REDUCTION IN NITRITE LEVEL IN COOKED LOIN MANUFACTURED WITH VEGETABLE POWDER AS SOURCE OF NITRITE

M. Vaquero, B. Rubio, B. Martínez and Mª. J. Sánchez

Consejería de Agricultura y Ganadería de Castilla y León. Instituto Tecnológico Agrario. Estación Tecnológica de la Carne. C/ Filiberto Villalobos s/n. 37770 Guijuelo (Salamanca). Spain

* Begoña Rubio (phone: +34-923580688; fax: +34-923580353; e-mail: rubherbe@itacyl.es)

Abstract— **The aim of this study was to determine the type and the amount of plant extract necessary to reduce the residual nitrite level in cooked loin manufactured with vegetable powder without modify its sensory quality. To achieve this objective four batches of cooked loin were manufactured: (1) 0.60% white wine extract, (2) 1.80% white wine extract, (3) 0.60% rosemary, sage and oregano extract, (4) 1.80% rosemary, sage, oregano extract. The following analyses: pH, nitrite and nitrate content, instrumental measurement of colour and sensory evaluation were carried out. The results showed that the use of white wine extract was a more suitable strategy than the use of rosemary, sage and oregano extract for reducing the residual nitrite level in cooked loin manufactured with vegetable powder. The inclusion of 1.80% of white wine extract involved a nitrite reduction of 40%. However, the cooked loins manufactured with this percentage were the least appreciated.**

Index Terms— **cooked loin, nitrite, spices extract, wine extract.**

I. INTRODUCTION

Today, consumers demand healthier meat products. In this sense, the use of natural compounds is more accepted than the use of chemical additives in their manufacture. Several authors (Sindelar, Cordray, Sebranek, Love & Ahn, 2007; Sánchez-Iglesias, Vaquero-Martín, Rubio & Martínez, 2009; Vaquero-Martín, Sánchez-Iglesias, Martínez & Rubio, 2009) have pointed that the use of vegetable powder is a technologically viable alternative for manufacturing cooked meat products without nitrite added. However, cooked meat products made with this strategy can present a high residual nitrite level. These nitrites could act as precursors of nitrosamines, therefore, it is necessary the reduction of these levels. Viuda-Martos, Fernández-López, Sayas-Barbera, Sendra, Navarro & Pérez-Álvarez (2009a) proposed the incorporation of citrus co-products as strategy to reduce residual nitrite content in meat products. Besides, authors as Krishnaswamy (2001) and Garrote, Cruz, Moure, Domínguez & Parajó (2004) have pointed that different biocompounds occurring in plants, such as polyphenols, flavonoids, terpenes can act as nitrite scavengers. But their scavenging capacity towards nitrite differs according to the nature and quantity of these compounds. Therefore, the objective of this work was to determine the type and the amount of plant extract necessary to reduce the residual nitrite level in cooked loin manufactured with vegetable powder without modify its sensory quality.

II. MATERIALS AND METHODS

To achieve this objective 12 loins were divided in four batches. Each batch of loins was injected with a brine containing 1.32% of vegetable powder and the different type and concentration of plant extracts:

- *Batch 1*: 0.60% white wine extract (25% polyphenols).
- *Batch 2*: 1.80% white wine extract (25% polyphenols).
- *Batch 3*: 0.60% rosemary, sage and oregano extract (20% diterpenes).
- *Batch 4*: 1.80% rosemary, sage and oregano extract (20% diterpenes).

All the loins were pumped to 120% of their green weight with the different brine solutions using a multi-needle brine injector (Ogalsa CH-14). The injected loins was transferred to a tumbling machine (SM-Pulmax) and massaged with cycles of time on and time off, at 2 ºC. After tumbling, the loins were introduced into elastic mesh and the pieces were cooked in an oven (Industrial Junior 1100, Verinox) to a core temperature of 68ºC. During the cooked, a smoking step was applied. Finally, cooked loins were cooled in a chilling room at 2ºC.

On all cooked loins manufactured, different analyses were carried out. The **pH** were determined with a pH meter model 507 (Crison Instruments, Barcelona, Spain) equipped with a glass electrode and a temperature control in the brines, raw matter and final product. **Nitrate content** was determined by high performance liquid chromatography (HPLC), using a method proposed by Merino, Edberg, Fuchs & Aman (2000). **Nitrite content** was determined according to the ISO method 3091:1975. **Instrumental measurement of colour** was performed on the surface of cooked loin using a reflectance spectrophotometer (CM-2600d/2500d Konica Minolta, Aquateknia S.A., Spain). The illuminant used was D65 and the standard observer position was 10º. Colour coordinates were determined in the CIE-LAB system and the results were expressed as lightness (L*), redness (a*), yellowness (b*), hue angle (H*) and chroma (C*). Finally, **sensory evaluation** was carried out on cooked loins by a trained sensory panel (UNE 87-024-1, 1995; UNE EN ISO 8586-2, 2009). The slices (1cm thickness) were roasted in an oven (model HT-610-ME, Teka, Santander, Spain) set at 170ºC to reach a core temperature in the slice of 68ºC. Then, samples (4×2×1 cm) were obtained, wrapped with aluminium foil and kept at 45ºC to present to the panellists one at a time. Triangle (UNE 4120:2008) and preference (UNE 87023, 1995) tests were conducted.

Data sets were statistically analyzed using one-way variance analysis (ANOVA) in order to determine any significant differences among the different batches. The means were separated by Tukey-honest significant difference test at 5% level. Data analyses were conducted using STATISTICA 7.0 statistical package.

III. RESULTS AND DISCUSSION

The results obtained in the pH evaluation are showed in the table 1. The brine containing a high amount of white wine extract showed the lowest pH value ($p<0.05$). This could be due to the composition of this extract, which contains a high level of acidic compounds. No differences were found in the pH of raw matter. On the other hand, the use of a higher percentage of plant extract had not effect on the pH of the cooked loins.

Table 1.- Results (means ± SD) obtained in the pH evaluation.

*Batch	**Brine**	**Raw matter**	**Cooked loin**
1	^{B}5.88 ± 0.03	^{A}5.68 ± 0.08	^{B}6.12 ± 0.04
2	^{A}5.62 ± 0.01	^{A}5.66 ± 0.12	^{AB}6.00 ± 0.09
3	^{C}6.03 ± 0.02	^{A}5.71 ± 0.24	^{A}5.97 ± 0.03
4	^{B}5.82 ± 0.03	^{A}5.79 ± 0.17	^{A}6.00 ± 0.02

**Batch 1:* 0.60% white wine extract. *Batch 2*: 1.80% % white wine extract. *Batch 3*: 0.60% rosemary, sage and oregano extract. *Batch 4*: 1.80% rosemary, sage and oregano extract.
$^{A\text{-}C}$Means of each parameter with different letter in the same column are different ($p<0.05$).

The residual nitrite levels obtained in the cooked loins are presented in the table 2. In the case of cooked loins manufactured with white wine extract (batches 1 and 2), the nitrite levels fell ($p<0.05$) in a dose-dependent manner while in the case of cooked loins manufactured with rosemary, sage and oregano extract (batches 3 and 4) the nitrite level was not statistically reduced. Viuda-Martos, Ruiz-Navajas, Fernández-López & Pérez-Álvarez (2009b and 2010) reported a reduction of nitrite level in bologna sausage and mortadella due to the addition of citrus co-products and essential oils. These authors pointed out that such reductions in residual nitrite were probably due to the reaction of the different biocompounds present in the orange dietary fibre, especially polyphenols and flavonoids and in the essential oils, especially terpenes and alcohols. In our work, the addition of extract containing polyphenols was more effective than the use of extract containing diterpenes in the reduction of residual nitrite levels. Regarding nitrate, the cooked loins manufactured with 1.80% of rosemary, sage and oregano extract (batch 4) presented a higher nitrate content than the cooked loins made with 0.60% (batch 3). This result could indicate that the nitrite formation was lower in batch 4 than in the batch 3, what may confirm that, in our study, terpenes were less effective in the decrease of residual nitrite.

Table 2.- Residual nitrite and nitrate content (means ± SD) in the cooked loins manufactured.

*Batch	**Nitrite** (ppm)	**Nitrate**(ppm)
1	^{B}141.37 ± 39.21	^{A}18.05 ± 0.07
2	^{A}84.59 ± 19.99	^{AB}23.95 ± 0.35
3	^{B}143.54 ± 58.13	^{A}17.75 ± 0.64
4	^{AB}94.48 ± 17.54	^{B}31.60 ± 5.09

**Batch 1:* 0.60% white wine extract. *Batch 2*: 1.80% % white wine extract. *Batch 3*: 0.60% rosemary, sage and oregano extract. *Batch 4*: 1.80% rosemary, sage and oregano extract.
$^{A\text{-}B}$Means of each parameter with different letter in the same column are different ($p<0.05$).

Colour values (L*, a*, b*, C* and H*) of the different cooked loins are given in the table 3. In spite of the differences observed in the pigmentation of the brines, the addition of different types of plant extract had not effect on the colour parameters measured in the final product. On the contrary, some studies reported changes in the final product when natural antioxidants were added due to the pigmentation of these natural compounds (Deda, Bloukas & Fista, 2007); Mitsumoto, O´Grady, Kerry & Buckley, 2005). In general, the values obtained in this study for colour parameters were similar than those found by Rubio, Sánchez, Vaquero & Martínez (2009) in cooked loin.

Table 3.- Results (means ± SD) of instrumental measurement of colour in the cooked loins manufactured.

Batch*	**L***	**a***	**b***	**C***	**H***
1	71.31 ± 2.94	6.25 ± 0.59	8.42 ±1.18	10.51 ± 1.12	53.24 ± 3.77
2	73.08 ± 3.04	5.44 ± 1.24	8.31 ± 0.61	10.04 ± 0.84	56.05 ± 6.55
3	73.39 ± 2.02	5.73 ± 0.59	7.85 ± 0.16	9.73 ± 0.28	53.93 ± 3.20
4	72.33 ± 1.71	6.22 ± 0.31	8.12 ± 0.82	10.24 ± 0.73	52.44 ± 2.60

**Batch 1:* 0.60% white wine extract. *Batch 2*: 1.80% % white wine extract. *Batch 3*: 0.60% rosemary, sage and oregano extract. *Batch 4*: 1.80% rosemary, sage and oregano extract.

With respect to the results of the triangle tests (table 4), consumers distinguished between cooked loins processed with different concentration of the same extract type (between batches 1 and 2 and between batches 3 and 4). According to the panellists, the differences found between cooked loins manufactured with different percentage of white wine extract were due to the colour. This result is not in agreement with those found in the instrumental measurement of colour (table 3), probably due to sensory evaluation was carried out on cooked loins roasted. On the other hand, the taste was the parameter that allowed differs between cooked loins made with different amount of diterpenes. When different extract type was used the panellists could establish differences between the cooked loins manufactured with 0.60% of white wine extract (batch 1) and those manufactured with 0.60% of rosemary, sage and oregano extract (batch 3) and between the cooked loins manufactured with 1.80% of white wine extract (batch 2) and those manufactured with 0.60% of rosemary, sage and oregano extract (batch 3).

Concerning to the results of the preference test, the cooked loins more appreciated ($p<0.05$) were those processed with 0.60% of white wine extract (batch 1) and those with 1.80% of rosemary, sage and oregano extract (batch 4).

Table 4.- Discriminant analysis between cooked loins manufactured. (Number of judges in triangle tests n = 21).

*Compared batches	Number of correct judgements	Significance level
1 and 2	15	0.01
1 and 3	15	0.01
1 and 4	11	n.s
2 and 3	13	0.1
2 and 4	9	n.s
3 and 4	12	0.5

**Batch 1:* 0.60% white wine extract. *Batch 2*: 1.80% % white wine extract. *Batch 3*: 0.60% rosemary, sage and oregano extract. *Batch 4*: 1.80% rosemary, sage and oregano extract.

IV. CONCLUSION

The use of white wine extract was a more suitable strategy than the use of rosemary, sage and oregano extract for reducing the residual nitrite level in cooked loin manufactured with vegetable powder. The inclusion of 1.80% of white wine extract involved a nitrite reduction of 40%. However, the cooked loins manufactured with this percentage were the least appreciated.

ACKNOWLEDGEMENT

This research was supported by the FEDER project (PEP- 2006.001934) and Consejería de Agricultura y Ganadería de Castilla y León. Instituto Tecnológico Agrario de Castilla y León (ITACyL). Montserrat Vaquero gratefully acknowledges of a grant from Instituto Nacional de Investigaciones Agrarias y Alimentarias (INIA).

REFERENCES

Deda, M. S., Bloukas, J. P., Fista & G. A. (2007). Effect of tomato paste and nitrite level on processing and quality characteristics of frankfurters. *Meat Science*, 76, 501-508.

Garrote, G. Cruz, J. M., Moure, A., Domínguez, H., & Parajó J. C. (2004). Antioxidant activity of byproducts from the hydrolytic processing of selected lignocellulosic materials. *Trends in Food Science & Technology*, 15, 191-200.

ISO (International Organization for Standarization) (1975). Determination of nitrite content, ISO 2918:1975 standard. In International standards meat and meat Product-Geneva, Switzerland: International Organization for Standarization.

Krishnaswamy, K. (2001). Nonnutrients and cancer prevention. ICRM Bulletin, 31. http://www.icmr.nic.in

Merino L., Edberg U., Fuchs G., & Aman P. (2000). Liquid chromatographic determination of residual nitrite/nitrate in foods: NKML Collaborative Study. *Journal of AOAC International*, 83: 365- 375.

Mitsumoto, M., O´Grady, M. N., Kerry J. P., & Buckley, D. J. (2005). Addition of tea catechins and vitamin C on sensory evaluation, colour and lipid stability during chilled storage in cooked or raw beef and chicken patties. *Meat Science*, 69, 773-779.

Rubio, B., Sánchez, Mª. J., Vaquero, M., & Martínez, B. (2009). Elaboración de productos cárnicos cocidos ecológicos. *Eurocarne*, 180, 1-9.

Sánchez-Iglesias, M. J., Vaquero-Martín, M., Rubio, B. & Martínez, B., (2009). Physico-chemical and sensory characteristics of cooked ham manufactured with vegetable juice powder *vs* sodium nitrite. In Proceedings 55th International Congress of Meat Science and Technology , parallel session 4, part 2, PE4.44, 16-21 August 2009, Copenhagen, Denmark.

Sindelar J. J., Cordray, J. C., Sebranek, J. G.; Love J. A., & Ahn, D. U. (2007). Effects of vegetable juice powder concentration and storage time on some chemical and sensory quality attributes of uncured, emulsified cooked sausages. *Journal of Food Science*, 72, S324-S332.

UNE. (1995). Análisis sensorial. Guía general para la selección, entrenamiento y control de jueces. Norma española 87-024-1.

UNE. (2009). Análisis sensorial. Guía general para la selección, entrenamiento y control de evaluadores. Norma española 8586-2.

UNE. (2008) Análisis sensorial. Metodología. Prueba triangular. Norma española 4120. Asociación Española de Normalización y Certificación (AENOR).

UNE. (1995). Análisis sensorial. Ensayo de clasificación por ordenación. Norma española 87023. Asociación Española de Normalización y Certificación (AENOR).

Vaquero-Martín, M., Sánchez-Iglesias, M. J., Martínez, B., & Rubio, B. (2009). Effect of manufacturing with vegetable juice powder as source of nitrites of cooked loin. In Proceedings 55th International Congress of Meat Science and Technology parallel session 4, part 2, PE4.41, 16-21 August 2009, Copenhagen, Denmark.

Viuda-Martos, M., Fernández-López, J., Sayas-Barbera, E., Sendra, E., Navarro, C., & Pérez-Álvarez J. A. (2009a). Citrus co-products as technological strategy to reduce residual nitrite content in meat products. *Journal of Food Science*, 74, R93-R100.

Viuda-Martos, M., Ruiz-Navajas, Y., Fernández-López, J., & Pérez-Álvarez, J. A. (2009b). Effect of adding citrus waste water, thyme and oregano essential oil on the chemical, physical and sensory characteristics of a bologna sausage. *Innovative Food Science and Emerging Technologies*, 10, 655-660.

Viuda-Martos, M., Ruiz-Navajas, Y., Fernández-López, J., & Pérez-Álvarez, J. A. (2010). Effect of added citrus fibre and spice essential oils on quality characteristics and shelf-life of mortadella. *Meat science*, 10.1016/j.meatsci.2010.03.007.