EFFECTS ON BEEF MICROFLORA OF A THREE-STEP SOUS-VIDE METHOD

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Abstract – The objective of this study was to determine the effect of a three-step sous-vide cooking method on the natural microflora present on beef at the time of cooking. Steaks were cooked in waterbath under three conditions: aerobically at 70°C to an internal temperature of 59°C (C1), single-step sous-vide at 59°C with a holding time of 4 h (C2), and three-step sous-vide in which steaks were cooked sequentially at 39°C for 1 h (C3), at 49°C for 1 h (C4) and at 59°C with a holding time of 4 h (C5). Seven groups, including total aerobes, lactic acid bacteria (LAB), Pseudomonads, *Brochothrix thermosphacta*, *Enterobacteriaceae*, coliforms and *Escherichia coli* were determined for raw and cooked steaks. The numbers of aerobes, LAB, Pseudomonads, *B. thermosphacta*, *Enterobacteriaceae*, and coliforms on C3 and C4 steaks were not significantly different ($p>0.05$) from raw steaks. All bacterial groups except aerobes and LAB on C2 and C5 steaks were not recoverable.

Key Words – sous-vide, single-step, three-step, inactivation, meat microflora

I. INTRODUCTION

Sous-vide refers to slow-cooking of vacuum-packed food in water held at a predefined temperature. Sous-vide cooking of beef is often carried out by a one-step cooking procedure in which meat is cooked in the temperature and time ranges of 65 to 90°C and 2-48 h, respectively [1]. The tenderness of beef thus cooked can be significantly increased, by gelation of collagen and reduction of inter-fiber adhesion as a function of heat and time [2]. Inherent proteolytic enzymes in meat which break down the structure of muscle fiber can remain largely active at temperatures up to 50°C. At higher temperatures, these enzymes start to degrade and are mostly inactivated at ≥ 65°C [3, 4]. It would then be possible to include in sous-vide cooking steps of incubation at temperatures below 50°C to increase the tenderness of meat. However, the microbiological effect of cooking meat at temperatures permissible to bacterial proliferation was uncertain. In this study, the effect on the natural microflora of steaks of a three-step sous-vide was determined and compared against a typical single-step sous-vide and a research-level waterbath cooking method.

II. MATERIALS AND METHODS

Chilled vacuum-packed beef primal cuts were obtained from a local slaughter plant with the storage time varying from 2 to 5 weeks. The cuts were trimmed and made into 36 steaks, each measuring approximately 3 cm thick, 10 cm long and 8 cm wide. Steaks were tossed in large vacuum bags for 2 min to equilibrate the initial microbial load and were each placed into a vacuum pouch. Six packs were placed in a cooler operated at 2°C. Six packs were cooked in a 70°C waterbath to an internal temperature of 59°C (C1). The remaining 24 packs were vacuum-packed, 6 of which were cooked in a 59°C waterbath and held for 4 h (C2). Upon completion of cooking, the packs were cooled in ice water. The remaining 18 vacuum packs were cooked with a three-step method in which packs were cooked in a 39°C waterbath for 1 h (C3), the waterbath temperature was increased to 49°C and held for 1 h (C4) and finally increased to 59 °C and held for 4 h (C5). Six packs were withdrawn after completion of each of C3, C4, and C5 and cooled in ice water as before. The cooled packs were stored at 2°C. During cooking and cooling, the internal temperatures of steaks were monitored. The next day, the packs of steaks (raw and cooked) was each aseptically opened and rinsed with 10 ml of 0.1% peptone water. Appropriate dilutions of each rinse fluid were used to determine total aerobes, Pseudomonads, and *B. thermosphacta* [5], and LAB, *Enterobacteriaceae*, coliforms and *E. coli* [6]. Three independent sets at quarterly intervals were conducted. In total, 108 steaks were used in this study, with each condition including 18 steaks. All bacterial counts were converted to log cfu/100 cm^2. Mean log values were separated by a Dunn test SAS (SAS institute, Cary, NC), with a significance level of $p<0.05$.

III. RESULTS AND DISCUSSION

The mean initial bacterial counts on steaks from different sets varied largely, particularly for total aerobes and LAB, likely resulting from the difference in storage time before the meat was used for cooking [7]. *E. coli* was only sporadically found at very low numbers. The numbers of bacteria except *E. coli* recovered from C3 and C4 steaks were not significantly different from raw steaks ($p>0.05$), though the numbers from C4 steaks were somewhat lower (Table 1). The numbers of total aerobes and LAB were similar on C1, C2 and C5 steaks and were significantly lower ($p<0.05$) than the respective numbers on raw steaks. None of the C2 and C5 steaks yielded bacteria of any of the other 5 groups.

Table 1 Numbers of various groups of bacteria recovered from raw and cooked steaks*

Microorganism	Bacteria (log cfu/100 cm^2)					
	Raw steaks	After cooking				
		C1	C2	C3	C4	C5
Aerobes	7.19±2.04A	3.71±1.21BC	2.05±1.73B	7.21±2.17A	4.49±3.41AC	2.41±1.69B
Lactic acid bacteria	5.53±2.38A	2.24±1.84B	1.18±1.78B	5.44±2.57A	3.65±3.57AB	1.57±1.77B
Pseudomonads	5.52±0.98A	1.34±1.92BC	-	5.32±1.25A	3.01±2.72AC	-
B. thermosphacta	2.78±2.44A	- §	-	2.58±2.35AC	0.05±1.26BC	-
Enterobacteriaceae	3.68±0.85A	-	-	3.07±1.95A	1.86±2.04A	-
Coliforms	2.67±1.89A	-	-	2.45±2.26A	1.21±2.08AB	-
E. coli	-0.27±0.68A	-	-	-0.39±0.45A	-	-

*The cooking treatments: cooked aerobically at 70°C to 59°C (C1), vacuum-packed steaks cooked at 59°C and held for 4 h (C2), and cooked sequentially for 1 h at 39°C (C3), at 49°C for 1 h (C4), finally at 59°C for 4 h (C5).
§-, no bacteria were recovered.

IV. CONCLUSION

The findings show that bacterial proliferation on beef was unlikely to occur during the first two steps where temperatures were ≤ 50°C of the three-step sous-vide method and the overall reduction by the three-step sous-vide method of the natural microflora on beef was comparable to that by typical single-step sous-vide.

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