Positive modulation of rabbit meat microbial ecology using feeding strategies

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Introduction: Highlights

World population: 10 billion people in 2050

Animal proteins

**Meat consumption in Canada, 1991 to 2014**

<table>
<thead>
<tr>
<th></th>
<th>1991</th>
<th>2014</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Red meat</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Beef and veal</td>
<td>25.5</td>
<td>20.1</td>
</tr>
<tr>
<td>Pig</td>
<td>19.7</td>
<td>15.7</td>
</tr>
<tr>
<td><strong>White meat</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chicken, hen and turkey</td>
<td>28.3</td>
<td>37.5</td>
</tr>
</tbody>
</table>

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Introduction

Rabbit meat consumption

<table>
<thead>
<tr>
<th></th>
<th>Italy: 5.2 kg capita per year</th>
<th>Spain: 2.7 kg capita per year</th>
<th>France: 2.0 kg capita per year</th>
<th>Canada: 0.025 kg capita per year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total fat (g)</td>
<td>5.55</td>
<td>15.97</td>
<td>14.95</td>
<td>10.43</td>
</tr>
<tr>
<td>Protein (g)</td>
<td>20.05</td>
<td>18.33</td>
<td>18.95</td>
<td>20.71</td>
</tr>
<tr>
<td>Calories (Kcal)</td>
<td>136</td>
<td>222</td>
<td>216</td>
<td>185</td>
</tr>
<tr>
<td>Omega 3 (g)</td>
<td>0.32</td>
<td>0.20</td>
<td>0.03</td>
<td>0.29</td>
</tr>
<tr>
<td>Phosphorus (mg)</td>
<td>213</td>
<td>132</td>
<td>200</td>
<td>176</td>
</tr>
<tr>
<td>Selenium (µg)</td>
<td>23.7</td>
<td>15.5</td>
<td>28.4</td>
<td>18.8</td>
</tr>
<tr>
<td>Vitamin B12 (µg)</td>
<td>7.16</td>
<td>0.32</td>
<td>0.66</td>
<td>2.16</td>
</tr>
<tr>
<td>Fer (mg)</td>
<td>1.57</td>
<td>0.95</td>
<td>0.86</td>
<td>1.97</td>
</tr>
</tbody>
</table>
Problematic

- Meat contamination: skin of the animal, gut content, workers’ hands and the slaughter environment (Saucier, 1999);

- *Listeria monocytogenes* is ubiquitous in nature and remains a safety concern (Wheatherill, 2008);

- High amount of PUFA in rabbit meat: more susceptible to oxidation,

- Challenges: foods, as natural as possible, without compromising quality and shelf life and safety.
Solutions

Plant extracts: (polyphenols)
Strawberry, onion, cranberry, etc.

Essential oils:
Eugenol, thymol, carvacrol, etc.

Lactic Acid Bacteria and bacteriocin
Nisin, Pediocin etc.
Hypothesis

Addition of plant extracts (Nutra Canada Inc.), essentials oils (XtractTM Instant, Pancosma) or a protective culture, to the feed modulate positively the microflora on rabbit meat.

Research Objectives

To develop new feeding strategies to improve the safety and preservation of rabbit meat without major investment for the producers.
Addition of plant extracts and essential oils in the feed


- Poster N° 10: Improvement of rabbit meat microbial quality by feed supplementation with natural sources of polyphenols (higher doses).

- Poster N° 9: Improvement of rabbit meat oxidative status using feeding strategies.
Microflora management
Did you know that?

- Micocin® is approved for use in Canada to reduce the risk of *Listeria* in ready-to-eat foods;
- Micocin® has been granted GRAS status;
- Micocin® is also approved in Mexico, Costa Rica and Colombia;
- Micocin® (Griffith Foods, Toronto, Canada) contains « *Carnobacteria maltaromaticum CB1* »;
- *Carnobacteria maltaromaticum CB1* produces three bacteriocins, namely carnocyclin A (CclA), piscicolin 126 and carnobacteriocin BM1.

(Health Canada, 2010)
Materials and methods: Animals and housing conditions

- 144 weaned Grimaud rabbits
- Commercial Control diet
- Micocin ® (8 log CFU/kg of feed)
- 72 rabbits/group
- 6 rabbits/cage as in commercial farms
- The cage was the experimental unit (EU)
- 12 cages per experimental group
- Different but similar rooms under strict biosecurity measures
- Slaughter on two different days (control first) after a total feed withdrawal of 15h.

CFU: colony forming unit
Materials and methods: after Slaughter

Cage = 6 rabbits

Whole carcass (1 rabbit) → Thighs → Deboning, grounding the rest of the meat, stored at -30°C

Quality measurement of meat: pH 1 and 24h, Drip loss, Colour, Cooking loss

Microbial analysis:
- Aerobic (0, 3, 6, 8 days)
- Anaerobic (0, 5, 10, 15, 20 days)
Materials and methods:
Microbial enumeration (log CFU/g)

• *Enterobacteriaceae* (Petrifilm™ 3M; 24 hours, 35-37°C)
• Presumptive *Pseudomonas* (CFC; 48 hours, 25°C)
• *E. coli* et coliforms (Petrifilm™3M; 24 hours, 35-37°C)
• Presumptive *S. aureus* (Petrifilm™3M; 24 hours, 35-37°C)
• Total aerobic mesophilic (PCA, 48 hours, 35°C)
• Presumptive lactic acid bacteria LAB (MRS, 48 hours, 25°C)
• **All Purpose Tween** (APT, 48 hours, 25°C)
• *Listeria* spp. (Palcam, 48 hours, 30°C)
Materials and methods: Experimental inoculation with *L. monocytogenes* strains

Ground rabbit meat

Uninoculated ground meat (control and Micocin® supplemented groups)

Aerobic and anaerobic conditions (0, 3, 6, 9, 12, 15 days) at 4 and 10 °C

Inoculated ground meat by using a cocktail of five *Listeria monocytogenes* strains (4 log CFU/g) in the control and Micocin® groups
Table 1. Presence of *C. maltaromaticum* producing CclA in rabbit ground meat stored at 4 and 10 °C under aerobic and anaerobic conditions (0, 3, 6, 9, 12, 15 days) as determined by PCR analysis of three specific genes:16S-cpg, ISR and CclA.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Aerobic</th>
<th>Anaerobic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td></td>
<td></td>
</tr>
<tr>
<td>16S-cpg</td>
<td>7</td>
<td>6</td>
</tr>
<tr>
<td>ISR</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>CclA</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Micocin®</td>
<td></td>
<td></td>
</tr>
<tr>
<td>16S-cpg</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>ISR</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>CclA</td>
<td>8</td>
<td>8</td>
</tr>
</tbody>
</table>

*a Number of positive gene identification out of 11 sample of ground meat for each storage conditions (n=11; one sample per temperature and storage time).

Results and discussion

- **Meat quality**: Adding Micocin® to the feed did not influence meat quality significantly.
Results and discussion

- **On thighs at 4 °C:**
  - **Aerobic**
    Presumptive *Pseudomonas* spp. is the prevailing microflora and log difference between the two groups is below 1 log.
  - **Anaerobic**
    Presumptive LAB is the prevailing microflora. *Listeria* spp., coliforms and *Enterobacteriaceae* counts were **0.93 to 1.19** log below for the Micocin® supplemented group.
Results and discussion

- **Uninoculated ground meat** at 4 and 10 °C

- *Aerobic conditions*: no effect of treatment

- *Anaerobic conditions*: no effect of treatment, but *Enterobacteriacea*, coliforms and presumptive *S. aureus* counts were $\geq 1 \log$ (1.01-1.86) unit below for the Micocin® supplemented group.

Competitive exclusion was most effective against presumptive *S. aureus* at 10 °C.
*L. monocytogenes* in ground meat with or without Micocin® in the feed

\[ P = 0.02 \]

\[ P > 0.05 \]
Conclusion

- By feeding a ration supplemented with *C. maltaromaticum CB1* (Micocin®) we were able to modulate its presence in the faeces, on the thighs and in ground meat.

- The presence of *C. maltaromaticum CB1* (Micocin®) in rabbits feed provided a better control of *L. monocytogenes* on ground meat.

- The search for other more powerful LAB must continue to better control undesirable organisms (*Listeria, coliforms, Enterobacteriaceae, S. aureus*) that will lead to safer food/meat.
Acknowledgements

« Let food be thy medicine »
Hyppocrate