**Effect of production process and high-pressure processing on viability of *Listeria innocua* in traditional Italian dry-cured *coppa***

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**Abstract**

In this study the effect of the application of High Pressure Treatment (HPP) combined with four different manufacturing processes on the inactivation of *Listeria innocua*, used as a surrogate for *L. monocytogenes*, in artificially contaminated *coppa* samples was evaluated in order to verify the most suitable strategy to meet the *Listeria* inactivation requirements needed for the exportation of dry-cured meat in the U.S. Fresh anatomical cuts intended for *coppa* production were supplied by four different delicatessen factories located in Northern Italy. Raw meat underwent experimental contamination with *Listeria innocua* using a mixture of 5 strains. Surface contamination of the fresh anatomical cuts was carried out by immersion into inoculum containing *Listeria* spp. The conditions of the HPP treatment were: pressure 593 MPa, time 290 seconds, water treatment temperature 14°C. *Listeria innocua* was enumerated on surface and deep samples post contamination, resting, ripening and HPP treatment. The results of this study show how the reduction of the microbial load on *coppa* during the production process did not vary among three companies (P>0.05) ranging from 3.73 to 4.30 log CFU/g, while it was significantly different (P<0.01) for the fourth company (0.92 log CFU/g). HPP treatment resulted in a significant (P<0.01) deep decrease of *L. innocua* count with values ranging between 1.63-3.54 log CFU/g with no significant differences between companies. Regarding superficial contamination, HPP treatment resulted significant (P<0.01) only in *coppa* produced by two companies. The results highlight that there were processes less effective to inhibit the pathogen; in particular for company D an increase of *L. innocua* count was shown during processing and HPP alone cannot be able to in reaching the *Listeria* inactivation requirements needed for exportation of dry-cured meat in the U.S. According to the data reported in this paper, HPP treatment increases the ability of the manufacturing process of *coppa* in reducing *Listeria* count with the objective of a lethality treatment.

**Introduction**

*L. monocytogenes* is a foodborne pathogen that causes listeriosis, a relatively rare but potentially fatal illness. Healthy individuals are usually not susceptible to *L. monocytogenes*, unlike those with compromised immune system such as elderly, pregnant women and newborns, in which mild to severe consequences can be observed (Jordan and McAuliffe, 2018). In 2018 there were 2,549 confirmed human cases of listeriosis in the EU, corresponding to a notification rate of 0.47 cases per 100,000 population. In the last years (period 2014–2018) there has been a statistically significant increasing trend of confirmed listeriosis cases in the EU/EEA, with a high case fatality (15.6%), which makes listeriosis one of the most serious foodborne diseases under EU surveillance (EFSA and ECDC, 2019). In the United States it has an incidence of 0.3 cases per 100,000 population (Tack et al., 2019).

*L. monocytogenes*, being ubiquitous in the environment, can contaminate food-processing plants, survive for long periods due to its capability to resist to various stresses, such as exposure to sanitizers, pH and temperature, and form biofilm. It mainly represents a problem for the Ready-To-Eat (RTE) food industry, as there is no lethal treatment in the EU (Zanardi et al., 2000; Busconi et al., 2014). The production process includes, after deboning, half-slicing, and trimming the anatomical cut, one or two salting procedures, using a mixture of salt, additives, and spices. Meat is massaged manually or mechanically, by a meat tumbling machine, in order to distribute the salting mixture evenly. Salting is generally followed by storage at low temperatures for a few days on steel trays (cold rest). After cold resting the meat is wrapped and tied and then exposed to higher temperatures and lower relative humidity, in order to reduce moisture. The last step consists in ripening.
which takes several weeks, at a lower temperature and higher relative humidity than drying, to reach the desired characteristics of the product. The production processes applied in this work are summarized in Table 1.

Contamination of dry-cured meat products by *L. monocytogenes* may result from superficial contamination of the fresh anatomical cuts, both during slaughtering and production and/or from cross-contamination in case of manipulation by contaminated operators or contact with contaminated equipment or surfaces. The current EU regulation for *L. monocytogenes*, Regulation (EC) No. 2073/2005 (European Commission, 2005), admits different levels of presence depending on whether or not the RTE product supports the growth of *L. monocytogenes* (products with pH ≤ 4.4 or *a*<sub>w</sub> ≤ 0.92, products with pH ≤ 5.0 and *a*<sub>w</sub> ≤ 0.94 and products with a shelf-life of less than five days are considered non supporting its growth). In products supporting its growth EU regulation requires the absence (in 5x25g samples) before the food has left the immediate control of the food business operator, and levels <100 CFU/g (in 5x25g samples) for products already placed on the market during their shelf life; in RTE samples, in order to verify the ability of the combined processes to meet the requirements needed for exportation to the U.S.

In this study it was evaluated the effect of the application of HPP combined with different manufacturing processes on the inactivation of *Listeria innocua*, used as a surrogate for *L. monocytogenes* (Hu and Gurtler, 2017), in artificially contaminated *coppa* samples, in order to verify the ability of the combined processes to meet the requirements needed for exportation to the U.S.

**Materials and Methods**

**Inoculum composition**

The *L. innocua* inoculum culture was prepared using a mixture of 5 strains: IZSLER 111373/1 and IZSLER 111373/2 isolated from industrial site (superficial swab collected in pork meat transformation plant), IZSLER 257529/1 isolated from fresh pork sausages, IZSLER 257529/2 isolates from fresh swine meat and the reference strain ATCC 33090, 100 µL of a stock culture (stored in 20% glycerol at -80°C) of each strain were transferred to 10 ml Brain Heart Infusion (BHI) broth and incubated for 24 h at 30°C. Subsequently, an aliquot of 100 µl was transferred to 1000 mL BHI broth and incubated at 12°C for 72 h (Merialdi et al., 2015).

Just before contamination, the 5 subcultures of *L. innocua* were mixed in equal volume and the resulting culture was checked by enumeration on selective agar.

**Samples contamination and production process**

Fresh anatomical cuts intended for Coppa production were supplied by four different artisanal delicatessen factories located in Northern Italy herein named A, B, C and D. Raw meats, with a weight ranging from 2.5 to 3 kg, underwent experimental contaminations with *L. innocua*. The anatomical cuts were contaminated by immersion for 10 minutes into the inoculum containing *L. innocua*, then dried for 30 minutes at room temperature.

The detailed processing procedures applied are detailed in Table 1. The protocols included one (company A and D) or two (company B and C) salting procedures with salting mixtures being supplied by the four companies. In all the protocols, meat samples underwent one or more steps in meat tumbling machine in order to get a homogenous distribution of the salting mixture. *Coppa* samples were processed according to the producer’s specifications (Table 1) undergoing a resting phase (9 to 32 days at 1-8°C), a drying phase (3 to 7 days at 12-27°C) and a ripening phase (44 to 69 days at 14-21°C).

After salting, *coppa* samples were singularly packed in synthetic casing and, at the end of the maturation period, they were separately transferred to nylon-polyethylene bags and vacuum sealed.

**HPP treatment**

For each contamination study, 5 vacuum-packed Coppa samples were exposed to HPP treatment and 5 samples acted as control. For the HPP treatment the following settings were applied: pressure 593 MPa, time 290 seconds, water treatment temperature 14°C, product temperature at the time of treatment 4°C. The pressure holding treatment time in this study did not include the pressure increase time or the decompression time. The water temperature during the process started from 14°C, grew until 32°C during the treatment, and

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**Table 1. Experimental scheme including the number of analyzed test units for each processing step, sampling characteristics and scheduled analyses.**

|                      | Company A | Company B | Company C | Company D |
|----------------------|-----------|-----------|-----------|-----------|
| Anatomic cut weight (Kg) | 2.7       | 2.5       | 3         | 2.5/3     |
| Number of salting    | 1         | 2         | 2         | 1         |
| Resting length (days)/temperatures | 14/3-5°C  | 32/3-5°C  | 27/1-4°C  | 9/6-8°C   |
| Drying length (days)/temperatures | 5/20°C    | 7/27°C to 14°C | 6/22°C to 16°C | 3/12-27°C |
| Ripening length (days)/temperatures | 51/15°C   | 40/14-18°C + 24/17-21°C | 69/14-16°C | 44/14°C to 16°C |
Sampling procedure
After the inoculation with *L. innocua* strains, for each challenge test, a total of 3 samples for each sampling time, inoculum (T0), post resting (T1), and a total of 5 samples post ripening (Tend) and post HPP treatment (THPP) were collected and analyzed for the determination of pH and *a*<sub>w</sub> values and the evaluation of *L. innocua* count. The analyses carried out for this study were made both on the surface and in depth of coppa samples. Superficial samples consisted of three squares of approximately 3x3 cm length and about 0.3 cm thickness to get a final weight of 25 g, excided from apical, central and terminal positions of each coppa. For deep samples, coppa was immersed for 60 seconds in boiling water, then a 25 g sample was extracted from the depth of coppa (Bonilauri et al., 2004).

Physicochemical analysis
Water activity (*a*<sub>w</sub>) was measured with AquaLab series 4 Model TE instrument, in accordance with ISO 21807:2004 (ISO, 2004). pH was evaluated through Mettler Toledo LE427 glass electrode probe connected to pHenomenal PCS000 L (VWR) pH/conductivity meter. Weight loss values (expressed as percentage of the initial weight) were determined throughout the production process, on three samples for each contamination study.

Microbiological analysis
Before inoculation anatomical cuts were controlled for the absence of *Listeria* spp. following ISO 11290-1:1996/Amend 1:2004 (ISO, 2004) protocol intended for *L. monocytogenes* detection, excepting that suspected colonies were confirmed by biochemical miniaturized tests (API Listeria kit; BioMérieux, France). For *L. innocua* enumeration, samples were diluted 1/10 in Buffered Peptone Water (homemade) and homogenized in stomacher for 60 s. Ten-fold serial dilutions were plated onto ALOA agar (Biolife, Milan, Italy) and incubated at 37°C for 48 h. Suspected colonies were confirmed by biochemical miniaturized tests (API Listeria kit; BioMérieux, France). In samples below the quantification limit (10 CFU/g), the qualitative analysis was carried out as described before. Results of *L. innocua* counts were expressed in CFU/g and converted into Log10 CFU/g.

Data analysis
For comparison between control and treated samples, if the pathogen resulted detectable but not quantifiable in enumeration analysis (under the limit of quantification: LOQ=10 CFU/g), it was assigned the value of 9 CFU/g (corresponding to log<sub>10</sub> 9 = 0.95 log CFU/g) (EFSA, 2010).

To compare the level of the pathogen observed during processing steps and post HPP treatment two way ANOVA test, was used; level 1 was Company productive process (A, B, C, D) and level 2 consisted in productive phases; (T0) inoculum, (T1) post resting, (Tend) post ripening, and (THPP) post HPP treatment. When statistically significant differences were detected, one-way ANOVA and post hoc pairwise comparison across levels were performed by using Tukey’s test. Surface and deep contaminations were compared separately.

The statistical analyses were carried out using the computer software program STATA 7.0 (STATA Corporation, College Station, TX, USA). Significance was established at P<0.05.

Table 2. Results of chemico-physical analysis differentiated for manufacturing company carried out in superficial (Sup) and deep (Deep) samples: it is reported the mean value of the obtained measurements followed by the standard deviation into brackets.

|               | Company A | Company B | Company C | Company D |
|---------------|-----------|-----------|-----------|-----------|
| pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> | pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> | pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> |
| T0            | 5.95 (0.07) | 6.01 (0.12) | 5.97 (0.03) | 5.99 (0.02) | 5.59 (0.12) | 5.60 (0.01) | 5.57 (0.01) | 5.55 (0.12) | 5.60 (0.01) | 5.57 (0.01) | 0.95 (0.01) |
| T1            | 6.04 (0.10) | 0.979 (0.005) | 6.05 (0.13) | 0.981 (0.004) | 5.92 (0.12) | 0.977 (0.003) | 5.84 (0.13) | 0.980 (0.003) | 5.62 (0.18) | 0.977 (0.002) | 5.58 (0.05) | 0.972 (0.001) |
| Tend          | 5.61 (0.18) | 0.970 (0.008) | 5.61 (0.12) | 0.965 (0.002) | 5.61 (0.12) | 0.977 (0.003) | 5.61 (0.12) | 0.965 (0.002) | 5.58 (0.18) | 0.977 (0.003) | 5.58 (0.05) | 0.972 (0.001) |

Table 3. Mean value log CFU/g (standard deviation) of *L. innocua*. (L) enumeration analyses carried out in superficial (Sup) and deep (Deep) Samples.

|               | Company A | Company B | Company C | Company D |
|---------------|-----------|-----------|-----------|-----------|
| pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> | pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> | pH<sub>sup</sub> | a<sub>sup</sub> | pH<sub>dee</sub> | a<sub>dee</sub> |
| T0            | 7.11 (0.19) | 7.34 (0.24) | N.D. | N.D. | 7.54 (0.20) | N.D. | 7.60 (0.12) |
| T1            | 6.65 (0.27) | 5.30 (0.56) | 5.95 (0.23) | 4.23 (0.29) | 6.02 (0.39) | 4.51 (0.20) | 5.78 (0.20) | 2.31 (0.58) |
| Tend          | 3.85 (0.51) | 4.37 (0.80) | 4.34 (0.51) | 4.20 (0.58) | 4.65 (0.56) | 3.94 (0.25) | 7.62 (0.24) | 4.54 (1.52) |
| THPP          | 3.38 (0.45) | 1.80 (0.91) | 3.60 (0.30) | 1.71 (0.26) | 3.24 (0.41) | 2.31 (0.51) | 6.68 (1.37) | 1.00 (0.10) |

N.D.: Not Determined; * in 2 out of 5 replicates value 0.35 was assumed since pathogens were detected but not countable; ** in 4 out of 5 replicates value 0.50 was assumed since pathogens were detected but not countable. In each column, different capital letters mean significant differences between *L. innocua* contamination evaluated at each sampling step. Significant (x,y) or not significant (x,x) differences between surface and deep contamination, separately evaluated for each step and each company.
Throughout the early phases of the four production processes, the progressive contamination of deeper parts of the anatomical cuts by inoculated bacteria took place, seemingly facilitated by the use of the meat tumbling machine in concomitance with salting. The first examinations were made at the end of the resting phase, obtaining values for deep contamination comprised between 2.31 and 5.30 log CFU/g (Table 3).

The overall superficial reduction resulted of 3.73 to 4.30 log CFU/g for companies A, B, C and 0.92 log CFU/g for company D. Table 4 reports details of L. innocua count reduction showing how the reduction of the microbial load on the surface of the coppa during the production process was not different among companies A, B and C (P>0.05) ranging from 2.89 to 3.26 log CFU/g, while it was significantly different (P<0.01) (-0.02 log CFU/g) for company D (Table 4). Similarly, the microbial reduction occurred in depth during drying and ripening process, resulting between 0.03 and 0.93 log CFU/g (P>0.05) for companies A, B, C while an increase of L. innocua count (2.23 log CFU/g) was shown for company D (P<0.01).

HPP treatment resulted in a significant (P<0.01) deep decrease of L. innocua with values ranging between 1.63-3.54 log CFU/g (Table 4) with no significant differences between companies. Regarding superficial contamination HPP treatment resulted significant (P<0.01) only in companies B and C.

### Discussion and Conclusions

This study reports data of 4 challenge tests performed on Italian dry-cured coppa produced by following four different companies’ procedures, and subsequently treated with HPP in order to verify the ability of the overall procedure to obtain a lethal process for Listeria.

L. innocua reduction during curing did not show any significant difference between companies A, B, C (P>0.05) with values ranging from 2.89 to 3.26 log CFU/g (Table 4); similar results have been shown by other authors in several dry-cured meat products. In particular a reduction of 4.0 log CFU/g in dry-cured ham after 69 days of curing was observed by Reynolds et al. (2001), and a 2.5 log CFU/g decrease was reported in dry-cured Serrano ham after 60 days of ripening (Montiel et al., 2020), while Barbuti et al. (2009) reported after 108 days of ripening a decrease of L. monocytogenes of 4.5 CFU/g in Italian Parma ham. Differences with the values obtained in this study can be addressed mainly to different products characteristics and different production processes.

In the early phase of the production process, the decrease of L. innocua in companies A, B and C could primarily be related to the salting treatment to which the anatomical cuts were exposed as it was observed a higher reduction in the two processes that included double salting, while in the subsequent phases the loss of microbial vitality could mainly be related to the progressive reduction of the a.s values correlated with the weight loss of the product.

Differently, values obtained from company D, in terms of L. innocua reduction after resting (-1.84 log CFU/g) and at the end of the process (-0.02 log CFU/g), were significantly lower than the others, (see Table 4), probably because of shorter resting, drying and ripening phases. Several authors reported how a longer maturation period leads to higher reduction of pathogenic bacteria: it was demonstrated that a short ripening period in fermented sausages was associated with greater survival of L. monocytogenes (Gonzales-Barron et al., 2015; Nightingale et al., 2006) and that the length of the drying period may be particularly important in the control of L. monocytogenes. Reynolds et al. (2001) found that in dry-cured ham L. monocytogenes populations continued to decline to undetectable levels after the hams completed the dry-aging process highlighting the importance of ageing in the control of L. monocytogenes.

In the present study, after HPP treatment at 593 MPa for 290 sec., a reduction of 0.47-1.41 log CFU/g and 1.63-3.54 log CFU/g on surface and in depth respectively; there are no data in literature on HPP of coppa to compare our results. In studies regarding HPP treatment of fermented dry meat products at time/pressure parameters similar to those applied in this study, a 1.6 – 5.0 Log CFU/g reduction at 600MPa for 5 min in Genoa salami (Porto-Fett et al., 2010), a 1.79-3.15 Log CFU/g reduction in Spanish chorizo at 600 MPa for 5-10 minutes (Rubio et al., 2018) and a 0.9 Log CFU/g reduction in slightly fermented sausages at 400MPa for 10 minutes (Garriga et al., 2005) were reported.

Various authors have tested the inactivation of L. monocytogenes in other unfermented meat products by HPP (Garriga et al., 2004; Hugas et al., 2002; Merialdi et al., 2015), many of them in dry-cured ham (Morales et al., 2006; Bover-Cid et al., 2011; Hereu et al., 2012; Bover-Cid et al., 2015; Hereu et al. 2012) have reported a reduction of L. monocytogenes from 1.82 to 3.85 log CFU/g in artificially contaminated sliced ham after treatment at 600 MPa for 5 min, Bover-Cid et al. (2011), through a HPP treatment at 613 MPa for 5 min., achieved the 2.39D proposed by Hoz et al. (2008) to meet the USA zero tolerance policy, Bover-Cid et al. (2015) showed a reduction of L. monocytogenes in dry-cured ham at 600MPa for 5 minutes ranging from 2.24 log to 6.82 Log CFU/g depending on the a.s. and fat content of the ham.

In our study a general lower effect on L. innocua count reduction was observed probably due to variables as a result of intrinsic characteristics of the product (pH, a.s. fat, protein and solute content) that influence the efficacy of HPP treatment on Listeria spp.; also the different bacterial baroresistance of different strains used in different studies should be taken into account (Possas et al., 2017).

### Table 4. Logarithmic unit reductions of L. innocua (L) in superficial and deep samples after each sampling step.

| Sampling Step          | Company A | Company B | Company C | Company D |
|------------------------|-----------|-----------|-----------|-----------|
|  | L<sub>Sup</sub> | L<sub>Deep</sub> | L<sub>Sup</sub> | L<sub>Deep</sub> | L<sub>Sup</sub> | L<sub>Deep</sub> | L<sub>Sup</sub> | L<sub>Deep</sub> |
| Resting - Δ(T0-T1)  | 0.46      | -         | 1.39      | -         | 1.52      | -         | 1.82      | -         |
| Drying and Ripening - Δ(T1-Tend) | 2.80 | 0.93 | 1.61 | 0.03 | 1.37 | 0.57 | -1.84 | -2.23 |
| Production process - Δ(T0-Tend) | 3.26 | - | 3.00 | - | 2.89 | - | -0.02 | - |
| HPP - Δ(Tend-THPP) | 0.47 | 2.57 | 0.74 | 2.52 | 1.41 | 1.63 | 0.94 | 3.54 |
| TOTAL - Δ(T0-THPP) | 3.73 | - | 3.74 | - | 4.30 | - | 0.92 | - |

- values were not calculated since T0 was not determined.
According to the data reported in this paper, to achieve the USDA/FSIS requirement, for sanitation, resulted to be a decisive factor in the treatment used as a post-production process.

For products intended for exportation to countries with zero tolerance policy for Listeria, in particular the United States, HPP treatment used as a post-production process for sanitation, resulted to be a decisive factor in reaching the superfluous Listeria inactivation requirements needed for exportation of dry-cured meat in the U.S. Processors should assess the ability of their process to control incoming pathogens, to predict the L. innocua load at the end of the process, and to evaluate the need for a process modification and/or for the addition of a final lethal process.

References

Barbuti S, Grisenti M, Frustoli M, Parolari G, 2009. Validation of the manufacturing process of Italian dry-cured ham (prosciutto) for the inactivation of Listeria monocytogenes and Salmonella spp. Int Congr Meat Sci Technol PE6.10.

Black EP, Huppertz T, Fitzgerald GF, Kelly AL, 2007a. Barprotection of vegetative bacteria by milk constituents: A study of Listeria innocua. Int Dairy J 17:104–10. doi:10.1016/j.idairyj.2006.01.009.

Black EP, Setlow P, Hocking AD, Stewart CM, Kelly AL, Hoover DG, 2007b. Response of spores to high-pressure processing. Compr Rev Food Sci Food Saf 6:103–119.

Bonilauri P, Liuzzo G, Merialdi G, Bentley S, Poeta A, Granelli F, Dottori M, 2004. Growth of Listeria monocytogenes on vacuum-packaged horsemeat for human consumption. Meat Sci 68:671-4.

Bover-Cid S, Belletti N, Aymerich T, Garriga M, 2015. Modeling the protective effect of a w and fat content on the high pressure resistance of Listeria monocytogenes in dry-cured ham. Food Res Int 75:194–9.

Bover-Cid S, Belletti N, Garriga M, Aymerich T, 2011. Model for Listeria monocytogenes inactivation on dry-cured ham by high hydrostatic pressure processing. Food Microbiol 28:804–9.

Busconi M, Zaccioni C, Scolari G, 2014. Bacterial ecology of PDO Coppa and Pancetta Piacentina at the end of ripening and after MAP storage of sliced product. Int J Food Microbiol 172:13–20.

EFSA, 2010. Management of left-censored data in dietary exposure assessment of chemical substances EFSA J 8:1557.

EFSA and ECDC, 2019. The European Union One Health 2018 Zoonoses Report. EFSA J. 17:5926, 276.

European Commission, 2005. Commission Regulation (EC) No 2073/2005 of 15 November 2005 on microbiological criteria for foodstuffs. OJ EU:1–26.

Food Safety and Inspection Service/ United States Department of Agricultural (USDA/FSIS), 2017. FSIS Salmonella Compliance Guidelines for Small and Very Small Meat and Poultry Establishments that Produce Ready-to-Eat (RTE) Products and Revised Appendix A.

Food Safety Inspection Service (FSIS), 2015. Control of Listeria monocytogenes in ready-to-eat meat and poultry products. Fed Regist 80:35178–35188.

Garriga M, Grébol N, Aymerich MT, Monfort JM, Hugas M, 2004. Microbial inactivation after high-pressure processing at 600 MPa in commercial meat products over its shelf life. Innov Food Sci Emerg Technol 5:451–7.

Garriga M, Marcos B, Martin B, Veciana-Nogués MT, Bover-Cid S, Hugas M, Aymerich T, 2005. Starter cultures and high-pressure processing to improve the hygiene and safety of slightly fermented sausages. J Food Prot 68:2341–8.

Gonzales-Barron U, Cadavez V, Pereira AP, Gomes A, Araújo JP, Saavedra MJ, Estevínho L, Butler F, Pires P, Dias T, 2015. Relating physicochemical and microbiological safety indicators during processing of linguica, a Portuguese traditional dry-fermented sausage. Food Res Int 78:50–61.

Gray JA, Chandy PS, Kaur M, Kocharanichtt C, Bowman JP, Fox EM, 2018. Novel biocontrol methods for Listeria monocytogenes biofilms in food production facilities. Front Microbiol 9:605.

Hayman MM, Kouassi GK, Ananthewaran RC, Flores JD, Knabel SJ, 2008. Effect of water activity on inactivation of Listeria monocytogenes and lactate dehydrogenase during high pressure processing. Int J Food Microbiol 124:21–6.

Hereu A, Bover-Cid S, Garriga M, Aymerich T, 2012. High hydrostatic pressure and biopreservation of dry-cured ham to meet the Food Safety Objectives for Listeria monocytogenes. Int J Food Microbiol 154:107–12.

Hoz L, Cambero MI, Cabeza MC, Herrero AM, Ordóñez JA, Ordóñez O, Ordóñez O, 2008. Elimination of Listeria monocytogenes from Vacuum-Packaged Dry-Cured Ham by E-Beam Radiation. doi: 10.4315/0362-028X-71.10.2001.

Hu M, Gurtler JB, 2017. Selection of surrogate bacteria for use in food safety challenge studies: A review. J Food Prot 80:1506–36.

Hugas M, Garriga M, Monfort JM, 2002. New mild technologies in meat processing: High pressure as a model technology. Meat Sci 62:359–71.

ISO, 2004. Microbiology of food and animal feeding stuffs — Horizontal method for the detection and enumeration of Listeria monocytogenes. — Part 1: Detection method AMENDMENT 1: Modification of the isolation media and the haemolysis test, and inclusion of precision data. ISO 11290-1:1996/AMD 1:2004.

ISO, 2004. Microbiology of food and animal feeding stuffs. Determination of water activity ISO 21807:2004.

Jordan K, McAuliffe O, 2018. Listeria monocytogenes in Foods. Adv Food Nutr Res 86:181–213.

Meloni D, Galluzzo P, Mureddu A, Piras F, Griffiths M, Mazzette R, 2009. Listeria monocytogenes in RTE foods marketed in Italy: Prevalence and automated EcoRI ribotyping of the isolates. Int J Food Microbiol 129:166–73.

Merialdi G, Ramini M, Ravanetti E, Gherri G, Bonilauri P, 2015. Reduction of listeria innocua contamination in vacuum-pack-aged dry-cured Italian pork products after high hydrostatic pressure treatment. Ital J Food Saf 4:101–3.

Montiel R, Peirotén Á, Ortiz S, Bravo D, Gaya P, Martinez-Suárez J V, Tapiañor J, Nuñez M, Medina M, 2020. Inactivation of Listeria monocytogenes during dry-cured ham processing. Int J Food Microbiol 318:108469.

Morales P, Calzada J, Nuñez M, 2006. Effect of high-pressure treatment on the survival of Listeria monocytogenes Scott A in sliced vacuum-packaged Iberian and Serrano cured hams. J Food Prot 69:2539–43.
Morales-Partera ÁM, Cardoso-Toset F, Jurado-Martos F, Astorga RJ, Huerta B, Luque I, Tarradas C, Gómez-Laguna J, 2017. Survival of selected foodborne pathogens on dry-cured pork loins. Int J Food Microbiol 258:68-72.

Nightingale KK, Thippareddi H, Phebus RK, Marsden JL, Nutsch AL, 2006. Validation of a traditional Italian-style salami manufacturing process for control of Salmonella and Listeria monocytogenes. J Food Prot 69:794–800.

Patterson MF, 2005. Microbiology of pressure-treated foods. J Appl Microbiol 98:1400–9.

Porto-Fett ACS, Call JE, Shoyer BE, Hill DE, Pshebniski C, Cocoma GJ, Luchansky JB, 2010. Evaluation of fermentation, drying, and/or high pressure processing on viability of Listeria monocytogenes, Escherichia coli O157:H7, Salmonella spp., and Trichinella spiralis in raw pork and Genoa salami. Int J Food Microbiol 140:61–75.

Possas A, Pérez-Rodríguez F, Valero A, García-Gimeno RM, 2017. Modelling the inactivation of Listeria monocytogenes by high hydrostatic pressure processing in foods: A review. Trends Food Sci Technol 70:45–55.

Rastogi NK, Raghavarao KSMS, Balasubramaniam VM, Niranjan K, Knorr D, 2007. Opportunities and Challenges in High Pressure Processing of Foods. Crit Rev Food Sci Nutr 47:69–112.

Reynolds AE, Harrison MA, Rose-Morrow R, Lyon CE, 2001. Validation of Dry-cured Ham Process for Control of Pathogens. J Food Sci 66:1373–9.

Rubio B, Possas A, Rincón F, García-Gimeno RM, Martínez B, 2018. Model for Listeria monocytogenes inactivation by high hydrostatic pressure processing in Spanish chorizo sausage. Food Microbiol. 69:18–24.

Sandora S, Stanford MA, Goddik LM, 2004. The Use of High-pressure Processing in the Production of Queso Fresco Cheese. J Food Sci. 69:FEP153–FEP158.

Tack DM, Marder EP, Griffin PM, Cieslak PR, Dunn J, Hurd S, Scallan E, Lathrop S, Muse A, Ryan P, et al., 2019. Preliminary incidence and trends of infections with pathogens transmitted commonly through food — foodborne diseases active surveillance network, 10 U.S. sites, 2015-2018. Morb Mortal Wkly Rep. 68:369–373.

Tao Y, Hogan E, Kelly AL, 2014. Chapter 1 – High-Pressure Processing of Foods: An Overview. In: Emerg Technol Food Process. Elsevier; p. 3–24.

Van Hekken DL, Tunick MH, Farkye NY, Tomasula PM, 2013. Effect of hydrostatic high-pressure processing on the chemical, functional, and rheological properties of starter-free Queso Fresco1. J Dairy Sci 96:6147–60.

Zanardi E, Novelli E, Ghiretti GP, Chizzolini R, 2000. Oxidative stability of lipids and cholesterol in salame Milano, coppa and Parma ham: Dietary supplementation with vitamin E and oleic acid. Meat Sci 55:169–175.

Zhang H, Mittal GS, 2008. Effects of High-pressure Processing (HPP) on bacterial spores: An overview. Food Rev Int 24:330–51.