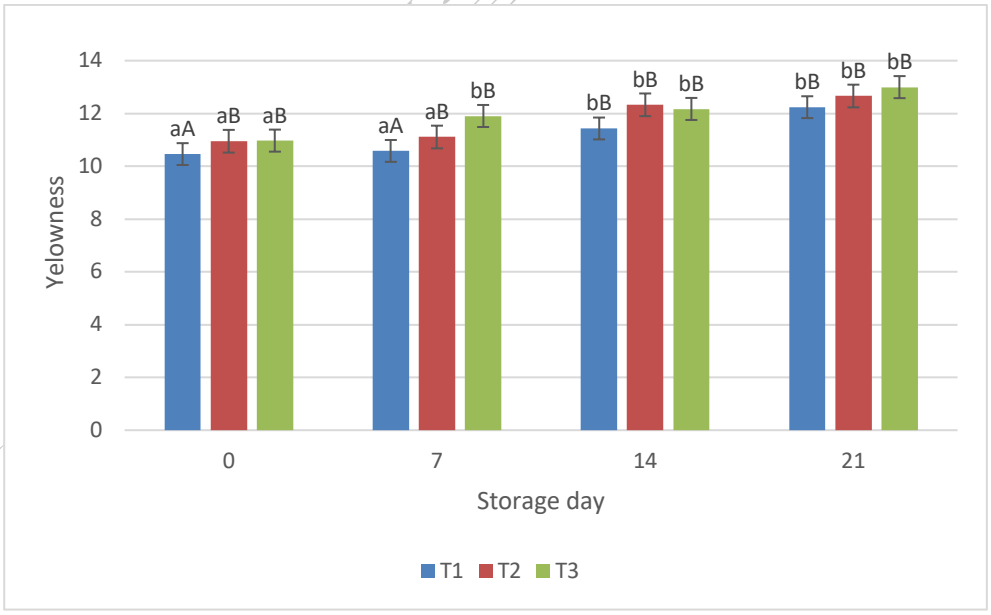

Figure 23. Yellowness value (b*) of sous-vide cooked chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$)..

According to the results of ANOVA and Tukey's post-hoc test, there was a statistically significant difference ($p<0.05$) in yellowness value (b*) between the single-step sous vide (T1) and double-step sous vide (T2 and T3) treatments, with T3 and T2 having higher values. This is as a result of lowered exposure of meat to 60°C during cooking in the two-step sous vide treatments (Hasani et al., 2022). This rise in b* values may be attributable to an increase in metmyoglobin concentrations (Ayub and Ahmad, 2019).

Regarding storage temperature, there was no statistically significant difference between 4°C and 10°C ($p = 0.309$). However, for storage days, there was a statistically significant difference ($p<0.05$) where, for storage at 4°C, the yellowing began on day 21 and on day 14 for storage at 10°C, indicating higher yellowness values. This result was in line with the findings of Pizato et al. (2015). According to Genot (2003), the discoloration of meat during storage was caused by the oxidation of pigment in the muscle tissue, and the pigment's stability depended on the animal species and the biochemical properties of the muscle.

Table 8. Total colour difference (ΔE) between chicken breasts cooked with different combinations of treatment and storage days at 4°C storage.

Treatment		**T1**				**T2**				**T3**			
	Storage day	**0**	**7**	**14**	**21**	**0**	**7**	**14**	**21**	**0**	**7**	**14**	**21**
T1	**0**		I	II	III	I	II	III	III	II	III	III	IV
	7	0.99		I	II	I	I	II	III	I	II	II	III
	14	1.74	0.81		II	II	I	I	II	I	I	II	III
	21	3.01	2.05	1.27		III	II	I	I	II	I	I	II
T2	**0**	0.65	0.38	1.15	2.39		I	II	III	I	I	III	I
	7	1.17	0.20	0.68	1.89	0.53		II	III	I	II	II	III
	14	2.37	1.51	0.72	0.78	1.81	1.38		I	I	I	I	II
	21	3.18	2.23	1.44	0.19	2.57	2.07	0.90		II	I	I	I
T3	**0**	1.69	0.97	0.56	1.49	1.18	0.89	0.76	1.64		I	II	III
	7	2.29	1.37	0.60	0.74	1.69	1.22	0.32	0.90	0.77		I	II
	14	2.73	1.84	1.05	0.47	2.15	1.69	0.39	0.56	1.81	0.49		II
	21	3.95	3.03	2.23	1.02	3.36	2.88	1.59	0.83	2.32	1.67	1.22	

T1: 60 ◦C/120 min; T2: 50 ◦C /40 min and 60 ◦C/80 min; T3: 50 ◦C/60 min and 60 ◦C/60 min. The total colour difference (ΔE) categories using Roman numerals, where I ($0 < \Delta E < 1$) indicates no difference, II ($1 < \Delta E < 2$) indicates only experienced observers can detect a difference, III ($2 < \Delta E < 3.5$) indicates inexperienced observers can detect a difference, and IV ($3.5 < \Delta E < 5$) indicates a clear difference in colour is detectable.

Table 9. Total colour difference (ΔE) between chicken breasts cooked with different combinations of treatment and storage days at 10°C storage .

Treatment		T1				T2				T3			
	Storage day	0	7	14	21	0	7	14	21	0	7	14	21
T1	0		I	III	IV	I	II	IV	IV	II	II	IV	IV
	7	0.78		II	III	I	I	II	IV	I	II	III	IV
	14	2.51	1.76		II	II	I	II	III	I	I	II	III
	21	4.12	3.38	1.62		IV	III	I	I	III	II	I	I
T2	0	0.65	0.54	1.98	3.56		II	III	IV	II	II	IV	IV
	7	1.60	0.87	0.93	2.54	1.06		III	III	I	I	III	III
	14	3.67	2.96	1.24	0.58	3.08	2.10		I	III	II	I	II
	21	4.60	3.88	2.12	0.54	4.02	3.02	0.94		III	III	I	I
T3	0	1.69	0.97	1.00	2.55	1.18	0.33	2.13	3.03		II	III	III
	7	2.48	1.84	0.73	1.82	1.86	0.99	1.29	2.22	1.03		II	III
	14	4.21	3.47	1.73	0.35	3.65	2.62	0.72	0.58	2.93	1.88		I
	21	4.93	4.22	2.48	0.92	4.34	3.36	1.26	0.39	3.37	2.52	0.93	

T1: 60 ◦C/120 min; T2: 50 ◦C /40 min and 60 ◦C/80 min; T3: 50 ◦C/60 min and 60 ◦C/60 min. The total colour difference (ΔE) categories using Roman numerals, where I ($0 < \Delta E < 1$) indicates no difference, II ($1 < \Delta E < 2$) indicates only experienced observers can detect a difference, III ($2 < \Delta E < 3.5$) indicates inexperienced observers can detect a difference, and IV ($3.5 < \Delta E < 5$) indicates a clear difference in colour is detectable.

Total colour differences for storage at 4°C and 10°C are listed in Tables 8 and 9. According to Tomasevic et al. (2019), when $E > 3.5$, there is a clear colour difference between meat products. The comparison of storage temperatures reveals that category IV (indicates a clear difference in colour is detectable) differences were perceived more strongly at 4°C than at 10°C. The most noticeable colour difference was observed between days 0 and 21 of storage. The colour characteristics of the examined sous vide treatments were influenced by storage temperatures and days.

5.4 Lipid Oxidation

To determine the rate of lipid oxidation in sous vide-cooked chicken breast, the concentration of secondary lipid oxidation compounds was measured. TBARS values are used to quantify the amount of secondary lipid oxidation products, particularly malonaldehyde, which can substantially contribute to the aroma degradation of stored meat products. Figures 24 and 25 illustrate the results of this analysis.

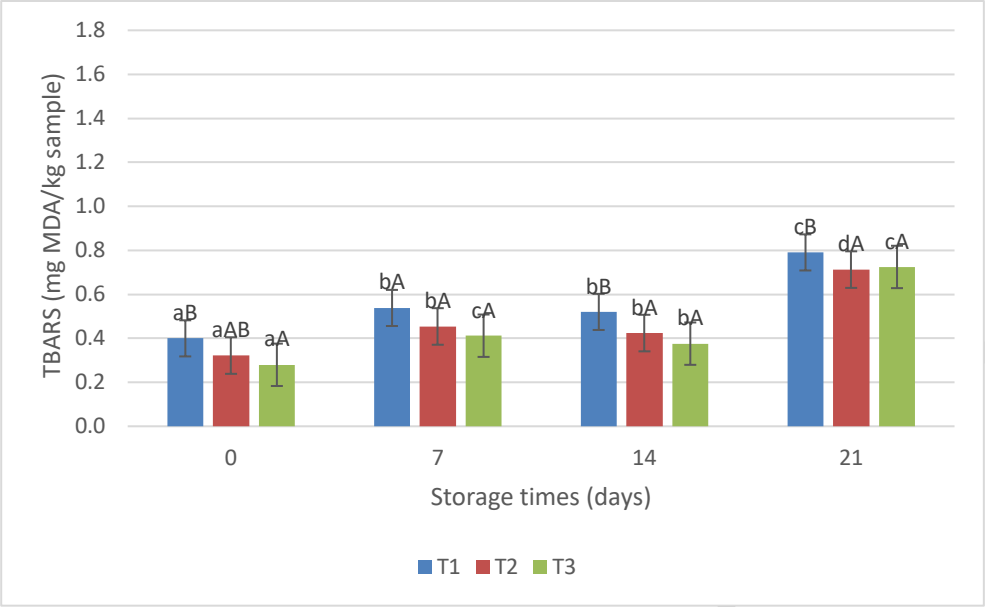

Figure 24. TBARS (mg MDA/kg sample) of sous-vide chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

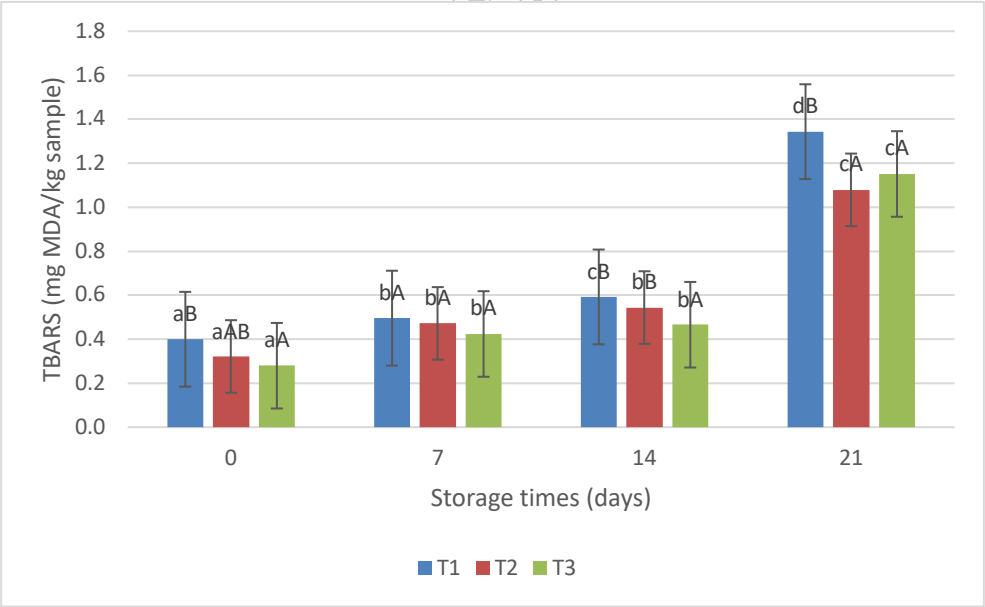

Figure 25. TBARS (mg MDA/kg sample) of sous-vide chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

ANOVA and post-hoc Tukey analysis revealed that on day 0, the single-step treatment (T1) demonstrated a statistically significant difference ($p<0.05$) compared to the double-step treatments (T2 and T3). Compared to the single-step treatment, the double-step treatment yielded a lower value. This result is consistent with the findings of Karpinska-Tymoszczyk et al. (2020) and Hasani et al (2022). This difference may be due to the fact that cooking at higher temperatures increases protein denaturation and the release of iron ions, which have a powerful pro-oxidant effect (Estevéz et al., 2011). According to Hong et al. (2015), cooking meat can accelerate lipid oxidation while deactivating compounds or enzymes that function as antioxidants.

Regarding the storage temperature, there was a significant difference between 4°C and 10°C with a p-value of 0.044, indicating that the value was greater at 10°C than at 4°C. During storage, there was a significant difference in lipid oxidation for both temperatures (4°C and 10°C) ($p<0.05$), indicating that lipid oxidation increased over time. Liu et al. (2019) reported that higher storage temperatures could lead to a greater oxidation of lipids. During storage at various temperatures, alterations in the fatty acid and amino acid compositions were observed, and this trend continued until the fourth week. This may be because the initial phases of lipid oxidation result in the formation of hydroperoxide, which converts into secondary oxidation products including ketones and aldehydes.

5.5 Odour Acceptability

To evaluate the odour acceptability of sous-vide chicken breast, meat were scored on a five-point scale based on their aroma, with a score of 1 indicating that the odour was acceptable, 2 indicating slight acceptability, 3 indicating neutrality (neither acceptable nor unacceptable), 4 indicating slight unacceptability, and 5 indicating complete unacceptability. This method enabled a standardized and objective evaluation of the chicken's odour, allowing researchers or evaluators to draw conclusions about the product's sensory attributes and prospective consumer appeal. The outcomes are depicted in figures 26 and 27.

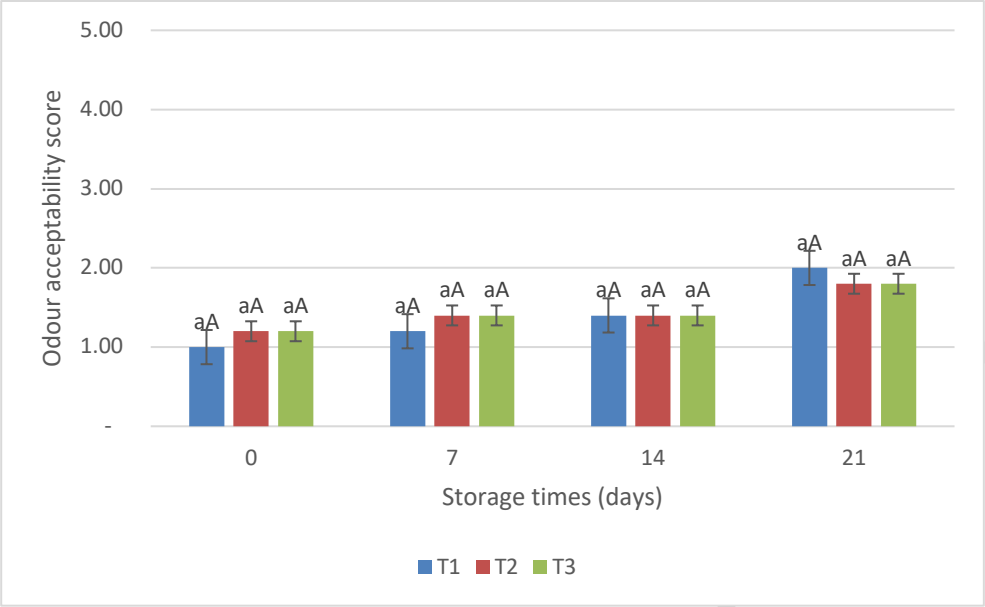

Figure 26. Odour acceptability score oof sous-vide chicken breasts stored at 4◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

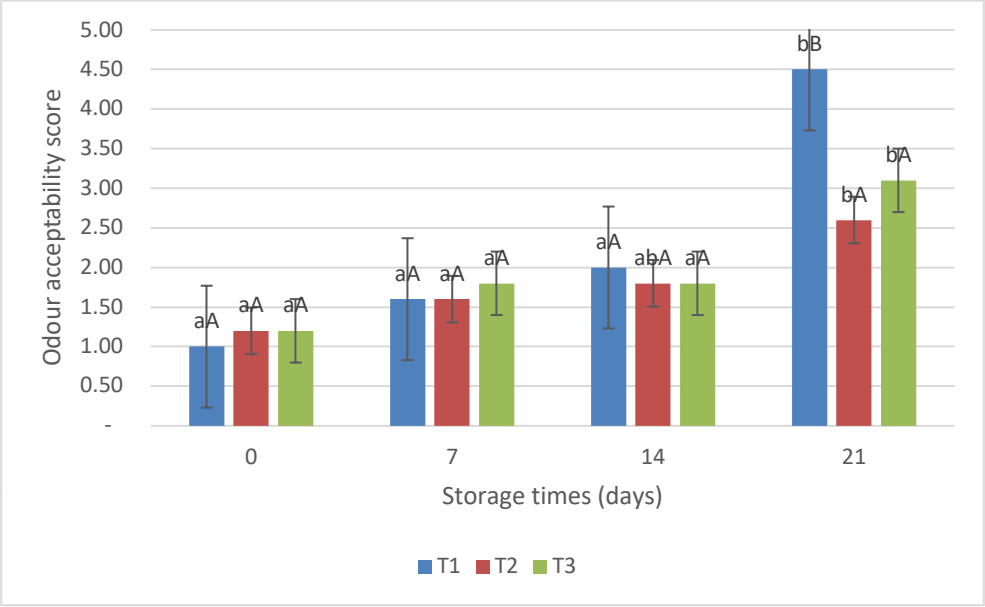

Figure 27. Odour acceptability score of sous-vide chicken breasts stored at 10◦C temperatures for 21 days. Three different cooking temperature-time combinations: T1 (60 ◦C/120 min), T2 (50 ◦C/40 min + 60 ◦C/80 min), and T3 (50 ◦C/60 min + 60 ◦C/60 min). The letters a, b, and c represent significant differences in values for different storage days within treatment types, while A, B, and C represent significant differences in values for different treatment types within storage days (with $p < 0.05$).

According to the results of the ANOVA and post-hoc Tukey test, there were no significant differences ($p>0.05$) between the three treatments (T1, T2, and T3), and they remained acceptable until the fourteenth day of storage. On the 21st day of storage at 10°C, however, a significant ($p<0.05$) unpleasant odour was detected. As reported by Akoglu et al. (2018), this could be attributed to the formation of secondary lipid oxidation products during 10°C storage, which can exceed the sensorial threshold limit of 1 mg MDA/kg sample.

6. SUMMARY

Each cooking treatment (T1: 60 ◦C/120 min; T2: 50 ◦C/40 min + 60 ◦C/80 min; and T3: 50 ◦C/60 min + 60 ◦C/60 min), differs significantly in terms of moisture content (%) and cooking loss (%) ($p<0.05$). the double step treatments (T3 and T2) had substantially lower in cooking loss (%) and higher in moisture content (%). There was a significant difference in moisture content (%) ($p = 0.016$) in storage temperature where 4°C showed higher moisture content. While in cooking loss, storage days had significant effect which there was an increasing during storage times for both 4°C and 10 °C.

Regarding color, double stage (T2 and T3) and single step (T1) differed significantly in terms of lightness (L*), redness (a*), and yellowness. In T1, lightness was greater than in T2 and T3, but the opposite for redness and yellowness. During storage at both temperatures (4°C and 10°C), lightness tended to decrease significantly while yellowness increased substantially. However, only redness decreased significantly under storage conditions of 10°C. In terms of storage temperature, only redness exhibited a statistically significant (p-value 0.003) difference, with the 4°C storage condition being higher to the 10°C storage condition. The storage temperatures reveal that category IV (indicates a clear difference in colour is detectable) were perceived more significantly at 4°C than at 10°C for the total colour difference. The most indicate a clear difference in colour is was observed between days 0 and 21 of storage. Regarding the TBARS value (mg MDA/kg of meat product), double stage (T2 and T3) and single phase (T1) significantly differed ($p<0.05$), with T1 having a higher value. With a p-value of 0.044, there was a significant difference between 4°C and 10°C regarding the storage temperature, indicating that the value was greater at 10°C than at 4°C. During storage at both temperatures (4°C and 10°C), there was a significant difference in lipid oxidation ($p<0.05$), implying that lipid oxidation increased over time. On the 21st day of storage at 10°C, an unacceptable odour was detected significantly ($p<0.05$) due to the formation of secondary lipid oxidation products that exceeded the sensorial threshold limit of 1 milligram MDA/kg sample. This research suggests conducting additional analysis, such as advanced sensory analysis with panellists, in order to better observe the significant differences between the three treatments. In addition, economic and efficiency calculations of the double stage sous-vide treatment must be performed before it can be implemented in the food industry.

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"Man Jadda Wa jada" means "whoever strives shall succeed."

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