Technological approach to reduce NaCl content of traditional smoked dry-cured hams: effect on quality properties and stability

Maria Martuscelli¹ · Laura Lupieri² · Clemencia Chaves-Lopez¹ · Dino Mastrocola¹ · Paola Pittia¹

Revised: 28 June 2015 / Accepted: 14 July 2015 / Published online: 25 July 2015
© Association of Food Scientists & Technologists (India) 2015

Abstract The modification of the salting procedure (from a three- to a two-salt coverage steps) and its effects on quality and stability properties has been investigated to reduce NaCl content of traditional dry-cured ham. The study was applied on green hams of small-S and large-L weight classes. Results evidenced that a two-salt coverage steps salting could be applied to reduce significantly NaCl content of S-size hams and to reach the physico-chemical conditions required for microbial stability at the end of ripening. The final salt content of the products results ($p < 0.05$) to depend on salting procedure and initial weight of the hams, while limited differences on quality properties have been observed being the latter mainly associated to the pattern of the volatile compounds. In particular, aldehydes and hexanal content were lower in hams undergone to a 2-steps salting. Sensory analysis evidenced that the hams with reduced NaCl (2s-S and 2s-L) were less easy to chew, less salty and with a lower intensity of the smoky flavour in respect to the 3s- ones. The study confirmed the feasibility of salt content reduction of traditional dry-cured hams by modifying the salting process. However, the weight of the initial tights resulted a critical factor in affecting salting diffusion, salt content and water activity of the ripened products, their quality and stability properties.

Keywords Dry-cured ham · NaCl reduction · Salting · a_w · Stability · Quality

✉ Paola Pittia
ppittia@unite.it

¹ Faculty of Bioscience and Technology for Food, Agriculture and Environment, University of Teramo, Via C.R. Lerici 1, 64023 Mosciano S. Angelo, TE, Italy

² Prosciuttificio Wolf S.p.A., Sauris di Sotto, UD, Italy

Introduction

Salting is an ancient technology widely used in combination with other actions to preserve meat by water activity depression and to develop the desired sensory properties (Toldrá 2006).

Recent health and nutritional concern about sodium intake has led to a reduction of the salt content also in processed meat products (Albarracín et al. 2011). However, this task could be non easy to be achieved as in some products salt plays important technological roles and is implied in their quality and stability. Its presence and concentration are influencing the physical processes related to the water loss and dehydration and the corresponding microbial stability; influence and modulate the activity of the endogenous enzymes implied in the development of sensory properties (i.e. flavour, consistency) (Toldrá 2006; Desmond 2006). Thus, NaCl decrease has to avoid undesired changes of the key quality attributes of the final ripened product. Several downsides have been observed in low salt content meat products including poor flavour and aroma development (Virgili et al. 1995); lack of cohesiveness of the slice and excessive softness due to a no-inhibited proteolysis (Ruiz-Ramírez et al. 2006); poor aroma due to the salt acting as flavour enhancer (Laureati et al. 2014); poor colour formation and stability in nitrate-free dry cured hams (Benedini et al. 2008).

Technological actions aimed to decrease the salt content of dry cured meat products include: (i) partial replacement of NaCl with other chloride salts; (ii) processing changes to decrease the quantity of salt that diffuses into the meat (Andrés et al. 2005). The use of alternative salts (e.g. KCl, MgCl₂, CaCl₂) has been associated to undesired sensory properties of the ripened products, occurrence of bitter taste and off-flavours (Armenteros et al. 2012). Moreover, this procedure is not allowed in typical Protected Designation of Origin

(PDO) and Protected Geographical Identity (PGI) products where sea salt is used as mandatory ingredient.

The change of processing conditions to limit the overall salt uptake resulted successful when the decrease of the salting time (Coutron-Gambotti et al. 1999) and/or of the salt quantity used in the salting step (Benedini et al. 2012) have been applied.

For many traditional dry ham products, the PDO and PGI designation of origin procedures define the salt content, or its range, based on the peculiar, traditional, but empirically developed, process conditions and the specific quality properties. In fact, beside salt content, processed meat products must comply with other quality indices (e.g. proteolysis index) that partly could depend on salt content, while pH and a_w are critical parameters for microbial growth according to Commission Regulations (EU) No. 2073/2005 and No. 852/2004.

In the last decade, to sort out consumers health and nutritional concerns, the reduction of salt content in traditional meat products has been undertaken but producers are facing main issues as they have to accomplish quality and stability indices according to the regulations and official procedures.

Dry cured hams are traditional meat products whose microbial stability is accomplished according to the well-known “hurdle technology” approach (Leistner 1992) by a combination of several actions (salt, water activity, pH, nitrate/nitrite when used) that at different level and intensity contribute to the control of the pathogens as well as alternative microorganism growth. The modification of processing and product parameters that could affect the efficacy of one factor could compromise the quality and stability of the final product and in dry-cured meats salt and a_w are the most important ones to be considered in this respect.

It is also worth to evidence that salt content of dry cured meat products is affected also by variables other than those related to processing (e.g. initial pH hams, size, fat-lean meat ratio) that need to be taken into account when a salt reduction has to be achieved.

This study is aimed at evaluating the feasibility of the modification of a conventional dry salting procedure to reduce the NaCl content of traditional dry-cured hams. Dry salting is generally carried out by an initial coverage of the fresh tights by salt and leaving them in contact with it for a period of time sufficient to allow a proper solute diffusion into the matrix. During this process step salt is renewed one or more times by hand-sprinkling onto the surface not covered by the skin and in particular onto the knob region, more prone to surface microbial spoilage, to allow a continuous uptake of the solute by the ham.

In this study, a two salt-coverage steps (initial salt coverage plus one additional renewal) salting procedure was applied to reduce the salt content of the final dry-cured hams and compared with a conventionally applied one based on a three steps

coverage (one initial followed by two following salt renewal), while keeping constant the overall salting time. The evaluation of its effects on quality properties and stability indices was carried out. The study was carried out starting from fresh tights that were initially grouped in two classes according to the initial weight, critical factor for mass transfer kinetics.

Material and methods

Dry-ham processing

Processing was carried out according to the production procedure of the dry-cured ham Prosciutto di Sauris PGI (Commission Regulation (EU) No. 320/2010) (Fig. 1) in a dry-cured ham factory (Prosciuttificio Wolf Sauris SpA, Sauris di sotto, Udine, Italy).

One hundred-ten green fresh tights (crossbreed of Landrace, Large White and Duroc) were selected and divided in two groups (55 hams/each) based on the initial weight as follows: small size (S): 13.5 ± 0.5 kg; large size (L): 15.0 ± 0.5 kg.

S- and L-batches were then further divided in two subgroups, each of 25 green hams, that separately underwent either the conventional (3 s-) or the modified (2 s-) salting procedure. The 3 s- salting process involves an initial complete coverage of the hams with dry salt carried out by a salting machine. This automated equipment conveys tights located on a steel belt through floating rollers enabling vein and artery purging and then to a second sector where they skin is rubbered with salt. At exit, tights are manually sprinkled with salt to assure complete coverage of specific critical areas (e.g. knob region-femur bone). Placed on shelves, salt-covered hams are then stored in a salting room at 3 ± 1 °C and 95 % RH for 19 days. During this time, at regular intervals, hams were manually sprinkled with tiny amounts of salt twice to keep them always covered by salt (in total three coverage salt steps).

The modified salting procedure (2 s- salting) was performed as the conventional one above described but only one additional salt coverage step was performed at the middle of the salting time (i.e. after 10 days).

At the end of salting, hams were cleaned of residual superficial not adsorbed salt by washing. After salting hams underwent to the same process conditions until the end of ripening (Fig. 1) as described in Martuscelli et al. (2009).

Sampling

Samples were taken at arrival of the green hams, end of salting (19 days), pre-resting (35 days), resting (90 days), middle ripening (180 days) and end of ripening (420 days).

Fig. 1 Flow-sheet of the dry cured ham process

At each sampling time five hams per size and salting condition were collected, deboned, cut on the cross section and two slices (thickness: 3 cm) were taken, according to Grau et al. (2008). After vacuum packaging, one slice was frozen and stored at -30°C until analysis; the second one was immediately analysed for mechanical analysis. Before analysis slices were equilibrated at 20°C for at least 2 h.

Analyses were carried out distinctly on *Semimembranosus* (SM) and *Biceps femoris* (BF) muscles. For hams taken after salting and resting steps, a sample representative of the external (e) and inner (i) SM muscle portions were analysed.

Chemical analysis

All reagents were provided by SIGMA (Bellefonte, USA). Distilled water was used throughout the study when required for analytical purposes.

Moisture content was determined by drying about 3 g of sample to constant weight, following the AOAC procedure (AOAC 2002). NaCl content was determined as chloride

concentration by Volhard titration according to AOAC official procedures (2002). Salt Index (S.I., %) was then computed according to the formula: $\text{S.I. (\%)} = \text{NaCl (\%)} * 100 / \text{moisture (\%)}$.

For the differently processed hams, the values of z^{NaCl} and X_{ff}^{w} indices were also calculated (Grau et al. 2008). Namely, z^{NaCl} index corresponds to the salt content as referred to the sum of the salt and water content (g salt/g salt + g water) while the X_{ff}^{w} corresponds to the moisture content as referred to the free fat dry matter (g water/g dry matter free of fat).

Fat content was determined by the Soxhlet method, using 40–60 petroleum ether (AOAC 2002).

Ashes were determined by the calcination of 2 g of sample in a furnace at $525 \pm 25^{\circ}\text{C}$ until a constant weight was reached (ISO 936 1998).

Total nitrogen content (TN, % w/w) was determined by the Kjeldahl method and proteins by multiplying $\text{TN} \times 6.25$ (AOAC 2002). Non-protein nitrogen content (NPN, % w/w) was measured by precipitation of proteins with trichloroacetic acid followed by determination of the nitrogen in the extract

by the Kjeldahl method. A Proteolysis Index (PI, %) was calculated as the ratio between NPN and TN ($P.I. \% = 100 \times NPN \times TN^{-1}$) (Careri et al. 1993).

Physico-chemical and physical properties analysis

Water activity (25 °C) was measured by a dew point hygrometer AquaLab CX 2 (Aqualab Scientific Pty Ltd., Castle Hill, NSW). The pH were determined with a pH penetration electrode on a pH-meter (3510 model, JENWAY, Bibby Scientific Limited, Staffordshire, UK).

Mechanical properties were evaluated at 20 °C on BF and SM muscles of half- and fully-ripened products (180 and 420 days) by an Instron dynamometer (mod. 5542-H5036, Instron International Limited, High Wycombe, UK) equipped with a Warner-Bratzler cell (Obuz et al. 2004) on samples of parallelepiped shape (10 × 10 × 15 mm) at a crosshead speed of 100 mm·min⁻¹; cutting force was applied perpendicularly to the muscle fibre length (Martuscelli et al. 2009). At least ten samples per batch were analysed. Maximum cutting force (N) and total energy (J) were obtained from the force vs. deformation graphs (Bourne 2002).

Volatile compounds analysis

Volatile compounds were determined by Solid Phase Microextraction GC-MS (SPME GC-MS) method. Aliquots of 2.5 g of BF and SM m. of half- and fully-ripened products, individually ground, were placed into 10 mL glass vials (Supelco Inc., Bellafonte, PA, USA), hermetically sealed. After equilibration for 45 min at 40 °C, the SPME fiber (2 cm-50/30 µm DVB/CAR/PDMS, Supelco, Bellafonte, PA, USA), pre-conditioned 10 min at 240 °C, was exposed to the sample vial headspace for 50 min at 40 °C by using a device for fibre housing (Supelco, Bellafonte, PA, USA).

A gas chromatograph (AutoSystem XL Gas Chromatograph; Perkin Elmer, Boston, MA, USA), coupled with a quadrupole mass spectrometer (Perkin Elmer TurboMass Mass Spectrometer), equipped with a fused-silica capillary column (60 m long, I.D. 0.32 mm, 1.0 µm film thickness; Supelco, Bellafonte, PA, USA) was used with ultra-high purity He as the carrier gas at 1 mL min⁻¹. The SPME fibre was desorbed at the injector at 240 °C for 1 min operating in a splitless mode. The analysis conditions were: initial oven temperature: 40 °C for 10 min, then increased to 200 °C at 5 °C/min and subsequently to 250 °C at 20 °C/min and held at that temperature for 10 min. The MS analysis conditions were: electronic ionization mode at an ionization voltage of 70 eV, m/z range of 20–250. Compounds identification was carried out by matching of mass spectral data with those contained in the mass spectral database (Perkin Elmer TurboMass Ver 4.1.1 software) and by comparison with pure standard compounds. Results were computed as relative

percentage area of each compound referred to the total chromatographic area.

Sensory analysis

Quantitative descriptive sensory analysis of the differently processed and ripened hams was carried out by a panel of 10 trained judges (aged between 22 and 53) (ISO 13299 2010). A preliminary training of the panel was carried out by testing samples of dry-cured hams at different ripening degree and salt content to acquire familiarity with descriptors and scale (Laureati et al. 2014). The analysis has been carried out in the sensory laboratory of the University of Teramo; judges received the samples in a randomized presentation order; two repetitions on different days for each sample were performed. Slices (thickness: 1 mm) were obtained by cross-cutting the deboned ham; two slices were posed on glass plates, covered with aluminium film and equilibrated for 15 min at 20 °C, before testing.

Panellists were asked to score sensory indices of appearance (colour intensity of meat and fat), aroma (olfactory intensity) taste and flavour (salted, acid, bitter, smoky, flavour intensity) and texture at mastication (chewiness, dryness) on a graduate scale (from 1–less - to 5–high).

Statistical analysis

All determinations were done in triplicate, except where differently indicated. Means and relative standard deviations were calculated.

One-way analysis of variance (ANOVA), with a confidence interval of 95 %, was run to evaluate the effect on the measured physical, chemical, physico-chemical parameters and sensory characteristics of ripened dry-cured hams of the four experimental batches (2s-L, 3s-L, 2s-S, 3s-S). Difference among means were separated by the least significant differences (LSD) test. Data were processed by using Statistica 8.0 (StatSoftTM, Tulsa, UK).

Principal Component Analysis (PCA) has been performed with the Single Value Decomposition (SVD) algorithm after data autoscaling, using MatLab R2009b (Mathworks, Natick, MA, USA).

Results and discussion

Green hams composition

Chemical composition and water activity of *Biceps femoris* (BF) and *Semimembranosus* (SM) muscles of the green hams of L- and S-weight class resulted not significantly different. Protein content ranged between 88.10 and 92.39 % on dry matter (d.m.) while pH values were between 5.50 and 5.64.

A higher variability of the fat content of BF *m.* was found in the L- and S- weight green hams, with values of 8.32 ± 4.40 % d.m. for the former and 5.13 ± 3.54 for the latter, respectively.

These data allowed to attribute the results only to the effect of the experimental variables (number of salt coverage steps; weight class) and to exclude that of the composition and physico-chemical properties of the green hams.

Effect on salt and moisture content

Weight measurements and mass transfer calculations during processing allow to determine the overall salt uptake of hams that, at the end of the salting period, resulted equal to 2.90 % (2s-S), 2.78 % (2s-L), 3.41 % (3s-S) and 3.14 % (3s-L) with statistical difference ($p < 0.05$) due to salting type and ham weight class.

During salting, osmotic phenomena occur at the ham surface that determine water loss and salt diffusion from the outer to the inner portions of the product. In Fig. 2, the change of the Salt Index (S.I.) % as a function of process time in BF and SM *m.* is shown. The different process conditions applied to hams affected water and salt mass transfer. In general, during the salting step, the NaCl concentration increases promptly and significantly in the SM *m.* due to its external position in contact with the coverage salt and the salt saturated solution at the

surface. Thereafter, during resting and ripening the S.I. in this muscle showed a progressive and almost linear increase but of a limited extent mainly due to water loss. At the end of ripening a different final S.I. value was achieved depending on the initial weight and salting process type. In particular, the 3s-S ripened samples due to the combined effect of partial drying and salt diffusion caused by the higher salt available onto the ham surface and corresponding higher osmotic potential, presented the highest S.I. % value.

The different salt concentration and water chemical potential between BF *m.* and the surrounding muscles determine a linear increasing trend of the S.I. in the former inner muscle until the middle of the ripening step (180 days) when it reaches the value of the SM *m.* and an equilibrium state among the two muscles seems to be achieved. Thereafter, the S.I. of BF *m.* showed an increase similar to that SM *m.* until the end of ripening.

After 180 days of ripening, S.I. % as well as moisture loss (data are not shown) in SM *m.* resulted statistically influenced by both number of salting steps and initial ham weight class ($p < 0.05$), while in BF *m.* the salt content was significantly affected ($p < 0.05$) only by salting steps.

The Salt Index (%) is a technical index generally referred to the BF *m.* and included in some product specification of protected designation products (e.g. for the PGI Prosciutto di

Fig. 2 Change of the Salt Index (S.I., %) in *Biceps femoris* (BF) and *Semimembranosus* (SM) muscles, as a function of process time (days, d) of hams produced from tights of different weight/size (S, L) and to

different salting procedure (2s-, 3s-). S-hams subjected to two (a) and three (b) steps of salting process; L-hams, subjected to two (c) and three (d) steps of salting process

Sauris official procedures -taken as reference for the dry-cured ham processing in this study- it has to be in the range from 7.2 to 11.2). Under the experimental conditions, it could be noticed that all the ripened products (420 d) were within the legal limits, even if a significant difference ($p < 0.05$) was observed between the values found in 2s-L (7.57 ± 0.61 %) and those in 3s-S (9.70 ± 0.48 %) hams.

To highlight the driving forces and the salt diffusion mechanisms and occurring in the ham muscle portions, in Table 1 the values of the z^{NaCl} and X^{w}_{ff} indices of the differently processed hams at the end of the salting, resting and ripening of SM *m.* and BF *m.*, are reported.

In general, after salting and resting the values of z^{NaCl} and X^{w}_{ff} of the SM *m.* are higher and lower, respectively, than those of the BF *m.* in agreement with their salt content. After the post-salting step, the BF z^{NaCl} values for the 2s-products resulted lower than those of the 3s-ones, independently on the weight. The products undergone to a reduced salting procedure showed the higher increase of z^{NaCl} in the post-salting step, being thus this process step determinant for the achievement of an equilibrium state in the water and salt transfer and the achievement of the final moisture and salt content that will affect the quality and stability of the ripened product.

On the contrary the 3s-S products presented at the same processing steps the lowest X^{w}_{ff} in respect to the other ones. This indicates that the salt concentration, and the corresponding thermodynamic conditions, allowed an early water transfer from the inner BF *m.* to the external SM *m.* that, in fact,

presented, quite unexpectedly a quite high X^{w}_{ff} at the beginning of the post-salting step.

In the case of the 2s-L products during the post-salting step a significant decrease of z^{NaCl} occurs in the SM *m.* due to the diffusive process induced by the different water potential and osmotic pressure in the muscle that depletes the salt penetrated during the modified 2-steps salting process and this may result a limiting critical factor in the hams of higher weight.

Effect on water activity

In dry-cured hams, water activity (a_w) is one of the main hurdles that in combination with other actions affect quality and stability by the control of the microbial growth and the rate of the desired biochemical reactions.

From a starting initial mean a_w value of 0.987 ± 0.003 , a progressive decrease during processing due to the combined effect of salt diffusion and water loss occurred (Table 1). The a_w decrease resulted higher in the external muscle portions (SMe vs SMi, SM vs BF) than in the inner ones with a trend similar to that reported by other authors (Blesa et al. 2008). At the end of ripening, weight class and salting process (2s- vs 3s-) significantly affected the a_w values of the SM and BF *m.* ($p < 0.01$), while no meaningful differences were observed in the water activity values of the two different muscle portions within hams of the same batch, due to the attainment of an equilibrium of the chemical potential of water (Albarracín et al. 2011).

Table 1 Values of z^{NaCl} , X^{w}_{ff} and a_w at the end of the salting, resting and ripening steps of *Biceps femoris m.* (BF) and *Semimebranosus m.* (external and internal part, SMe and SMi) of the differently processed hams

	Sample	Post-salting (19 days)			Post-resting (90 days)			End ripening (420 days)	
		BF	SMi	SMe	BF	SMi	SMe	BF	SM
z^{NaCl}	2s-S	0.006 ± 0.002^a	0.020 ± 0.008	0.043 ± 0.012	0.032 ± 0.002^b	0.048 ± 0.005^a	0.056 ± 0.008^{ab}	0.076 ± 0.008^{ab}	0.074 ± 0.004^a
	3s-S	0.010 ± 0.004^b	0.024 ± 0.007	0.055 ± 0.003	0.030 ± 0.001^{ab}	0.056 ± 0.006^a	0.059 ± 0.004^b	0.088 ± 0.004^c	0.084 ± 0.004^b
	2s-L	0.006 ± 0.003^a	0.019 ± 0.004	0.056 ± 0.005	0.029 ± 0.001^a	0.049 ± 0.005^a	0.051 ± 0.004^a	0.069 ± 0.005^a	0.074 ± 0.007^a
	3s-L	0.026 ± 0.002^c	0.027 ± 0.009	0.053 ± 0.011	0.036 ± 0.001^c	0.069 ± 0.005^b	0.072 ± 0.006^c	0.080 ± 0.006^{bc}	0.076 ± 0.003^{ab}
	sign.	**	n.s.	n.s.	*	**	**	**	*
X^{w}_{ff}	2s-S	3.083 ± 0.065	2.889 ± 0.080	2.271 ± 0.158^a	2.703 ± 0.265	2.661 ± 0.050^b	2.176 ± 0.124	1.755 ± 0.084^a	1.381 ± 0.079^{bc}
	3s-S	2.987 ± 0.046	2.861 ± 0.188	2.445 ± 0.073^b	2.838 ± 0.137	2.380 ± 0.144^a	2.189 ± 0.173	1.734 ± 0.035^a	1.271 ± 0.041^{ab}
	2s-L	3.172 ± 0.064	2.780 ± 0.169	2.150 ± 0.096^a	2.772 ± 0.021	2.267 ± 0.035^a	2.089 ± 0.070	1.885 ± 0.041^b	1.423 ± 0.131^c
	3s-L	3.055 ± 0.211	2.815 ± 0.153	2.268 ± 0.159^b	2.867 ± 0.051	2.426 ± 0.114^a	2.139 ± 0.048	1.847 ± 0.029^b	1.268 ± 0.074^a
	sign.	n.s.	n.s.	*	n.s.	**	n.s.	**	**
$a_w^{\S}$	2s-S	0.981 ± 0.004	0.976 ± 0.008	0.965 ± 0.008^b	0.971 ± 0.006	0.960 ± 0.001^{bc}	0.956 ± 0.009^{ab}	0.933 ± 0.003^c	0.930 ± 0.009^b
	3s-S	0.982 ± 0.002	0.976 ± 0.004	0.960 ± 0.003^b	0.965 ± 0.001	0.950 ± 0.001^a	0.949 ± 0.005^a	0.914 ± 0.007^a	0.911 ± 0.006^a
	2s-L	0.985 ± 0.008	0.976 ± 0.005	0.953 ± 0.005^a	0.975 ± 0.001	0.961 ± 0.003^c	0.959 ± 0.001^b	0.924 ± 0.004^b	0.916 ± 0.003^a
	3s-L	0.979 ± 0.006	0.972 ± 0.007	0.957 ± 0.005^{ab}	0.970 ± 0.009	0.954 ± 0.005^{ab}	0.949 ± 0.007^a	0.931 ± 0.008^{bc}	0.928 ± 0.001^b
	sign.	n.s.	n.s.	*	n.s.	**	*	**	**

[§] Values with different small letters along column results significantly different (LSD test, $p < 0.05$)

sign. significance level: *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$; n.s. no significance

Only the a_w value of the ripened 3s-S product has a direct relation with the corresponding salt content. The water state of a complex food system, besides moisture and salt content, depends also on presence and concentration of other solutes (e.g. peptides, free aminoacids) with water binding ability in the matrix whose formation and presence could have been differently occurred due to the salting processes.

The a_w values of the ripened products of this study are higher than those reported in a previous study performed on PGI Sauris dry-hams subjected to a conventional 3-steps salting process (Martuscelli et al. 2009); this disagreement has to be related to the different initial weight of the fresh tights, being lower in those used in the precedent investigation.

It should be noted that BF *m.* of the 2s-S and 3s-L ripened hams presented a_w of ca. 0.93, higher than the limit values recommended and/or critical for the microbial stability (Blesa et al. 2008), trade (e.g. commerce outside EU, dry-cured ham a_w value minimum: 0.92), as well as safety. The microbiological criteria for the official control of *Listeria monocytogenes* and *Salmonella*, indicates that foods considered non permissive for the growth of *L. monocytogenes* are those with pH <4.4 or $a_w \leq 0.92$, while for *Salmonella enterica* subsp. *enterica* pH ≤ 3.7 and $a_w \leq 0.94$.

Prencipe et al. (2012) reported the significant reduction of the contamination levels of *L. monocytogenes* during curing and ripening of Parma ham and attributed to the a_w decrease at the ham surface as also occurred in the hams under study (Table 1, see SMe data). Reynolds et al. (2001) observed that generally dry-cured ham manufacturing process did not permit the growth of pathogens such as *L. monocytogenes* and *Salmonella* due to the interactive effect of salt concentration (8 %), pH value (5.5) and a_w (0.92); similarly, other studies evidenced a progressively decline of *Listeria*, *Salmonella* and *E. coli* (Portocarrero et al. 2002) during ham processing. The effect of the modification of the salting procedure on microbial growth and state was out of the scope in this study. However, the sensory analysis carried out on the ripened hams did not detect defective products due to alterative microorganisms and/or degradation enzymatic activities.

The growth of bacteria in dry cured meat products may be controlled by the interactive effect of environmental and nutritional factors including, besides a_w , NaCl content, pH and moisture to protein ratio (Portocarrero et al. 2002) also in agreement with the well-known “hurdle technology” approach (Leistner 1992). The use of NaCl as ingredient in foods is aimed at its effective antimicrobial and preservative effects that involves more than only a dehydrating capacity (Taormina 2010).

According to the conventional process hams underwent to a light smoking step of ca 48–60 h that could represent an additional action towards the overall product stability (Pittia et al. 2005; Portocarrero et al. 2002). Smoke components such as formaldehyde, acetic acid, creosote and phenols may

exhibit bactericidal properties (Daun 1979), thereby contributing to inhibit development of spoilage microorganisms.

Effect on quality properties

In Table 2, proximate composition, pH values and mechanical properties data of the differently processed and ripened hams are reported. The pH values ranged from 5.71 to 5.87 with no significant differences due to salting procedure and class weight. Moisture content was affected by the salting process only for the small-size hams and in the case of the BF *m.*

The modification of the salting process allowed to reduce salt content of significant amounts and between 11 % (SM *m.*) and 13 % (BF *m.*) in respect to that determined in the dry-cured hams obtained by the conventional salting procedure (3s).

The proteolysis index (P.I., %) of the BF and SM *m.* of hams after 180 and 420 processing days are reported in Fig. 3. The main protein breakdown in dry cured hams occurs during the initial steps of the process (salting, resting and drying) when salt content, free water availability and process conditions (e.g. temperature) allow a more intense enzymatic activity.

At middle ripening, all the P.I. of the hams were not significantly different. At the end of ripening, a significantly lower proteolysis index value in the SM *m.* in comparison with that in the earlier ripening step has been evidenced in all the samples (Fig. 3b). On the contrary, in the BF *m.* (Fig. 3a) similar P.I. decrease has been evidenced within the same ripening time only for the smaller ones (S) independently from the salting procedure.

The decrease of the non-protein nitrogen (NPN) during dry cured ham processing, and in particular during the ripening step, has been already observed (Sárraga et al. 1989; Virgili et al. 1995) and attributed to a main inhibitory effect of the salt content on proteolytic activity. During ripening, endogenous enzymes cause the formation of peptides and aminoacids and an increase of the NPN is expected. In parallel, other chemical and biochemical reactions may occur that use the degradation protein products as substrates to produce volatiles compounds and Strecker aldehydes. The decrease of the proteolysis index may be caused by a rate of degradation of the proteins and peptides higher than that of their formation (Zhao et al. 2008). Moreover, free aminoacids can also react with some components of the smoke adsorbed during the smoking process via Maillard-like reaction (Ruiter 1979). In the ham, the SM muscle, is characterised by a higher salt content and lower a_w than BF *m.* that could inhibit proteolysis to a higher extent. Moreover, its external position makes it more able to adsorb compounds during the smoking process that could then react with the aminoacids during ripening. These factors could, thus, synergistically contribute to decrease the NPN content.

Table 2 Compositive, physico-chemical and physical parameters in *Biceps femoris* (a) and *Semimembranosus* (b) muscles of ripened Sauris hams (420 d), obtained from small (S) and large (L) size green hams, subjected to two (2s-) and three (3s-) steps of salting process

	2 s-L	2 s-S	3 s-L	3 s-S	F
a) <i>Biceps femoris</i> m.					
Moisture (%)	62.74 ± 0.51 ^b	62.36 ± 0.73 ^b	62.48 ± 0.68 ^b	61.30 ± 0.19 ^a	6.16**
Total proteins (%)	27.47 ± 0.79 ^a	30.77 ± 1.07 ^b	27.01 ± 0.79 ^a	27.66 ± 0.42 ^a	23.06**
Ashes (%)	5.83 ± 0.17 ^a	6.10 ± 0.56 ^b	6.83 ± 0.38 ^c	7.71 ± 0.37 ^d	22.53**
Total fat (%)	3.97 ± 0.85 ^b	2.16 ± 0.01 ^a	3.68 ± 1.33 ^b	3.33 ± 0.73 ^b	4.11*
NaCl (%)	4.68 ± 0.38 ^a	5.15 ± 0.53 ^b	5.44 ± 0.40 ^b	5.93 ± 0.29 ^c	8.22**
pH	5.84 ± 0.06	5.71 ± 0.25	5.85 ± 0.05	5.85 ± 0.05	n.s.
Maximum shear force (N)	120.92 ± 10.28	106.77 ± 22.99	104.87 ± 23.12	107.58 ± 32.77	n.s.
Maximum shear work (mJ)	920 ± 134	1088 ± 306	1352 ± 292	1412 ± 586	n.s.
b) <i>Semimembranosus</i> m.					
Moisture (%)	56.78 ± 1.89	55.91 ± 1.51	54.73 ± 2.26	54.88 ± 1.48	n.s.
Total proteins (%)	34.36 ± 1.94 ^a	34.97 ± 1.74 ^a	37.23 ± 0.86 ^b	35.99 ± 0.70 ^a	3.91*
Ashes (%)	5.3 ± 0.29 ^a	5.59 ± 0.42 ^{ab}	6.00 ± 0.33 ^b	6.63 ± 0.27 ^c	15.04**
Total fat (%)	4.21 ± 2.35	3.53 ± 1.63	2.45 ± 1.09	2.91 ± 0.54	n.s.
NaCl (%)	4.55 ± 0.40 ^a	4.49 ± 0.28 ^a	4.51 ± 0.23 ^a	5.04 ± 0.22 ^b	4.07*
pH	5.75 ± 0.07	5.79 ± 0.05	5.87 ± 0.07	5.77 ± 0.10	n.s.

F calculated Fisher's F, * Significant for $p < 0.05$, ** significant for $p < 0.01$; *** significant for $p < 0.001$; data followed by different letters in the same row are significantly different (LSD test, $p < 0.05$); n.s. not significant

As regards the mechanical properties, hardness and toughness of the SM and BF mm. of the ripened dry-cured hams resulted not significantly affected neither by the initial weight nor by salting procedure mainly due to the high variability of the data (data are not shown). However, at the same ripening time, 2 s-L ham showed the higher maximum shear force and work in respect to the other products. The mechanical properties of dry-cured hams are the result of the combined effect of myofibrillar protein cleavage, which causes the reduction of the hardness and cohesiveness, and dehydration that, conversely, tends to increase the product consistence. Moreover,

salt content and other compositional factors (e.g. lipid content) may be involved and differently affect the textural properties of the ripened dry-cured ham.

Aroma volatile compounds composition was determined on half- and fully-ripened hams. During processing chemical reactions and enzyme activity are involved in the generation of volatile compounds affecting the sensory properties of hams. In all samples the presence of volatile compounds typical of dry-cured hams and of the smoking process were determined (Wittkowski et al. 1990; Barbieri et al. 1992; García-Gonzalez et al. 2008; Pinna et al. 2013). In particular, 34

Fig. 3 Proteolysis index (P.I., %) on *Biceps femoris* (a) and *Semimembranosus* (b) m. of hams at 180 and 420 days. Legend of figure: samples from small (S) and large (L) size thighs subjected to two (2s) and three (3s) steps of salting process

volatile compounds including alcohols (n. 10), aldehydes (n. 4), esters (n. 3), ketones (n. 6), aliphatic hydrocarbons (n. 7), aromatic hydrocarbons (n. 2), sulfates compounds (n. 1) and furans (n. 1), have been detected and identified.

The main volatiles compounds derived from: a) lipid oxidation (hexanal, heptanal, 1-pentanol, 1-hexanol, 1-octen-3-ol, hexane, heptane, nonane, and 2-pentanone); b) aminoacids degradation via Strecker degradation and other chemical pathways (3-methyl- 2-butanol, 3 methyl butanol, 3-methylbutanal); c) endogenous and exogenous (microbial) enzymatic activity (i.e. ethanol, 1,3 octanol); d) smoke that could diffuse into the product (2-propanone, ethyl benzene, 1,2 dimethylbenzene).

Significant differences in the pattern of the volatile aroma compounds of the dry-cured hams due to ripening time (180 days vs. 420 days) and muscle type were found in agreement with other studies (Ruiz et al. 1998; Sánchez-Peña et al. 2005) (*data not shown*). Salt content differently affected the evolution of the lipid oxidation of the differently processed ripened hams and the formation of the corresponding secondary oxidation products. In particular, some aldehydes such as hexanal, were found in a lower concentration in the ripened products if compared with that in the same products after 180 days of process and this occurred in both muscles (BF and SM). At the same processing step, a higher relative content of hexanal has been determined in general in the BF m. of all hams and, in particular, in those undergone to a 3-s salting steps (and thus, with a higher salt content) by confirming the

role of salt as pro-oxidant factor favouring lipid auto-oxidation.

Sensory quality characteristics

Results of the descriptive sensory analysis are presented in Fig. 4 where the spider profiles of the 2s- and 3s- products of the S- (4a) and L- (4b) weight class-hams are shown.

Limited differences on the main descriptors were perceived between the 2s-L and 3s-L hams (Fig. 4b); 2s-L hams resulted less salty and acid ($p < 0.05$), in agreement with their composition and ripening indices (e.g. salt and moisture content, P.I.) (Table 2, Fig. 3). Moreover, panellists attributed to 2s-L hams a lower overall aroma score (2.6 ± 0.1) than 3s-L ones (3.0 ± 0.3). Salt content, being positively associated with some volatile compounds generated by oxidation compounds (salt-ing-out effect), may enhance aged-ham aroma perception (Pinna et al. 2013).

When S-hams are considered, the 2s- products resulted less easy to chew, less salty and with a lower intensity of the smoky flavour than the 3s- ones. The same products showed a more intense red colour and white colour of the coverage fat in disagreement with other studies that did not find significant effects on colour attributes of hams with a lower salt content (Andrés et al. 2005; Costa-Corredor et al. 2009). In dry cured hams, where salt is used as sole preservative agent, the stable red-colour and correspondent redness is due to the formation of a lipophylic molecule (Zn-protoporphyrin IX), whose

Fig. 4 Spider diagram of the sensory evaluation results of ripened hams obtained by the traditional (3s-) and modified (2s-) salting procedures (scale: 1 to 5). **a** products obtained from initial lower weigh hams (S); **b** products obtained from initial higher weight hams (L)

development seems not be related to the salt content (Adamsen et al. 2006). In this study hams, however, underwent to a smoking step and other reactions could have been occurred between myoglobin and some smoke compounds affecting the colour of the ripened product (Ruiter 1979).

Chewiness results influenced by salt concentration, with 2s-S ripened hams exhibiting the least average scores (2.0 ± 0.01) in agreement with Benedini et al. (2012). Regardless the chemical composition and the proteolysis degree of the product, this result could be also associated to the higher a_w of this product in respect to the 3s-S (Table 2) (Serra et al. 2005) and the related higher activity of the enzymes affecting the protein matrix (Fig. 3).

Statistical analysis

Statistical analysis has been carried out to determine significant correlations between quality attributes and chemical and

physico-chemical properties of differently processed dry-cured hams.

The data of all quality parameters (total: 66 variables) of the ripened hams were processed by Principal Component Analysis (PCA). In Fig. 5 the scores (5a, 5b) and loadings (5c, 5d) plot of the samples are shown. The PCA explained four interpretable components that describe about 60 % of the total variability), allowing the discrimination of the products based on muscle, green ham weight and number of salt coverage steps (Fig. 5a, b).

The first component (PC 1, Fig. 5c), was positively associated with salt and ashes content but negatively with sensory attribute (redness intensity), volatile compounds (2-heptanone, 1-pentanol) and protein content, allowing to discriminate products of small size (s) as a function of muscle type and salting procedure. The second component (PC 2, Fig. 5c) was positively associated with P.I., moisture and presence of some aromas (nonanone and 2-heptanone), while negatively associated with 3-ethyl 2,2-dimethylpentane and

Fig. 5 Plot of the Principal Component Analysis (PCA) of chemical, physical, physico-chemical and sensory data of *Biceps femoris* (BF) and *Semimembranosus* (SM) muscles taken from ripened hams (420 days) of initial different weight (L, S) and differently salted (2s-, 3s-)

protein content, thereby discriminating SM-3s from the other samples. PC 3 (Fig. 5b, d) results negatively associated with sensory descriptors (white intensity of subcutaneous fat, acid taste) and a_w and positively with chewiness, ethylbenzene and smoky perception. The fourth component (PC 4) presents negative loadings for some sensory attributes (bitter taste, adhesion to the palate-dryness) and positive for overall aroma intensity, some volatile compounds and meat colour intensity allowing to discriminate L-size ripened hams from the S-ones. It should be noted that hams processed with reduced salting steps (2s-) resulted associated with scarce presence of hydrocarbons and ketones and high salt content. The optimal development of the ripening process influences the oxidation and degradation reactions limiting the formation of undesirable volatile compounds (short-chain hydrocarbons, aldehydes, ketones, acids and furans) (Ruiz et al. 2005); on the contrary, the alcohols, products of oxidative decomposition of lipids, are considered suppliers of floral notes and, finally, the esters can be correlate to the typical flavour of ripened meat (Barbieri et al. 1992).

Conclusion

This study confirmed the feasibility of a decrease of the salt content in traditional dry-hams by a modification of the salting process based on the decrease of the salt-coverage steps number. The achieved reduced salt content (11–13 %) determined limited changes in the quality properties of the ripened products with impact also on some sensory properties (like aroma and texture).

However, the weight of the green hams resulted a critical process factor to be considered to reach the limiting values of the parameters related to safety and microbial stability (a_w). When values above the minimum official and/or recommended limits (like 2s-S and 3s-L hams) occur, a process optimisation has to be developed by the adoption of proper actions and change of technological parameters (e.g. longer ripening time, increased ripening temperature).

Acknowledgments Authors are grateful to Prosciuttificio Wolf Sauris Spa (Sauris, Italy) for financial support and quality control during the dry ham processing as well as the ham samples provided for this study.

References

- Adamsen CE, Møller JKS, Laursen K, Olsen K, Skibsted LH (2006) Zn-porphyrin formation in cured meat products: effect of added salt and nitrite. *Meat Sci* 72:672–679
- Albarracín W, Sánchez IC, Grau R, Barat JM (2011) Salt in food processing: usage and reduction: a review. *Int J Food Sci Technol* 46:1329–1336
- Andrés AI, Ventanas S, Ventanas J, Cava R, Ruiz J (2005) Physicochemical changes throughout the ripening of dry cured hams with different salt content and processing conditions. *Eur Food Res Technol* 221:30–35
- AOAC (2002) Official methods of analysis, 17th edn. Association of Official Analytical Chemists, Gaithersburg
- Armenteros M, Aristoy MC, Barat JM, Toldrá F (2012) Biochemical and sensory changes in dry-cured ham salted with partial replacements of NaCl by other chloride salts. *Meat Sci* 90:361–367
- Barbieri G, Bolzoni L, Parolari G, Virgili R, Buttin R, Careri M, Mangia A (1992) Flavour compounds of dry-cured ham. *J Agric Food Chem* 40:2389–2394
- Benedini R, Raja V, Parolari G (2008) Zinc-protoporphyrin IX promoting activity in pork muscle. *Lebensm-Wiss u-Technol Food Science Technol* 41:1160–1166
- Benedini R, Parolari G, Toscani T, Virgili R (2012) Sensory and texture properties of Italian typical dry-cured hams as related to maturation time and salt content. *Meat Sci* 90:431–437
- Blesa E, Aliño M, Barat JM, Grau R, Toldrá F, Pagán MJ (2008) Microbiology and physico-chemical changes of dry-cured ham during the post-salting stage as affected by partial replacement of NaCl by other salts. *Meat Sci* 78:135–142
- Bourne MC (2002) Food texture and viscosity. Concept and Measurement. Academic Press, New York
- Careri M, Mangia A, Barbieri G, Bolzoni L, Virgili R, Parolari G (1993) Sensory property relationships to chemical data of Italian-type dry-cured ham. *J Food Sci* 58(5):968–972
- Commission Regulation (EU) No 320/2010 of 19 April 2010 entering a name in the register of protected designation of origin and protected geographical indications (Prosciutto di Sauris (PGI)). *Off J Eur U L* 98/1
- Costa-Corredor A, Serra X, Amau J, Gou P (2009) Reduction of NaCl content in restructured dry-cured hams: post-resting temperature and drying level effects on physicochemical and sensory parameters. *Meat Sci* 83:390–397
- Coutron-Gambotti G, Gandemer S, Rousset O, Maestrini F, Casabianca (1999) Reducing salt content of dry-cured ham: effect on lipid composition and sensory attributes. *Food Chem*, 64: 13–19.
- Daun H (1979) Interaction of wood smoke components and foods. *Food Technol* 33(66–71):83
- Desmond E (2006) Reducing salt: a challenge for the meat industry. *Meat Sci* 74:188–196
- García-Gonzalez DL, Tena N, Aparicio-Ruiz R, Morales MT (2008) Relationship between sensory attributes and volatiles compounds qualifying dry-cured hams. *Meat Sci* 80:315–325
- Grau R, Albarracín W, Toldrá F, Antequera T, Barat JM (2008) Study of salting and post-salting stages of fresh and thawed Iberian hams. *Meat Sci* 79:677–682
- ISO 13299 (2010) Sensory analysis—methodology—general guidance to establish a sensory profile. International Organization for Standardization, Geneva
- ISO 936 (1998) Meat and meat products. Determination of Total Ash (Reference method) Geneva, Switzerland: International Organization for Standardization
- Laureati M, Buratti S, Giovanelli G, Corazzin M, Lo Fiego D, Pagliarini E (2014) Characterization and differentiation of Italian Parma, San Daniele and Toscano dry-cured hams: a multi-disciplinary approach. *Meat Sci* 96:288–294
- Leistner L (1992) Food preservation by combined methods. *Food Res Int* 25:151–158
- Martuscelli M, Pittia P, Casamassima LM, Manetta AC, Lupieri L, Neri L (2009) Effect of intensity of smoking treatment on the free amino acids and biogenic amines occurrence in dry cured ham. *Food Chem* 116:955–962
- Obuz E, Dikeman ME, Grobbel JP, Stephens JW, Loughin TM (2004) *Beef longissimus lumborum, biceps femoris, and deep pectoralis*

- Warner–Bratzler shear force is affected differently by endpoint temperature, cooking method, and USDA quality grade. *Meat Sci* 68: 243–248
- Pinna A, Simoncini N, Toscani T, Virgili R (2013) Volatile compounds of Parma dry-cured ham as markers of ageing time and aged ham aroma. *Int J Food Sci* 24:321–331
- Pittia P, Martuscelli M, Munari M, Chavez-Lopez C (2005) Effect of smoking on quality and microbial characteristics of dry cured ham. *Industrie Alimentari* 44:1235–1243
- Portocarrero SM, Newman M, Mikel B (2002) Reduction of *Listeria monocytogenes*, *Salmonella* spp., and *Escherichia coli* O157:H7 during processing of country-cured hams. *J Food Sci* 67:1892–1898
- Prencipe VA, Rizzi V, Acciari V, et al (2012) *Listeria monocytogenes* prevalence, contamination levels and strains characterization throughout the Parma ham processing chain. *Food Control* 25: 150–158
- Reynolds AE, Harrison MA, Rose-Morrow R, Lyon CE (2001) Validation of dry cured ham process for control of pathogens. *J Food Sci* 66:1373–1379
- Ruiter A (1979) Color of smoked foods. *Food Technol* 33:54–63
- Ruiz J, Cava R, Ventanas J, Jensen MT (1998) Headspace solid phase microextraction for the analysis of volatiles in a meat product: dry-cured Iberian ham. *J Agric Food Chem* 46:4688–4694
- Ruiz-Ramirez J, Arnau J, Serra X, Gou P (2006) Effect of pH 24, NaCl content and proteolysis index on the relationship between water content and texture parameters in biceps femoris and semimembranosus muscles in dry-cured ham. *Meat Sci* 72: 185–194
- Sánchez-Peña CM, Luna G, Aparicio R (2005) Characterization of French and Spanish dry-cured hams: influence of the volatiles from the muscles and the subcutaneous fat quantified by SPME-GC. *Meat Sci* 69:635–645
- Sárraga C, Gil M, Arnau J, Monfort J, Cussò R (1989) Effect of curing salt and phosphate on the activity of porcine muscle proteases. *Meat Sci* 25:241–247
- Serra X, Ruiz-Ramírez J, Arnau J, Gou P (2005) Texture parameters of dry-cured ham m. biceps femoris samples dried at different levels as a function of water activity and water content. *Meat Sci* 69:249–254
- Taormina PJ (2010) Implications of salt and sodium reduction on microbial food safety. *Crit Rev Food Sci Nutr* 50:209–227
- Toldrá F (2006) The role of muscle enzymes in dry-cured meat products with different drying conditions. *Trends Food Sci Technol* 17:164–168
- Virgili R, Parolari G, Schivazappa C, Soresi Bordini C, Borri M (1995) Sensory and texture quality of dry-cured ham as affected by endogenous cathepsin B activity and muscle composition. *J Food Sci* 60: 1183–1186
- Wittkowski R, Baltes W, Jennings WG (1990) Analysis of liquid smoke and smoke meat volatiles by head space chromatography. *Food Chem* 37:135–144
- Zhao GM, Tian W, Liu YX, Zhou GH, Xu XL, Li MY (2008) Proteolysis in *biceps femoris* during jinhua ham processing. *Meat Sci* 79:39–45