PS2.05 Effect of pork meat pH on iron release from heme molecule during cooking 253.00

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Abstract—Non-heme iron (NHI) is one of the most important catalyst of the lipid and protein oxidation process. This study investigated the relationship between the level of iron chemical forms and the initial raw meat pH values during cooking, including the effect of the sodium nitrite presence. The pH value, the heme iron (HI) and non-heme iron (NHI) concentrations were measured on one ground portion of five loin, five masseter, five semimembranosus, five neck muscles and five shoulder muscles. Afterwards, all raw meats were halved and mixed with NaCl (2%), ascorbic acid (0.05%) and 0 mg/Kg of sodium nitrite (mix 1) or 150 mg/Kg of sodium nitrite (mix 2). Both mixes were divided into 50g-portions, vacuum packed in cryovac bags and cooked in thermostatic bath up to F(10, 71°C)=30 minutes at the core. The cooked weight loss percentage was calculated and the NHI and HI concentrations were determined both on the cooked meats and on the juice lost during cooking. The cooking process caused the release of NHI and HI on all cooked sample juices. The HI overall percentage was significantly lower than raw meat one ($p < 0.005$) and the NHI significantly higher ($p < 0.005$) in all cooked mix 1. The raw and cooked HI percentage variances depended on the pork meat pH values ($r2 = 0.70$). The overall HI percentage was unchanged respect to raw meats on cooked mix 2, while the NHI amount was not quantitatively estimated. These results emphasized the role of sodium nitrite on tying up NHI in the cooked meats and safeguarding the oxidative stability of cooked meat products.

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Index Terms — pork cooked meat, heme-iron, non-heme iron, pH

I. INTRODUCTION

THE lipid and protein oxidation is one of the main causes of quality loss on meats and meat products. Lipid oxidation reduces food nutritional and sensory properties decreasing the content of essential fatty acids and vitamins and generating some toxic compounds together with off-flavors [1]. The protein oxidation contributes to meat degradation causing a protein solubility and functionality decrease in model system [2] and a texture and color deterioration on the muscles [3]. Non-heme iron (NHI) is thought the most important catalyst of the lipid and protein oxidation processes on meat products [4,5]. The NHI concentration varies from species and meat cuts [6]. The thermal process usually increases the NHI amount on the cooked meat products, because of the heme molecule breakdown and the iron release during cooking [7]. The heme-iron stability depends on the cooking methods and the added chemical molecules [8-10]. The sodium nitrite stabilizes the heme iron by forming the nitrosylhemochrome in cooked cured meats, but it is not clear how its antioxidant activity is carried out [11]. The effectiveness of alternative chemical and natural antioxidants is contradictory on cooked meat and it depends on the meat composition and the antioxidant concentration [12,13]. The cooked meat products are manufactured with pork raw cuts very different for composition, pH and initial proportion of iron chemical forms. Since the myoglobin denaturation is due to the cooking process and is influenced by the meat pH value [14], it is likely that there is a relationship between the iron release from heme molecule and the raw meat pH value. Moreover, the knowledge of the iron amount released from heme molecule during cooking on the different raw meats could help to better understand the meat product oxidative stability. The aim of this study is to investigate the relationship between the level of iron chemical forms and the pH value in pork meats during the cooking process, including the effect of the sodium nitrite presence.

II. MATERIALS AND METHODS

A. Experimental design

Five loin, five masseter, five semimembranosus, five neck muscles and five shoulder muscles were obtained by the local markets. All samples were vacuum-packed, frozen at -18°C and stored until the analyses. Therefore the meats were tempered until -2 °C, trimmed from visible fat and connective tissue and ground. The pH value, the heme iron and non-heme iron concentrations were measured on one portion of all raw meats. The other was halved and mixed with NaCl (2%), ascorbic acid (0.05%) and 0 mg/Kg of sodium nitrite (mix 1) or 150 mg/kg of sodium nitrite (mix 2). Both mixes were divided into 50g-portions, that were vacuum packed in cryovac bags and cooked in thermostatic bath up to F(10, 71°C)=30 minutes at the core. The cooked weight loss percentage was calculated and the heme and non-heme iron concentrations were determined both on the cooked meats and on the juice lost during cooking.

B. Analytical methods

Iron analysis Non-heme iron (NHI) was determined by spectrophotometry following the ferrozine method described by Ahn et al. [15]. The amounts of iron were expressed as milligrams of iron per 100 grams of sample (mg/100g). Heme iron (HI) was calculated using the following formula: HI (mg/100g)= hematin (mg/100g) x AW/MW where the hematin concentration was determined using the Hornsey method [16], AW was the iron atomica weight and MW the hematin molecular weight. The cooked meat NHI and HI percentages were calculated by using the cooking juice loss in order to compare the cooked meat data with those of raw meats. pH determination The pH was measured on the minced samples using a glass pH electrode (Ingold pH-meter) Cooking weight loss The percentage cooking weight loss (CWL%) was calculated using the formula: weight raw mix - weight cooked mix CWL(%) = x 100 weight raw mix

C. Statistical analysis

The one-way Anova analysis was carried out with the SPSS 12 statistical package, using the Least Significant Difference comparison test (LSD) to identify the differences between the means.

III. RESULTS AND DISCUSSION ‘

Table 1 shows the raw meat pH values, the raw and cooked meat heme iron (HI) and non-heme iron (NHI) concentrations, the corresponding heme iron/non-heme iron ratios (HI/NHI) and the mix 1 and 2 cooking weight loss percentages. The HI and NHI concentrations were very variable among the raw meats ($p<0.001$); the loin, semimembranosus and neck muscle pH values and HI/NHI ratios were significantly lower ($p<0.01$) than others. The applied cooking procedure was mild, therefore the cooked mix HI concentrations were slightly different compared to the corresponding raw meats ($p > 0.05$), except for the shoulder muscles. Instead, the NHI concentration slightly increased on cooked mix 1, but significantly decreased on cooked mix 2 compared to the corresponding raw meat. The last results were in accordance with the greater oxidative stability of nitrite added cooked meats respect to the raw and nitrite less cooked meats. The raw and cooked meat data of table 1 couldn’t be directly used in order to estimate the changes of heme and non heme iron distribution percentage before and after cooking because of the moisture loss during cooking and the iron part release in cooking juice. Therefore, the cooking juices were analyzed and the iron content was calculated on the cooked meats by taking into account the cooking weight loss. Figure 1 shows the HI and NHI mean percentages in the raw meats and the corresponding HI and NHI mean percentages in cooked mix 1 and mix 2. The cooking process caused the release of non-heme and heme iron on all cooked sample juices. However, the cooked meat and juice HI percentage sum was significantly lower than raw meat one ($p <0.05$), and, at the same time, the NHI percentage sum was significantly higher ($p <0.05$) in all samples made without nitrite (mix 1). Moreover, the iron release quantity from heme iron depended on the pork meat pH; in fact the raw and cooked HI percentage variances correlated with the pork meat pH values (Figure 2). This result underlines that the lower pH values of raw meats increases not only the pigment denaturation, but also the iron release from heme molecule. As we expected, the overall HI percentage was unchanged respect to the raw meats on cooked samples with nitrite (mix 2), but the NHI amount was not quantitatively estimated in all cooked mixes 2 (Figure 1). Some tests were carried out in our laboratory and showed that sodium nitrite did not interfere with the ferrozine method. In fact the iron was fully detected in heated watery solution containing sodium nitrite and Fe(II) and in meat samples first cooked then added with sodium nitrite. These results support the hypothesis that nitrite both converts heme proteins to stable and

catalytically inactive nitric oxid proteins and ties up non-heme iron during cooking process of the meat. Therefore, the lower non-heme iron amounts could make the nitrite added cooked meats more stable to oxidation process during storage.

IV. CONCLUSION

The non-heme iron is one of the most important catalyst of lipid and protein oxidation in meats and meat products. This work emphasizes the double antioxidant function of sodium nitrite on cooked meats by tying up the non-heme iron and preserving heme-iron. If the sodium nitrite is not added in cooked products, the iron release from heme is influenced by the meat pH. Therefore, these results should be taken into account in order to safeguard the oxidative stability of new cooked meat products manufactured without sodium nitrite and by adding new antioxidant molecules.

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Table 1- means and standard deviations of the raw meat pH, the raw and cooked meat heme-iron (HI) and non-heme iron (NHI) and the cooking weight loss percentage (CWL%)

Sample		pH		HI		NHI		HI/NHI		CWL%	
		mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.	mean	s.d.
Loin	Raw meat	5,64 a	0,13	0,46 a	0,10	0,25 a,y	0,04	1,84 a,x	0,39	-	-
	Mix1			0,45	0,13	0,31 y	0,08	1,5 x	0,58	14,36	2,94
	Mix2			0,48	0,11	0,19 x	0,04	2,79 y	1,28	12,86	3,35
Semimembranosus	Raw meat	5,77 a	0,12	0,71 a	0,12	0,37 b,y	0,02	1,91 a,x	0,27	-	-
	Mix1			0,68	0,13	0,39 y	0,03	1,74 x	0,14	15,25	1,96
	Mix2			0,73	0,16	0,25 x	0,07	2,99 y	0,61	14,66	2,83
Shoulder muscles	Raw meat	5,80 b	0,08	1,06 b,x	0,03	0,32 a,y	0,01	3,36 b, x	0,12	-	-
	Mix1			1,13 y	0,01	0,35 z	0,02	3,24 x	0,16	17,28	0,53
	Mix2			1,16 z	0,01	0,20 x	0,01	5,66 y	0,01	17,63	0,56
Neck muscles	Raw meat	5,76 a	0,07	1,16 c	0,05	0,77 d,y	0,01	1,54 a,x	0,06	-	-
	Mix1			1,24	0,21	0,83 y	0,01	1.50 x	0,28	16,08	0,31
	Mix2			1,20	0,25	0,46 x	0,30	2.04 y	0,03	16,41	0,81
Masseter	Raw meat	5,92 b	0,10	2,08 d	0,65	0,46 c,y	0,13	4,53 b,x	0,58	-	-
	Mix1			2,04	0,58	0,49 y	0,11	4,16 x	0,44	12,17	3,17
	Mix2			2,14	0,75	0,25 x	0,10	8,52 y	0,28	8,6	1,24

a,b,c,d: different letters in the same column correspond to significant differences among the raw meats

x,y,z: different letters in the same column correspond to significant differences among the raw meat and corresponding cooked mix 1 and 2 of single sample meat

Figure 1- HI and NHI percentages on raw meat and cooked mix 1 (0 mg/Kg of sodium nitrite) and mix 2 (150 mg/Kg of sodium nitrite) of loin (a), Semimembranosus (b), Shoulder muscles (c), neck muscles (d) and masseter (e).

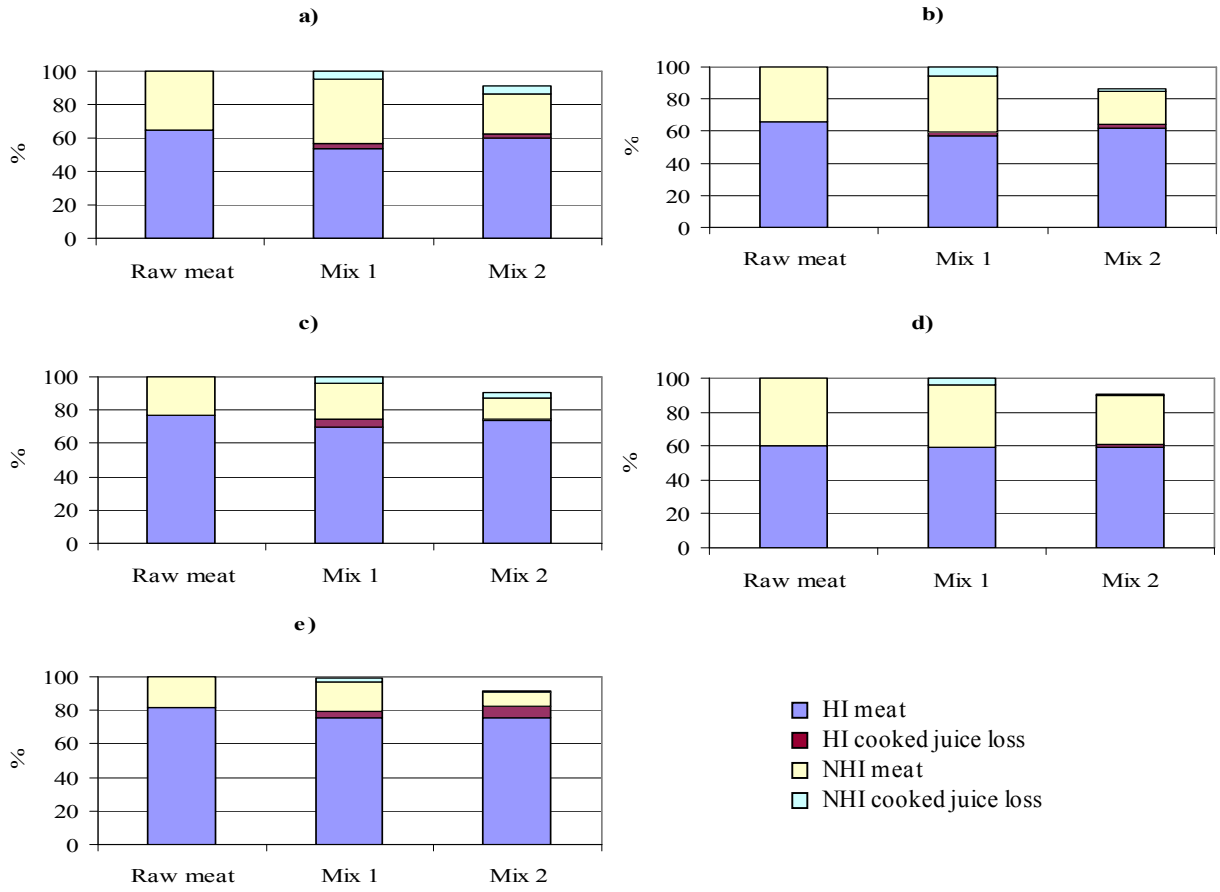

Figure 2 – Effect of raw meat pH on iron release from heme molecule on cooked meats without sodium nitrite