Growth probability of *Listeria monocytogenes* and classification of pork meat products

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Abstract

The performance of a growth probability model taking into account the pH, the water activity, and the storage temperature of food to predict the growth or the no growth of *Listeria monocytogenes* in pork meat products was assessed by comparing predictions with the behaviour observed in these products by challenge-testing. The predictive positive value of the model was 100% for cooked pork products characterized by high pH/water activity combinations and that always supported the growth of *L. monocytogenes*. The negative predictive value of the model was 100% for the dried, fermented or not, pork products characterized by low pH/water activity combinations and that never supported the growth of the pathogen. Raw brined pork products have intrinsic characteristics locating them near the growth/no growth boundary of *L. monocytogenes* and its growth was effectively sometimes observed in these products. In this case, taking into account the CO_2 present in the modified atmosphere and sodium nitrite concentration of the products increased the predictive values of the model and no false negative prediction was obtained.

Keywords

Growth/no growth modelling, *Listeria monocytogenes*, pork meat products

Introduction

Ready-to-eat pork meat products were implicated in listeriosis outbreaks several times during these last 15 years in France. It is then essential to have available tools allowing a rapid identification of products supporting or not the growth of *Listeria monocytogenes*. Recent growth/no growth models developed in predictive microbiology allow the classification of foods in connection with their sensitivity to the growth of pathogen micro-organisms. The aim of this study was to assess the use of a growth probability model to accurately predict the growth of *L. monocytogenes* in pork meat products knowing their storage temperature and their main intrinsic characteristics.

Materials and methods

Classification of pork meat products and growth / no growth data

96 pork meat products were included in this study and were classified into three categories depending on their processing. The first category groups together 43 cooked products such as cooked ham, cooked chicken, 'pâtés', 'rillettes', frankfurters. The second category groups together 18 dried, fermented or not, products such as slicing and dried sausages, salami, raw hams. The third category groups together 35 raw brined products such as bacon, bacon cubes, raw sausages. The products were characterized by their pH, water activity (aw) and, when known, by their concentrations of some preservatives (CO_2 in the modified atmosphere and added sodium nitrite).

L. monocytogenes growth/no growth observations were obtained in the 96 pork meat products by challenge testing. The products were artificially contaminated with relatively high initial concentrations (between 10^2 and 10^3 cfu per g of product) of single or pooled strains and were stored at 8°C or the first third of their shelf-life at 4°C and, then, the two thirds remaining at 8°C. The storage durations used were consistent with the shelf-lives of the products and were typically comprised between 30 and 90 days. *L. monocytogenes* was periodically enumerated during the storage and the growth was considered significant if an increase of more than 0.5 $\log_{10}$ cfu/g of the microbial population was observed for several counts.

Growth probability model

A growth probability model based on a logistic function and including the effects of environmental factors and their interactions (Augustin et al., 2005) was used to predict the growth of *L. monocytogenes*:

$$P = \frac{1}{1+\exp(-10\cdot\theta)} \quad \text{and} \quad \theta = 1 - \sum_i \left(\frac{X_{i,opt} - X_i}{X_{i,opt} - X_{i,\min}} \right)^3 - \sum_j \frac{c_j}{MIC_j} \qquad (1)$$

where P is the growth probability, X_i is the temperature, the pH or the aw of the product, $X_{i,opt}$ is the optimal temperature, pH or aw for the growth of *L. monocytogenes* (37°C, 7.1, and 0.997, respectively), $X_{i,min}$ is the minimal temperature, pH or aw for the growth of *L. monocytogenes* (-1.72°C, 4.71, and 0.913, respectively), c_j is the proportion of CO_2 in the modified atmosphere or the nitrite concentration of the product, MIC_j is the minimal inhibitory concentration for CO_2 or sodium nitrite for the growth of *L. monocytogenes* (3.04 and 25 μmol l^{-1}, respectively).

Growth/no growth data observed in pork meat products were compared with predictions obtained with this model by using a storage temperature of 8°C in Eq. (1). Growth was predicted when P was above 0.9, no growth was predict when P was below 0.1, and no prediction was performed when P lied between these two limits. The performance of the model to predict the growth of *L. monocytogenes* in pork meat products was assessed by calculating its positive and negative predictive values for each category of products.

Results and discussion

The pH/aw combinations of cooked products are presented in Fig. 1. The pH and aw ranges for these products were [5.80-6.55] and [0.956-0.988], respectively. The pH/aw combinations of these products are deeply located in the predicted growth area of *L. monocytogenes* (Fig. 1) and a significant growth of the pathogen was effectively observed by challenge testing for all these products. By taking into account temperature, pH, and aw (and possibly CO_2 and nitrite), the growth probabilities were between 0.97 and 1. The positive predictive value (PPV) of the model was thus equal to 100% for this category (Table 1).

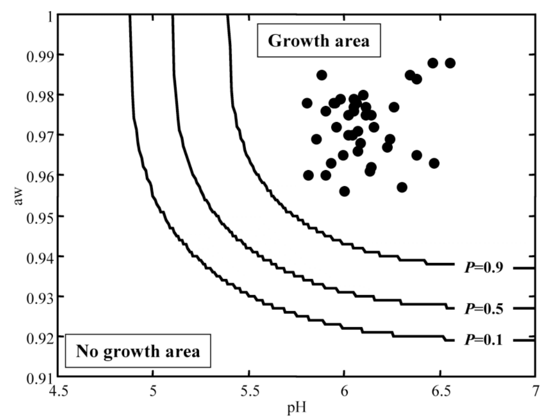

Fig. 1. pH/aw characteristics of cooked pork products and predicted growth/no growth areas for *L. monocytogenes* at 8°C with Eq. (1). (●) Growth observed, (■) No growth observed.

The pH/aw combinations of dried products are presented in Fig. 2. The pH and aw ranges for these products were [4.63-6.01] and [0.831-0.943], respectively. No growth of *L. monocytogenes* was observed for the 18 products that were effectively located in the predicted no growth area (Fig. 2). By taking into account temperature, pH, and aw, the growth probabilities lied between 0 and 0.01. The negative predictive value of the model was thus equal to 100% for this category. By using the criteria proposed by the European Commission Regulation No 2073/2005 on microbiological criteria for foodstuffs (Anon., 2005), *i.e.*, pH ≤ 4.4 or aw ≤ 0.92, or pH ≤ 5.0 and aw ≤ 0.94, no growth was predicted in only 15 of the 18 products (Table 1).

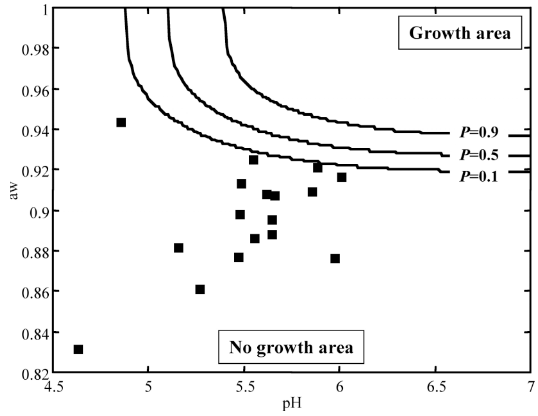

Fig. 2. pH/aw characteristics of dried pork products and predicted growth/no growth areas for *L. monocytogenes* at 8°C with Eq. (1). (●) Growth observed, (■) No growth observed.

The pH/aw combinations of raw brined products are presented in Fig. 3. The pH and aw ranges for these products were [5.18-5.95] and [0.939-0.975], respectively. These products displayed physico-chemical characteristics locating them near the predicted growth/no growth boundary (Fig. 3) and growth of *L. monocytogenes* was effectively only observed for 9 products out of 35. By taking into account temperature, pH, and aw, the growth probabilities were between 0.46 and 0.99. The PPV of the model was only equal to 32%. By taking into account CO_2 proportion and sodium nitrite concentration, the NPV of the model was 100% and no growth was accurately predicted for 10 products out of 18 (Table 1).

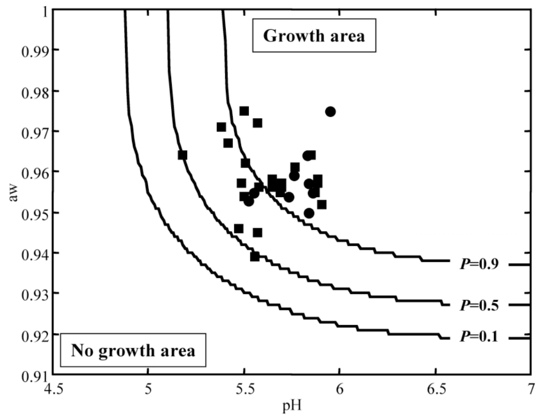

Fig. 3. pH/aw characteristics of raw brined pork products and predicted growth/no growth areas for *L. monocytogenes* at 8°C with Eq. (1). (●) Growth observed, (■) No growth observed.

Table 1. Growth/no growth observed and predicted for *L. monocytogenes* in pork meat products.

Pork meat products	Model	Predictions	Observations	
			Growth	No growth
Cooked	Eq. (1) with T, pH, aw	Growth	43	0
		No growth	0	0
		No prediction	0	0
Dried	Eq. (1) with T, pH, aw	Growth	0	0
		No growth	0	18
		No prediction	0	0
	Regulation (EC) No 2073/2005	No growth	0	15
		No prediction	0	3
Raw brined	Eq. (1) with T, pH, aw	Growth	7	15
		No growth	0	0
		No prediction	2	11
	Eq. (1) with T, pH, aw, CO_2, nitrite	Growth	0	0
		No growth	0	10
		No prediction	3	5

Conclusion

Growth/no growth models are very useful for food business operators and for official authorities to quickly assess the susceptibility of foodstuffs to microbial proliferation. This is especially the case for ready-to-eat foods regarding the contamination by *L. monocytogenes* for which the microbiological limits at the end of the production process specified in the regulation (EC) No 2073/2005 depend on their ability to support the growth of the pathogen. We observed that the growth probability model assessed in this study could be safely (no false no growth predicted) used for pork meat products, knowing their pH, aw, and storage temperature, to quickly classify them. Cooked products supported the growth of *L. monocytogenes*, dried products did not support the growth of the micro-organism, and raw brined products characteristics located them near the growth/no growth boundary of *L. monocytogenes*. In this last case, a thorough knowledge of the physico-chemical characteristics of the foods can improve the predictions and often challenge-testing is necessary to precisely assess the behaviour of the micro-organism. The model presented on this example is usable for other foodborne pathogens and is included in the Sym'Previus software (www.symprevius.org).

References

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