

**Diploma Thesis**

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**2024**



**Hungarian University of Agriculture and Life Sciences**

**Institute of Food Science and Technology**

**Evaluation of Physical Attribute Changes in Minimally  
Processed Meat during Frozen Storage**

**MSc**

**DIPLOMA THESIS**

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**Budapest, 2024**

# Hungarian University Of Agriculture And Life Sciences

**Specialization name:** Food technology and product development

**Place of thesis preparation:** Department of Livestock and Food Preservation Technology

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**Thesis title:** Evaluation of Physical Attribute Changes in Minimally Processed Meat during Frozen Storage

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**Date of issuing the thesis:** 22. April 2024



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**Year of publication:** 2024

**Name of the Department:** Department of Livestock and Food Preservation Technology

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# **I. Introduction**

In recent years, the demand for minimally processed meat products, particularly those prepared using the sous-vide cooking method, has been steadily rising, driven by consumer preferences for convenience, quality, and nutritional value (Zavadlav et al, 2020). However, ensuring the maintenance of physical attributes and quality of these products during storage, especially under frozen conditions, remains a critical challenge for producers and regulators alike. This study focuses on evaluating the physical attribute changes in minimally processed meat, specifically chicken breast prepared using the sous-vide method, during frozen storage, with a particular emphasis on its relevance within the Hungarian market.

Sous-vide cooking, characterized by vacuum-sealing and precise temperature control, has gained popularity for its ability to preserve the natural flavors, textures, and nutrients of meat while ensuring consistent cooking results (Schellekens, 1996). However, the effects of frozen storage on the physical properties of sous-vide cooked meat, particularly in the context of the Hungarian food market, have not been extensively studied yet.

By addressing this gap in knowledge, the study aims to inform producers, regulators, and consumers about the optimal storage practices for maintaining the quality and safety of minimally processed meat products, thus contributing to the sustainable growth of the industry in Hungary.

With a growing emphasis on convenience and quality in the food industry, understanding the physical attribute changes in minimally processed meat during frozen storage is crucial for meeting consumer expectations and ensuring product integrity throughout the supply chain. Through this research, we aim to provide practical recommendations and insights that can support informed decision-making and promote the continued success of the minimally processed meat industry.



## **II. Aim of the study**

This study aims to comprehensively evaluate the physical attribute changes in minimally processed meat, focusing on chicken breast prepared using the sous-vide method, during frozen storage. Through detailed analysis of factors such as color, pH, texture, and weight loss during 4 weeks of storage time, the research seeks to provide insights into the impact of freezing on the quality and sensory properties of these products.

### **III. Literature review**

#### **3.1 Chicken meat**

Chicken meat is highly regarded for its multitude of health benefits stemming from its rich nutritional composition characterized by high protein levels, as well as low cholesterol, calorie, and fat contents. Moreover, chicken meat offers a cost-effective alternative compared to other meats such as pork, beef, and lamb (Sujiwo et al., 2018).

According to the Organization for Economic Cooperation and Development (OECD), in countries with a GDP per capita of \$30,000 or more, chicken consumption in 2017 reached 30.2 kg per capita, exceeding that of pork (23.6 kg) and beef (14.5 kg) (OECD, 2019). It is expected that the consumption of chicken meat, especially breast meat, will increase due to the increasing awareness of its health benefits and the rising demand for cost-effective protein sources.

The growing demand for chicken breast meat can be attributed to its nutritious profile, versatile sensory properties suitable for various cooking styles at home and in processing, as well as its mild flavor and tender texture, which allow for customization to meet diverse consumer preferences. Moreover, the convenience of quick and easy preparation aligns with the lifestyle demands of modern societies where time for meal preparation is limited. Consequently, the poultry industry has been driven to enhance breast yield and breed heavier birds to meet the escalating demand for processed products (Petracci et al., 2013a; Brewer et al., 2012).

In particular, the energetic value of meats varies between chicken breast raw and chicken breast cooked, as shown in Table 1 (European Institute of Oncology 2008). It must be noted that cooking also affects energetic value, which increases by 30% for meat (essentially due to a loss of water during the cooking process) (Caballero, 2005).

Poultry meat, alongside other animal-derived products like meat, milk, and eggs, is generally recognized for its high-quality protein content. These animal-based foods typically have a Protein Digestibility Corrected Amino Acid Score (PDCAAS) value close to or slightly below one (Caine et al., 1997). Conversely, plant-based foods, despite containing considerable protein, often exhibit a less favorable protein

profile. They may lack one or more essential amino acids and/or pose digestion challenges, leading to significantly lower PDCAAS values, for example there is 0.75 for beans and 0.5 for wheat.

Another advantageous trait of poultry meat is its low collagen content, which is a structural protein. Collagen has been found to hinder the digestibility of meat, and elevated levels of this protein in muscular meat correlate with a decreased percentage of digested product over time (Marangoni et al., 2015).

The lipid content associated with poultry meat varies depending on the specific cut being considered. However, fats are predominantly located in the skin and can be easily removed. For instance, the leanest cuts like chicken breast typically contain around 1% lipid content, which can increase with the inclusion of skin (Alagawany et al., 2019). Although cooking methods may marginally increase fat content by removing water from meat or incorporating fats from condiments, poultry typically maintains a relatively low fat content compared to other types of meat.

From a nutritional standpoint, the fat composition of poultry is advantageous, characterized by a significant presence of monounsaturated fatty acids, while only one-third comprises saturated fatty acids. In addition to its rich nutrient profile, chicken also serves as a potential source of omega-3 fatty acids (Alagawany et al., 2019). Omega-3 fatty acids possess anti-inflammatory properties, mitigating cytokine release, whereas excessive omega-6 levels are associated with conditions like depression and heart disease. Nonetheless, both fatty acids offer diverse health benefits, including improved cholesterol levels and reduced risk of coronary heart disease. They are acknowledged for their role in reducing inflammation, promoting heart health, and safeguarding against certain types of cancer. Furthermore, evidence suggests that omega-3s effectively lower triglyceride levels in the bloodstream, highlighting their significance in maintaining overall wellness (Djricic et al., 2021).

## **3.2 Minimal processed meat and Sous-vide Method**

### **3.2.1 Overview of Sous-vide Cooking Method**

Food technologists face hurdles as a result of consumers' increased demand for minimally processed foods with little to no synthetic ingredients (Siddiqui et al. 2011). These requirements compel the development of healthy foods using the fewest

possible processing steps (Gilbert 2000). Creating minimally processed foods that meet safety standards and possess an optimal shelf life poses a significant challenge.

The aims of minimal processing is to effortlessly and rapidly prepare ready-made meals. The benefits of minimal processing include: (1) convenient and swift meal preparation, (2) utilizing gentle processing techniques, such as multi-hurdles or multi-preservation methods in most cases, (3) preserving the quality to resemble fresh or nearly fresh meals, (4) retaining the nutritional value of the products, and (5) offering diverse shelf-life options based on the types and intensity of preservation methods employed. (Banerjee et al., 2014).

Food processing is crucial to both food preservation and the provision of wholesome, safe, and palatable food to the general public. In recent years, there has been a particular focus on the use of low heat processing techniques, such as sous vide technology, to enhance the quality of meat-based dishes. The ability of sous vide meat to hold water, maintain its texture, and maintain its juiciness all depend on the ideal mix of temperature and time parameters (Singh et al., 2023).

Sous vide, which is a French term meaning "under vacuum", refers to a cooking method where raw ingredients or partially cooked foods are placed inside heat-stable vacuum-sealed pouches and cooked under controlled conditions of temperature and time (Schellekens, 1996).

Sous vide cooking diverges from conventional methods primarily through two key processes: Firstly, the food undergoes precise heating under controlled conditions, followed by vacuum sealing in food-grade plastic pouches that withstand heat (Baldwin, 2012). The initial cooking phase within the vacuum-sealed environment not only minimizes bacterial contamination risks but also inhibits anaerobic microorganism growth, owing to the combined effects of temperature and pressure. Consequently, cooked food remains viable for extended storage periods and cools rapidly post-cooking. This approach enables precise management of both cooking duration and heat levels.

Research indicates that any viruses present in sous-vide foods post-cooking, such as rotavirus, Norwalk virus, and hepatitis viruses, likely originate from raw ingredients, as they withstand the cooking process (Choi et al., 2018; Aguilera, 2018). Additionally, the precisely controlled temperature and duration of sous vide cooking

mitigate adverse effects on nutrient integrity, preserving proteins, lipids, and vitamins while enhancing total polyphenols and antioxidant activity. Furthermore, this method improves overall texture and color of the food (Kilibarda et al., 2018).

Vacuum sealing offers several advantages in practical application: it extends the shelf life of food by minimizing the risk of re-contamination during storage, prevents off-flavors resulting from oxidation, and reduces evaporation losses of flavor compounds and moisture during cooking. Additionally, vacuum sealing facilitates efficient heat transfer from the water or steam to the food (Church and Parsons, 2000).

### **3.2.2 Importance of Frozen Storage in Minimally Processed Meat Industry**

Frozen storage is essential in the minimal processed meat for various reasons. First of all, it preserves meat product by inhibiting bacterial growth and enzymatic activity, extending the shelf life of products without additives. Secondly, it helps maintain the quality of meat by minimizing damage to its cellular structure, ensuring texture, flavor, and nutritional value are retained. Additionally, frozen storage allows for seasonal availability, stabilizes supply and demand, and offers safety assurance by reducing the risk of food-borne illnesses. Lastly, it is cost-effective, requiring less energy and infrastructure compared to other preservation methods (Elansari, 2014).

The majority of the changes in meat quality brought on by freezing and frozen storage, particularly those involving physical attributes, are associated with the meat's water content. Drip losses and thawing of meat exudes are significantly impacted by frozen storage (Vieira et al., 2009).

Throughout frozen storage, significant changes in various physical and chemical parameters were exhibited, including an increase in total exudation, pH, and lightness ( $L^*$ ), alongside a decrease in shear force and yellowness ( $b^*$ ). Additionally, research by Helga Medic in 2018 demonstrated a decrease in water content and an increase in protein content in ham samples.

### **3.3. Physical Attributes of Minimally Processed Meat**

In the global meat export business, which is valued at over \$13 billion annually, freezing has emerged as the favoured technique of food preservation (Leygonie et al., 2012). Although freeze methods can preserve the safety and quality of meat, processors, and consumers are nonetheless highly concerned about related

issues. Frequent freezing and thawing also happens in home kitchens, restaurants, retail stores, and sometimes even during storage or transit (Srinivasan et al., 1997).

### **3.3.1 Temperature**

Temperature fluctuations, a fundamental concern within the meat cold chain industry, particularly in emerging economies, are associated with physiological and biochemical alterations in muscle systems. (Soottawat & Friedrich, 2001). The longevity of meat is typically gauged by its visual appeal, texture, color, taste, microbial presence, and nutritional content, which can be affected by frozen storage and subsequent thawing processes (Leygonie et al., 2012). The primary deterioration of frozen meat during storage is attributed to lipid and protein degradation processes (Dasuri et al., 2013). Additionally, distinguishing between truly fresh meat and previously frozen-thawed meat is of significant interest to the meat industry, given the substantial price disparities between the two. Consumers frequently encounter difficulty in identifying quality alterations in meat products that have been subjected to freezing (Heo et al., 2016).

Most alterations in meat quality induced by freezing and subsequent storage, especially those related to physical characteristics, are closely linked to the meat's water content. Processes like thawing and drip losses, which are part of meat exudates, are significantly influenced by frozen storage (Ngapo et al., 1999).

Additional physical alterations in frozen meat consist of a decrease in pH (Leygonie et al., 2012) and a rise in tenderness when thawed compared to fresh meat (Muela et al., 2015). The optimal temperature at which frozen meat should be stored is estimated at  $-40^{\circ}\text{C}$  (Estévez, 2011).

### **3.3.2 Color and pH**

When consumers choose poultry products, they consider color as a crucial quality factor. Various elements influence the color of poultry skin and muscles, such as age, environment, nutrition, and feed availability. Poultry skin can vary from creamy to yellow in color. Raw muscle may appear pink or red due to the presence of myoglobin and hemoglobin. The myoglobin content in muscles increases with their usage, resulting in darker meat, commonly found in the thighs or legs of birds. Conversely, lighter meat, like the breast, is characterized by muscles with a lighter color, indicating less frequent usage (Guerrero et al., 2010).

The pH of the meat plays a crucial role in determining its color. Lower pH levels, which result from changes in the structure of the muscle's myofibrils, are associated with a reduced ability to retain water. Consequently, muscles with higher water-holding capacity typically exhibit a lighter color. Comparing organically raised broilers to those raised conventionally, the meat from organically raised birds appeared less red and more yellow. This difference could be attributed to their lower pH levels and diminished water-holding capacity (Castellini et al., 2002).

Insufficient oxygen availability prompts the muscle to generate lactic acid, thereby reducing the pH. If the muscle's pH drops rapidly while the carcass retains a high temperature, protein denaturation within the muscle fibrils can lead to pale meat. The paleness arises from heightened denaturation of sarcoplasmic proteins, resulting in amplified scattered light, thereby causing the meat to appear lighter (Sams, 2004).

The vacuum-packaged turkey breasts exhibited a deeper red hue compared to the aerobically packaged ones. This intensity of redness remained stable over a two-week storage period and continued to increase thereafter. Interestingly, the lightness of the samples remained unaffected by the irradiation treatment, irrespective of the dosage. The irradiation process produces carbon monoxide gas, which impacts the heme pigments present in turkey breasts. The interaction between carbon monoxide gas and myoglobin within the muscle is responsible for the pink or red coloration observed in irradiated turkey breasts, which is typically considered a flaw (Nam and Ahn, 2002).

Cooking alters the color of both poultry skin and muscle due to Maillard browning, a reaction involving amino acids, reducing sugars, and moisture. Unlike other foods, there are no enzymes involved in this process. However, this reaction contributes to the appealing color of cooked meat. Irradiation influences not only flavor and aroma but also color by altering heme pigments, potentially leading to off-flavors. Additionally, irradiation affects meat quality by generating free radicals that impact lipid and protein molecules, resulting in the formation of volatile compounds responsible for off-odors, with dimethyltrisulfide being the most prominent in irradiated raw chicken meat. (Patterson and Stevenson, 1995).

The microbial and nutritional quality of meat is significantly influenced by the irradiation dose, among other factors (Thayer et al., 1993). The chemical transformations induced by oxidation in meat are contingent upon the irradiation dose, with the presence of oxygen exerting a significant influence on the rate of oxidation (Katusin-Razem et al., 1992).

Studies have noted that raw irradiated chicken meat emits a bloody and sweet aroma (Heath et al., 1990). Hashim et al. (1995) assessed the effect of irradiation on refrigerated and frozen chicken skinless boneless breasts and leg quarters through sensory evaluation. They found that raw irradiated chicken exhibited higher intensities of "fresh chickeny," bloody, and sweet aromas compared to non-irradiated samples. Additionally, they observed that cooked irradiated frozen dark meat had a more pronounced chicken flavor than nonirradiated samples (Hashim et al., 1995).

### **3.3.3 Texture Properties**

The acceptance of meat is contingent upon numerous factors, with texture, especially tenderness, emerging as one of the most crucial. Processing conditions, including temperature, handling, and stunning methods, play a significant role in shaping the texture and overall quality of meat.

The transition from muscle to meat brings about substantial postmortem changes in both the physical and biochemical aspects of the muscle. This transformation, known as rigor mortis development, is characterized by stiffening, loss of extensibility and elasticity, muscle shortening due to the formation of permanent actomyosin bonds, and a decrease in muscle pH and adenosine triphosphate (ATP) concentration (Hedrick et al., 1989). The pH level drops from 7.4 in living muscle to 5.5 to 5.7 after rigor development (Hedrick et al., 1989; Pearson and Young, 1989). The decline in pH, as well as its rate of decline, holds significance as these changes can influence numerous meat quality attributes, including color, water-holding capacity, and texture.

Connective tissue, which is more prevalent in older animals like spent fowl, is often utilized in soup products with small meat portions and subjected to high thermal treatments. Collagen, the primary protein in connective tissue, boasts a distinctive structure designed for high tensile strength (Hultin, 1985; Bechtel, 1986). It consists of three polypeptide chains forming a triple helix called tropocollagen, which serves



as the structural unit of collagen fibrils. These fibrils are assembled with adjacent tropocollagen molecules in a quarter-stagger parallel pattern, featuring an end overlap of 25 nm and stabilized by ionic and hydrophobic interactions. Collagen's fibrous structure gains its strength from enzyme-induced intermolecular cross-links (Pearson and Young, 1989).

### **3.4. Application in the Hungarian Market**

The poultry industry plays a significant role in Hungarian meat production, contributing to two-thirds of the total slaughtered meat output. Chicken holds the largest production volume, followed by turkey, duck, and goose. In terms of value, the poultry sector stands as the leading producer within animal production. Hungary demonstrates remarkable self-sufficiency in poultry products, reaching close to 180% (Kálmán et al, 2023).

When considering live animals and eggs, the production value amounts to €920-€950 million, which doubles when including processed products. The processing industry is predominantly domestically owned, with processors effectively integrating farmers. Employment within the sector, both directly and indirectly, spans approximately 50,000 to 60,000 individuals. Chicken production operations are highly automated, whereas goose and duck production necessitate a substantial manual workforce.

Hungary's domestic goose and duck sector holds a prominent position globally, ranking second in Europe for meat production and first worldwide for goose liver production (Slabock et al, 2016). According to Poultry sector report in 2019, Poultry meat production of the European Union (28 Member States) totalled 15.3 million tonnes in 2019, with Hungary having a 3 percent share, thereby ranking as the eighth largest poultry meat producer (Gergely et al, 2019).

The consumption data seem somehow contradicted to production figures since annual per capita poultry meat consumption in Hungary has been steadily increasing, reaching 35.7 kilograms in 2019, compared to the EU average of 23.3 kilograms in 2020 (Poultry sector report in 2019).

### 3.5 Regulatory Standards and Quality Assurance Practices

According to research institute of agricultural economics (AKI) report of Poultry sector in Hungary 2019, Hungarian poultry farmers are obligated to adhere to EU regulations, which incurred additional costs estimated at around 6 percent of total production costs in 2017. These costs encompass various aspects such as environmental protection, food safety, and animal welfare standards, including compliance with regulations on nitrate pollution, ammonia emissions, salmonella control, and the ban on certain feed additives and genetically modified crops.

Furthermore, two new EU regulations, Regulation (EU) 2019/4 on medicated feed and Regulation (EU) 2019/6 on veterinary medicinal products, are set to come into effect on January 28, 2022, to support the responsible use of antibiotics.

Government Decrees No. 49/2001 and No. 219/2004 specify technical requirements for manure storage, adding to the environmental burdens on farmers, particularly by limiting the locations for manure storage.

Many processing plants conduct daily nutritional and organoleptic tests on their products, in compliance with regulations and consumer demands, with additional analysis conducted in accredited laboratories. Adherence to ISO and HACCP standards is crucial for meeting both domestic and foreign market demands. Quality assurance certificates, such as the IFS Food Standard and BRC Global Standards, are prerequisites for exports to Western European markets. Additionally, special religious standards (halal, kosher) have been introduced by many Hungarian poultry processors to access markets in the East and cater to the growing number of religious minorities in Western Europe. Although these certificates increase production costs, they enable producers to charge higher prices for these specialized products.

Relevant regulations: □

- ◆ 11/2019. (IV. 1.) Decree AM on the conditions of animal welfare subsidies available in the poultry sector.
- ◆ 128/2009. (X. 6.) FVM decree on veterinary medicinal products. □
- ◆ 148/2007. (XII. 8.) FVM decree on the procedure for requesting and paying subsidies related to the prevention and control of certain animal diseases.
- ◆ 188/2019. (VII. 30.) Government decree on animal husbandry.

- ◆ XXVIII of 1998 law on the protection and welfare of animals.
- ◆ Act CXL of 2004 on the general rules of administrative authority procedure and service.
- ◆ Act XLVI of 2008 on the food chain and its official supervision.
- ◆ 22/2012. (II.29.) Government Decree on the National Food Chain Safety Office.□
- ◆ 314/2005. (XII. 25.) Government Decree on the environmental impact assessment and uniform environmental use licensing procedure.□
- ◆ 32/1999. (III. 31.) FVM decree on animal protection rules for keeping agricultural farm animals.
- ◆ 41/1997. (V. 28.) FM decree on issuing the Animal Health Regulations.
- ◆ 63/2012. (VII. 2.) VM Decree on the amount of administrative service fees to be paid in procedures initiated before the National Food Chain Safety Office and the agricultural administrative bodies of county government offices, as well as the rules for paying the administrative service fee.
- ◆ 65/2012. (VII. 4.) VM decree on certain rules for the production, placing on the market and use of animal feed.

## IV. Material and Methods

### 4.1 Sample Selection and Preparation

Fresh broiler chicken breast meat was collected from a commercial meat processing plant (Hungarit Meat Company, 6600 Szentes, Alttilla út 2.), as shown in Figure 1. All visible fat and connective tissues were removed prior to the experiment. Breast meat, about 90-120 g per replicate, was randomly selected and cut into 10 x 7.5 x 2.5 cm<sup>3</sup> pieces.

There are 2 groups of sample preparation before sous-vide cooking. Group 1: These pieces were packed in moisture impermeable polyethylene bags, vacuum sealed (Figure 2). Group 2: These pieces were packed in moisture impermeable polyethylene bags, sealed and frozen at -25°C (Figure 3). For a day, a set of frozen samples was thawed at ambient temperature 1 hour before vacuum sealing .

Then the raw material was subjected to thermal treatment and was investigated. Before heat treatment, the color parameters L\* (lightness), a\* (redness), and b\* (yellowness), weight, texture and pH were measured in raw material.



*Figure 1. Fresh broiler chicken breast meat was collected from a commercial meat processing plant (Hungarit Meat Company, 6600 Szentes, Alttilla út 2.)*

### 4.2 Sous-Vide Cooking Process

In this study, for the sous-vide (SV) cooking method, the temperature of 72°C and time 45 min was used. Prior to cooking, each breast muscles was weighed, placed into a vacuum polyamide/polyethylene pouches (thickness of 92 µm, heat resistance (HR) of -40°C/+120°C, O<sub>2</sub> permeability of 9 cm<sup>3</sup>/m<sup>2</sup> per 24 h at 4°C/80% HR and water steam permeability of 1.2 g/m<sup>2</sup> per 24 h) and further using vacuum sealing

machine in Figure 4. (Multivac C200, Multivac Sepp Haggenmüller, Germany) with extent of vacuum 99.6%.



**Figure 2. Raw refrigerated chicken breast meat were packed in moisture impermeable polyethylene bags and vacuum sealed.**



**Figure 3. Raw chicken breast meat packed in moisture impermeable polyethylene bags, sealed and frozen at  $-25^{\circ}\text{C}$  in 24 hours before vacuum sealing for Sous-vide treatment**

After that, the samples were submerged in a thermostated water bath (*model SW 22, Julabo GmbH, Seelbach, Germany*), as shown in Figure 5. that was preheated to  $72^{\circ}\text{C}$ . The heating time of 45 minutes was applied once the core temperature of the muscles reached the water bath temperature of  $72^{\circ}\text{C}$  (*the hand-held thermometer was used in an additional control sample-Thermometer, DT-34, Termoprodukt, Bielawa, Poland*) as shown in Figure 5. Immediately after the heat treatment, the pouches were removed from the water bath and rapidly chilled with ice-cold water ( $2^{\circ}\text{C}$ ) for 30minutes, as shown in Figure 6. Thereafter the packed breast muscles were put in experiemental designed storage condition.

### **4.3 Experimental Design**

There are 2 variables: Material freshness and Storage condition as shown in Table 1 and 2.

- 2 type of chicken meat sample: Refrigerated Raw (3°C) and Frozen (-25°C),
- 2 type of storage condition: Frozen (-25°C) or Refrigerated (3°C).



*Figure 4. Packaging machine (Multivac C200, Germany)*



*Figure 5. Thermostated water bath of 72°C detected by a hand-held thermometer*

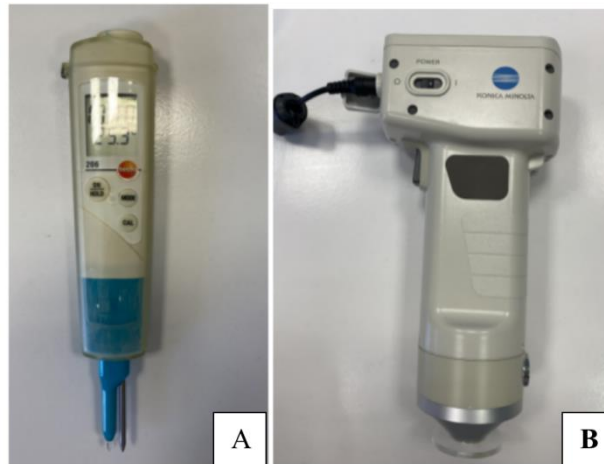


*Figure 6. Chilling process - chicken breast samples with ice-cold water after sous-vide cooking*

## 4.4 Analytical Methods

### 4.4.1. pH analysis

The pH of meat samples was measured with a digital pH metre (*Testo 206-pH2 pH meter*) after equilibration of the meat to room temperature, as shown in Figure 7. The assessments were carried out on 3 preselected locations at the surface of each sample.



*Figure 7. Testo 206-pH2 pH meter on the right (A) and Konica Minolta colorimeter CR-400 (Japan) on the left (B).*

### 4.4.2 Color analysis

The colour of the breast of raw and cooked samples was assessed for lightness, redness and yellowness using a Konica Minolta colorimeter CR-400 (Japan), as shown in Figure 7. The assessments were carried out on 3 preselected locations at the surface of each sample. the individual differences ( $\Delta E$ ) in  $L^*$ ,  $a^*$  and  $b^*$  values were calculated using the following equations (CIE, 1986):

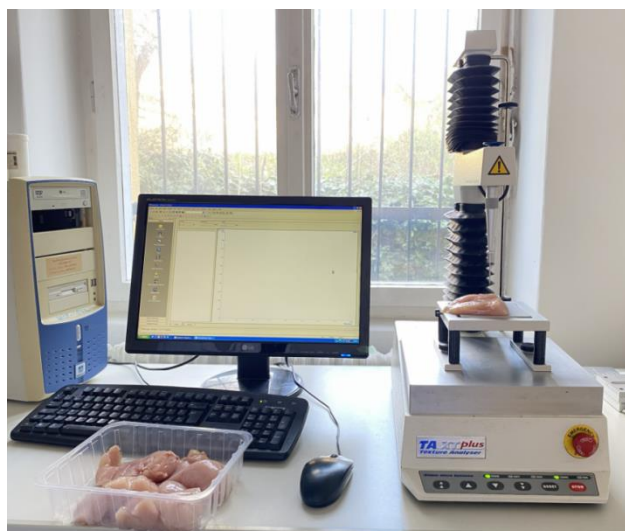
$$\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

### 4.4.3 Texture analysis using texture analyzers or penetrometers

Texture analysis was conducted at room temperature with TA.XT Plus, Texture Technologies, Scarsdale, NY, as shown in Figure 8. Texture profile analysis (TPA) includes hardness, springiness, cohesiveness, gumminess, chewiness. The assessments were carried out on 3 preselected locations of each sample to the longitudinal orientation of the muscular fibers were taken.



A 20mm diameter needle punch-ended probe was attached to a texture analyzer and the sample was oriented to ensure fibers were perpendicular to the direction of the probe. The force required and work done to drive a flat-ended probe to compress the sample to 70% of its height was recorded. The instrument was set with a cross head speed of 2 mm/min and wait time of 5s in a double-bite compression test. The TPA parameters were calculated using Bluehill 3—testing Software Instron.



*Figure 8. Texture Analyzer (TA.XT Plus, Texture Technologies, Scarsdale, NY)*

#### **4.4.4 Cooking Loss**

Breast samples were weighed and the bags were unsealed once the sample had reached room temperature in order to determine the percentage of cooking loss. The weights before and after heat treatment (Wh) were compared in order to compute CL. Every muscle was used three times (Wołoszyn et al., 2020).

$$\text{CL (\%)} = (\text{W}-\text{Wh})/\text{W} \times 100\%$$

Which is:

CL: Cooking loss (%)

W: initial weight before cooking (g)

Wh: after heating weight (g)

#### **4.4.5 Weight loss during Storage time**

The meat was weighed before and after storage (Frozen or Cold), and weekly during storage. Measurements of Kern EMB 200-2 weight were made by a 200g



capacity balance with 0.01 g divisions, as shown in Figure 10. The results of weight losses were statistically analysed by a multiple step-by-step regression.

$$WL(\%) = (W - W_{st}) / W \times 100\%$$

Which is:

CL: Cooking loss (%)

W: initial weight before cooking (g)

W<sub>st</sub>: after certain time of storage weight (g)



*Figure 9. Kern EMB 200-2 weight*

#### **4.4.6 Sensory Evaluation**

Each breast muscles was evaluated in terms of flavor and aroma (typical for chicken meat), tenderness, juiciness, cohesiveness, springiness and overall repeatability. The samples were analyzed for the intensity of sensory descriptors. It was 3 repetitions from each muscle.

#### **4.4.7 Statistical analysis**

The basic descriptive statistics (mean, standard deviation) were calculated. The significance of differences between the groups in terms of the analyzed methods of storage treatment was determined using the Student's t-test. The significance of differences was estimated with the statistical significance coefficient  $p < 0.05$ . To determine the colour, pH, and TPA data, each replicate was measured three times.

## V. Results and discussion

### 5.1. Physical attribute changes of raw chicken breast with different storage conditions

The quality characteristics of raw material intended for testing are presented in Table 1. The data that characterized the physical quality of a commercial chicken breast meat selected for Sous-vide treatment using refrigerated and frozen storage for preparation did not differ significantly (t-test,  $p < 0.05$ ).

*Table 1. Characteristics of raw chicken breast samples (n=9)*

Feature	Average $\pm$ SD		Significant level
	Refrigerated	Frozen	P
Color parameters: L*	49.02 $\pm$ 2.32	51.69 $\pm$ 4.99	0.132
a*	2.52 $\pm$ 1.04	1.99 $\pm$ 1.34	0.188
b*	3.25 $\pm$ 0.6	7.18 $\pm$ 2.14	0.0003**
pH	5.97 $\pm$ 0.18	5.77 $\pm$ 0.12	0.004**
Texture - Shear force(N)	1.67 $\pm$ 0.36	2.04 $\pm$ 0.50	0.028*

All values are presented as means $\pm$ SD (n=9).

#### 5.1.1. Color of raw chicken breast

The difference in color of the fresh-cut chicken breast is clearly discernible to the human eye, as depicted in Figure 2. According to the color measurements provided in Table 1, the L\* value is 49.02  $\pm$  2.32, indicating that it is slightly darker than a neutral gray but still relatively light. The a\* value is 2.52  $\pm$  1.04, suggesting a slight tendency towards redness, while the b\* value is 3.25  $\pm$  0.6, indicating a slight yellowish hue. In essence, the chicken breast appears to have a slightly darkened, reddish-yellow coloration.

In the case of 4-week-frozen raw chicken breast, the color was assessed, revealing an L\* value of 51.69  $\pm$  4.99, which is relatively lighter than that of the refrigerated raw meat, which is not statistically significantly different with a p-value of 0.132. This indicates a higher light reflectance, resulting in an overall lighter appearance. Additionally, the a\* value of 1.99  $\pm$  1.34 suggests a subtle inclination

towards redness in the frozen raw meat, albeit lower than the  $a^*$  value of the refrigerated counterpart ( $a^* = 2.52$ ), indicating a slightly diminished redness. However, there is not statistic significantly different with p-value is 0.132. Significantly, with a  $b^*$  value of  $7.18 \pm 2.14$ , the frozen raw meat displays a distinct yellowish hue, indicating a more pronounced yellow coloration compared to the refrigerated raw meat ( $b^* = 3.25$ ), representing a more vivid yellow appearance. Oppositely, The decreasing of  $L^*$  value could be caused by a reduction of water retention which leads to a lower surface light reflectivity (Hunghe et al.,2014). The accumulation of metmyoglobin (MetMb) at the surface of meat during storage contributes significantly to its discoloration (Bekhit et al., 2007), which could explain the changes in  $a^*$  value. The increasing lipid oxidation and the formation of MetMb are the main factors leading to the changes in  $b^*$  value as well (Xiong YL et al., 2000).



**Figure 10. Chicken breast thawed in 1 hours using warm water following 4 weeks of frozen storage.**

Therefore, the color measurements indicate that the frozen raw meat appears lighter with slightly less redness but a more intense yellow hue compared to the refrigerated raw meat. Delta E values are typically used to quantify the perceptible difference between two colors. In this case, the Delta E value between raw chicken meat and frozen raw chicken meat is given as 4.78, as shown in Table 3. This value suggests a significant perceptible difference in color between the two states of the chicken meat. However, it's worth understanding that the standard deviation (SD) value is indicative of a wide range, signifying high variability within the data set.

### **5.1.2. pH of raw chicken breast**

The statistical analysis showed significant differences in pH after heat treatment of 2 raw chicken breast. In this case, both refrigerated and frozen chicken breasts have slightly acidic pH values, with the refrigerated one being slightly more neutral at  $5.972 \pm 0.18$  compared to the frozen one at  $5.77 \pm 0.12$ . This meat is acceptable in normal ultimate pH (pHu) range for chicken meat, between 5.7 and 6.1 (Beauclercq S et al, 2022). One potential explanation is the effect of freezing on the cellular structure of the meat. Freezing can cause ice crystal formation within the muscle fibers, which may disrupt cell membranes and lead to changes in pH.

### **5.1.3. Texture of raw chicken breast**

In refrigerated conditions, the shear force required to cut the chicken breast samples was measured at  $1.67 \pm 0.36$  Newtons. While in frozen conditions, the shear force required was higher, measured at  $2.04 \pm 0.50$  Newtons. The p-value associated with this comparison is 0.028, indicating that the difference in shear force between refrigerated and frozen samples is statistically significant. This factor is crucial, as temperature fluctuations can trigger ice recrystallization, resulting in the growth of ice crystals and increased damage to the structural integrity of the meat. Past research has indicated that a higher number of freeze-thaw cycles resulted in more significant alterations in texture, protein oxidation, color, and water-holding capacity of meat (Xia et al., 2009; Ali et al., 2015).

### **5.1.4 Thawing loss of frozen raw chicken breast**

This type of chicken meat has Thawing loss is  $3.25 \pm 1.52\%$ , which was in agreement with the findings of Zhang et al. (2015), the values varied between 1.63 and 4.08% for the  $-2^{\circ}\text{C}$  samples. Weight loss during storage is closely linked to water loss, impacting the quality and yield of both fresh and cooked meat. Storage temperature and duration, along with microbiological growth, are primary factors influencing the water retention capacity of myofibrils in meat during storage under cold conditions (Cheng & Sun, 2008).

## 5.2. Physical attribute changes of Sous-vide cooked chicken breast from different stored materials.

This Table 2. compares the characteristics of raw chicken breast meat after Sous-vide treatment, specifically examining the differences between storage in refrigerated and frozen conditions.

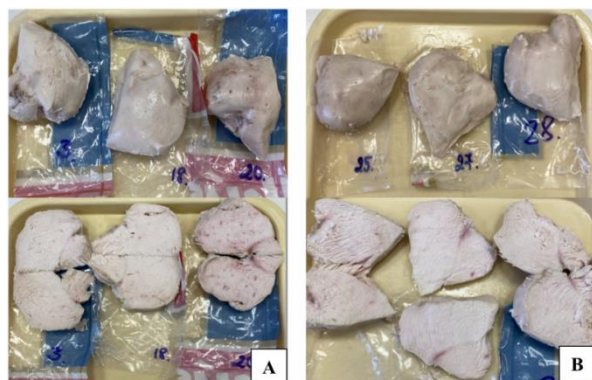
*Table 2. Characteristics of Sous-vide chicken breast (n=9)*

Feature	Average $\pm$ SD		Significant level
	Refrigerated	Frozen	P
Color parameters: L*	82.73 $\pm$ 1.82	82.96 $\pm$ 1.97	0.033*
a*	4.28 $\pm$ 1.02	4.11 $\pm$ 1.00	0.153
b*	10.06 $\pm$ 0.8	10.20 $\pm$ 0.75	0.0055**
pH	6.42 $\pm$ 0.33	6.45 $\pm$ 0.36	0.333
Texture (N)	3.43 $\pm$ 1.08	3.47 $\pm$ 0.39	0.457
Cooking loss (%)	18.32%	16.70%	0.3393

*All values are presented as means $\pm$ SD (n=9).*

### 5.2.1. Changing in color of Sous-vide chicken breast

In case of the L\* value, chicken breast under Sous-vide cooking was obtained whiter. From  $49.02 \pm 2.32$  for refrigerated raw meat to  $82.73 \pm 1.82$  for cooked meat and from  $51.69 \pm 4.99$  for frozen raw meat to  $82.96 \pm 1.97$  for cooked meat. Although there's a slight brightening between the two raw samples, the difference between refrigerated and frozen raw material conditions is statistically significant, with a P-value of 0.033 ( $<0.05$ ).



*Figure 11. Visual comparison of sous-vide chicken breast appearance: Refrigerated sample on the right (A) and Frozen sample on the left (B).*

Regarding the  $a^*$  value, which represents redness, a similar trend is observed. The value increases after cooking from  $2.52 \pm 1.04$  for refrigerated raw meat to  $4.28 \pm 1.02$  and from  $1.99 \pm 1.34$  for frozen raw meat to  $4.11 \pm 1.00$ . There's a statistically significant difference in redness level between raw meat and uncooked-cooked meat, with P-Values of 0.06. The difference in  $a^*$  values between refrigerated ( $4.28 \pm 1.02$ ) and frozen ( $4.11 \pm 1.00$ ) storage conditions was not statistically significant ( $p = 0.153$ ), indicating that there was no significant difference in the redness of the chicken breast meat between the two storage conditions.

In case of the  $b^*$  value, The cooked meat is more yellowish than raw meat. The difference between the two storage conditions was statistically significant ( $p = 0.0055$ ), suggesting that there was a slight difference in the yellowness of the chicken breast meat between refrigerated and frozen storage. In other words, Frozen cooked meat was obtain more yellowish.

Sous-vide cooking results in chicken breast appearing whiter, more yellowish, and reddish compared to raw meat. Moreover, it's imperceptible to human eyes to discern differences in cooked meat between frozen and refrigerated material (Table 3), as their Delta E is 0.29. Conversely, the Delta E between refrigerated and frozen raw meat is 4.78, which is perceptible at a glance.

**Table 3. Delta E Values for Raw and Sous-vide States under Various Storage Conditions of Raw Meat**

Feature		Delta E
Raw meat	Refrigerated	4.78
	Frozen	
Cooked meat	Refrigerated	0.29
	Frozen	

Following sous vide cooking, the color metrics ( $L^*$ ,  $a^*$ ,  $b^*$ ) significantly increase. Likewise, the color of chicken breast meat post-sous vide (cooked at  $76^\circ\text{C}$  for 60 minutes) is reported by Przybylski, W. in 2021 as follows:  $L^*$  is  $84.26 \pm 0.63$ ,  $a^*$  is  $2.54 \pm 0.69$ , and  $b^*$  is  $15.12 \pm 0.69$ , compared to the raw meat with  $L^*$  of  $53.07 \pm 1.93$ ,  $a^*$  of  $0.08 \pm 0.83$ , and  $b^*$  of  $9.55 \pm 1.45$ .

In this study, cooking loss is 18.32% for refrigerated sample and 16.07% for frozen sample, which is acceptable in range from 10.23% to 28.08% reported by Haghghi et al. in 2020. There is no favorable different between them with p value is 0.3393.

### **5.2.2. Changing in pH of Sous-vide chicken breast**

The pH values provided indicate the acidity or alkalinity level of the samples, with a slightly higher pH observed in the frozen samples compared to the refrigerated ones. The p-value of 0.333 suggests that this difference is not statistically significant at the commonly used significance level of 0.05. The mean pH for the refrigerated samples is  $6.42 \pm 0.33$ , while for the frozen samples, it is  $6.45 \pm 0.36$ . Increasing temperature to 72 °C caused an increase in pH value. Similarly, Bıyıklı et al. found that elevating the cooking temperature from 65°C to 75°C and extending the cooking time from 20 minutes to 60 minutes resulted in a rise in the pH of sous vide turkey cutlet. Additionally, Becker et al. observed that increasing the temperature led to a pH increase primarily attributed to protein denaturation and alterations in protein charge.

### **5.2.3. Changing in texture of Sous-vide chicken breast**

The shear force values for Sous-vide chicken breast samples were  $3.43 \pm 1.08$  N for refrigerated samples and  $3.47 \pm 0.39$  N for frozen samples. In contrast to the raw samples, there was no statistically significant difference in shear force between refrigerated and frozen samples after Sous-vide cooking ( $p = 0.457$ ). This suggests that the texture of Sous-vide chicken breast samples was not significantly affected by the storage state (refrigerated or frozen) prior to cooking. The shear force values for raw chicken breast samples were generally lower than those for Sous-vide chicken breast samples, regardless of whether they were refrigerated or frozen. This indicates that the Sous-vide cooking process resulted in a firmer texture compared to raw chicken breast samples. Thus, while storage state (refrigerated or frozen) had a significant impact on the texture of raw chicken breast samples, this effect was not observed in Sous-vide chicken breast samples. Additionally, the Sous-vide cooking process led to an increase in texture firmness compared to raw chicken breast samples, which is supported by study of Kerdpi boon in 2019, regardless of the storage state prior to cooking.

#### 5.2.4. Cooking loss of Sous-vide chicken breast

In this study, cooking loss is 18.32% for refrigerated sample and 16.07% for frozen sample, which is acceptable in range from 10.23% to 28.08% reported by Haghghi et al. (2020). There is no significant difference in cooking loss between Sous-vide chicken breast samples cooked under refrigerated versus frozen conditions with p value is 0.3393. Both cooking methods appear to result in similar levels of moisture retention during the cooking process.

#### 5.2.5. Sensory

The remarkable sensory characteristics, including vibrant colors, robust flavors, and intense tastes, are preserved exceptionally well. After pan-frying, the meat remains juicy and tender, devoid of the dry texture often associated with traditional cooking methods like frying raw chicken breast.



*Figure 12. Pan-frying Sous-vide chicken breast.*

Plastic foil acts as a barrier, preventing the loss of aromatic volatile compounds and moisture during the sous vide cooking process. This preservation enhances the sensory experience, leading to increased juiciness and tenderness in meat. Furthermore, the compression of meat during sous vide packaging helps maintain its desirable attributes, including flavor, natural color, and original shape, resulting in a fresh appearance that appeals to consumers, similar as results of Kerdpiboon et al., 2019; Park et al., 2020 and Haghghi et al., 2021.



### 5.3. Physical attribute changes of Sous-vide cooked chicken breast during 4 weeks of refrigerated storage.

After Sous-vide treatment, cooked chicken meat samples were stored at 3°C and frozen -25°C and measured every week. Therefore, it is possible to indicate the change of physical parameters of Sous-vide meat during storage, as shown in Figure 13-16. The Table 4-9 show the average values for cold storage conditions of 2 different sample conditions after 4 weeks.



*Figure 13. Visual comparison of sous-vide chicken breast appearance following 1 week of cold storage: Refrigerated sample (A-above) and frozen sample (B-below).*



*Figure 14. Visual comparison of sous-vide chicken breast appearance following 2 weeks of cold storage: Refrigerated sample (A-above) and frozen sample (B-below).*

#### 5.3.1. Evaluation of the color during cold storage of Sous-vide chicken breast

During refrigerated storage, as depicted in the provided Figure 17 and Table 4, the luminosity (L) values exhibit a significant resilience despite signs of frustration, while still maintaining the characteristic white tint of the surface coloration, as cooked chicken color. During the third week of refrigerated storage, cooked meat exhibits the

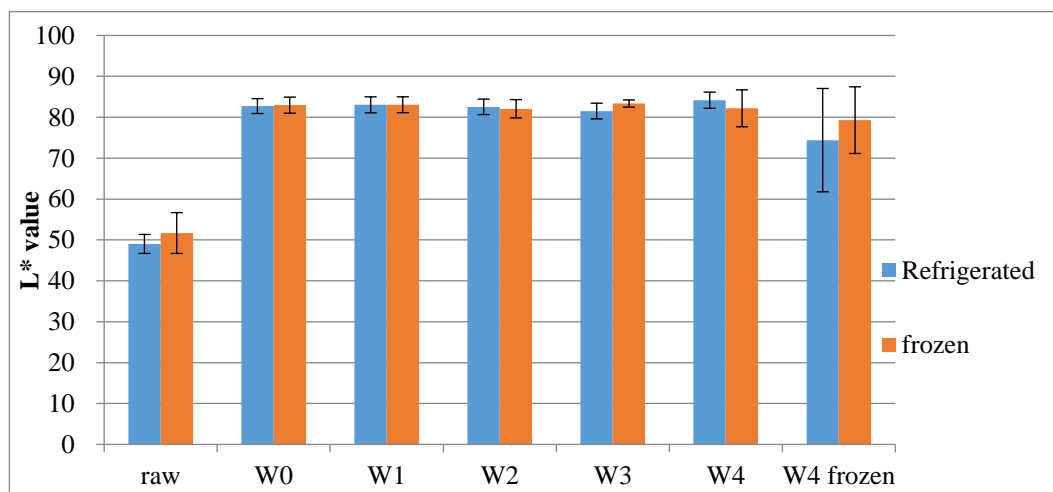
lowest  $L^*$  values throughout the observation period, measuring at  $81.51 \pm 1.93$  for refrigerated samples, whereas the frozen counterpart records the highest value at  $83.35 \pm 0.88$ .



**Figure 15.** Visual comparison of sous-vide chicken breast appearance following 3 weeks of cold storage: Refrigerated sample (A-below) and frozen sample (B-above).



**Figure 16.** Visual comparison of sous-vide chicken breast appearance following 4 weeks of cold storage: Refrigerated sample (A-below) and frozen sample (B-above).



**Figure 17.** Variation in  $L^*$  values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) conditions with two pre-treatment groups.

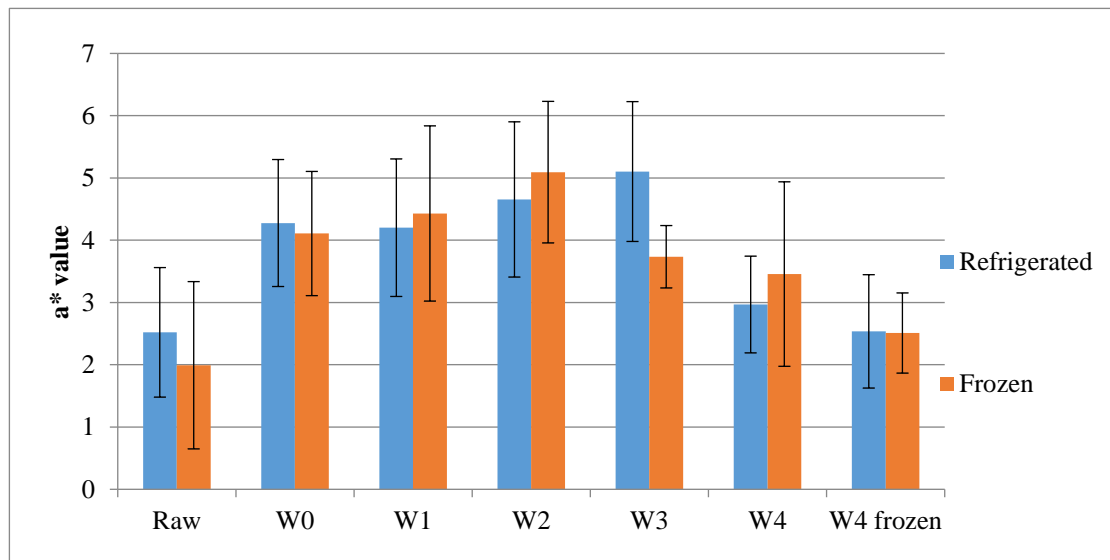
This disparity is statistically significant, with a p-value of 0.003. This indicates a pronounced darkening effect on the surface. Especially, frozen cooked meat exhibits a comparatively slower rate of darkening than its refrigerated counterpart. However, at the 4th stage of cooking, there's a significant drop in L\* values for frozen samples ( $82.18 \pm 4.51$ ) compared to refrigerated ones ( $84.17 \pm 1.98$ ), indicating a darker appearance for the frozen samples at this stage.

**Table 4. Change in L\* values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) conditions with two pre-treatment groups.**

L*	Average $\pm$ SD		Significant level
	Refrigerated	Frozen	P
raw	$49.02 \pm 2.32$	$51.69 \pm 4.99$	0.132
W0	$82.73 \pm 1.82$	$82.96 \pm 1.97$	0.033*
W1	$83.03 \pm 1.96$	$83.05 \pm 1.94$	0.079
W2	$82.54 \pm 1.89$	$82.07 \pm 2.24$	0.025*
W3	$81.51 \pm 1.93$	$83.35 \pm 0.88$	0.003**
W4	$84.17 \pm 1.98$	$82.18 \pm 4.51$	0.085
W4 Frozen	$74.40 \pm 12.64$	$79.28 \pm 8.15$	0.189

Similarly to L\*, the a\* values of both refrigerated and frozen samples fluctuated throughout the cold storage period (Figure 18 and Table 5) with no consistent trend observed in the differences between refrigerated and frozen conditions over time.

In the second week, frozen samples ( $5.09 \pm 1.14$ ) exhibited a slightly higher a\* value compared to refrigerated samples ( $4.66 \pm 1.25$ ), indicating a slightly redder appearance for the frozen samples at this stage. However, this variance was not statistically significant, with a p-value of 0.235. In the third week, a twist occurred: the frozen samples experienced a decline to  $3.73 \pm 0.50$ , while the refrigerated ones increased to  $5.10 \pm 1.12$ , with a p-value of 0.001. Finally, all samples showed a similar drop, with a non-significant p-value of 0.242.



**Figure 18.** Variation in  $a^*$  values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) conditions with two pre-treatment groups.

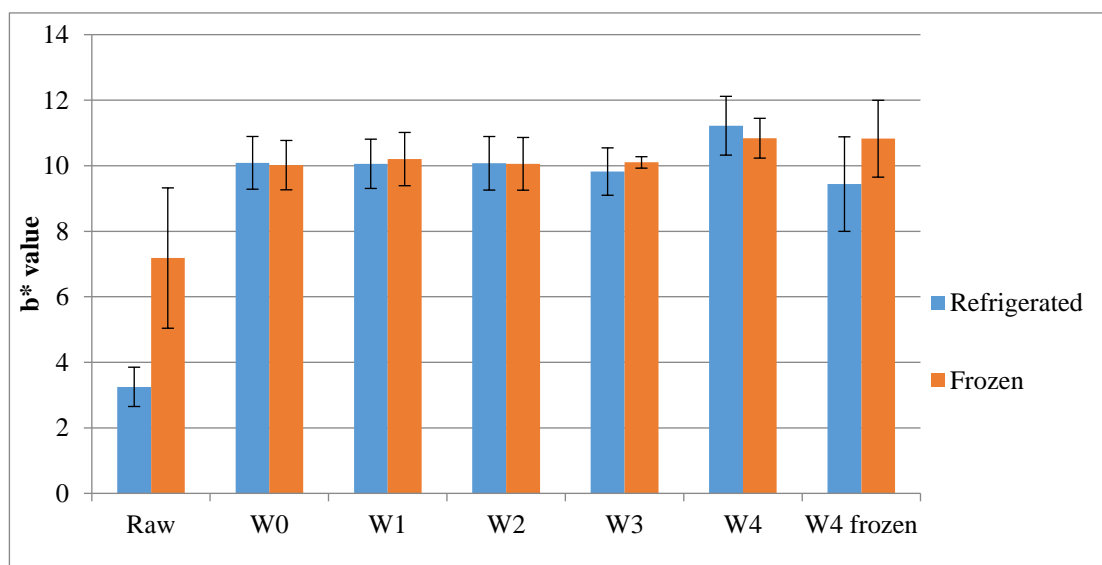
**Table 5.** Change in  $a^*$  values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) conditions with two pre-treatment groups.

$a^*$	Average $\pm$ SD		Significant level P
	Refrigerated	Frozen	
raw	2.52 $\pm$ 1.04	1.99 $\pm$ 1.34	0.188
W0	4.28 $\pm$ 1.02	4.11 $\pm$ 1.00	0.153
W1	4.20 $\pm$ 1.10	4.43 $\pm$ 1.41	0.398
W2	4.66 $\pm$ 1.25	5.09 $\pm$ 1.14	0.235
W3	5.10 $\pm$ 1.12	3.73 $\pm$ 0.50	0.001**
W4	2.97 $\pm$ 0.78	3.46 $\pm$ 1.48	0.242
W4 Frozen	2.54 $\pm$ 0.91	2.51 $\pm$ 0.64	0.440

*All values are presented as means  $\pm$  SD (n=9).*

Continuing with  $L^*$  and  $a^*$ , during the fourth week of cold storage, both refrigerated and frozen samples exhibited fluctuations in  $b^*$  values (Figure 19 and Table 6), with no consistent trend observed in the differences between them. In the

third week, there was a slight drop in  $b^*$  values for refrigerated samples ( $9.82 \pm 0.72$ ) and an increase for frozen samples, with the latter displaying a higher value ( $10.10 \pm 0.17$ ), indicating a yellower appearance for the frozen samples at this stage. Subsequently, both  $b^*$  values increased significantly in the fourth week, with refrigerated samples measuring at  $11.22 \pm 0.90$  and frozen samples at  $10.84 \pm 0.61$ , with a p-value of 0.1359. Taken together, these findings provide detailed insights into the increasing yellowness during the cold storage of Sous-vide chicken breast.



**Figure 19.** Variation in  $b^*$  values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen conditions (W4 frozen) with two pre-treatment groups.

After four weeks of storage under cold conditions for two types of raw material-treated cooked chicken breast, discernible differences in coloration are apparent. Between the initial and fourth week of cold storage, refrigerated cooked chicken breast exhibits more pronounced changes compared to its frozen counterpart, with a delta E value 2.25 higher than 1.41, indicating a more substantial shift in color perception (Table 7).

This trend persists after cooking and cooling, with the color disparity becoming even more pronounced, particularly by approximately fourfold. Specifically, Sous-vide chicken breast after four weeks of storage displays darker hues, reduced redness, and increased yellowness, suggesting a significant alteration in its color profile which are shown in Figure 16.

**Table 6. Change in  $b^*$  values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen conditions (W4 frozen) with two pre-treatment groups.**

<b><math>b^*</math></b>	<b>Average <math>\pm</math> SD</b>		<b>Significant level</b>
	<b>Refrigerated</b>	<b>Frozen</b>	<b>P</b>
raw	3.25 $\pm$ 0.6	7.18 $\pm$ 2.14	0.000**
W0	10.09 $\pm$ 0.8	10.20 $\pm$ 0.75	0.005**
W1	10.06 $\pm$ 0.75	10.20 $\pm$ 0.81	0.048*
W2	10.07 $\pm$ 0.82	10.06 $\pm$ 0.80	0.000**
W3	9.82 $\pm$ 0.72	10.10 $\pm$ 0.17	0.121
W4	11.22 $\pm$ 0.90	10.84 $\pm$ 0.61	0.136
W4 Frozen	4.11 $\pm$ 1.68	3.93 $\pm$ 0.56	0.048*

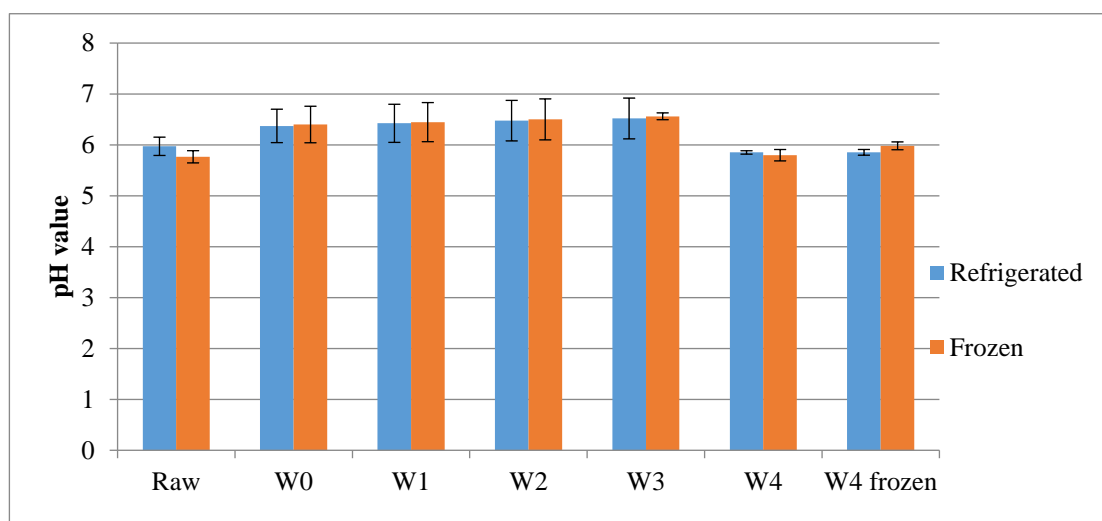
*All values are presented as means  $\pm$  SD (n=9).*

**Table 7. Delta E value during storage time after Sous-vide treatment**

<b>Material treatment</b>	<b>Storage treatment</b>	<b>Delta E</b>
Refrigerated	W0-W1	0.32
Frozen	W0-W1	0.38
Refrigerated	W1-W2	0.67
Frozen	W1-W2	1.19
Refrigerated	W3-W4	3.68
Frozen	W3-W4	1.41
Refrigerated	W0-W4 - Refrigerated	2.25
Frozen	W0-W4 - Refrigerated	1.41
Refrigerated	W0-W4 - frozen	8.53
Frozen	W0-W4 - frozen	4.09

### 5.3.2. Evaluation of the pH during cold storage of Sous-vide chicken breast

Over four weeks of storage under cold conditions, there are fluctuations in pH values for both refrigerated and frozen samples, as shown in Figure 19. At the 1st week, the pH values remain similar to the initial readings, with the refrigerated samples at  $6.42 \pm 0.37$  and the frozen samples at  $6.45 \pm 0.38$ . The p-value (0.115) suggests a trend towards significance but does not reach the conventional threshold.



**Figure 20.** Variation in pH values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen conditions (W4 frozen) with two pre-treatment groups.

**Table 8.** Change in pH values of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen conditions (W4 frozen) with two pre-treatment groups.

pH	Average $\pm$ SD		Significant level
	Refrigerated	Frozen	P
raw	$5.97 \pm 0.18$	$5.77 \pm 0.12$	0.004**
W0	$6.42 \pm 0.33$	$6.45 \pm 0.36$	0.333
W1	$6.42 \pm 0.37$	$6.45 \pm 0.38$	0.115
W2	$6.48 \pm 0.40$	$6.50 \pm 0.40$	0.092
W3	$6.52 \pm 0.40$	$6.56 \pm 0.07$	0.393
W4	$5.85 \pm 0.03$	$5.80 \pm 0.11$	0.069
W4 Frozen	$5.85 \pm 0.06$	$5.98 \pm 0.08$	0.002**

All values are presented as means  $\pm$  SD (n=9).

Then, in the next week, pH values slightly increase for both refrigerated and frozen samples, with the refrigerated samples at  $6.48 \pm 0.40$  and the frozen samples at  $6.50 \pm 0.40$ . The p-value (0.092) is close to significance but still not statistically significant. pH values continue to rise at the 3rd week, with the refrigerated samples at  $6.52 \pm 0.40$  and the frozen samples at  $6.56 \pm 0.07$ . However, the p-value (0.393) indicates that the difference is not statistically significant. However, there is a significant decrease in pH values at week 4 for both refrigerated ( $5.85 \pm 0.03$ ) and frozen samples ( $5.80 \pm 0.11$ ). However, the p-value (0.393) suggests that this difference is not statistically significant.

Overall, the pH values fluctuate slightly over the four-week period for both refrigerated and frozen samples, with no consistent pattern observed. The p-values indicate that the differences in pH between refrigerated and frozen samples at each week are not statistically significant. Similarly, O Baston reported a continuous increase in the pH of the meat from 5.92 on the first day to 7.33 by the 20th day of refrigerated storage. However, after a month of cold storage, a remarkable decrease in pH values in both groups by the fourth week, which is not statistically significant either.

### **5.3.3. Evaluation of the texture during refrigerated storage of Sous-vide chicken breast products.**

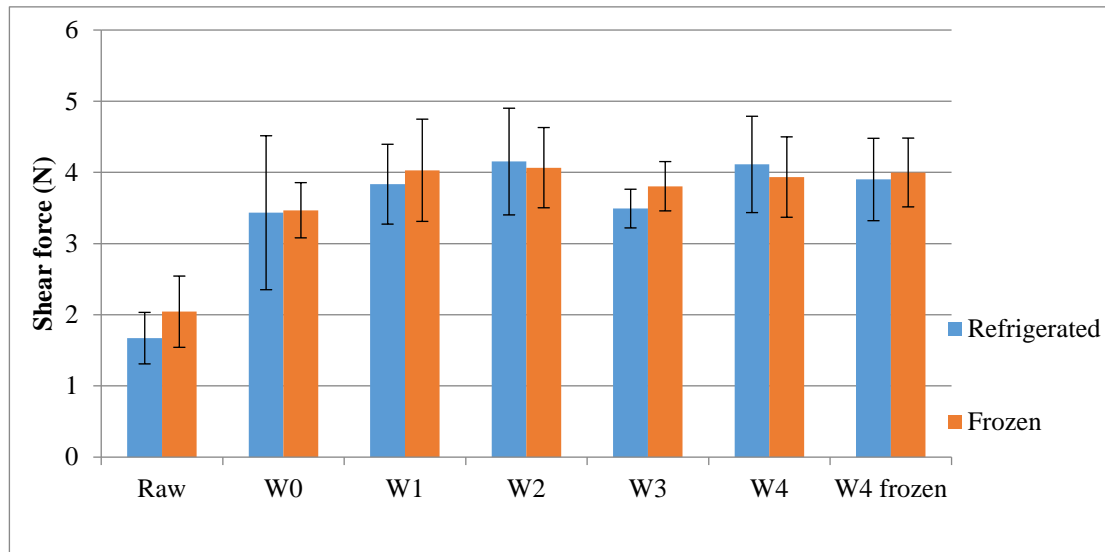
At the initial sous vide stage of the refrigerated sample, the average hardness measured 3.43 N. Over the 4-week storage period, hardness exhibited fluctuations, peaking at W2 ( $4.15 \pm 0.75$ ) and W4 ( $4.11 \pm 1.68$ ), indicating tougher meat (Figure 21 and Table 9). Conversely, the frozen sample displayed a slightly higher initial average hardness of 3.47 N compared to the refrigerated samples.

Similar to the refrigerated samples, hardness fluctuated throughout the storage period, reaching its peak at W2 ( $4.07 \pm 0.56$ ). Both refrigerated and frozen samples showed an overall increase in hardness over the 4-week storage period. While frozen samples initially tended to have slightly higher hardness values, they followed a similar fluctuation pattern to the refrigerated samples over time.

At the initial sous vide stage of the refrigerated sample, the average hardness measured 3.43 N. Over the 4-week storage period, hardness exhibited fluctuations, peaking at W2 ( $4.15 \pm 0.75$ ) and W4 ( $4.11 \pm 1.68$ ), indicating tougher meat. Conversely,



the frozen sample displayed a slightly higher initial average hardness of 3.47 N compared to the refrigerated samples.



**Figure 21.** Variation in texture of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) condition with two pre-treatment groups.

**Table 9.** Change in texture of raw, sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) condition with two pre-treatment groups.

Shear force (N)	Average $\pm$ SD		Significant level P
	Refrigerated	Frozen	
raw	1.67 $\pm$ 0.36	2.04 $\pm$ 0.50	0.028*
W0	3.43 $\pm$ 1.08	3.47 $\pm$ 0.39	0.457
W1	3.83 $\pm$ 0.56	4.03 $\pm$ 0.72	0.137
W2	4.15 $\pm$ 0.75	4.07 $\pm$ 0.56	0.406
W3	3.49 $\pm$ 0.27	3.81 $\pm$ 0.35	0.036*
W4	4.11 $\pm$ 1.68	3.93 $\pm$ 0.56	0.358
W4 Frozen	3.90 $\pm$ 0.58	4.00 $\pm$ 0.48	0.304

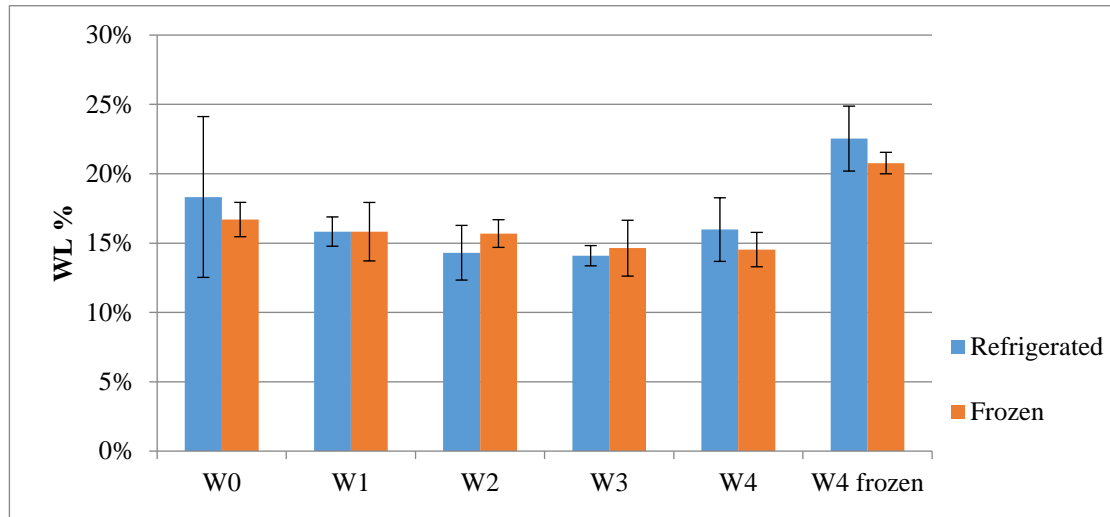
*All values are presented as means  $\pm$  SD (n=9).*

Similar to the refrigerated samples, hardness fluctuated throughout the storage period, reaching its peak at W2 (4.07  $\pm$  0.56). Both refrigerated and frozen samples showed an overall increase in hardness over the 4-week storage period. While frozen

samples initially tended to have slightly higher hardness values, they followed a similar fluctuation pattern to the refrigerated samples over time.

### 5.3.4. Evaluation of weight loss during refrigerated storage of Sous-vide chicken breast

This experiment demonstrated how cold storage time affected weight loss (Figure 22 and Table 9).



**Figure 22.** Variation in weight loss sous-vide treated, and stored chicken breast after 4 weeks under refrigerated (W0-W4) and frozen (W4 frozen) conditions with two pre-treatment groups.

**Table 10.** Weight loss changes in sous-vide chicken breast during 4 weeks storage: Comparing refrigerated and frozen conditions.

Weight loss (%)	Average ± SD		Significant level (P)
	Refrigerated	Frozen	
W0	18.32 ± 0.06	16.70 ± 0.01	0.339
W1	15.83 ± 0.01	15.82 ± 0.02	0.498
W2	14.30 ± 0.02	15.69 ± 0.01	0.226
W3	14.09 ± 0.01	14.63 ± 0.02	0.379
W4	15.97 ± 0.02	14.53 ± 0.01	0.275
W4 frozen	22.54 ± 0.02	20.77 ± 0.01	0.203

In this instance, the weight loss decreases over four weeks, with the frozen sample decreasing more gradually. By week four, the refrigerated sample had increased to  $15.97 \pm 0.02$ , which was still much less than the sample that had been treated sous vide. The weight loss percentages of frozen and refrigerated Sous-vide chicken breast samples over a 4-week period, exhibit statistically insignificant differences in weight loss between the two storage regimes.

#### 5.4. Physical attribute changes of Sous-vide cooked chicken breast after 4 weeks of frozen storage.

After Sous-vide treatment, cooked chicken meat samples were stored at frozen  $-25^{\circ}\text{C}$  and measured after 4 week. Therefore, it is possible to indicate the change of physical parameters of Sous-vide meat during storage. The Table 10 and Figure 23 shows the average values for 2 different storage conditions of 2 different sample conditions after 4 week.

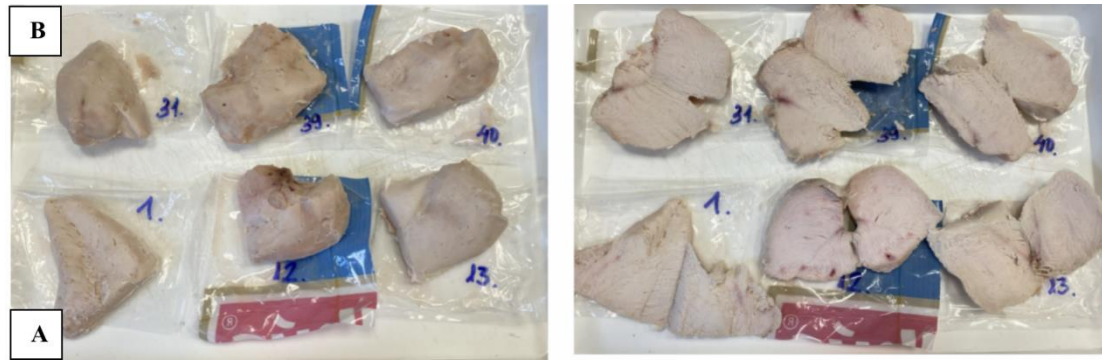
##### 5.4.1. Evaluation of the color after frozen storage of Sous-vide chicken breast

After being stored in a frozen state for one month, there was a significant difference in the colour parameters between the samples that were refrigerated and those that were frozen.

*Table 11. Characteristics of Sous-vide chicken breast in 4 weeks frozen storage (n=9)*

Feature	Average $\pm$ SD		Significant level P
	Refrigerated	Frozen	
Color parameters: L*	$74.40 \pm 12.64$	$79.28 \pm 8.15$	0.189
a*	$2.54 \pm 0.91$	$2.51 \pm 0.64$	0.440
b*	$9.44 \pm 1.44$	$10.82 \pm 1.17$	0.0481*
pH	$5.85 \pm 0.06$	$5.98 \pm 0.08$	0.002**
Texture - Shear force (N)	$3.90 \pm 0.58$	$4.00 \pm 0.48$	0.304

*All values are presented as means  $\pm$  SD (n=9).*



**Figure 23. Visual comparison of sous-vide chicken breast appearance following 4 weeks of frozen storage: Refrigerated sample (A-below) and frozen sample (B-above)**

The initial  $L^*$  value of the refrigerated samples was  $82.73 \pm 1.82$ , and after four weeks, it decreased to  $74.40 \pm 12.64$ . In contrast, after the same amount of time, the  $L^*$  value of the frozen samples dropped from  $82.96 \pm 1.97$  to  $79.28 \pm 8.15$ . Over the course of four weeks, there was an unfavourable decrease in the  $L^*$  value for both groups of samples ( $p = 0.189$ ), which is suggestive of flesh darkening. Significantly, the drop in the refrigerated samples was more noticeable, indicating that frozen storage would provide better retention of beef lightness. Similarly, after four weeks, the initial  $a^*$  value for chilled beef was  $4.28 \pm 1.02$  and decreased slightly to  $2.54$ . On the other hand, frozen meat started off with an  $a^*$  value of  $4.11 \pm 1.00$  and remained rather stable at  $2.51$  for the same amount of time. Over the course of four weeks, the  $a^*$  value decreased in both frozen and refrigerated samples, signifying a loss of redness. Between the frozen and chilled samples, there was, nevertheless, a small and statistically insignificant difference ( $p = 0.440$ ). Regarding the green/yellow colour, after four weeks, the  $b^*$  value of the chilled samples dropped from  $10.06 \pm 0.8$  to  $9.44 \pm 1.44$ . On the other hand, after the same duration, the frozen samples showed an increase to  $10.82 \pm 1.17$  from an initial  $b^*$  value of  $10.20 \pm 0.75$ . Changes in the  $b^*$  value during a four-week period were evident in both sets of samples. But the difference was more noticeable in the frozen samples, suggesting that freezing storage can cause a more noticeable increase in yellowness than refrigeration. In frozen storage, Sous-vide chicken breast after four weeks of storage displays darker hues, reduced redness, and increased yellowness.

#### **5.4.2. Evaluation of the pH after frozen storage of Sous-vide chicken breast**

After one month of storage under frozen conditions, a remarkable difference emerged in the pH values between the refrigerated and frozen samples. The

refrigerated samples showed a decrease in pH to  $5.85 \pm 0.06$ , while the frozen samples exhibited a higher pH of  $5.98 \pm 0.08$ . Importantly, the p-value associated with this comparison is 0.002, indicating a statistically significant difference between the pH values of the refrigerated and frozen samples after one month of freezing. Besides, Sous-vide chicken breast under store of refrigerator was obtain a decrease of pH after 1 month as well. Additionally, Previous studies have shown that freezing with subsequent exudate release and the loss of water from the meat may cause an increase in the concentration of the solutes, resulting in a decrease in the pH of thawed meat (Leygonie C et al., 2012).

#### **5.4.3. Evaluation of the texture after frozen storage of Sous-vide chicken breast**

The average shear force values are  $3.90 \pm 0.58\text{N}$  for refrigerated samples and  $4.00 \pm 0.48\text{N}$  for frozen samples, which exhibit the harder and tougher in texture of cooked chicken breast. These values suggest that the frozen samples require slightly more force to shear compared to the refrigerated ones. However, the p-value associated with the comparison (0.304) indicates that this difference is not statistically significant. In other words, there is no strong evidence to suggest that the observed difference in texture between refrigerated and frozen samples is not due to random chance alone.

These findings suggest that the freezing technique produced a similar meat texture compared to that achieved by the chilling method. This result aligns with a previous study conducted on superchilled chicken breast meat (Kerdpi boon et al., 2019).

#### **5.4.4. Evaluation of the weight lost after frozen storage of Sous-vide chicken breast**

Compared to the 4-week cold storage, there was an apparent rise in weight loss in the frozen/thawing condition over time. The average percentage of weight loss increased to  $22.54\% \pm 0.02$  for chilled meat and  $20.77\% \pm 0.01$  for frozen meat after four weeks of storage. With a p-value of 0.203, there was, however, no statistically significant impact seen in relation to the pre-stored chicken meat material's attributes on this front. Bahuaud et al. (2008) reported that in superchilled fish fillets (stored at  $-1.5^\circ\text{C}$ ), myofiber detachment and breakage increased with storage time due to ice crystal formation, resulting in heightened water loss during storage. Liquid loss

comprises both substances and water leaking from cells as they undergo disruptive processes such as storage and thawing. Consequently, these losses impact the flavor, texture, and appearance of fresh meat, with the liquid exudate serving as an excellent nutritive source for bacteria growth (Duun & Rustad, 2008; Liu et al., 2013).

## **VI. Conclusion**

Color analysis revealed significant differences between refrigerated and frozen storage conditions. Sous-vide chicken breast exhibited darker hues, reduced redness, and increased yellowness over time. Frozen storage generally resulted in a lighter appearance with a more pronounced yellow hue and a slower rate of darkening in cooked meat compared to refrigerated storage. Although pH levels showed a decrease, particularly after prolonged storage, the differences were not statistically significant, indicating that both storage methods maintained acceptable pH levels for chicken meat. Texture analysis demonstrated that frozen storage led to a slightly firmer texture in cooked chicken breast compared to refrigerated storage, although the difference was not statistically significant. Both methods exhibited an increase in observed hardness. Weight loss analysis showed no significant difference between refrigerated and frozen storage conditions pre-treatment, indicating that chilling chicken breast before Sous vide does not affect the quality of the product. However, there was an increase in weight loss observed over time in frozen storage, which may affect the final product and consumer acceptance. Based on the color, pH, texture and weight loss results in the present study, both refrigerated and frozen storage methods are viable options for preserving minimally processed meat, particularly with Sous-vide treatment. While frozen storage may offer slightly better color retention and texture firmness over prolonged storage periods, both methods maintain acceptable quality characteristics of the meat. The freezing process damages the structural integrity of chicken meat and decreases its ability to retain water, which could affect consumer acceptance. Therefore, the choice between refrigerated and frozen storage should be based on factors such as convenience, storage space, and specific quality preferences.

## **VII. Summary**

This study aimed to assess the changes in physical characteristics of sous vide-cooked chicken breast during refrigerated storage at 3°C and frozen storage at -25°C before and after sous-vide cooking for a duration of 4 weeks. Cooking loss and shear force significantly increased, while expressible drip decreased along with a reduction in water holding capacity in both storage conditions. Redness of meat juice decreased significantly during storage, while yellowness and darkness increased notably in both groups after sous-vide cooking at week 4. The chicken breast samples became more acidic and tougher, indicating the impact of storage conditions and duration on the final product, regardless of chilling or freezing. These findings provide preliminary insights for further research on sous-vide cooking of chicken breast and frozen storage conditions to enhance quality control measures



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