Challenge-testing Belgian artisanal cheeses for *Listeria monocytogenes*

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### INTRODUCTION

Numerous listeriosis outbreaks have been associated with cheese consumption in developed countries. Various types of cheese have been incriminated, including fresh cheeses and mold-ripened soft cheeses. As a consequence, the absence of *Listeria monocytogenes* in 25g of cheese allowing its growth is imposed by Regulation (CE) No 2073/2005. Three situations not allowing the growth of the pathogen have been identified: (a) pH ≤ 4.4, (b) aw ≤ 0.92, and (c) pH ≤ 5.0 and aw ≤ 0.94. A pH or a aw above these threshold values does not necessary mean that *L. monocytogenes* is able to grow. Challenge-test is a powerful tool allowing producers to understand the real fate of the pathogen in their cheeses.

### OBJECTIVES

The main goal was to perform challenge-tests in order to assess the growth potential (δ) of *L. monocytogenes* in Belgian artisanal cheese, and to identify safe products. Physicochemical characteristics of these products were also studied.

### METHODOLOGY

**Day 0**
- Sampling of 12 cheeses for each batch (1 or 3 batches, depending on growth simulation).

**Day 0**
- Inoculation of a cocktail of three strains of *L. monocytogenes* in 6 samples/batch (remaining samples considered as standards).
- **L. monocytogenes** enumeration and physicochemical analyses in 3 standards and 3 inoculated samples. Other cheeses stored at 7°C for 2/3 of their shelf-life.

**2/3 shelf-life**
- Samples transferred to a fridge at 9°C for the last 1/3 of their shelf-life.

**End of shelf-life**
- **L. monocytogenes** enumeration and physicochemical analyses on the remaining samples. Calculation of δ.

### RESULTS, DISCUSSION & PERSPECTIVES

<table>
<thead>
<tr>
<th>Type of cheese</th>
<th>Number of challenge-tests</th>
<th>Number of positive δ</th>
<th>pH range</th>
<th>aw range</th>
<th>Dry matter range (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maquette</td>
<td>8</td>
<td>0/8</td>
<td>4.4 – 4.5</td>
<td>0.98 – 1.00</td>
<td>13.6 – 27.5</td>
</tr>
<tr>
<td>Moulded unripened</td>
<td>4</td>
<td>0/4</td>
<td>4.3 – 4.5</td>
<td>0.97 – 0.99</td>
<td>30.6 – 41.2</td>
</tr>
<tr>
<td>cheeses</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mold-ripened soft</td>
<td>4</td>
<td>4/4</td>
<td>5.3 – 7.1</td>
<td>0.96 – 0.98</td>
<td>44.5 – 56.9</td>
</tr>
<tr>
<td>cheeses</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smear-ripened soft</td>
<td>4</td>
<td>3/4</td>
<td>5.0 – 7.1</td>
<td>0.97 – 0.98</td>
<td>43.5 – 56.3</td>
</tr>
<tr>
<td>cheeses</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Semi-hard/hard cheeses</td>
<td>12</td>
<td>4/12</td>
<td>5.4 – 6.1</td>
<td>0.92 – 0.97</td>
<td>46.6 – 69.0</td>
</tr>
</tbody>
</table>

**Fig. 1 – Results of the challenge-tests**

**No growth in unripened cheese**

Although almost all unripened samples had pH > 4.4, it seems that *L. monocytogenes* is unable to grow in these cheeses. Federal Agency for the Safety of the Food Chain (FASFC) is currently evaluating the eventuality of a regulatory relaxation for acidic unripened cheeses.

**Contrasted results for semi-hard/hard cheeses**

Huge differences of δ were observed between batches of a given cheese, like a δ > 0 for batch 1, δ = 0 for batch 2 and δ < 0 for batch 3. This observation remains unexplained, given that no significant differences in pH, aw, dry matter or fat content were reported. However, all δ were < 1 log cfu/g. Further studies should be performed to understand this inter-batch variability.

**5 log cfu/g**

The final contamination level for some mold- and smear-ripened soft cheeses. Globally, this kind of product could represent a major threat for food safety. One exception was observed for Herve cheese. An hypothesis is that the endogenous microflora of this product was particular. This idea is currently investigated.

### ACKNOWLEDGEMENTS

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