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The effects of sous-vide cooking on the physicochemical, microbiological, and carbon footprint of buffalo meat at various temperatures and times

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Abstract The effects of temperature (55, 59, or 70°C) and time (24 or 48 h) on pH, surface color, browning index (BI), cooking loss (CL), color changes (Δ), muscle shrinkage, shear force (SF), toughness, textural profile analysis (TPA), microbial content, and carbon footprint in sous-vide-cooked buffalo meat were examined. These quality parameters were compared with traditional cooking (TC; 80°C for 30 min). The results showed that meat cooked by TC had higher SF, toughness, hardness, springiness, cohesiveness, gumminess, and chewiness than the other sous-vide treatments. A temperature and time combination showed a major increase in pH, a*, b*, ΔE, BI, and cohesiveness but a reduction of Δa*, toughness, and microbial content. The CL, transversal shrinkage (TS), SF, hardness, gumminess, and carbon footprint increased with increased temperature. Almost all instrumental texture values, particularly hardness, were decreased with prolonged time. Among sous-vide treatments, the lower TS, shear tests, and almost all texture profiles were found in samples cooked at 55 ℃ for 48 h. Compared to raw meat, there was no risk of inadequate pasteurization in cooked treatments. Thus, the results concluded that sous-vide cooking had a key advantage in retaining moisture, supplying tender meat (55 ℃-48 h and 59 ℃-24 h), and minimizing carbon footprint (55- and 59 ℃-24h).

Keywords: Sous-vide, Meat quality, Tenderness, Carbon footprint, Buffalo

Introduction

Sous-vide cooking, low temperatures–long times (LT-LT), is a method used to cook foods at precisely controlled temperatures and times under vacuum conditions (Schellekens, 1996). This cooking technique has been used by restaurants, chefs, and others (Myhrvold *et al.*, 2011). The major benefit of the sous-vide vacuum sealing bag is that it allows heat to be efficiently

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transferred from the water bath to the meat (Baldwin, 2012). The sous-vide method has the potential to shorten the time required for preparation and to increase convenience. Moreover, it is well known that this cooking has many benefits, including palatability, tenderization effects, moisture retention, nutrient retention, microbiological safety, and extended shelf-life (Church, 1998; Ohlsson, 2003). However, the conditions recommended for different kinds of meat are very different.

Tough cuts of beef (Bouton and Harris, 1981) and pork (Christensen *et al.*, 2011) were the most tender when cooked at lower temperatures, around 55–70 ℃, for a long time. Cooking the beef at 55 and 60 ℃ for 24 h markedly reduced shear force (SF) by around 26–72%, while the SF in tough beef cooked at 59°C for 4 h decreased by around 17–21% (Uttaro *et al.*, 2019), allowing for more tenderness by decreasing the myofibrillar traction strength. In addition, cooking at 50–55 ℃ can induce gelatin-dissolving collagen (Neklyudov, 2003), and at 60 ℃ for up to 6 h the meat may tenderize, as the collagenase enzyme remains active (Tornberg, 2005). During heating, myofibrillar protein and connective tissue (60–70% of muscle protein) were slight, and sarcoplasmic protein (30–34%) were extended (Baldwin, 2012). This denaturation process can cause muscle shrinkage, collagen solubilize, and water loss, all of which affect the tenderness of the meat.

Beyond the palatability and safety of sous-vide products, and carbon emission should be of concern as well. Meat carbon emissions are derived from the growing, farming, processing, transportation, storage, cooking, and disposal of the food (Environmental Working Group, 2011), and meat production is responsible for nearly a quarter of all greenhouse gas (GHG) emissions, contributing to climate change (Weber and Matthews, 2008). Hence, monitoring the carbon footprint in all food production areas will be a feature of the processing sectors that prioritize reducing carbon emissions. There are many ways to reduce GHGs, such as eating organic, eating less meat, cooking smartly, using water efficiently, reusing, and recycling.

Buffalos are one of the leading domestic animals in Thailand. Although their meat quality, particularly tenderness, is less acceptable than beef, they are still excellent sources of particular nutrients (Uriyapongson, 2013). To get the same level of tenderness as meat from younger animals, meat obtained from older animals must be cooked at a higher temperature and for a longer period of time (Naqvi *et al.*, 2021). It is currently unknown if sous vide cooking affects the quality of low-value buffalo meat. Therefore, this study aimed to evaluate the physicochemical, textural, microbiological characteristics and carbon footprint of sous-vide cooked culled buffalo meat at 55, 59, or 70℃ for 24 or 48 h compared to traditional cooking (80℃ for 30 min).

Materials and methods

Sample preparation

The *longissimus dorsi* (LD) muscles of a local, natural, grass-fed swamp buffalo were purchased from a local market in Nakhon Phanom Province, Thailand. The cuts had the following characteristics: up to 5 years of age, 350–400 kg live weight, 9 h post-mortem. The samples were packed, kept in an icebox, and transferred to the research lab for 3 h. The meat was kept cool at 4℃ in the refrigerator for 12 h prior to analysis. All excessive visible fats, ligaments, and connective tissues of the muscles were trimmed, and a total of seven parts of the muscles were then cut into 1-inch-thick steaks. Before the study, the samples' weight (77.2 ± 15.9 g), pH (5.39 ± 0.11), color, and microbiological characteristics were measured.

Thermal treatment

The samples were randomly divided into three sous-vide cooking temperatures of 55, 59, and 70 ℃, and two durations, namely, 24 and 48 h, as well as a group for traditional cooking (TC) at 80 ℃ for 30 min. All samples were packed in an LLDPE vacuum bag (14 × 12 cm), vacuum-sealed (DZ-500, Sammi Packing Machine Ltd., China), and cooked using an immersion cooker (800 W, SVJ-1000, Sous-vide Precision Cooker, China). The sous-vide cooking steps in this study proceeded follow: (1) preheat precision cooker, (2) weigh the beef (1^{st}), (3) bag the beef, (4) seal and vacuum the bag, (5) heat or pasteurize the beef, (6) remove the beef from the bag, (7) keep the beef at room temperature, (8) weigh the beef (2^{nd}), and (9) test. Finally, all packages were removed, kept at room temperature for a while, and immediately chilled at 4 ℃ overnight prior to analysis of the percentage of cook loss, color properties, shear force, and texture profile.

pH measurement

Meat samples were collected and evaluated using a portable pH meter supplied with an FC2323 pH/ temperature probe with an FC099 stainless steel blade tipped in triplicate (HI99163, Hanna Instruments, USA). Before measuring, the pH meter was calibrated with standard pH 4.01 (HI5004) and pH 7.01 (HI5007).

Instrumental color measurement

The instrumental surface color was measured using a Minolta Color Meter (CR-400, Konica Minolta Sensing Inc., Japan) equipped with a standard D65 illuminant, 2° observers, and 11-mm measuring field and calibrated with a standard whiteboard (L = 94.71, a = -0.25, b = 2.57) after 30 minutes of blooming at room temperature. The colors lightness, L*; redness, a*; and yellowness, b*; of each meat sample was taken from five random positions. Pre- and post-cooked differences in surface color were used to determine color changes (ΔL*, Δa*, Δb*), total color difference (ΔE) (AMSA, 2012), and browning index (BI) (Mohammadi *et al.*, 2008) with Equation 1 and 2 below:

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

$$\text{Browning Index (BI)} = [100 \times (x - 0.31)]/0.17 \quad (2)$$

where $x = (a^* + 1.75L^*)/(5.645L^* + a^* - 3.012b^*)$

Cooking loss measurement

Cooking loss (CL) was calculated from the difference of the weights before (W_1) and after (W_2) sous-vide cooking, following Equation 3:

$$\text{CL (\%)} = \frac{W_1 - W_2}{W_1} \times 100 \quad (3)$$

Muscle shrinkage measurement

Muscle shrinkage consists of two types: longitudinal shrinkage (LS) describes the difference in sample length before (L_1) and after (L_2) cooking, and transversal shrinkage (TS) identifies the difference in circumference of the sample before (C_1) and after (C_2) cooking (Becker *et al.*, 2016). The equations to calculate LS and TS are shown in Equation 4 and 5 below:

$$\text{LS (\%)} = \frac{L_1 - L_2}{L_1} \times 100 \quad (4)$$

$$\text{TS (\%)} = \frac{C_1 - C_2}{C_1} \times 100 \quad (5)$$

Shear force measurement

The shear force was carried out following the procedures of AMSA (2016). In short, all samples were transferred for overnight cooling in the refrigerator at 2–5 ℃ before coring. The testing was performed at room temperature, around 23 ℃. A 1.27-cm round core with diameter was removed in parallel diameter with the longitudinal muscle fibers using a hand-held coring device in stainless steel and determined using a Warner Bratzler Blade Set with a V slot blade (HDP/WBV) connected to the TA-XT *plus* Texture Analyzer (Stable Micro System Ltd., Surrey, UK). Peak force was obtained using a 50-kg load cell at a test speed of 240 mm/min. The maximum peak force and shear work were recorded as SF (kg) and toughness (kg.s).

Texture profile analysis

The texture profile analysis (TPA) as described by Bourne (1978) used the Texture Analyzer equipped with a P/50 cylindrical aluminum probe to evaluate the meat texture. Six samples from the steaks were cut into 1-cm^3 cubes, as described above, but using a knife. The setting conditions of TPA were as follows. Test speed: 1.0 mm/s; strain: 75%; time: 5.0 s; triggers type: auto; trigger force: 5 g; load cell: 50 kg. Seventy-five percent of the original portion height was compressed by a double compression cycle test to determine hardness, adhesiveness, cohesiveness, springiness, gumminess, and chewiness values (Exponent software version 6.1.16.0). Each sample's texture attribute value was an average of six core values.

Microbiological analysis

For total plate count (TPC) in aseptic conditions, a total of 11 g of each sample was taken to quantify the anaerobic count of treatment following the instruction guidelines of the microbial test kit. After the piece was mixed and diluted with the final solution until $\times 10^6$ on the Compact Dry™ TC (Aerobic, Nissui Pharmaceutical Co. Ltd., Japan), 1 mL was spread. The plate was incubated for 24 h at 35 ℃. The TPC was expressed as a unit forming the log colony ($\times 10^6$ CFU/g).

E. coli and coliform bacteria were measured using the microbial test kit Compact Dry™ EC (Nissui Pharmaceutical Co. Ltd., Japan). The exact amount of sample was collected, mixed, and diluted with the solution, as described in the instruction guidelines. One mL of each dilution was spread on the center of the Compact Dry™ EC, which was put in the sterile bag, zipped, and incubated, as described above.

Carbon footprint calculation

Indirect emissions of GHG from sous-vide cooking in this study include buffalo meat production, electricity used, vacuum plastic used, tap water used, and meat transportation. Thus, the carbon footprint (CF) could basically be calculated using Equation 7:

$$CF = [(M \times EF_m) + (E \times EF_e) + (P \times EF_p) + (W \times EF_w) + (T \times EF_t)] \quad (6)$$

where carbon footprint is the total GHG emissions by summarizing all individual GHG emissions, M is the amount of meat used for sous-vide cooking, EF_m is the emission factor for buffalo meat production ($kgCO_2e/kg$ meat produced), E is the amount of electricity used for sous-vide cooking, EF_e is the emission factor for electricity production ($kgCO_2e/kWh$), P is the amount of vacuum plastic used, EF_p is the emission factor for LLDPE plastic production ($kgCO_2e/kg$), W is the amount of tap water used for sous-vide cooking, EF_w is the emission factor for tap water production ($kgCO_2e/L$), T is the amount of meat transportation, and EF_t is the emission factor for transportation ($kgCO_2e/km$). In addition, the emission factors are obtained from a report of Thailand Greenhouse Gas Management Organization (TGO) (TGO, 2020), except buffalo meat was estimated based on the work of Gerber *et al.* (2013).

Statistical analysis

All data from meat pH, color, CL, shrinkage, shear values, texture profile analysis, microbiological content, and carbon footprint were analyzed by 3 × 2 (+1) augmented factorial experiments in a completely randomized design using PROC GLM (SAS, 2015), as the following model:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \alpha\beta_{ij} + \varepsilon_{ijk}$$

Where Y_{ijk} are the experimental response for the i^{th} level of factor A (temperature), the j^{th} level of factor B (time), and the k^{th} replication; μ is the overall mean effects; $\alpha_1, ..., \alpha_i$ are the main effects of factor A; $\beta_1, ..., \beta_j$ are the main effects of factor B; $\alpha\beta ij$ are the interaction effects between factors A and B; and ε_{ijk} is experimental error. Moreover, orthogonal contrast was followed to determine the response effects. Data were averaged and expressed as mean ± SD (n = 9). A Tukey HSD post-hoc test was followed by all ANOVA to determine significant differences at the 5% level.

Results

The results of sous-vide cooking at 55, 59, or 70 ℃ for 24 or 48 h and traditional cooking at 80 ℃ for 30 min on pH, surface color, and total color differences (ΔE) of buffalo beef are shown in Table 1. Cooking at 59 ℃ for 48 h was highest in pH values (P<0.05), followed by 70 ℃ for 24 h, 55 and 59 ℃ for 24 h, 70 ℃ for 48 h, and 55 ℃ for 48 h. Nonetheless, the pH of TC was lower than other treatments (P<0.05). Increasing the cooking temperature from 55 to 70 ℃ quadratically increased (P<0.05) the pH values.

As seen from Tables 1, after sous-vide cooking, the color values show that there were no significant variations in L* (P>0.05) for all treatments. However, the lightness change (ΔL*) of sous-vide treatments was lower than that of TC (P<0.05). In addition, the a* and b* values were affected by different temperature and time interactions (P<0.05): the sous-vide cooking at 70 ℃ for 48 h had the highest a* and b* colors and the lowest Δ*a** and Δb* compared to other treatments.

Table 2 shows the effects of temperature and time on the browning index (BI), CL, TS, LS, SF, and toughness of sous-vide-cooked buffalo beef. A significant interaction was found in BI in this study, where values increased with temperatures and time (P<0.05). The TC sample had a higher CL than sous-vide cooking. Only cooking temperature affected CL; the values were 24.24, 34.62, and 44.73 when raising the heat from 55 to 70 ℃ (P<0.05). However, none of the LS parameters considered in this analysis was influenced by either cooking temperature or time. Our findings regarding LS were comparable between sous-vide cooking and TC. An ANOVA two-way study showed a significant temperature effect on the TS, where the samples cooked at 70 ℃ for 48 h had a greater TS, and samples cooked at 55 ℃ for 24, and 48 h showed the lowest TS values (P<0.05). Only temperature, not time, affected SF, while combined effects of both temperature and time altered toughness (P<0.05). TC resulted in a higher SF and toughness values, by 37% and 30%, respectively, compared to any other treatment (P<0.05). Raising the cooking temperature from 55 to 70 ℃ resulted in a quadratic increase in SF values (P<0.05; 4.02, 3.90, and 5.31 kg).

The effects of temperature and time on the TPA of sous-vide-cooked buffalo beef is shown in Table 3. Interaction effects of temperature and time on cohesiveness were found (P<0.05). Samples in TC treatments were tougher than in other treatments due to higher values in hardness, springiness, cohesiveness, gumminess, and chewiness (P<0.05). The values of hardness (lin), adhesiveness (lin), cohesiveness (qua), and gumminess (lin) increased with increasing temperatures (P<0.05). Moreover, cooking for 48 h produced meat tendered by lowering hardness, springiness, gumminess, and chewiness values (P<0.05).

Table 1. Effects of temperature and time on pH, surface color, color changes (Δ), and total color differences (ΔE) of sous-vide cooked buffalo meat

Treatment	pH	L*	a*	b*	ΔL*	Δa*	Δb*	ΔE
Raw	5.35 ± 0.06	38.79 ± 2.25	20.37 ± 1.60	9.05 ± 1.31	-	-	-	-
TC (80 ℃ – 30 min)	5.62 ± 0.24^{b}	45.18 ± 0.77	11.64 ± 1.35^{abc}	14.63 ± 1.39^{d}	-6.27 ± 2.10	8.68 ± 1.72^{ab}	-4.99 ± 2.28^{a}	12.19 ± 1.54^{b}
55 ℃ - 24 h	5.73 ± 0.08^{ab}	44.11 ± 0.86	11.21 ± 0.65^{bc}	14.44 ± 0.65^{d}	-4.21 ± 4.79	8.62 ± 2.79^{ab}	-5.63 ± 1.57^{ab}	12.26 ± 1.87^{b}
55 ℃ - 48 h	5.61 ± 0.12^{b}	42.93 ± 1.19	9.69 ± 0.99^{c}	15.61 ± 0.96^{cd}	-3.80 ± 2.47	10.61 ± 1.47^{a}	-6.66 ± 1.49^{ab}	13.37 ± 1.45^{ab}
59 ℃ - 24 h	5.75 ± 0.07^{ab}	42.90 ± 1.68	12.24 ± 2.02^{ab}	16.56 ± 2.59^{bc}	-4.19 ± 2.19	7.97 ± 1.04^{ab}	-7.85 ± 1.12^{bc}	12.13 ± 1.06^{b}
59 ℃ - 48 h	5.88 ± 0.16^{a}	42.65 ± 1.61	10.38 ± 0.84^{bc}	16.05 ± 1.18^{cd}	-3.24 ± 3.46	9.42 ± 1.79^{ab}	-7.47 ± 1.77^{abc}	13.03 ± 1.39^{ab}
70 ℃ - 24 h	5.81 ± 0.05^{ab}	43.72 ± 2.80	11.49 ± 0.84^{bc}	18.25 ± 1.74^{ab}	-4.47 ± 2.41	8.46 ± 4.76^{ab}	-9.53 ± 1.72^{cd}	14.41 ± 1.71^{a}
70 ℃ - 48 h	5.70 ± 0.13^{ab}	42.87 ± 0.46	13.83 ± 0.86^{a}	19.23 ± 0.77^{a}	-8.87 ± 1.82	6.42 ± 1.58^{b}	-10.44 ± 1.90^{d}	12.99 ± 1.05^{ab}
Orthogonal contrast								
TC vs. others	0.016	0.108	0.758	<0.001	0.026	0.903	<0.001	0.117
Temperature (A)	0.013	0.797	<0.001	<0.001	0.952	0.035	<0.001	0.059
A (lin)	0.078	0.842	<0.001	<0.001	0.945	0.010	<0.001	0.073
A (qua)	0.015	0.521	0.587	0.129	0.762	0.841	0.418	0.112
Time (B)	0.404	0.411	0.413	0.120	0.311	0.472	0.274	0.621
Interaction	0.012	0.916	<0.001	0.058	0.936	0.034	0.409	0.024
A (lin) × B	0.847	0.888	<0.001	0.818	0.734	0.017	0.912	0.012
A (qua) × B	0.003	0.695	0.012	0.018	0.901	0.284	0.184	0.220

$^{a–d}$ Means with different superscripts within the same column are significantly different (P<0.05).
Values display mean ± standard deviations; TC: traditional cooking.

Table 2. Effects of temperature and time on browning index, cooking loss, muscle shrinkages, shear force, and toughness of sous-vide cooked buffalo meat

Treatment	BI	%CL	%TS	%LS	Shear force (kg)	Toughness (kg.s)
TC (80 ℃ – 30 min)	57.54 ± 2.42[d]	36.58 ± 2.42[a]	19.44 ± 3.44[ab]	28.14 ± 5.38	6.98 ± 1.53[a]	14.49 ± 2.96[a]
55 ℃ - 24 h	57.72 ± 3.37[d]	25.11 ± 3.36[c]	8.48 ± 1.20[c]	35.01 ± 18.79	4.22 ± 0.61[bc]	12.46 ± 3.04[ab]
55 ℃ - 48 h	61.09 ± 4.14[cd]	23.38 ± 4.13[c]	7.83 ± 3.56[c]	32.70 ± 4.58	3.82 ± 0.67[c]	8.72 ± 0.61[b]
59 ℃ - 24 h	68.94 ± 3.43[bc]	33.30 ± 3.43[b]	16.37 ± 3.76[abc]	29.38 ± 5.91	3.79 ± 0.71[c]	8.68 ± 1.93[b]
59 ℃ - 48 h	65.08 ± 1.53[bcd]	35.93 ± 1.53[b]	12.28 ± 5.76[bc]	27.49 ± 16.86	4.02 ± 1.00[bc]	8.52 ± 2.33[b]
70 ℃ - 24 h	72.64 ± 0.92[b]	44.16 ± 0.92[a]	21.78 ± 6.53[ab]	33.45 ± 6.52	5.48 ± 1.43[b]	11.26 ± 4.06[ab]
70 ℃ - 48 h	82.63 ± 5.19[a]	45.30 ± 5.19[a]	25.88 ± 3.20[a]	38.99 ± 9.60	5.13 ± 0.75[bc]	11.29 ± 3.08[ab]
Orthogonal contrast						
TC vs. others	<0.001	0.023	0.051	0.701	<0.001	0.023
Temperature (A)	<0.001	<0.001	<0.001	0.370	<0.001	0.014
A (lin)	<0.001	<0.001	<0.001	0.673	<0.001	0.462
A (qua)	0.425	0.893	0.374	0.182	0.012	0.005
Time (B)	0.079	0.451	0.904	0.922	0.537	0.091
Interaction	0.009	0.142	0.177	0.730	0.589	0.078
A (lin) × B	0.133	0.198	0.275	0.485	0.940	0.045
A (qua) × B	0.007	0.130	0.128	0.718	0.307	0.307

[a–c] Means with different superscripts within the same column are significantly different ($P<0.05$).

Values display mean ± standard deviations; BI: browning index; CL: cooking loss; TS: transversal shrinkage; LS: longitudinal shrinkage; TC: traditional cooking.

Table 3. Effects of temperature and time on texture profile analysis (TPA) of sous-vide cooked buffalo meat

Treatments	**Hardness (kg)**	**Adhesiveness (g.s)**	**Springiness**	**Cohesiveness**	**Gumminess**	**Chewiness**
TC (80 ℃ – 30 min)	1.15 ± 0.77[a]	-5.53 ± 1.82	0.12 ± 0.03[a]	0.71 ± 0.08[a]	0.79 ± 0.50[a]	0.11 ± 0.09[a]
55 ℃ - 24 h	0.65 ± 0.45[ab]	-9.23 ± 10.9	0.11 ± 0.02[ab]	0.61 ± 0.02[bc]	0.40 ± 0.28[ab]	0.05 ± 0.04[ab]
55 ℃ - 48 h	0.34 ± 0.26[b]	-7.06 ± 4.77	0.08 ± 0.02[b]	0.64 ± 0.04[b]	0.22 ± 0.18[b]	0.02 ± 0.02[b]
59 ℃ - 24 h	0.93 ± 0.52[ab]	-5.10 ± 1.41	0.10 ± 0.01[ab]	0.66 ± 0.03[ab]	0.61 ± 0.34[ab]	0.07 ± 0.04[ab]
59 ℃ - 48 h	0.72 ± 0.34[ab]	-5.53 ± 1.47	0.10 ± 0.01[ab]	0.67 ± 0.02[ab]	0.48 ± 0.23[ab]	0.05 ± 0.03[ab]
70 ℃ - 24 h	1.07 ± 0.28[a]	-4.26 ± 1.21	0.10 ± 0.01[ab]	0.65 ± 0.03[ab]	0.69 ± 0.21[a]	0.07 ± 0.03[ab]
70 ℃ - 48 h	0.69 ± 0.17[ab]	-4.47 ± 1.65	0.09 ± 0.01[ab]	0.57 ± 0.05[c]	0.39 ± 0.10[ab]	0.04 ± 0.01[b]
Orthogonal contrast						
TC vs. others	0.010	0.810	0.002	<0.001	0.002	0.001
Temperature (A)	0.023	0.051	0.701	<0.001	0.023	0.203
A (lin)	0.011	0.019	0.534	0.201	0.018	0.142
A (qua)	0.281	0.491	0.572	<0.001	0.154	0.307
Time (B)	0.016	0.691	0.032	0.115	0.011	0.031
Interaction	0.837	0.658	0.612	0.001	0.649	0.853
A (lin) × B	0.812	0.452	0.534	<0.001	0.506	0.657
A (qua) × B	0.586	0.604	0.442	0.235	0.508	0.732

[a–c] Means with different superscripts within the same column are significantly different (P<0.05).
Values display mean ± standard deviations. NE: not estimated; TC: traditional cooking.

As shown in Table 4, to validate whether sous-vide cooked buffalo meat with different temperatures and times would guarantee microbiological safety for the consumer, the microbial content was evaluated before and after sous-vide cooking. The microbial content of raw meat was, for the total plate count (TPC), around 12.25 ± 5.40 × 10^6 CFU/g; for *E. coli*, 177 ± 143 CFU/g; and for coliform, 146.00 ± 132.00 CFU/g. The TPC and *E. coli* contents were above regular reference of the Department of Medical Sciences, Ministry of Public Health, Thailand.

Table 4. Effects of temperature and time on microbial content of sous-vide cooked buffalo meat

Treatment	**TPC (×10^6 CFU/g)**	***E. coli* (CFU/g)**	**Coliform (CFU/g)**
Raw	12.25 ± 5.40	177.00 ± 143.00	146.00 ± 132.00
All sous-vide treatments	ND	ND	ND

Values display mean ± standard deviations; ND: not detected

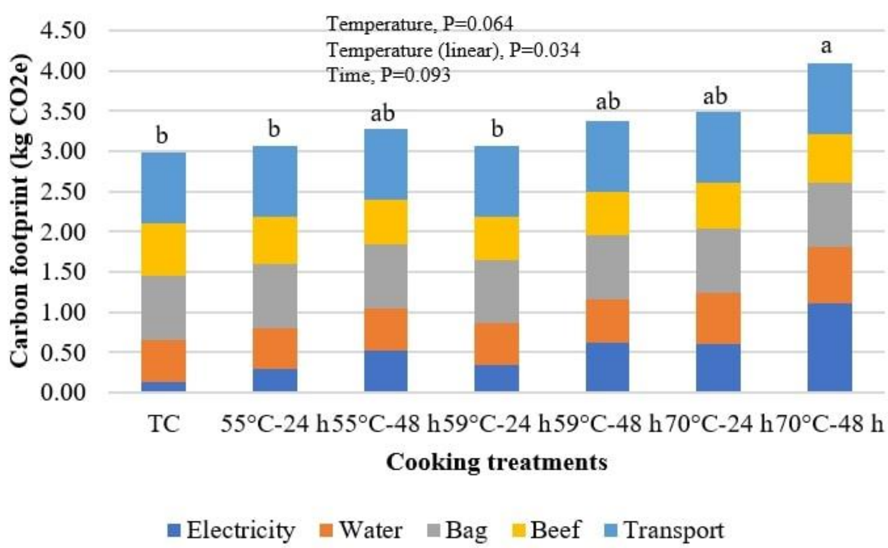

Figure 1. Carbon footprint of sous-vide cooked buffalo meat

The results in Figure 1 showed that the carbon footprints of different cooking methods were 2.98 ± 0.29 kgCO_2e for traditional cooking at 80 ℃ for 30 min (TC), 3.94 ± 0.33, and 4.15 ± 0.40 kgCO_2e for sous-vide cooking at 55 ℃ for 24 and 48 h, 3.95 ± 0.32 and 4.26 ± 0.34 kgCO_2e for sous-vide cooking at 59 ℃ for 24 and 48 h, and 4.36 ± 0.37 and 4.97 ± 0.27 kgCO_2e for sous-vide cooking at 70 ℃ for 24 and 48 h. The carbon footprint of TC was the lowest due to the shorter time involved. Cooking temperature (P=0.064) and time (P=0.093) showed a trend of increasing carbon footprint. The carbon footprint of sous-vide cooking at 70 °C for 48 h had the highest values compared to the other approaches. However, no significance of temperature and time combination were observed (P>0.05). Most of the carbon footprint of sous-vide-cooked buffalo beef steaks in this study was a result of carbon emissions from transport (26.43), followed by material use (23.94), beef steaks (17.25), water use (16.89), and electric consumption (15.48%).

Discussion

The increase in pH when heating is primarily associated with the denaturation of protein and the reduction of acidic groups (Hamm and Deatherage, 1960). Sous-vide cooking generally presents a greater L* and lower a* when the temperature increases (Sánchez del Pulgar *et al.*, 2012). The results of color changes during sous-vide cooking and prolonged heating are questionable (Becker *et al.*, 2015).

While there was no significant variation in lightness values between treatments, heating and time increased the browning index. This means that the meat gets browner as it cooks. In relation to fresh meat, the lightness values increased by 16.47 and 11.37% for TC and sous-vide treatments, respectively. This finding was the opposite of results reported by Christensen *et al.* (2011) and Ismail *et al.* (2019b), in which increasing the cooking temperature resulted in increased L* values. Regarding the percentage changes of a* and b* in TC and sous-vide treatments compared to raw meat, the reduction in values for a* were 42.81% and 43.64% and 61.77% and 81.44% increase for b* values, respectively. Metmyoglobin heat denaturation can explain the changes in a* and b*, leading to a rise in the formation of brown colors (Roldán *et al.*, 2013). Cooking at a higher temperature resulted in significantly higher values of ΔE (70 °C for 24 h) and BI (70 °C for 48 h) than with other cooking treatments (P<0.05). The lowest values of ΔE and BI were observed in samples cooking at 55 or 59 °C for 24 h and were comparable to TC (P<0.05). The higher heme-iron may explain the higher ΔE and BI in myoglobin oxidation during cooking (Hunt *et al.*, 1999).

It is commonly understood that heating meat reduces its water content due to the contraction of muscle fiber bundles, which in effect facilitates the release of water from the meat cut (Sims and Bailey, 1992). This result is in line with earlier research, which has found that sous-vide heated-samples reduce myofibrillary bulk, promotes hypercontraction and wrinkling of muscle fibers, and damages some mitochondria (Supaphon *et al.*, 2021). According to Becker *et al.* (2015), samples cooked at lower temperatures result in lower CL than those cooked at higher temperatures. The cooking time (24 vs. 48 h) at each cooking temperature showed a similar CL. This finding is the opposite of other results in pork (Becker *et al.*, 2015) and beef (Phoemchalard *et al.*, 2019), which found that extending cooking time increases the CL. However, sous-vide cooking produced lower CL than the TC samples (34.53 vs. 36.59).

Sous-vide cooking changes the structural structure of muscle fibers, which shows a contraction of the meat, which loses water. The changes are associated with the temperature and the length of time the meat is cooked (Supaphon *et al.*, 2021). In the conventionally-cooked meat, longitudinal shrinkage percentage was especially obvious in the reduction of the entire muscle after heating (Becker *et al.*, 2016). Moreover, they explained that the muscle fiber was higher in TS when the heat was under 60 ℃, while the LS was affected by heat between 60–90 ℃. The higher levels of TS in the present study are strongly associated with CL. This result is consistent with Becker's findings in pork samples, in which TS increased with increasing temperatures (Becker *et al.*, 2016).

The application of the sous-vide methods to tenderize meat, weakening myofibrillar, and connective tissues, is optional because tenderness plays a key role in customer choice. Increased cooking temperatures from 55 to 70 ℃ led to higher SF values. This was similar to findings reported by Ismail *et al.* (2019a). A sample cooked at 45 ℃ showed a significantly lower value of SF than a sample cooked at 65 ℃ or by TC. However, extended heat from 48 to 63 ℃ for 0–17 h in pigs and sows resulted in decreased SF (Christensen *et al.*, 2011). Although the samples cooking at 55 ℃ for both 24 and 48h were better in their water-holding capacity and TS (Table 2), producing tender meat, whereas cooking at 59 ℃ for 24h formed less SF. The denaturation of the myofibrillar proteins, particularly the actomyosin complex, can explain the difference in SF (Palka and Daun, 1999). Meat toughness refers to the ability of meat to withstand any force exerted. Our findings showed that toughness values were lower in samples cooking at 55 ℃ for 48 h and 59 ℃ for 24 and 48 h ($P<0.05$). Collagen denaturation and myofibrillar structure changes may also cause a variation in toughness values (Vaudagna *et al.*, 2002).

TC-treated samples are harder than samples from other treatments, which is in accordance with the result of sous-vide-cooking beef at the same temperature, but the hardness of buffalo beef in this study was lower than that of crossbred beef (Phoemchalard *et al.*, 2019). Following our previous result, beef that was sous-vide cooked for 48 h produced softer meat. In addition, the surface of the samples was higher in gel, confirming that the increase in meat softness occurred when the collagen became gelatinized (Tornberg, 2005).

The higher initial germ rates were comparable to other studies (Becker *et al.*, 2015, 2016); sous-vide removed significant amounts of microorganisms while cooking at 53 or 58 ℃ for up to 5 h. In addition, our analysis showed that all sous-vide treatments and TC eliminated TPC, E. *coli*, and coliform. Thus, in terms of microbiological safety, these meat samples should be safe after cooking treatments.

The average emission intensity of buffalo beef steaks in this study was higher than our previous study in beef (Phoemchalard *et al.*, 2019); sous-vide cooking at 55, 59, and 70 ℃ generated a carbon footprint of 1.50, 1.54, and 2.24 $kgCO_2e$, and a prolonged time of 24 and 48 h was associated with a carbon footprint of 2.07 and 3.05 $kgCO_2e$, respectively. The marked difference in values (3.4-fold higher) was due to the carbon emission of producing and transporting buffalo meat. In fact, poorer productivity, lower feed quality, lower dressing percentages, and longer rising period may explain the high carbon footprint of buffalo meat in the Southeast Asia region (Gerber *et al.*, 2013). Additionally, one of the critical carbon emissions added is the distance of meat transport and the volume of meat per load. When compared to the typical meat preparation method, the carbon dioxide emissions from braised (3.72), deep fried (4.67), grilled (17.46), and smoked (41.19 kg) are higher. For instance, the gas emissions from a propane-grilled and charcoaled-grilled turkey are equivalent to 7.21 to 17.76 kgCO2e. However, sous vide cooked turkey releases only 1.49 kgCO2e (Chester, 2018).

In conclusion, cooking buffalo steaks sous-vide at 55, 59, or 70 ℃ for 24 or 48 h can alter quality and safety compared to TC. The prolonged temperature and time combination increased pH, a*, b*, ΔE, BI, and cohesiveness but reduced Δa*, toughness, and microbial content. When increasing temperature, CL, TS, SF, hardness, gumminess, and carbon footprint (trend) increased, but Δb* and adhesiveness values decreased. Longer cooking time also produced a decrease in almost all the texture values, particularly hardness. Thus, we recommended cooking at 55 ℃ for 48-h and 59 ℃ for 24-h, given the better quality of buffalo steaks due to lower CL, TS, shear test, and texture, despite lower redness. In addition, cooking at a low temperature is better to minimize the carbon footprint (55 or 59 ℃). However, more research is required to determine the effects of sous vide cooking on buffalo meat quality, eating quality, and carbon emissions at varying temperatures and times.

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