High hydrostatic pressure treatment for the assessment of quality in sliced skin-packaged cooked and cured hams

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Abstract: In this study, a pressure of 600 MPa was applied to sliced skin-packaged cooked ham and cured hams. Different lots were spiked with different food pathogens and spoiling microorganisms: slime-producing lactic acid bacteria (LAB), Staphylococcus aureus, Listeria monocytogenes, Yersinia enterocolitica, Escherichia coli, Salmonella spp., Campylobacter jejuni and Debaryomyces hansenii and compared to non-spiked samples. All samples were treated at high pressures and compared to parallel treatments non pressurized. Samples were stored up to 120 days at 4°C and periodically analysed.

The microbial evaluation of non-spiked control and pressurized samples consisted in aerobic total count, psychrophilic total count, Enterobacteriaceae, Escherichia coli, Lactic acid bacteria, Staphylococcus aureus and yeasts. After 60 days of storage, lactic acid bacteria were the predominant microorganisms. At this sampling time, pressurized samples yielded a 6 log cycles drop of LAB compared to the non-pressurized samples. However, by the end of the sampling time, the difference between the pressurized and non-pressurized samples was only 1 log cycle.

In cooked ham, S.aureus was the most resistant bacteria to the high pressure treatment. 600 MPa were most efficient in killing yeasts, LAB, and E.coli. However, in some samples presence of Salmonella and L.monocytogenes was detected. After 30 days of storage, LAB recovered in pressurized samples achieving the same levels than the non-pressurized spiked samples (10^8 cfu/g) while yeasts were kept below the detection level.

Keywords: high hydrostatic pressure, pathogenic bacteria, cooked ham, cured ham

Introduction: Sliced cooked ham is a highly perishable meat product. The low salt content (2% on average), a pH of around 6.0 and a water activity higher than 0.945
are only small hurdles to inhibit the usual types of organisms associated with post-
processing contamination. The application of high hydrostatic pressures (HHP) to
meat products for the assessment of quality and enhancement of shelf life is a new
preservation process. Sliced cured ham has higher shelflife dates due to its salt
content and a_w, however the hurdles in this product may not be enough to
completely inhibit some food pathogens. The effectiveness of HHP on the
destruction of cells of several foodborne pathogens in phosphate buffer and in
some foods has been published. In general, the higher the pressure and time of
treatment the higher the bacterial destruction.
The aim of this study was to assess the efficiency of HHP in pork products when
contaminated with pathogens of industrial concern.

Materials and Methods: The strains (Staphylococcus aureus, Listeria
monocytogenes, Yersinia enterocolitica, Escherichia coli, Salmonella spp,
Campylobacter jejuni, Debaryomyces hansenii and slime-producing lactic acid
bacteria(LAB)) used for spiking samples were isolated from meat products,
characterized and stored in BHI or MRS broth plus 25% glycerol at -80°C.
The meat products (cooked and cured ham) were processed in a commercial plant,
sliced and spiked with 10^3 or 10^4 cfu/g of each microorganism between two slices
and skin-packaged.
Presurization at 600 Mpa was carried out at 20°C for 6 min. Control and spiked
samples were enumerated for pathogen growth at different sampling times: before
presurization, after presurization and during storage at 4°C for 120 days.
Microbial growth was recorded following ISO procedures for determination of the
microorganisms inoculated.

Results:
Cooked Ham In HPP^- and HHP^+ non-inoculated samples, LAB were the main
flora along storage. Presurization was very effective for (<10 cfu/g). Enterobacteriaceae
counts were under 10 cfu/g along storage in the HHP^+ samples. In the non-presurized samples, a great variability was recorded after 90
and 120 days of storage. E.coli and Staph aureus were below the detection limit
(<10 and <10^2 cfu/g) during the process. No L.monocytogenes, Campylobacter
spp. and Salmonella spp. were detected (Figure 1).
In the presurized and artificially contaminated samples, no Salmonella and Listeria
were detected in 25 and 10 g respectively after 60 days and 120 days of chilling
storage (Table 1). Staph aureus was resistant to presurization, however in the
presurized samples, its viability decreased along time. E. coli was very sensitive to
presurization (Figure 2).
Cured Ham Non-inoculated and presurized samples yielded lower counts of aerobic and psychrophilic microorganisms than non-presurized samples. Enterobacteriaceae, *E. coli* and *Staph. aureus* were not detected. Presurization was very effective for yeasts, being <10 cfu/g in presurized samples while the non-presurized yielded $10^2$ cfu/g along storage (Figure 1). The investigation of the rest of the pathogenic bacteria showed absence in 10 or 25 g. One out of three non-presurized samples showed presence of *L. monocytogenes* at the beginning of the process.

In spiked samples, *Salmonella* and *L. monocytogenes* counts decreased but still presence of this microorganisms was found after presurization and along storage (Table 1). *E. coli* was also, in this product, very sensitive to presurization and *Staph aureus* achieved 1 log$_{10}$ cfu/g reduction compared to non-presurized samples.

**Discussion and Conclusions:** The use of high hydrostatic pressure in pork products is a way to enhance its quality. The performance of a given pressure against microorganisms is determined by the meat matrix and the ingredients and additives used for the manufacture. The physico-chemical conditions of a certain product mainly pH, $a_w$, nitrifying agents, salt content can protect bacteria from the bactericidal effect of the HHP. In this study, it has been shown that certain microorganisms are more sensitive than others to HHP and that the effectivity of HHP in pork products needs to be assayed in each type of product and in every manufacturing technology.

New studies to assess the efficacy of extra hurdles as food-grade bacteriocins from lactic acid bacteria are in progress to extent the preservation effect of high hydrostatic pressure.

**Acknowledgements**
This work was supported by PROFIT and EUREKA programmes in cooperation with Esteban Espuña S.A:
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<tr>
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<th>Salmonella spp. (in 25g)</th>
<th>L. monocytogenes (in 10g)</th>
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<tr>
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<td>HPP- #</td>
<td>HPP+ ##</td>
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<tr>
<td><strong>Cooked ham</strong></td>
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<tr>
<td>Days</td>
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<tr>
<td>0</td>
<td>3.83 ± 0.10</td>
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<td>30</td>
<td>3.81 ± 0.30</td>
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<tr>
<td>60</td>
<td>3.66 ± 0.24</td>
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<tr>
<td>120</td>
<td>3.53 ± 0.18</td>
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<td><strong>Dry cured ham</strong></td>
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<td>Days</td>
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<tr>
<td>0</td>
<td>3.72 ± 0.53</td>
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<tr>
<td>30</td>
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<td>60</td>
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<tr>
<td>120</td>
<td>2.68 ± 0.51</td>
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Table 1 Counts or presence/absence of *Salmonella* and *L. monocytogenes* after HHP at 600 Mpa, 22°C, 6 min during storage at 4°C.

(#{}) Expressed as log CFU/g ± standard deviation. Triplicates for each sampling time, (##) Expressed as positive samples / investigated samples, (HPP-) non-pressurized, (HPP+) pressurized samples.
Figure 1 Behaviour of endogenous microflora after HHP at 600 Mpa, 22°C, 6 min and storage for 120 days at 4°C in cooked and cured ham. (■, stand for presurized samples and ◆, for non-presurized samples)

COOKED HAM

CURED HAM
Figure 2. Behaviour of *Staph. aureus* and *E. coli* after HHP at 600 Mpa, 22°C, 6 min and storage for 120 days at 4°C in cooked and cured ham. (■, stand for presurized samples and ♦, for non-presurized samples)