Effect of high hydrostatic pressure on lactic bacteria growth in turkey ham stored at normal and abusive refrigeration temperatures

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Abstract

The lactic acid bacteria (LAB) involved a group of microorganisms responsible for food fermentation which can also cause deterioration in meat products packaged under vacuum. As a result, inadequate quality conditions are developed, which includes unpleasant odour and slime green colour associated with high population counting (generally above 10^7CFU/g) (Slongo et al., 2009). High hydrostatic pressure (HHP) treatment consists of a non-thermal process which uses pressure instead of heating for food preservation, and is currently considered a major attraction for the industry of meat products, mainly for the high potential to inactivate microorganisms leading to an increased commercial validity. Such a process usually result in negligible changes in the product quality in comparison to usual preservation processes. Storage temperature consists in the main extrinsic factor related to microorganisms' multiplication. Therefore, in this study, turkey ham was treated under pressure at 400MPa for 15 minutes at 25℃ and then stored at refrigeration temperatures of 7℃ and abuse temperature of 13℃, aiming at evalua ting the hydrostatic high pressure treatment on the LAB growth in comparison to the control. For the ham manufacture turkey thighs it was added brine and additives, mixed in a cutter, formed and cooked up to 72℃, being finally refrigerated at 7℃ or 13℃. For the LAB isolation and plating serial decimal dilutions from 10^{-1} to 10^{-7} were carried out and pour-plated on an upper layer of MRS agar, followed by BOD incubation at 30℃ for 3 to 5 days. Microbial counting was carried out in duplicate using a colony counter and statistically assessed by variance analysis and Tukey test. The control sample stored at 7℃ reached the maximum commercial validity in 35 days ($1.46x10^7$CFU/g) while the control stored at 13℃ achieved it after 21 days ($1.25x10^7$CFU/g) of storage. The pressurized samples stored at 7℃ r esulted in 91 days ($1.70x10^7$CFU/g) of commercial validity, which represented a period of 56 days higher than the control, while the sample stored at 13℃ resulted in a commercial vali dity of 42 days ($1.79x10^7$CFU/g), 21 days greater than the control. It was evident from the study that the preliminary high pressure treatment resulted in higher commercial validity of the turkey ham, both at the usual refrigerated temperature and at the abusive refrigeration conditions. Even in the pressurized turkey ham stored at abusive temperature the limiting LAB population counting was only reached long after the common shelf life period for the product. The preliminary pressure treatment proved to be an interesting option for extending the shelf life of the turkey ham, by retarding the LAB growth and delaying reaching the limiting population level. By modelling the LAB growth at 8℃ and 12℃ good fit was achieve d by using the Baranyi and the Modifed Gompertz models, respectively.

Keywords: high hydrostatic pressure, lactic acid bacteria, cold storage, turkey ham.

1. Introduction

Consumers' demands on healthier and easy to prepare food have driven industry innovation in recent time. Such innovations include the search for technologies that bring about benefits, particularly in relation to increased commercial viability without undesirable sensory changes (Slongo et al., 2009).

High hydrostatic pressure (HHP) is an innovative clean technology that minimises harming the environment. It uses pressure instead of heating in the range of 100MPa to 900MPa, within various times and possibly in combination with mild temperature process, which ensures flexibility of work in accordance with the food. It has the great advantage of allowing the production of safe food with increased commercial viability, while maintaining the sensory characteristics and nutritional virtually unchanged due to the processing (Slongo et al., 2009).

The HHP produces morphological changes in cell membrane of microorganisms increasing its permeability, besides causing some other biochemical and genetic disturbances (Sangronis et al., 1997). It inhibits and denatures enzymes essential for growth and microbial reproduction (Calderón-Miranda et al., 1998). The HHP can ensure the destruction of up to 8 log units of bacterial cells, without altering the flavour and food nutritional (Dogman and Erkmen, 2004). The destruction or inactivation of microorganisms by HHP varies with the pressure level, time and temperature of pressurization; the type and stage of growth of the microorganism; as well as the characteristics of the food (composition, pH and water activity) (Calderón-miranda et al., 1998; Rosenthal and Silva, 1997).

When meat products are refrigerated stored under microaerophilic conditions, such inside vacuum or modified atmosphere packages, the lactic acid bacteria may predominate in the deteriorated product. Since products are commonly heated at 68 to 75℃ during the production, most vegetative cells are killed and recontamination of the post-heating determines their commercial validity (Borch et al., 1996; Vermeiren et al., 2004). The recontamination after cooking especially by the microbiota present in the industries' court is often the main factor affecting the shelf life of meat products, along with the storage temperature (Samelis et. al., 1998). Typically, the initial LAB counts in vacuum packaged meat products are low, but increase during storage under refrigeration and generally cause evident deterioration when the count reaches about 7 to 8 log 10 CFU/g (Santos et. al., 2005; Vermeiren et. al., 2005). Defects caused by LAB deterioration in that stage include unpleasant odour, sour taste, colour green and slimy (Chenoll et. al., 2007).

This study aimed at evaluating the LAB growth in pressurized turkey ham stored at refrigerated temperature (7℃) and abuse temperatur e (13℃). It also aimed at modelling LAB growth in nearby temperatures (8℃ and 12℃) in comparison with the non-pressurized ham (control) aiming at evaluating the trade validity in each case.

2. Methods

1.1. Methods

For the manufacture of ham, turkey thigs were submitted to a "toilet" for removing tendons, nerves, skin and bones, and then cut into smaller pieces. Commercial spice formulation including additives were diluted in cold water and added to the meat under constant stirring followed by cutter homogenization. After a 24-hour-refrigeration period, the mixture was vacuum packed using a high temperature resistant flexible plastic (cook-in) and placed in stainless steel forms. The cooking was carried out until the internal temperature of the product reached a maximum of 72℃, by monitoring di fferent portions with a temperature controller (model ELLAB). After cooking, the product was cooled down in ice bath for 40 minutes and then stored in the refrigerator at 4℃ for 24 hours. The turkey ham was sliced at 0.5 mm thick, vacuum packed and destined to the pressure treatment carried out in a laboratory high pressure model (Stansted Fluid Power and model S-FL-850-9-W). The

operational parameters (pressure, time and temperature) were controlled on a digital panel. A HHP treatment was carried out at 400MPa for 15 minutes at room temperature, based on Slongo et al. (2009) who in the case of pork ham managed to highly increase the commercial viability.

1.4. Microbiological analyses – commercial viability

To perform the microbiological testing, samples were handled in the cleaned flow chamber, divided in portions, vacuum packed in sterile bags and stored at 7℃ and 13℃ for 75 days. The commercial viability of turkey ham and pressurized control was determined based on the LAB research according to the methodology recommended by APHA (2001), by plating diluted sample on agar culture containing Man, Rogosa & Sharp (MRS), followed by incubation at 30℃ for 3 to 5 days. The tests were performed in duplicate and counts were expressed in Log (N) (N: colony forming unit end [CFU/g]), until the samples reached 10^7 CFU/g.

1.5. Predictive modelling and statistical analyses

The predictive models of Modified Gompertz (Baty and Delignette-Muller, 2004) and Baranyi (Baranyi and Roberts, 1994) were adjusted to the growth curves at 8℃ and 12℃, using the software DMFit and Matlab® (Math Works, Natick, MA, USA). The following indices were used to compare the performance of models were mean-squared error (MSE), regression coefficient (R^2), factor bias and accuracy factor.

2. Results and Discussion

The growth of Lactic Acid Bacteria along the storage, both for the control (7ºC) and abuse temperature (13ºC) are shown in Figure 1. It can be observed that the lag phase at 7℃ for the pressurized was highly enlarged up to 15 days in average, suggesting that high pressure lead to a sub lethal injury. Such a damage associated to the low temperature resulted in a more difficult adaptation turning slower the lactic acid bacteria growth and enhanced the commercial validity.

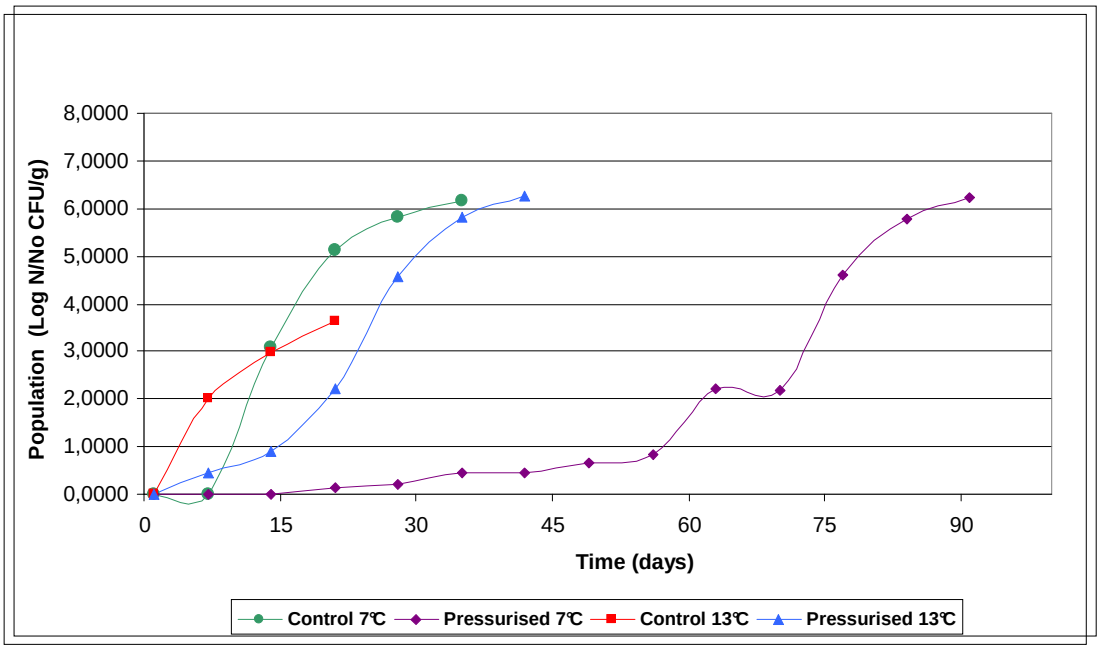

FIGURA 2: Lactic acid bacteria growth curves in pressurised and unpressurised turkey ham stored at 7 and 13℃.

The control sample stored at 7℃ reached commercial validity in 35 days ($1.46x10^7$CFU/g) while the control stored at 13℃ achieved 21 days ($1.25x10^7$CFU/g) of storage. The pressurized samples stored at 7℃ resulted in 91 days ($1.70x10^7$CFU/g) of commercial validity, which represented a period of 56 days higher than the control, while the sample stored at 13℃ resulted in a commercial validity of 42 days ($1.79x10^7$CFU/g), 21 days greater than the control. It was evident from the study that the preliminary high pressure treatment resulted in higher commercial validity of the turkey ham, both at the usual refrigerated temperature and at the abusive refrigeration conditions. Even in the pressurized turkey ham stored at abusive temperature the limiting LAB population counting was only reached long after the common shelf life period for the product. The preliminary pressure treatment proved to be an interesting option for extending the shelf life of the turkey ham, by retarding the LAB growth and delaying reaching the limiting population level

The parameters resulting from adjusting growth data to Baranyi and Gompertz Modified models are shown in Table 1, and the statistical related data are shown in Table 2. For the conditions of control and pressurized to 8℃, Baranyi model provided a better fit, while for the modified Gompertz model resulted more appropriated for adjusting the growth data at 12ºC.

TABLE 1: Acid Lactic Bacteria growth parameters in pressurised and control turkey ham stored at 8℃ and 12ºC resulting from adjustment to Modified Gompertz and Baranyi models.

	Control					Pressurized				
T (ºC)	CV (day)	A	λ (day)	μ (day^{-1})	r^2	CV (day)	A	λ (day)	μ (day^{-1})	r^2
8[1]	25	----	----	0,2387	0,976	65			0,0841	0,959
12[2]	24	6,208	6	0,4521	0,984	30	7,047	5	0,3170	0,999

1) Baranyi Model; 2) Modified Gompertz Model

TABLE 2: Statistical indexes related to the fit of Acid Latic Bacteria growth in pressurised and control turkey ham stored at 8℃ and 12ºC to Modified Gompertz and Baranyi models

Predictive Model	Statistical parameters			
	r^2	*MSE*	Bias factor	Accuracy factor
Control 8ºC[1]	0,9768	0,09382	1,00000	1,03960
Pressurized 8ºC[1]	0,9595	0,13940	1,00780	1,04370
Control12ºC[2]	0,9843	0,02540	1,00195	1,01812
Pressurized 12ºC[2]	0,9999	0,00017	0,99967	1,00203

1)Baranyi Model; 2) Modified Gompertz Model

In ham treated at 400MPa for 15 minutes at a temperature of 8℃, Slongo et. al. (2008) obtained commercial viability of 85 days compared with control, which lasted only 19 days. These results are similar to those obtained with turkey ham, in which under the same condition, the pressurized sample showed commercial viability of 65 days and the control sample reached commercial viability in 25 days. According to studies by Ruiz-Capellas (2007), the high-pressure treatment applied to vacuum packaged ham at 400MPa for 10 minutes, provided a validity of 77 commercial and 28 days for products stored at 2 and 12℃, respectively. However, López-CaLABlero et al. (1999) did not succeed in the same extent using pork ham treated at 200MPa and 400MPa, and managed to achieve a commercial viability of just 21 days at 3℃.

By using higher pressures Slongo et al. (2009) did not verify any LAB counting in sliced pork ham pressurized at 600MPa at 30℃ for 5 minutes followed by storage at 5℃ for 120 days. In similar treatment conditions (600MPa for 5 minutes and 25℃), Park et. al. (2001) obtained a reduction of ~ 4 log 10 CFU/g in pork ham. Garriga et al. (2004) reported that vacuum

packaged ham was treated at 600MPa at 16℃ for 4 mi nutes presented after 30 days a LAB count of 2.10 log10. Carpi et al. (1999) reported an increase up to 75 days in the commercial viability of sliced cooked ham treated at 600MPa for 5 minutes and stored at 4℃.

3. Conclusion

The HHP treatment at 400MPa for 15 minutes was effective to highly increase the validity of the vacuum packed turkey ham. The high hydrostatic pressure has managed to increase the validity of the product stored at a higher storage temperature (13℃) even longer when compared to the non-pressurised sample stored at a lower temperature (7℃). The technology proved to be interesting in extending the shelf-life by slowing down microbial deterioration. Predictive models fitted well to LAB growth data both for the control and pressurised sample in the two different storage temperatures evaluated.

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