ABSTRACT

This study evaluated the growth of naturally occurring _L. monocytogenes_ in sliced, vacuum-packed mortadella samples during storage at 5°C until the expiration date. Tukey’s test indicated that counts of _L. monocytogenes_ on 0, 10, 20, 30 and 40 days of storage were significantly different (p<0.05), indicating growth during shelf life. In three trials, the mean increase was 1.72 log cycles. Vacuum packing and storage under refrigeration were not effective in controlling the growth of _L. monocytogenes_ in sliced mortadella, indicating that good manufacturing practices and implemented HACCP programs are essential to assure safety of this product.

Key-words: *Listeria monocytogenes*; growth; sliced mortadella; vacuum-packing
transferred to plastic bags containing 450 mL of Listeria Enrichment Broth, UVM Formulation (LEB) (Oxoid, England). After homogenization, the mixture was submitted to four decimal dilutions (equivalent to 10g, 1g, 0.1g and 0.01g of product) in LEB, in triplicates, and incubated at 30°C for 24h. For identification of *L. monocytogenes*, the methodology of Farber et al. (6) was carried out. The experiments were run three times (trials A, B and C).

Results were submitted to variance analysis and Tukey’s test, using Statistic for Windows, release 5.0 (Stat Soft Inc, 1984-1995).

Table 1 shows the mean populations of *L. monocytogenes* (MPNg⁻¹) in the sliced, vacuum-packed mortadella samples, in the three trials, and the mean counts for each of the five days of analysis (days 0, 10, 20, 30 and 40). Final populations at the end of the shelf life were 89.1, 23.4 and 102.3 MPN.g⁻¹ for trials A, B and C, respectively. Variance analysis of the results indicated that differences in these populations were not significant (p>0.05). Tukey’s test, applied to the general means, showed that differences in populations on day 10 compared to day 0, on day 20 compared to day 10, on day 20 compared to day 30 and on day 40 compared to day 30 were significant (p < 0.05), indicating multiplication of *L. monocytogenes* during the shelf life of the product stored under refrigeration (5ºC).

Similar populations of *L. monocytogenes* at the end of shelf life were reported elsewhere. Examining sliced vacuum-packed frankfurters the authors found counts around 100 – 200 CFU.g⁻¹ of the product (16,17).

It is interesting to note that in two out of three trials the population of *L. monocytogenes* on the 30th day of storage was lower than that observed on the 20th day. This decrease in *L. monocytogenes* population may be explained by the presence of autochtonous lactic acid bacteria (LAB), well-known for their capability of inhibiting growth of *L. monocytogenes* during food storage (4). Contamination with LAB can occur during slicing and packaging procedures, mainly when hygiene conditions are precarious.

Table 2 shows that *L. monocytogenes* population increase (mean MPN.g⁻¹) during storage at 5ºC for 40 days was equal to 76.4, 22.1 and 89.0 for trials A, B and C, respectively. General mean increase of *L. monocytogenes* was 62.5 MPN. g⁻¹. According to several reports, *L. monocytogenes* population can increase up to 6 logs in vacuum-packed meat products stored under refrigeration (3,7,8,11,17). However these studies were conducted with spiked products and the conditions required for the growth of microorganisms are not necessarily the same as for naturally contaminated foodstuffs. In artificially contaminated products, the microbial cells are in optimal physiological conditions, favoring the competition with the normal microbiota of the product.

Refrigeration between 4 and 8ºC of sliced, vacuum-packed meat products does not seem to be an obstacle to *L. monocytogenes* growth, as observed in the present study and also by Rosso et al. (15). Temperatures below 2ºC are recommended for the control of *L. monocytogenes* in these products; however, these temperatures are not commonly achieved at retail level. It should be also taken into account that *L. monocytogenes* tolerates low O₂ tension, being able to survive and grow in anaerobic conditions, even at low temperatures (13), as encountered in vacuum-packed mortadella stored under refrigeration.

The low initial *L. monocytogenes* population in the products found in this study has also been observed by other authors (5,16,17). However, these low initial numbers are not an assurance of low risk for the product since the pathogen may grow and reach risky levels (≥10⁰g⁻¹) (13).

The method employed for enumeration of *L. monocytogenes* in a food product has strong influence in the results (2). Once

### Table 1. Mean populations of *L. monocytogenes* in sliced, vacuum-packed mortadella, during storage under refrigeration for 40 days.

| Day | Trial A (MPN.g⁻¹) | Trial B (MPN.g⁻¹) | Trial C (MPN.g⁻¹) | General mean population (MPN.g⁻¹) |
|-----|------------------|------------------|------------------|-------------------------------|
| 0   | 0.03             | 1.00             | 0.25             | 0.40***                       |
| 10  | 0.14             | 0.72             | 11.20            | 4.00a                         |
| 20  | 95.50            | 0.50             | 41.70            | 45.90b                        |
| 30  | 12.60            | 1.60             | 2.40             | 5.50a                         |
| 40  | 89.10            | 23.40            | 102.30           | 71.6b                         |

* mean of three determinations  
** mean of trials A, B and C  
*** same letters indicate values that are statistically similar (p>0.05)

### Table 2. Increment in population of *L. monocytogenes* in sliced, vacuum-packed mortadella during storage under refrigeration for 40 days

| Interval in days | Increment (MPN.g⁻¹) | General increment (MPN.g⁻¹) |
|------------------|---------------------|----------------------------|
| 0 – 10           | 0.10                | 10.95                      |
| 10 – 20          | 95.4                | 30.50                      |
| 20 – 30          | -82.9               | 99.9                       |
| 30 – 40          | 76.5                | 66.1                       |

Total increment 76.4 22.1 89.0 62.5 *mean of three determinations.
initial population of the pathogen can be as low as 0.03 MPN g⁻¹ (Table 1), one may not find L. monocytogenes in the product if the method lacks sensitivity.

In the present study L. monocytogenes was found in all sliced mortadella samples. This result differs from the one observed in a previous study (1), carried out with non-sliced mortadella, where the pathogen was detected in 26.7% of the samples. These findings indicate that slicing has an important role in the contamination process.

In view of the fact that L. monocytogenes can reach risky levels in vacuum-packed sliced mortadella stored under refrigeration, appropriate control measures are needed to minimize contamination of the product and/or inhibit the growth of the pathogen. Implementation of good hygiene practices (GHP), SSOPs and good manufacturing practices (GMP), together with HACCP is of uppermost importance to minimize environmental and product contamination. Reduction of shelf life, reformulation of the product by adding inhibitory ingredients, post-packing listericidal treatment (e.g. irradiation or ultra-high pressure) and active packaging are some options for the control growth of the pathogen in sliced meat products such as mortadella.

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