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Sous-vide cooking process and its effect on the quality and shelf life of beef steaks

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Abstract*– The present research aimed to characterize the effect of the sous-vide cooking process at moderate temperatures on the quality and shelf life of beef steaks. Thermal lethality studies were conducted using Staphylococcus aureus ATCC 25923 and Escherichia coli ATCC 25922 strains inoculated into vacuum-packaged steaks and cooked at 55 °C, 60 °C, and 65 °C for 0, 20, 40, 60, and 80 minutes. Based on microbial counts, decimal reduction times and lethally equivalent time–temperature combinations were determined: 55 °C × 60 minutes, 60 °C × 55 minutes, and 65 °C × 50 minutes. These conditions were applied to evaluate physicochemical quality (pH, water-holding capacity, and cooking loss) and sensory texture, with the treatment at 55 °C for 60 minutes being selected due to its higher water-holding capacity and lower weight loss. Subsequently, the treated steaks were stored under refrigeration (3 ± 2 °C) for 60 days, during which physicochemical (pH), sensory (color, odor, appearance, flavor, and texture), and microbiological parameters (mesophilic aerobes, Staphylococcus aureus, coliforms, Escherichia coli, and Salmonella spp.) were evaluated to estimate shelf life. The results indicated that pH did not vary significantly among cooking treatments and that the texture of steaks cooked at 55 °C and 60 °C was similar. The limiting parameter for shelf life was the mesophilic aerobic count, with an estimated shelf life of 67 days under refrigerated conditions. It is concluded that sous-vide cooking at 55 °C for 60 minutes provides the best quality and allows the shelf life of the steaks to be extended by 60 days compared to conventionally cooked products.

***Keywords*-- Lethality, D-value, vacuum cooking, and shelf life.**

I. INTRODUCTION

In recent years, trends in food consumption have undergone changes because of new lifestyles and the redefinition of the concept of nutrition. This has led to a renewed appreciation of philosophies such as that proposed by Hippocrates [1], who maintained that the primary function of the foods we consume is to preserve health and prevent disease, and that of Plato [2], who promoted a diet based on natural foods such as fruits, vegetables, and cereals in order to keep the soul in a state of harmony and control over physical desires.

Peru has not been immune to these changes. In 2013, Law No. 30021 on the Promotion of Healthy Eating for Children and Adolescents was enacted, incorporating the recommendations of both philosophers and aiming to promote a varied diet, preferably in its natural state or with minimal processing, that provides energy and all the essential nutrients required to maintain health and improve quality of life. In addition, the regulations of this law, approved by Supreme Decree No. 017-2017-SA, establish maximum values for technical parameters such as sodium, total sugar, saturated fats, and trans fats; when these limits are exceeded, food manufacturers are required to include warning labels on their packaging to discourage frequent consumption.

Currently, the food market is showing a growing demand for products that are easy to prepare, contain fewer or no additives, and at the same time maintain their nutritional and sensory characteristics as close as possible to traditional foods [3]. Likewise, there has been a considerable increase in interest in products that are quick and simple to prepare and have a long shelf life, such as precooked meals, fourth- and fifth-range products, and other “ready-to-eat” foods [4].

A recent innovation in food development and production, mainly driven by the HORECA sector (Hotels, Restaurants, and Catering), is the set of technologies known as Cook-Chill. This system involves the preparation of ready-to-eat foods, generally in individually vacuum-packed portions, which are then stored under refrigeration or freezing. Its main advantage is the ease of reheating, as the products can be heated in a microwave oven just before serving [5]. Products made using these technologies include preparations based on beef, chicken, fish, and seafood, as well as various fruit and vegetable products such as vegetables, fruits, legumes, and cereals, among others [6]. Cook-Chill technologies allow food to be cooked under controlled time and temperature conditions, separating the preparation process from the distribution of the final product. This is possible because foods can be kept under refrigeration or freezing for longer periods than traditional cooking methods, thereby ensuring their preservation and quality for an extended time [5]. Productivity and efficiency increase when tasks are planned and executed in advance and centralized in a main kitchen, where food preparation, cooking, cooling, and storage are carried out. This approach allows for a reduction in labor and ingredient consumption, while the available time can be used to portion products into standardized servings and manage their distribution. In addition, efficiency is improved through the optimized use of existing equipment and infrastructure. Often, sous-vide is used as the cooking phase in a Cook-Chill system [7].

The sous-vide cooking system is considered an emerging technology that originated in French cuisine under the name Cuisson en papillote, a technique that involved cooking food wrapped in paper. Chef Georges Pralus, from Briennon, France, was the first to apply this method in 1974 with the aim of reducing weight loss in foie gras, a traditional French dish. To achieve this, he placed the food in a vacuum-sealed plastic bag and cooked it in a water bath at a controlled temperature. This procedure reduced product shrinkage from 40% to 5%, while also enhancing its sensory qualities [8]. The sous-vide method employs the principle of low-temperature, long-time (LTLT) thermal treatment and provides better control over the texture, flavor, and doneness of the product [9]. The thermal treatment and moist environment break down myofibrillar proteins and solubilize connective tissue, resulting in more tender meat [10]. Other main advantages of sous-vide are economic—due to better use of labor and equipment through centralized production—and qualitative, including reduced need for flavor enhancers, better preservation of vitamins, and greater juiciness retention [11].

Vacuum cooking differs from traditional cooking methods in two fundamental ways: first, raw foods are vacuum-packed in heat-stable, food-grade bags; and second, the bags are cooked at controlled, relatively low temperatures [11, 12, 13]. The main advantages of vacuum packaging are the low cooking temperature and improved heat transfer during cooking and cooling, achieved by eliminating air bubbles between the package and the product, the inhibition of aerobic microorganism growth, and space savings. Precise temperature control is even more important than vacuum packaging because it allows for (a) exceptional reproducibility; (b) greater control over doneness (compared to traditional methods); (c) the ability to pasteurize foods at lower temperatures (i.e., food does not need to be overcooked to be safe); and (d) the ability to tenderize tough cuts while maintaining a medium doneness [8]. However, the long cooking time required for this method often makes it less cost-effective, which may explain why it has not been widely adopted in the meat industry.

In this context, the present research aimed to determine how the sous-vide cooking method affects the quality and shelf life of beef steaks.

II. MATERIAL AND METHODS

A. *Type of Research*

The research is applied with an experimental approach. Samples from each applied treatment were evaluated, and the data obtained were processed using statistical analysis, which allowed for determining the best sous-vide cooking time–temperature combination and estimating the shelf life of the treated steaks. These results will serve as a recommendation for collective food services and restaurants in general.

B. *Population*

The population consisted of 1,000 steaks, each weighing 150 g, obtained from processing 150 kg of Peruvian pasture-raised beef, narrow striploin cut, from the brand Llanuras de Oxapampa - Perú.

C. *Sample*

The sample size was 279 steaks, which was determined using the sample size equation for finite populations, considering a Z-value for a 95% confidence level, an estimated proportion of 0.5, and a 5% margin of error [15].

D. *Study Factor*

The microbial suspensions of *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 used to inoculate the steaks were obtained after enrichment in BHI broth until reaching a bacterial population with turbidity equivalent to 0.5 McFarland standard. A total of sixty steaks were inoculated with 10 mL of the *Staphylococcus aureus* ATCC 25923 suspension, with 4 repetitions for each temperature value (55 °C, 60 °C, and 65 °C) and sous-vide cooking time (0, 20, 40, 60, and 80 minutes) [16]. A similar procedure was carried out for the *Escherichia coli* ATCC 25922 strain.

E. *Determination of Decimal Reduction Time and Lethally Equivalent Cooking Times*

The D-value was calculated using the graphical method. For the cooking times (UP), the goal was to achieve a decimal reduction in the inoculated strains that would result in a cooking time between 30 and 50 minutes at 65 °C [17].

F. *Selection of the Best Sous-Vide Cooking Time–Temperature Combination*

The selection criteria used were water-holding capacity (WHC), cooking weight loss, and the texture of the cooked samples.

1) Water-Holding Capacity (WHC)

Water-holding capacity was determined by the weight difference of a steak sample before and after cooking [18]. This procedure was performed in six replicates, using the following equation:

$$WHC\,(\%) = \frac{Wraw\ steak\ -\ Wcooked\ steak}{Wraw\ steak} \times 100$$

2) Cooking Weight Loss (CWL)

Cooking weight loss was determined by weighing the water released inside the bag during the cooking of the steaks and dividing it by the weight of the raw steak before vacuum packaging [18]. This procedure was performed in six replicates, using the following equation:

$$CWL\,(\%) = Wreleased\ water\ \ x\ 100$$

$W_{raw\ steak}$

3) Texture

Texture was evaluated using a ranking test. Six trained judges were asked to rank the samples from lowest to highest hardness [19].

G. *Estimation of the Shelf Life of Steaks under Refrigeration Conditions*

The steaks were stored for 60 days under refrigeration (3 ± 2 °C) and evaluated on day 0, 30, 45, 50, 55, and 60 for physicochemical quality (pH), sensory attributes (color, odor, appearance, flavor, and texture), and microbiological parameters (mesophilic aerobes, *Staphylococcus aureus*, coliforms, *Escherichia coli*, and *Salmonella* spp.). Each parameter or attribute was assessed in six replicates to obtain average values.

Using the generated data, and applying chemical kinetics equations, the order of reaction, rate of deterioration, and estimated shelf life were determined. The failure criteria considered were a pH value greater than or equal to 6.8 for physicochemical quality [20], a value of 5.5 on the Karlsruhe scale for sensory quality [21], and the maximum allowable limits for microbiological parameters established by the sanitary standard NTS No. 071-MINSA/DIGESA.

1) pH**:** The pH evaluation was performed in six replicates using the potentiometric method [22].

2) Sensory Evaluation of Color, Odor, Appearance, Flavor, and Texture: This was carried out using the same trained panel that participated in the texture ranking test. This time, they were asked to assess the quality of each sensory attribute using the Karlsruhe quality rating scale, which consists of 9 points, with each value on the scale clearly described [21].

3) Mesophilic Aerobes: Determined by colony counting at 30 °C using the plate pour method [23].

4) Total Coliforms: Determined using liquid media, inoculated with serial dilutions of the samples and incubated at 37 °C to estimate the number of coliforms through the Most Probable Number (MPN) method, based on positive and negative results [24].

5) *Staphylococcus aureus*: Determined by colony counting on Baird-Parker agar plates, inoculated using a Drigalsky loop on the agar surface with horizontal, vertical, and circular movements, and incubated at 37 °C [25].

6) *Escherichia coli*: Determined using liquid media, inoculated with serial dilutions of the samples and incubated at 37 °C in Lauryl Sulfate broth for 48 hours. Tubes showing gas formation were then transferred to EC broth using an inoculating loop and incubated for 48 hours at 44 °C. After incubation, EC broth tubes with gas and turbidity were transferred to peptone water tubes, incubated at 44 °C for 48 hours, and confirmed using the indole test [26].

7) *Salmonella* spp.: Determined by isolation on Xylose Lysine Deoxycholate (XLD) agar, which allowed differentiation of typical *Salmonella* colonies based on their biochemical and fermentation characteristics. Finally, suspected colonies were subjected to confirmatory biochemical tests (TSI agar, lysine decarboxylase broth, and urea broth) and serological tests using somatic (O) and flagellar (H) antigens [27].

III. RESULTS AND DISCUSSION

A. *Determination of Decimal Reduction Time and Lethally Equivalent Cooking Times*

Tables 1 and 2 show that sous-vide cooking treatments at temperatures of 55 °C, 60 °C, and 65 °C for 50 minutes can achieve a reduction of up to 8 logarithmic cycles (8D) in viable cells of *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 inoculated in beef steaks.

Table 1
E. coli counts in beef steaks at different cooking times

Temperature	**Time (min)**				
	0	**20**	**30**	**40**	**50**
55°C	180000000	190000	930	48	<10
	120000000	180000	920	41	<10
	100000000	160000	900	33	<10
	110000000	170000	910	37	<10
Average	127500000	175000	915	39,75	
Log (Average)	8,11	5,24	2,96	1,60	
60°C	230000000	170000	580	19	<10
	210000000	160000	590	18	<10
	160000000	140000	550	11	<10
	190000000	150000	570	15	<10
Average	197500000	155000	572,50	15,75	
Log (Average)	8,30	5,19	2,76	1,20	
65°C	590000000	110000	130	13	<10
	520000000	100000	120	12	<10
	470000000	88000	110	11	<10
	500000000	95000	120	11	<10
Average	520000000	98250	120	11,75	
Log (Average)	8,72	4,99	2,08	1,07	

Table 3 shows that by using the maximum UP value (50 min), established by [17] for meat and cooked meat products at a temperature of 65 °C, it is possible to achieve a 10D reduction in the population of viable cells of *Staphylococcus aureus* ATCC 25923 and *Escherichia coli* ATCC 25922 in beef steaks. On the other hand, it is evident that there is no significant difference (P-value = 0.05) between the cooking times (UP) obtained for each indicator microorganism (*Staphylococcus aureus* ATCC 25923 and *Escherichia coli*

ATCC 25922) at each of the different temperatures evaluated, with the difference between them being 1 minute for the temperatures of 55 °C and 60 °C.

Table 2
Counts of S. aureus in beef steaks at different cooking times

Temperature	Time (min)				
	0	20	30	40	50
55°C	280000000	19000	910	46	<10
	210000000	16000	990	41	<10
	190000000	15000	880	40	<10
	240000000	18000	940	43	<10
Average	230000000	17000	930	42,50	
Log (Average)	8,35	4,23	2,97	1,63	
60°C	430000000	15000	420	20	<10
	490000000	17000	480	22	<10
	410000000	14000	470	18	<10
	450000000	16000	410	23	<10
Average	445000000	15500	445	20,75	
Log (Average)	8,65	4,19	2,65	1,32	
65°C	720000000	14000	150	12	<10
	710000000	10000	140	11	<10
	770000000	9600	120	12	<10
	760000000	9700	130	11	<10
Average	740000000	10825	135	11,5	
Log (Average)	8,87	4,03	2,13	1,06	

Table 3
Lethally Equivalent Sous-Vide Cooking Process Times to Ensure the Safety of Beef Steaks

Microorganism	Par (min)	Temperature (°C)		
		55	60	65
E. coli	Valor D	6.0	5.5	5.0
	UP	60	55	50
S. aureus	Valor D	5.9	5.4	5.0
	UP	59	54	50

There is considerable disagreement regarding both the indicator microorganism and the intensity of the thermal treatment to which different types of steam-cooked foods should be subjected. The sensory quality of these foods is considered a very important factor and must be considered; therefore, it is necessary to apply treatments aimed at inactivating vegetative cells of anaerobic psychotropics microorganisms. *Enterococcus faecalis* or *Listeria monocytogenes* have been proposed as indicator microorganisms of sanitary quality [11].

On the other hand, in the United States it is recommended that, for dishes containing beef and stored under refrigeration, the cooking treatment should be sufficient to achieve a 5D reduction of *Escherichia coli* O157:H7, whereas for poultry-based dishes a 7D reduction of *Salmonella spp*. is advised [28].

The thermal destruction values obtained for the microorganisms evaluated in the present study were well above the 6D reductions reported by [29] for L. monocytogenes in beef fillets obtained from the superficial pectoralis muscle treated with papain and subjected to sous-vide cooking at 65 °C for 9 min and at 70 °C for 2 min; as well as those reported by [30] for pork loins refrigerated at 4 °C for 24 h and frozen at −20 °C for 15 days, inoculated with *Listeria monocytogenes* Scott A and CECT 4031 strains and subjected to sous-vide cooking for 30, 60, and 90 min; and the 4D reductions of *Salmonella enteritidis*, *Salmonella senftenberg*, and a *Salmonella cocktail* achieved by [16] in pork loins treated at 50 °C and 60 °C for 30, 60, and 90 min. This is presumably because the sous-vide cooking processes in the present investigation were carried out in agitated water baths rather than in steam-injection ovens, as in the studies. In those cases, thermal treatments begin at sublethal temperatures and the increase in the internal temperature of the food is not sufficiently rapid, leading to physiological changes in bacterial cells that increase their resistance to subsequent stress. In this regard, [31] indicated that this situation constitutes a potential risk to food safety, as a greater-than-expected number of pathogenic bacterial cells could survive.

Another factor that may have contributed to the higher lethality observed in the present work compared with the results reported by [16, 30] can be explained by the deviations from the D/z model observed by [32], particularly at mild temperatures and for vegetative cells. The first reason is that the assumption of a linear relationship between log D and temperature (T) assigns disproportionate weight to low temperatures at the expense of high temperatures, where most of the inactivation occurs [33]. Especially at low temperatures, the semilogarithmic survival curves of many microorganisms and spores are not linear. Therefore, forcing a linear fit to experimental survival data that exhibit curved patterns may lead to an underestimation or overestimation of the thermal process, thereby increasing the risk to food safety.

B. Selection of the Best Sous-Vide Cooking Time–Temperature Combination

Table 4
Quality parameters of beef steaks cooked sous-vide at three temperatures

Quality Attribute or Parameter	Shelf Life (days) of Cooked Beef Steaks	
	Sous Vide 50°C	Conventionally
Color	158^a	122^a
Odor	147^a	112^a
Appearance	128^a	80^b
Flavor	197^a	65^b
Texture	214^a	95^b
pH	382^a	86^b
Mesophilic Aerobes	67^a	7^b

Table 4 shows that with a sous-vide cooking temperature of 55 °C for 60 minutes, there is sufficient statistical evidence (P-value = 0.05) to state that the physicochemical quality parameters—water-holding capacity, cooking weight loss, and the texture attribute—are higher.

The pH of the beef fillets cooked at the three temperatures evaluated (55, 60, and 65 °C) was 5.50, a value that falls within the ranges of 5.47–5.54 and 5.5–6.4 established by [34] and [35], respectively, but is slightly below the ranges of 5.67–5.78 and 5.91–6.11 reported by [20] and [36]. In this regard, [37] states that meats subjected to high temperatures experience an increase in pH, due to the release of amines present in animal tissues during the cooking process, which confer a more basic character to the meat.

With respect to water-holding capacity (WHC) and cooking losses, significant differences were observed among the three temperatures evaluated (55, 60, and 65 °C) ($P < 0.05$). Cooking loss values in the samples ranged from 1.3 to 2.1%, which are markedly lower than the 26.50 ± 6.73% reported by [38] for pork and the 28.54 ± 1.43% estimated by [39] for cooked chicken breasts, but very similar to the 0.6% reported by [40] for turkey breasts cooked for 10 h at 60 °C. According to [41], sous-vide cooking temperature is a critical factor in meat products, as it selectively denatures certain proteins while leaving others intact, thereby allowing the production of cuts with improved attributes such as juiciness and tenderness, which are enhanced at lower treatment temperatures. The increase in cooking losses with temperature has been widely reported in the literature; in this regard, [42] indicates that at approximately 65–66 °C the most significant change in water flow within the fillet is detected, as collagen contraction and muscle fiber contraction occur simultaneously. However, [43] concludes that there is no evidence that collagen contributes to an increase in cooking losses when it contracts at around 65 °C and that, above 42 °C, losses are mainly due to muscle fiber contraction. The longitudinal and transverse contraction of muscle structure during heating causes a reduction in fillet volume of up to 10%, leading to the expulsion of free water, since approximately 80% of the water in muscle is bound to myofibrillar proteins [8].

Sensory texture was significantly more tender (P-value = 0.05) in vacuum-cooked beef fillets treated at 55 °C and 60 °C than in those cooked at 65 °C. This finding is consistent with that reported by [44], who state that prolonged cooking can double meat tenderness by converting collagen entirely into gelatin and reducing fiber-to-fiber adhesion to nearly zero; indeed, this principle underlies the stewing of meats and has been applied since ancient times. Likewise, these authors found that tough cuts of beef (from animals aged 0 to 4 years) were more tender when cooked between 55 and 60 °C, a range that includes the temperature (55 °C) at which collagen dissolution begins.

C. *Estimation of the Shelf Life of Steaks under Refrigeration Conditions*

Table 5 shows that there is sufficient statistical evidence (P-value = 0.05) to state that the estimated shelf life of vacuum-cooked beef fillets stored under refrigerated conditions (3 ± 2 °C) is 67 days, and that this shelf life is determined by the mesophilic aerobic microorganism’s quality parameter.

Table 5

Estimated shelf life of beef steaks cooked sous-vide at 55 °C and conventionally, stored under refrigeration (3 ± 2 °C)

Parameter	Unit	Average Value at Cooking Temperature (°C):		
		55°	60°	65°
pH	-	$5,5^{a}$	$5,5^{a}$	$5,5^{a}$
Water-Holding Capacity	%	$98,7^{a}$	$98,5^{b}$	$97,9^{c}$
Cooking Weight Loss	~~%~~	$1,3^{a}$	$1,5^{b}$	$2,1^{c}$
Texture	-	$0,425^{a}$	$0,425^{a}$	-0.85^{b}

The shelf life of vacuum-cooked beef fillets processed at 55 °C for 60 min was significantly longer (P-value = 0.05) than that of conventionally cooked fillets. The sous-vide cooking treatment extended the shelf life of beef fillets stored under refrigerated conditions (3 ± 2 °C) from 7 to 67 days, considering aerobic mesophilic microorganisms as the critical parameter. This value is slightly higher than the 40 and 45 days proposed by [45] for vacuum-cooked cod fillets processed at 57.1 °C for 30 min, using the growth of aerobic mesophiles on TSA agar and saline TSA agar, respectively, as critical parameters, and markedly higher than the 23 and 29 days reported by [30] for pork fillets cooked at 60 °C for 30 min, in which total mesophilic counts and Listeria monocytogenes counts were considered the shelf-life-limiting factors.

In this regard, [46] points out that the microorganisms that limit the shelf life of vacuum-cooked products include spore-forming anaerobic bacteria such as *Clostridium botulinum* and *Clostridium perfringens*, as well as facultative anaerobic bacteria such as *Salmonella spp*., *Escherichia coli*, *Staphylococcus aureus*, *Yersinia enterocolitica*, and *Listeria monocytogenes*. Furthermore, the shelf life of this type of product depends primarily on the intensity of the thermal treatment, the time required to cool the product, and subsequent storage under appropriate refrigeration temperatures, and to a lesser extent on additional factors such as the initial microbial load, hygienic processing conditions, the type of packaging used, and the residual atmosphere within the package. Therefore, an adequate combination of time and temperature during cooking, together with rapid cooling and continuous storage under strict refrigeration, is essential to ensure not only an extended life shelf but also the microbiological safety of the product.

IV. Conclusions

It was determined that sous-vide cooking of beef fillets at 55, 60, and 65 °C for 60, 55, and 50 min, respectively, allows a reduction of up to 10 logarithmic cycles in the populations of Escherichia coli ATCC 25922 and Staphylococcus aureus ATCC 25923, the main microorganisms used as indicators of sanitary quality in processed foods, demonstrating that the applied treatments ensure microbiological safety.

The sous-vide cooking treatment at 55 °C for 60 min showed a significantly favorable effect (P-value = 0.05) on the physicochemical quality of beef fillets, as reflected by a higher water-holding capacity (WHC), a more tender texture, and a significant reduction in cooking losses. In addition, this treatment extended the product shelf life to up to 67 days, representing an increase of approximately 60 days compared with fillets cooked using conventional methods without oxygen reduction, thereby confirming the potential of the sous-vide process as an effective technological alternative for improving the quality and preservation of meat products.

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