Influence of temperature on conservability of chilled vacuum-packed beef from different origins

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Abstract— **The objective of this experiment was to study the conservability of chilled vacuum-packed meat depending on storage temperature (–1 °C *vs.* +4 °C) during the last third of their shelf life. Physicochemical parameters (pH and colour) and microbiological growth (total aerobic bacteria, lactic acid bacteria, *Enterobacteriaceae, Pseudomonas* spp. and *Brochothrix thermosphacta*) of *Longissimus dorsi* samples from different origins (United Kingdom and Ireland, Australia and Brazil) were measured at: i) 2/3 of their shelf life and ii) the end of their shelf life. Sample bacteria population growing on MRS was identified by API 50 CHL strips. Unlike Irish and British samples, pH of some Australian and Brazilian samples decreased during conservation. The colour of the samples remained stable and it did not seem to be influenced by temperature. All samples conserved at –1 °C presented a satisfactory microbiological quality at the end of their shelf life (British and Irish meat = 35~45 days; Australian meat = 140 days and Brazilian meat = 120 days). On Australian and Brazilian samples, temperature did not influence total aerobic bacteria growth, but conservation at +4 °C favoured lactic acid bacteria and *Enterobacteriaceae* growth. API 50 CHL strip identifications revealed the presence of bacteria like *Lactobacillus brevis, Carnobacterium maltaromaticum* and *Lactobacillus fermentum*, which occur naturally in fresh meat and are known for their bioprotective effect against other microorganisms. Further analyses are being carried out using molecular methods in order to study the initial bacteria population diversity and its evolution during storage.**

Keywords— **beef, vacuum-packed, conservability.**

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I. INTRODUCTION

The shelf life of fresh meat is limited by spoilage phenomena, which can degrade the organoleptic characters of the product, or by growth of pathogen microorganisms, which represents a health risk for the consumers. In this way, the main challenges of chilled meat preservation are to maintain the fresh appearance of the product and retard bacterial development.

The shelf life of raw chilled meat can be extended to many weeks by adequate monitoring of hygiene conditions and by selection and use of appropriated packaging [1, 2]. The lack of oxygen in vacuum packages is known for reducing oxidative reactions and aerobic bacteria growth [3]. Moreover, since shelf life is inversely proportional to the storage temperature, strict control of temperature is essential to extend shelf life. Meat storage just above freezing point has been used with success and allows the maximum colour stability [4].

The combination of low oxygen concentrations and low temperatures results in a microbial population selection, and let lactic acid bacteria be part of the dominant bacteria flora. This selection is particularly interesting since some lactic acid bacteria strains would have great technological and bioprotective importance on raw beef [5].

The objective of this experiment was to study the conservability of chilled vacuum-packed meat depending on storage temperature (–1 °C *vs*. +4 °C) during the last third of their shelf life.

II. MATERIALS AND METHODS

Samples: Vacuum-packed striploins from three different origins were supplied by a Belgian food wholesaler. Seven batches were used in this study (three from United Kingdom and Ireland, three from Australia and one from Brazil). For each batch, three

striploins were purchased. At arrival, striploins were stored at –1 °C. On the day corresponding to ⅔ of the shelf life stated by the supplier (35~45 days for British and Irish, 140 days for Australian and 120 days for Brazilian meat), they were cut into 2~3 cm thick steaks, which were vacuum-packed and stored at –1 °C or +4 °C until the end of the shelf life.

Analyses were performed on the day corresponding to ⅔ of the shelf life and at the end of the shelf life proposed by the supplier (Fig. 1).

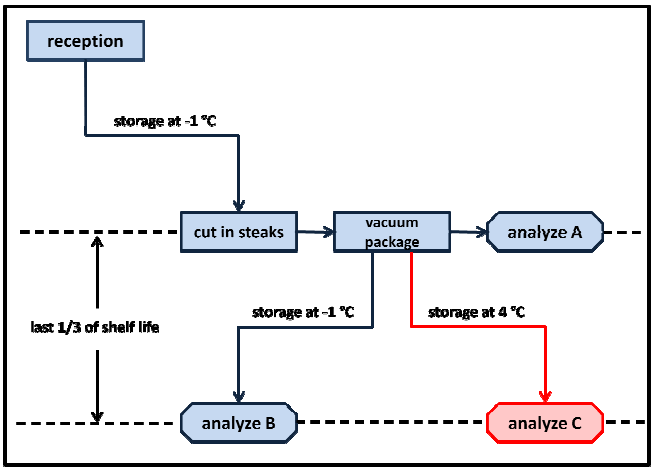

Fig. 1 Experimental design

pH measurement: pH was measured using a pH meter Knick type 765 and a combined pH electrode (Ingold ref. 104063123). Measurement was performed in five different zones of each sample and values were averaged.

Colour measurement: instrumental colour of samples was evaluated 1.5 h after removal from vacuum package bags using a Hunterlab Labscan II spectrophotometer (25 mm diameter aperture, D^{65} illuminant, 10° observation angle). Values for CIE L*, a* and b* were measured on five different zones of each sample and averaged.

Microbiological analysis: A sample of approximately 25 cm^2 × 1 cm (~25 g) was removed from each steak, diluted 10 times in peptone water, and stomached for 120 s. The enumeration of aerobic total flora (at +22 °C), lactic acid bacteria (at +22 °C) and *Enterobacteriaceae* (at +30 °C) was performed using BioMérieux's TEMPO® automated enumeration system. *Pseudomonas* spp. and *Brochothrix thermosphacta* were enumerated according to ISO 13720 and ISO 13722, respectively. Microbiological quality of samples presenting less than 5×10^6 CFU/g lactic acid bacteria, 5×10^3 CFU/g *Enterobacteriaceae*, 10^5 CFU/g *Pseudomonas* and 5×10^5 CFU/g *Brochothrix thermosphacta* was considered satisfactory [6]. Identification of bacteria growing on MRS was performed using API 50 CHL strips (BioMérieux, Belgium).

III. RESULTS AND DISCUSSION

Unlike other samples, one Australian and the Brazilian batch presented an important pH decrease when conserved at +4 °C during the last third of their shelf life (0.27 and 0.19 pH unit respectively). Lactic acid bacteria growth in meat was associated to pH decrease [7], and could be an explanation for this phenomenon in the batches mentioned before.

The colour of samples remained stable during conservation and did not seem to be influenced by temperature. This result is not surprising since the lack of oxygen in vacuum packaging limits oxidative reaction and thus preserves meat colour.

All samples conserved at –1 °C presented a satisfactory microbiological quality at the end of their shelf life, which confirms that vacuum package associated with sub-zero temperatures makes meat storage for several weeks possible [4]. On the other hand, conservation at +4 °C favoured lactic acid bacteria and *Enterobacteriaceae* growth, the latter being the limiting factor for satisfactory microbiological quality in various batches.

In order to evaluate the differences in lactic acid bacteria flora between samples, aliquots of samples of three batches were suspended in peptone water, plated on MRS agar and incubated at +22 °C for 72 h. The dominant colonies on each plate were then identified using API 50 CHL strips (Table 1).

Identification of lactic acid bacteria revealed that predominant species varied according to the origin of the sample. Lactic acid bacteria isolates frequently producing bacteriocins, such as *Lactobacillus brevis*, [7] and *Carnobacterium maltaromaticum* [8], were identified in Irish and Brazilian samples respectively. Surprisingly, *Brochothrix thermosphacta* was the dominant isolate in Australian samples in the end of

Table 1 Proportion of lactic acid bacteria isolated from three beef batches from different origins (Ireland, Australia and Brazil), identified by API 50 CHL strips

Lactic acid bacteria	⅔ shelf life	end of shelf life (–1 °C)	end of shelf life (+4 °C)
- Ireland			
Lactobacillus lactis	-	50%	-
Lactobacillus brevis	-	25%	50%
Lactobacillus delbrueckii	-	25%	-
Lactococus lactis	-	-	25%
Leuconostoc lactis	-	-	25%
- Australia			
Brochothrix thermosphacta	67%	100%	100%
Lactobacillus curvatus	33%	-	-
- Brazil			
Carnobacterium maltaromaticum	67%	-	-
Lactobacillus fermentum	33%	67%	67%
Lactobacillus acidophilus	-	33%	16%
Lactobacillus paracasei	-	-	16%

shelf life. Thus, this result is conflicting with the long shelf life of Australian meat.

In order to increase the reliability of bacterial population identification, metagenomic analyses were carried out on three different batches (an Irish one, an Australian one and the Brazilian one). This relatively new field of genetics allows studying the communities of microbial organisms in their natural environment, bypassing the need for isolation and laboratory cultivation [9].

Preliminary results reveal that at ⅔ of shelf life *Aquabacterium* was the dominant gender in Irish and Brazilian batches; *Pseudomonas* was the dominant gender in the Australian batch at ⅔ of shelf life. These genders were probably originated from initial environmental contamination. At the end of shelf life (after storage at +4 °C), *Aquabacterium* and *Escherichia* were the dominant genders in the Irish batch. Conversely, the order *Lactobacilalles* figured to be the dominant one in Brazilian and Australian batches at the end of shelf life (after storage at +4 °C). The identification until the gender was not possible with the existing databases. The presence of high proportions of lactic acid bacteria in Brazilian and Australian meat would partially explain their long shelf life especially if these strains were bacteriocin producers. Other researches on lactic acid bacteria identification are ongoing.

IV. CONCLUSIONS

Results confirmed that the long shelf life stated by suppliers is achievable as long as storage temperatures close to the freezing point of meat are strictly respected. Differences in meat microbiota composition, especially regarding lactic acid bacteria, are a possible explanation for the long meat shelf life found in some countries. Further research on identification of the initial lactic acid bacteria population and its dynamics during meat storage is still needed.

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