PE4.76 High pressure and freezing temperature effect on quality and microbial inactivation of pork carpaccio 270.00

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***Abstract*—Pork loins from 6 Large White gilts were used to evaluate the effect of high hydrostatic pressure (HHP: 0, 400, 600 MPa) and freezing temperature (-15° vs. -35°C) on quality and microbial inactivation of pork carpaccio. Treatment with HHP resulted in pork carpaccio with a lighter and yellower colour, a higher saturation index and a higher shear force compared with untreated samples. Treating samples at -35°C resulted in darker colour and more tender pork carpaccio compared with -15°C. HHP treated samples showed higher scores for pink colour, cooked and gelatine appearance and lower scores for rainbow-iridescence and raw meat appearance compared with control samples with no sensory differences between 400 and 600 MPa. Differences between control and treated samples were larger for pork carpaccio treated at -15 compared with -35°C. Sweetness of pork carpaccio was higher for 600 and -35 compared with 400 MPa and -15°C, respectively. Samples treated at -35°C showed higher rating for crumbliness and lower for fibrousness and chewiness compared with -15°C. HHP treated samples showed lower levels of lactic acid bacteria and psychrotrophs during shelf life compared with control carpaccio. There were no differences in microbial counts between -15 and -35°C or between 400 and 600 MPa treatments. Enterobacteriaceae counts were very low and there were no differences among treatments. While HHP is effective in microbial inactivation and shelf life extension of pork carpaccio, product quality may be decreased due to lower tenderness and a poorer appearance. Treating carpaccio at -35°C may have a lower impact on meat quality compared with -15°C, indicating that low freezing temperatures may reduce the effect of HHP on meat quality.**

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***Index Terms*— high pressure, quality, pork carpaccio**

I. INTRODUCTION

CARPACCIO has been considered traditionally a dish of raw beef, veal or tuna, thinly sliced and served as an appetizer with a dressing containing olive oil, Parmesan cheese and seasonings. The meat industry is currently preparing more convenient ready-to-eat carpaccio by curing pieces of meat, that are then frozen, sliced, packaged under vacuum or modified atmosphere without oxygen and marketed at refrigeration temperature. However, the consumption of this product is limited probably due to its fresh meat appearance as well as its sensory and safety concerns by consumers. High hydrostatic pressure processing has the potential to extend product shelf life through microbial inactivation. However, undesirable quality changes in meat products have been reported for meat colour [*1*] texture [*2*], lipid oxidation [*3*] and flavour [*4*]. High pressure treatment at different temperatures will induce different effects on meat properties and it has been shown that product treated at freezing temperatures may show lower quality changes [*5*]. The objective of this study was to evaluate the effect of HHP and freezing temperature on quality and microbial inactivation of pork carpaccio.

II. MATERIALS AND METHODS

F. Materials

Pork loins from 6 Large White gilts we used to produce pork carpaccio. Left- and right-side loins from each pig were marinated (sodium chloride, nitrite, dextrose, sucrose, sodium ascorbate and trisodium citrate), aged during 3 days at 2°C and kept frozen (-20°C) until slicing. Left-side loins were sliced (-8°C, 1.5 cm thick) and each individual slice was vacuum packaged and used for instrumental colour and tenderness evaluation. Right-side loins were sliced (-8°C, 1.5 mm thick) and vacuum packaged for sensory and microbial analysis. All samples were kept frozen in bags containing propylene glycol either at -15° or -35°C until HHP treatment. Six treatments using combinations of pressure (0, 400, 600 MPa) and

freezing temperature (-15° vs. -35°C) were applied during 6 min. on frozen samples kept in propylene glycol during processing. HHP was performed on an industrial high pressure system (Hyperbaric Wave 6500/120, Burgos, Spain) with a maximum capacity of 120 L. After HHP treatment, samples were kept overnight in a cooler (2°C) and instrumental colour and tenderness were determined on the thawed product at 24 h post-HHP treatment. Samples for microbial analysis were kept in a cooler (2°C) during evaluation (3 times: 24 h, 13 d and 41 d post-HHP treatment). Sensory samples were kept frozen (-20°C) until analysis.

G. *Instrumental colour and tenderness*

Instrumental colour measurements were recorded after HHP treatment for L*(lightness), a*(redness), and b*(yellowness) using a Minolta Chromameter (CR-400, Minolta Inc., Osaka, Japan) in the CIELAB space (CIE, 1976). Three readings were taken on one side of the 1.5 cm slices right after removing the packaging. Values were averaged for each sample and Chroma and Hue angle were calculated for each measurement.

Each slice was cut into 10 samples (1.5x2x1 cm) for instrumental texture analysis and samples were sheared perpendicular to muscle fibre direction. Warner-Bratzler shear force (WBSF) was measured using a texture analyzer Alliance RT/5 (MTS Systems Corp., Eden Prairie, MN, USA) equipped with a Warner-Bratzler blade with crosshead speed set at 2 mm/s.

H. *Sensory Analysis*

Six selected and trained assessors [*6- 8*] undertook the sensory analysis on 1.5 mm slices of pork carpaccio. The generation of the descriptors has been carried out by open discussion in three previous sessions. The visual and tactile descriptors retained were: pink colour intensity, brightness and iridescence as visual appearance attributes; cooked meat aspect, raw meat aspect and gel aspect as visual texture attributes and roughness as a tactile texture attribute. These attributes were evaluated in samples from all treatments. The flavour and texture sensory descriptors retained were: sweetness, saltiness, umami taste and metallic as flavour attributes and adhesiveness, hardness, crumbliness, stringiness and chewiness as texture attributes. These attributes were evaluated in HHP treated samples only. Although microbial data resulted in normal counts, control samples were not evaluated for safety reasons. A non-structured scoring scale [*9*] was used, where 0 meant absence of the descriptor and 10 meant high intensity of the descriptor.

I. *Microbial Analysis*

Enterobacteriaceae, lactic acid bacteria (LAB) and psychrotrophs counts were determined in 1.5 mm slices at 3 times during shelf life (time 1: 24 h post-HHP treatment, time 2: 13 d from time 1, time 3: 28 days from time 2).

At selected times each sample was diluted 10-fold in BPW (AES Chemunex España, S.A., Terrassa, Spain) and blended for 1 minute in a Masticator Classic (IUL S.A., Barcelona, Spain). Serial dilutions were made and plated onto appropriate culture media to determine Psychrotrophs Total Count (PCA, Merck, Darmstadt, Germany) at 20ºC for 5 days, LAB in Man, Rogosa and Sharpe agar (MRS, Merck) double-layered plates at 30ºC, 72 h in anaerobiosis and Enterobacteriaceae in VRBG Agar (Merck) double-layered plates at 30ºC, 24 h. Data were expressed as log CFU g^{-1} with a detection limit of 1 log CFU g^{-1}.

J. *Statistical Analysis*

Data were analyzed as a 3x2 factorial design with pressure (0, 400, 600 MPa), freezing temperature (-15 vs. -35°C), animal (block) and two-way interactions in the model using the GLM procedure of SAS (SAS Inst. Inc., Cary, NC). Microbial data were analyzed as repeated measures. Sensory evaluation was undertaken in 12 sessions and a complete block design was used (Steel and Torrie, 1983), where each taster assessed all the treatments in each session. Samples were coded with three random numbers and were presented to the assessors balancing the first order and the carry over effects according to MacFie et al. [*10*].

III. RESULTS AND DISCUSSION

A. INSTRUMENTAL COLOUR (L*, a*, b*) AND TENDERENSS (WBSF)

There was an interaction ($P<0.05$) between pressure and temperature for L* (Figure 1). Colour lightness was higher for samples treated with HHP than control samples when treated at -15°C with no differences between 400 and 600 MPa. However, there were no differences in L* among treatments when samples were treated at -35°C. These results may indicate a protection effect from the HHP of the muscle colour by the lower temperature.

There were no interactions ($P>0.05$) between pressure and temperature for a*, b*, Chroma or Hue angle, and there was no temperature effect on these color parameters. Samples treated at 600MPa showed higher redness compared with samples treated at 400

MPa with no differences in a* between the control sample and the HHP-treated samples (Table 1). Yellowness was higher for 400 and 600 MPa compared with untreated samples. Chroma was higher for pork carpaccio treated at 600 MPa compared with control, while there were no differences in Hue angle between treatments.

There were no interactions ($P>0.05$) between pressure and temperature for instrumental tenderness (WBSF, Figure 2). Samples treated with high pressure showed higher values of WBSF in kg compared with the control indicating toughening of the samples with HHP treatment regardless of the pressure level. Samples treated at -15°C tended ($P<0.10$) to show higher shear force values than samples treated at -35°C. Differences between temperatures may indicate a protection effect from the HHP of the muscle structure by the lower temperature.

B. SENSORY

There were interactions ($P<0.05$) between pressure and temperature for pink colour intensity, and all visual texture parameters (Table 2). Pink colour intensity was higher for HHP treated samples compared with control. These differences were significantly higher when samples were treated at -15°C than -35°C. These results agree with instrumental colour data which indicated greater colour changes when samples were treated at the higher temperature. Visual texture parameters showed similar trend to pink colour intensity. HHP treated samples showed higher aspect of cooked meat and gel aspect and lower aspect of raw meat compared with control samples. These differences were more significant at -15°C compared with -35°C indicating that a lower temperature would keep visual appearance and texture closer to control samples.

There were no interactions ($P>0.05$) between pressure and temperature for brightness, iridescence and roughness. There was no effect of pressure or temperature for roughness and there was no temperature effect on brightness or iridescence. Brightness was higher for control samples compared with samples treated at 400MPa, with carpaccio treated at 600 MPa being intermediate. HHP treatment resulted in higher iridescence compared with control.

Untreated pork carpaccio was not tasted by panelists due to safety reasons. There were no interactions between pressure and temperature for flavour or texture sensory parameters (Table 3). There were no effects ($P>0.05$) of pressure or temperature on saltiness, umami and metallic flavour or adhesiveness and chewiness. Sweetness was higher for samples treated at 600 MPa and -35°C compared with 400MPa and -15°C, respectively. There was no pressure effect on texture paramenters. However, higher temperature of treatment (-15°C) resulted in higher hardness and stringiness and lower crumbliness compared with the lower temperature (-35°C). These data agree with instrumental shear force values which tended to be higher in kg force at -15 compared with -35°C.

C. MICROBIAL INACTIVATION

There were no differences among treatments in Enterobacteriaceae counts which were under the detection limit (<1.0 log CFU g^{-1}) at the three evaluated times during shelf life (41 d). Pork carpaccio treated with HHP showed lower counts of lactic acid bacteria and psychrotrophs during shelf life compared with untreated carpaccio (Table 4). There were no differences in microbial counts between -15 and -35°C except for lactic acid bacteria at 41 d or between 400 and 600 MPa treatments. Control samples showed values for lactic acid bacteria and psychrotrophs close to spoilage levels at 41 d, while HHP was effective in inactivating lactic acid bacteria and psychrotrophs total counts extending shelf life of pork carpaccio.

IV. CONCLUSION

While HHP is effective in microbial inactivation delaying the growth of spoilage microorganisms and extending shelf life of pork carpaccio, product quality may be decreased due to lower tenderness and a poorer appearance. Treating carpaccio at -35°C may have a lower impact on meat quality compared with -15°C, indicating that low freezing temperatures may reduce the effect of HHP on meat quality. Further work will be carried out to evaluate the effect of HHP on inoculated pork carpaccio with selected pathogens.

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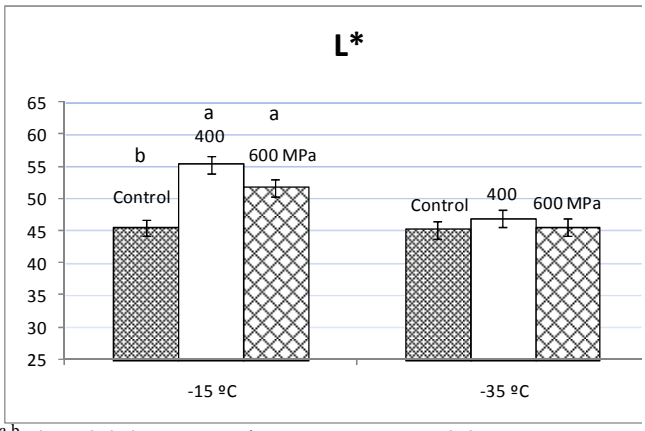


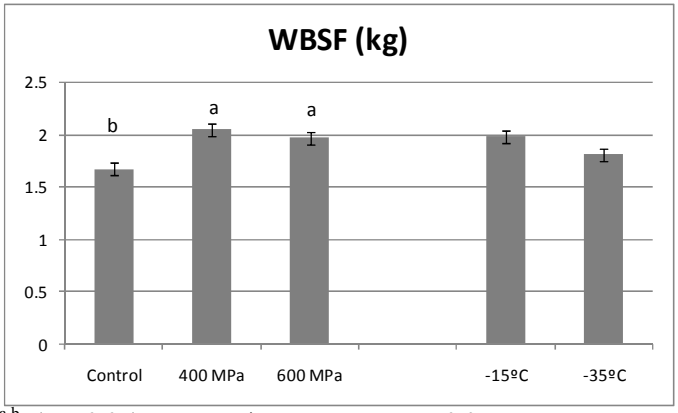
[a,b] (P < 0.05). Pressure*Temperature P < 0.05.

Figure 1. Effect of HHP (0, 400, 600 MPa) and freezing temperature (-15, -35°C) on colour lightness (L*) of pork carpaccio.

Table 1. Effect of HHP (0, 400, 600 MPa) on colour redness (a*), yellowness (b*), Chroma and Hue angle of pork carpaccio.

	0 MPa	400 MPa	600 MPa	SE
a*	7.56[ab]	7.53[b]	8.12[a]	0.21
b*	2.15[b]	3.49[ab]	3.7[a]	0.28
Chroma	7.90[b]	8.44[ab]	8.99[a]	0.25
Hue	25.43	30.01	23.91	4.33

Means within the same raw with different letters differ (P < 0.05). Pressure*Temperature P > 0.05.



[a,b] (P < 0.05). Pressure*Temperature P > 0.05.

Figure 2. Effect of HHP (0, 400, 600 MPa) and freezing temperature (-15, -35°C) on Warner-Bratzler shear force (WBSF, kg).

Table 2. Effect of HHP (0, 400, 600 MPa) and freezing temperature (-15, -35°C) on visual appearance and visual and tactile texture of pork carpaccio.

Temp (°C)	-15			-35			RMSE
Pressure (MPa)	**Control**	**400**	**600**	**Control**	**400**	**600**	
Visual Appearance							
Pink color intensity	1.8[d]	5.8[a]	5.0[b]	1.9[d]	3.4[c]	3.5[c]	0.63
Brightness	3.9	3.2	3.0	4.1	3.1	3.8	0.88
Iridescence	0.1	0.8	0.8	0.1	0.7	0.6	0.38
Visual Texture							
Aspect of cooked meat	0.8[c]	5.1[a]	4.7[a]	0.9[c]	2.9[b]	3.4[b]	0.65
Aspect of raw meat	6.6[a]	2.5[d]	2.9[cd]	6.5[a]	4.5[b]	3.7[c]	0.71
Gel aspect of the lean	1.6[c]	4.6[a]	4.4[a]	1.8[c]	3.3[b]	3.9[a]	0.57
Tactile texture							
Roughness	1.6	1.7	1.9	1.7	1.6	1.9	0.51

Means within the same raw with different letters differ (P < 0.01). Pressure*Temperature P < 0.05.

Table 3. Effect of HHP (0, 400, 600 MPa) and freezing temperature (-15, -35°C) on taste, flavor and texture parameters of pork carpaccio.

	Pres. (MPa)		Temp. (ºC)		RMSE
	400	**600**	**-15**	**-35**	
Taste and Flavour					
Sweetness	2.2^{b}	2.5^{a}	2.1^{b}	2.5^{a}	0.406
Saltiness	2.9	3.0	3.0	2.9	0.517
Umami	2.5	2.6	2.6	2.5	0.497
Metallic	1.5	1.7	1.5	1.7	0.639
Texture					
Adhesiveness	3.5	3.7	3.4	3.8	0.762
Hardness	3.6	3.6	3.9^{a}	3.3^{b}	0.5700
Crumbliness	5.1	5.0	4.8^{b}	5.3^{a}	0.714
Stringiness	4.0	4.1	4.5^{a}	3.6^{b}	0.677
Chewiness	4.3	5.0	4.8	4.5	2.068

Means within the same raw with different letters differ ($P < 0.05$). Pressure*Temperature $P > 0.05$.

Table 4. Effect of HHP (0, 400, 600 MPa) and freezing temperature (-15, -35°C) on lactic acid bacteria and Psychrotrophs total counts.

		Lactic acid bacteria					
		1d		**13d**		**41d**	
		Lsmean	SE	Lsmean	SE	Lsmean	SE
Pressure	**0 MPa**	1.43^{a}	0.08	2.45^{a}	0.12	6.75^{a}	0.23
	400 MPa	0.97^{b}	0.08	1.21^{b}	0.12	2.99^{b}	0.23
	600 MPa	0.95^{b}	0.08	1.25^{b}	0.12	3.20^{b}	0.23
Temp	***-15 ºC***	1.09	0.06	1.53	0.10	4.00^{b}	0.19
	-35 ºC	1.15	0.06	1.74	0.10	4.62^{a}	0.19
		Psychrotrophs					
		1d		**13d**		**41d**	
		Lsmean	SE	Lsmean	SE	Lsmean	SE
Pressure	**0 MPa**	3.78^{a}	0.08	4.52^{a}	0.12	7.25^{a}	0.19
	400 MPa	2.24^{b}	0.08	2.30^{b}	0.12	4.70^{b}	0.19
	600 MPa	2.06^{b}	0.08	2.18^{b}	0.12	4.74^{b}	0.19
Temp	**-15 ºC**	2.63	0.06	2.96	0.1	5.43	0.15
	-35 ºC	2.76	0.06	3.04	0.1	5.69	0.15

Means within the same raw with different letters differ ($P < 0.05$). Pressure*Temperature $P > 0.05$.